UNIVERSIDADE FEDERAL DE MINAS GERAIS

INSTITUTO DE CIÊNCIAS BIOLÓGICAS

PROGRAMA DE PÓS-GRADUAÇÃO EM ECOLOGIA, CONSERVAÇÃO E

MANEJO DE VIDA SILVESTRE

ECOLOGIA, COMPORTAMENTO E MANEJO DE

EMAS (*Rhea americana*, RHEIDAE, AVES)

Cristiano Schetini de Azevedo

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Tese apresentada ao Programa de Pós-graduação em Ecologia, Conservação e Manejo de Vida Silvestre do Instituto de Ciências Biológicas da Universidade Federal de Minas Gerais como requisito parcial para a obtenção do título de doutor.
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APOIOS E PARCERIAS

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RESUMO

A ema é uma ave ameaçada de extinção em Minas Gerais, conseqüentemente, ações que visem sua conservação e manejo são essenciais para a conservação da espécie. Este estudo foi dividido em 6 capítulos, assim distribuídos: (A) Ecologia: 1 - Avaliação do comportamento de grupos de emas na natureza; 2 – Atuação das emas na dispersão de sementes de plantas do cerrado; (B) Comportamento e manejo: 1 – Avaliação do comportamento e reconhecimento de predadores por emas cativas; 2 – Avaliação do enriquecimento ambiental de forrageio na diminuição da exibição de comportamentos anormais em emas cativas; 3 – Avaliação da influência do público no comportamento de emas cativas; (C) Etnozoologia: 1 – Opinião das comunidades rurais e urbanas sobre a conservação das emas. O comportamento das emas, bem como o uso do habitat, se modifica quando o ambiente é alterado pelo homem. A destruição do cerrado para a implantação de monoculturas é fator importante na exclusão das emas da área, especialmente se as monoculturas forem de plantas arbóreas (florestas). Pastagens, ao contrário, são bastante utilizadas pelas emas, que usam o capim como fonte principal de alimentos. As emas se mostraram boas dispersoras de sementes de algumas espécies de plantas do cerrado, e o seu desaparecimento pode comprometer a dinâmica e o equilíbrio das áreas onde ocorre. O conhecimento da população humana acerca do papel das emas na conservação do cerrado varia entre pessoas que vivem no campo e na cidade, mas, em geral, as pessoas vêem relação entre a destruição do cerrado e o desaparecimento das emas. A capacidade de reconhecer predadores deve ser avaliada, e caso constatado o não-reconhecimento, sessões de treinamento anti-predação devem ser conduzidas (como observado neste estudo). O uso do enriquecimento ambiental deve ser reforçado; pois, além de estimular a exibição de comportamentos naturais, diminui a chance de que comportamentos anormais se desenvolvam. Neste estudo, o público visitante não pareceu estressar as emas, já que seu comportamento não foi diferente quando o público estava presente ou ausente.

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APRESENTAÇÃO

Muitas espécies animais encontram-se atualmente ameaçadas de extinção. Uma delas é a ema (*Rhea americana*, Rheidae, Aves), ave endêmica da América do Sul (Sick, 1997). Uma das principais causas do declínio populacional desta espécie é a destruição do Cerrado, bioma principal de sua ocorrência (Del Hoyo *et al.* 1992). Estratégias de conservação *in-situ*, como a realização de estudos sobre a biologia da espécie e suas relações com o ambiente em que vivem, bem como a criação de áreas de proteção natural no Cerrado, têm sido realizadas pelos órgãos ambientais, institutos de pesquisa, universidades e ONG's brasileiras, mas as populações de emas continuam declinando ano a ano (Davies, 2002). Estratégias de conservação *ex-situ* precisam aliar-se às estratégias *in-situ* de maneira a aumentar as chances de que esta espécie não desapareça. A criação de emas em cativeiro é antiga no Brasil e a maioria dos zoológicos brasileiros possui a espécie em seus plantéis. Entretanto, ações conservacionistas, como a reintrodução destas aves na natureza, só foram conduzidas até o momento pelo jardim zoológico da Fundação Zôo-Botânica de Belo Horizonte; muito pouco se considerarmos a quantidade de zoológicos, o número de indivíduos em cativeiro e o *status* de conservação da espécie.

Um resultado desanimador observado nas tentativas de reintrodução de emas na natureza foi o fato de todos os indivíduos reintroduzidos terem sido mortos por cachorros domésticos pouco tempo após a soltura (Guimarães Filho & Faggioli, 1997). Este acontecimento deixou claro que a simples manutenção de plantéis em cativeiro e sua soltura sem intervenções educativas nas comunidades podem significar perda de tempo e dinheiro, além de não acrescentar nada na conservação da espécie. Portanto, técnicas melhoradas de manutenção em cativeiro, estudos sobre a capacidade de reconhecimento e

exibição de comportamentos antipredatórios adequados, a manutenção de recintos que permitam às aves experimentarem baixos níveis de estresse e a avaliação da percepção do público visitante e de sua influência no comportamento e bem-estar das aves são ações importantes de conservação *ex-situ* que devem ser implementadas antes dos programas de reintrodução.

A conservação *ex-situ*, embora sabidamente importante, tem sido pouco realizada no Brasil. Os zoológicos ainda são vistos pelos pesquisadores apenas como vitrines da vida silvestre. Seu papel na conservação fica comprometido por adotarem medidas de manejo muitas vezes vistas com desconfiança pela comunidade acadêmica. Animais domesticados, estressados, com altas taxas de endogamia e apresentando comportamentos anormais são comuns nos zoológicos brasileiros. Estes animais, sem a devida intervenção, não serviriam para fins conservacionistas e os zoos, então, não funcionariam como centros de conservação.

Este estudo teve como principais objetivos avaliar aspectos do comportamento das emas, tanto na natureza quanto em cativeiro, e a percepção humana sobre a importância das emas para o Cerrado.

ESTRUTURA DA TESE

A estruturação da tese acompanhou o seguinte formato: referencial teórico, objetivos geral e específicos do estudo, capitulo I, capítulo II, capítulo III, capítulo IV, capítulo V, capítulo VI, referências bibliográficas gerais e anexos. Todos os capítulos foram escritos em formato de artigo científico, estando a formatação do texto e das citações bibliográficas adequadas às regras de cada revista escolhida para a submissão (as revistas

para as quais os artigos serão/foram submetidos estão informadas no início de cada capítulo). Desta forma, algumas informações apresentadas no referencial teórico foram repetidas nos capítulos, desde que necessário.

No referencial teórico buscou-se caracterizar de forma geral a biologia e comportamento das emas, o bioma Cerrado e aspectos da conservação *ex-situ* de espécies animais. No final do referencial teórico estão apresentados os objetivos geral e específicos do presente trabalho.

O capítulo I aborda aspectos do comportamento das emas na natureza. Resultados de dois anos de coleta de dados comportamentais em campo, na região de Felixlândia, noroeste de Minas Gerais, são apresentados. Avaliou-se o comportamento das emas em relação ao ambiente (área florestada e aberta); ao tamanho do grupo; ao horário do dia às estações de seca e chuva.

O capítulo II aborda o papel das emas na dispersão de sementes de plantas do Cerrado. Vários frutos de espécies vegetais do Cerrado foram oferecidos às emas cativas. Sementes que passaram pelo trato digestório das aves (tratamento) e que não passaram pelo trato digestório das aves (controle) foram, então, colocadas para germinar e as taxas de germinação e as velocidades de germinação comparadas entre os dois grupos (tratamento X controle).

O capítulo III aborda o grau de conhecimento das populações humanas de um local de ocorrência das emas e de um centro urbano. Questionários semi-estruturados foram aplicados às comunidades humanas de Felixlândia (local de ocorrência das emas) e aos visitantes do zoológico de Belo Horizonte (centro urbano, local sem ocorrência de emas na natureza). As perguntas mediram o grau de conhecimento sobre a biologia e conservação

das emas e importância dada à essas aves pelo público entrevistado, e as respostas foram comparadas.

O capítulo IV aborda a capacidade de reconhecimento de predadores e de resposta antipredação pelas emas cativas do zoológico de Belo Horizonte. Modelos taxidermizados de predadores e não-predadores foram exibidos às emas e suas respostas comportamentais medidas.

O capítulo V aborda o uso de enriquecimento ambiental na diminuição de comportamentos anormais exibidos por emas cativas. A grande exibição de comportamentos anormais e estereotipados, como "andar de um lado para outro" e "comer fezes" foram detectados durante a coleta de dados do capítulo IV. Altas taxas de expressão de comportamentos anormais são indicativos de baixo bem-estar e elevados índices de estresse. A eficiência de itens de enriquecimento ambiental na diminuição e extinção destes comportamentos foi avaliada.

O capítulo VI aborda a influência do público visitante do jardim zoológico da Fundação Zoo-Botânica de Belo Horizonte no comportamento das emas cativas. O grau de habituação das emas ao público do zoológico foi medido através da comparação dos comportamentos exibidos pelas aves em dias com visitação e sem visitação. O estresse causado pela visitação pode ser responsável pela exibição dos comportamentos anormais previamente observados.

Ressalta-se que os dois primeiros capítulos avaliam aspectos do comportamento das emas *in-situ*, bem como o seu papel na manutenção do Cerrado. Parâmetros iniciais para a implementação de atividades e de um programa de educação ambiental são abordados no capítulo III (etnozoologia). Os capítulos IV, V e VI abordam aspectos do comportamento e manutenção das emas em cativeiro, visando melhorias nas técnicas de manejo para um

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aumento das chances de sucesso em reintroduções. Os estudos conduzidos, então, abordam os três pilares da conservação: *in-situ*, *ex-situ* e educação ambiental (aqui abordada em um estudo etnozoológico inicial).

REFERENCIAL TEÓRICO

Biologia e comportamento da ema

A ema, *Rhea americana* LINNAEUS, 1758, é a maior ave da América do Sul (Schauensee, 1982). O macho e a fêmea podem atingir 35 e 32 kg e 170 e 134 cm de altura, respectivamente (Sick, 1997), existindo um leve dimorfismo sexual: mais robusto, o macho adulto tem a cabeça mais perfilada e o pescoço mais grosso, sendo negros a base do pescoço, o peito e a parte mediana do dorso anterior; o restante da plumagem tem coloração cinza pardacenta (Brandt & Neto, 1999) (Figura: 1).

Figura 1: Dimorfismo sexual entre macho (centro) e fêmeas (direita e esquerda) de emas (*Rhea americana*). O macho adulto tem o pescoço mais grosso, sendo negros a base do pescoço, o peito e a parte mediana do dorso anterior.

Distribui-se pelo Brasil, Argentina, Paraguai, Uruguai e sul da Bolívia (Giannoni, 1996). No Brasil, ocupa as regiões sul do Pará, nordeste (incluindo o Maranhão), centro-oeste, sudeste e sul (Sick, 1997) (Figura 2). Vive em regiões campestres e Cerrados, desde que exista oferta de água (Brandt & Neto, 1999). As maiores populações naturais encontram-se nos estados do Mato Grosso e Goiás (Gunski, 1992).
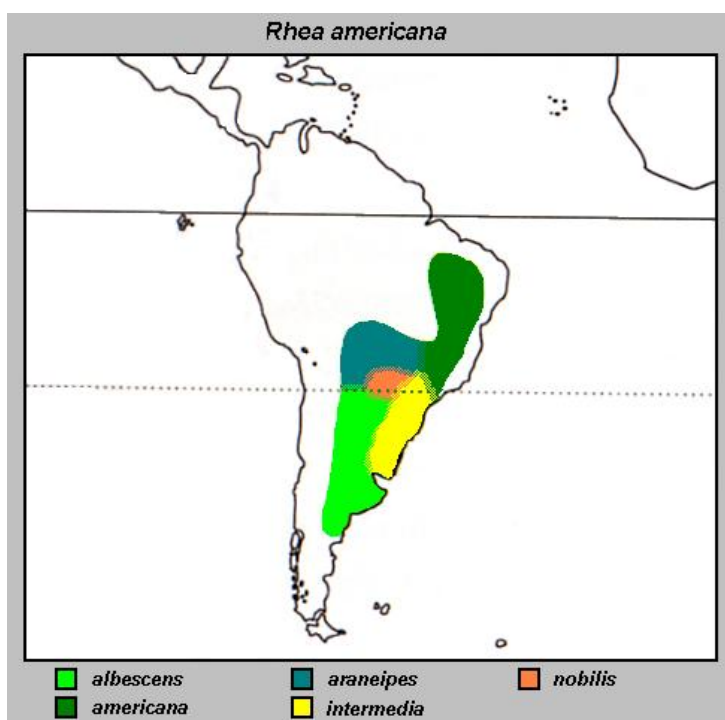
Figura 2: Distribuição da ema *Rhea americana* e suas cinco subespécies na América do Sul. Neste estudo, a subespécie investigada foi a *Rhea americana americana*.

Essencialmente terrícola, a espécie foge a grandes velocidades (a passos de 1,5 m de distância e média de 60 km/h), correndo em ziguezagues controlados pelas asas, que são

alternadamente abaixadas e levantadas. Corridas em linha reta, sem a utilização das asas também são comuns (Codenotti *et al*., 1995).

Apresenta uma dieta onívora. Adultos e filhotes se alimentam de folhas, frutos, sementes, insetos e pequenos vertebrados (lagartixas, rãs, cobras, etc.) (Martella *et al*., 1996; Azevedo *et al*., 2006). Pastam lentamente e ingerem pedras e outros objetos para auxiliarem a trituração dos alimentos; necessitam de extensas áreas para forrageio (Sick, 1997).

É uma ave gregária, vivendo em grupos mistos de machos e fêmeas, adultos e jovens, normalmente em número acima de cinco (5 a 30 indivíduos, mas grupos com mais de 100 aves já foram observados por Bruning, em 1974).

Reproduzem de julho a setembro. Nessa época, os grupos se separam em pequenos bandos e os machos tornam-se bastante agressivos uns com os outros. O macho expulsa os outros machos da área e reúne pequenos grupos de fêmeas (entre três e seis fêmeas) (Del Hoyo *et al.*, 1992). Os machos vocalizam durante a época reprodutiva, produzindo um som grave de duas notas "nnnhhhaaannn-dddúúú". Após a vocalização, correm em direção e ao redor das fêmeas (Raikow, 1969). *Display* de asas (cabeça abaixada, pescoço em forma de U e asas abertas) e de cabeça (movimentos do pescoço que elevam e abaixam a cabeça ou que levam a cabeça de um lado para o outro) são realizados pelos machos durante a corte (Davies, 2002). Após o acasalamento, os machos constroem os ninhos e aguardam pela postura das fêmeas; os ninhos podem chegar a conter 56 ovos (Fernandez & Reboreda, 1998), mas em média são observados 26 ovos por ninho (Bruning, 1974). Cada fêmea põe entre quatro e cinco ovos (Codenotti *et al*., 1995). Após a postura, as fêmeas se separam dos grupos e acasalam com novos machos. O sistema de acasalamento das emas, portanto,

é um misto de poliginia com poliandria seqüencial (Bruning, 1974). A incubação dos ovos e o cuidado parental é tarefa exclusiva dos machos (Sick, 1997).

Em geral, as ações humanas danosas dizimaram ou vêm reduzindo as populações de ema em toda a sua área de ocorrência, principalmente no nordeste, Brasil central e meridional (Dani, 1993; Sick, 1997). Além da caça intensiva, as atividades incluem queimadas, destruição do habitat, eliminação de ovos por maquinário agrícola durante a preparação do solo para plantio e utilização de agrotóxicos em áreas cultivadas (provocando o envenenamento dos exemplares). Em Minas Gerais, existem populações isoladas, sendo a espécie classificada como vulnerável (Machado *et al.*, 1998). Globalmente, a ema é classificada como quase-ameaçada pela IUCN (IUCN, 2009).

A predação de indivíduos também contribui para a diminuição das populações naturais de emas (Guimarães Filho & Faggioli, 1997). Na natureza, espécies como o lagarto teiú (*Tupinambis teguixin*), cachorro-do-mato-vinagre (*Speothos venaticus*), lobo-guará (*Chrysocyon brachyurus*), onça-pintada (*Panthera onca*), alguns gaviões e até cachorros domésticos são predadores das emas (Dani, 1993). Diante do quadro de ameaças referido, tornam-se necessárias medidas de proteção para a espécie. Criação de áreas de proteção natural e a implementação de técnicas adequadas de manejo em cativeiro são algumas medidas de conservação *in-situ* e *ex-situ* preconizadas.

O Bioma Cerrado

O Cerrado é o segundo maior bioma brasileiro (ocupa 21% do território nacional), sendo menor apenas que a Floresta Amazônica (Borlaug, 2002). Savanas, matas, campos e matas de galerias são fitofisionomias observadas dentro do bioma Cerrado (Ribeiro *et al.*,

1981). Duas estações climáticas são bem definidas no Cerrado, sendo uma seca, entre os meses de abril e setembro, e outra chuvosa, entre os meses de outubro e março (Klink & Machado, 2005).

O Cerrado é um dos *hotspots* mundiais de biodiversidade (Myers *et al.*, 2000); estudos com vegetais (Mendonça *et al.* 1998), invertebrados (Dias, 1992), peixes (Casatti, 2005), anfíbios e répteis (Colli *et al*., 2002), aves (Silva, 1995) e mamíferos (Marinho-Filho *et al.* 2002) mostraram grande diversidade de espécies e alto grau de endemismo para alguns grupos.

O Cerrado, bioma de ocorrência natural da ema, tem sido enormemente explorado: mais de 50% de sua área já foi alterada pela ação humana (Silva, 1995a) (Figura: 3) e apenas 0,7% de sua área está protegida legalmente no Brasil (Silva, 1997). Estimativas realizadas por Machado *et al.* (2004) sugerem o desaparecimento quase completo do Cerrado em 2030.
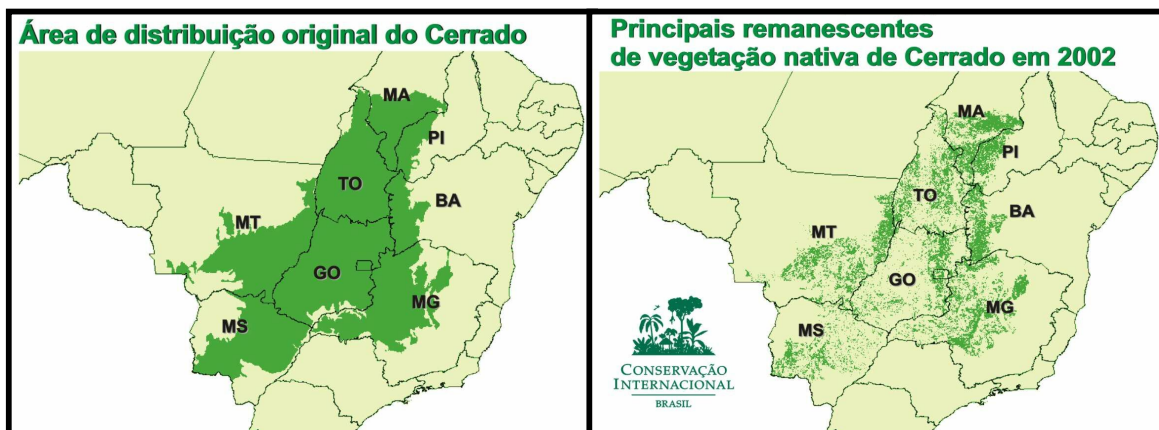
Figura 3: Área de distribuição original e remanescente (em 2002) do Bioma Cerrado no Brasil.

Com a destruição do Cerrado, várias espécies animais e vegetais encontram-se ameaçadas de extinção em algum grau (Klink & Machado, 2005). Espécies grandes, como a ema, parecem ser mais susceptíveis à extinção do que espécies pequenas [populações menores, baixas taxas reprodutivas, grandes áreas de vida e estar presente no topo das cadeias tróficas são fatores sugeridos por Gaston & Blackburn (1995) e Cardillo (2003) para o maior risco à extinção das espécies grandes em comparação com as espécies pequenas]; se estas espécies forem dispersoras de sementes, seu desaparecimento pode levar a um colapso de toda a rede ecológica do bioma (Memmot *et al.,* 2005), diminuindo as taxas de regeneração das áreas impactadas (Link & Di Fiore, 2006). Magnani & Paschoal (1990) sugerem que as emas são boas dispersoras de várias espécies de plantas do Cerrado; sendo assim, seu desaparecimento poderá desencadear uma cadeia de extinções vegetais no bioma, modificando todo o sistema (especialmente se forem as únicas dispersoras de sementes das espécies vegetais em questão).

O papel dos zoológicos na conservação ex-situ

Segundo a União Internacional para a Conservação da Natureza (IUCN), existem 17291 espécies ameaçadas de extinção no planeta (IUCN, 2009). Ações de conservação *in-situ* (ações que ocorrem na natureza), não são muitas vezes suficientes para garantir a sobrevivência das espécies (reservas com tamanhos pequenos, isolamento de populações e poucos recursos no ambiente são as principais causas da insuficiência das ações conservacionistas *in-situ*) (Armsworth *et al*., 2006). Para algumas destas espécies, os jardins zoológicos e botânicos se configuram nos últimos redutos protegidos (conservação

ex-situ, ou seja, fora da natureza); extintas da natureza, apenas indivíduos de cativeiro ainda sobrevivem (Ryder & Feistner, 1995; Ruokonen *et al.,* 2007).

Os zoológicos modernos são normalmente chamados de centros de conservação (Wemmer *et al.*, 2001, Mallinson, 2003; Tribe & Booth, 2003; Young, 2003; Conway, 2004; Primack, 2004) devido à sua capacidade de manter um grande número de indivíduos. No caso das espécies ameaçadas, informações sobre o comportamento, nutrição, genética, reprodução e doenças são reunidas pelos zoológicos e utilizadas na produção de guias de manejo e *studbooks*, ambas maneiras de se aumentar os esforços conservacionistas (Strahl *et al.,* 1995; Kaldenberg, 2004; Cornejo, 2006).

O papel conservacionista dos zoológicos não se restringe apenas à manutenção de populações saudáveis em cativeiro; a produção de conhecimento científico a partir de estudos em cativeiro, a promoção da educação ambiental, e a utilização de estoques cativos para reintrodução na natureza são outros três pilares que norteiam as ações destas instituições nos dias atuais (IUDZG/CBSG, 1993).

Zoológicos têm contribuído sobremaneira na aquisição de conhecimentos sobre a biologia de inúmeras espécies (IUDZG/CBSG, 1993); estudos comportamentais, reprodutivos, genéticos, nutricionais, veterinários, tecnológicos (testes de novos instrumentos, como rádio-colares e sensores de monitoramento fisiológico, por exemplo) só são possíveis de serem realizados em condições controladas de cativeiro (Cornejo, 2006). Para espécies crípticas, pequenas e/ou noturnas, difíceis de serem observadas na natureza, os zoológicos se tornam importantes locais de estudo.

Atividades de educação ambiental são realizadas pela grande maioria dos jardins zoológicos atuais; o número de visitantes destas instituições é enorme e cresce anualmente (a Fundação Zoo-Botânica de Belo Horizonte, por exemplo, recebe cerca de 1,2 milhões de

visitantes por ano – PBH, 2010). A maioria dos zoológicos está localizada em centros urbanos e normalmente se configuram como únicos locais de contato com a natureza de toda a população próxima (IUDZG/CBSG, 1993); a possibilidade de ver, ouvir, sentir, tocar e cheirar os animais, além de observar seus comportamentos, dá aos visitantes a chance de desenvolver respeito por estas criaturas e de entender o seu papel na natureza e a importância de sua existência no planeta (IUDZG/CBSG, 1993).

A função principal dos zoológicos modernos é a de atuarem como estoques de indivíduos para reintrodução (Ruokonen *et al.,* 2007). Entretanto, muita discussão sobre os prós e contras da utilização de animais cativos em reintroduções tem sido realizada no meio acadêmico; animais domesticados, exibindo comportamentos anormais, com baixa variabilidade genética e transmissores de doenças para os coespecíficos selvagens são problemas usualmente apontados para os animais de zoológicos pelos especialistas (IUDZG/CBSG, 1993). Medidas adequadas de manejo podem evitar todos estes problemas. O manejo comportamental é hoje em dia muito empregado neste aspecto: a construção de recintos que imitam o ambiente natural das espécies e fornecem estímulos variados, enriquecimento ambiental, condicionamento animal, e estudos cognitivos e de aprendizagem são realizados para se manter todo o repertório comportamental natural dos animais quando em cativeiro (Young, 2003).

A predação dos indivíduos soltos na natureza a partir de estoques cativos é um problema freqüente que leva ao insucesso dos programas de reintrodução (Beck *et al*., 1991; Short *et al*., 1992; Miller *et al*., 1994). Projetos anteriores de reintrodução de *R. americana* na Estação Ambiental de Galheiro (Perdizes, MG) e na área de proteção ambiental do Serra Azul (Juatuba, MG) falharam devido à predação dos indivíduos por cachorro doméstico (Guimarães Filho & Faggioli, 1997). Segundo Coss (1999), animais que foram isolados de

seus predadores por muitas gerações podem apresentar modificações em seu comportamento antipredação; este tipo de comportamento tem grande custo energético, e o tempo e energia gastos evitando-se predadores podem ser investidos em outras atividades, como forrageamento e reprodução (Ryer & Olla, 1998). Animais de cativeiro, que não tem contato com seus predadores por muitas gerações, portanto, são candidatos em potencial à perda de comportamentos antipredação adequados e à morte prematura após a soltura na natureza.

Testes para avaliação da capacidade de reconhecimento de predadores utilizando-se modelos taxidermizados (Curio, 1998) e técnicas de treinamento antipredação tem sido bastante utilizados antes da soltura dos animais na natureza (McLean *et al*., 1999; Azevedo & Young, 2006). Tem-se sugerido que tais intervenções aumentam as chances de sobrevivência dos animais reintroduzidos (Griffin *et al.,* 2000).

Portanto, o papel dos zoológicos na conservação *ex-situ* pode ter grande importância, desde que medidas adequadas de manejo das espécies e atividades de educação ambiental sejam adotadas por estas Instituições.

OBJETIVOS

O objetivo geral deste estudo foi o de fornecer subsídios para a conservação das emas a partir de pesquisas sobre sua biologia, comportamento e manejo, tanto na natureza quanto em cativeiro.

Os objetivos específicos deste estudo foram:

a) Avaliar como as emas usam seu ambiente natural, bem como a influência das estações do ano, do horário do dia, do tamanho do grupo e do tipo de ambiente no comportamento das aves;

b) Avaliar qual o papel das emas na dispersão de sementes de plantas do Cerrado;

c) Avaliar o grau de conhecimento das populações humanas sobre a biologia e papel das emas na manutenção do equilíbrio ambiental, comparando as opiniões de pessoas em áreas de ocorrência e não ocorrência desta espécie;

d) Avaliar a capacidade de reconhecimento de predadores por indivíduos cativos de emas e suas respostas comportamentais anti-predação;

e) Avaliar a eficiência do enriquecimento ambiental na diminuição da exibição de comportamentos anormais pelas emas;

f) Avaliar como o público visitante do zoológico influencia o comportamento das emas cativas.

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Capítulo 1

Time-activity budget of greater rheas (*Rhea americana*, Aves) on a human-disturbed area: the role of habitat, time of the day, season and group size.

Time-activity budget of greater rheas (*Rhea americana*, Aves) on a human-disturbed area: the role of habitat, time of the day, season and group size.

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Abstract

The aim of this study was to evaluate activity-time budget, habitat use and how seasonality and group size influences the expression of greater rhea behaviours. Greater rheas are threatened South American birds; habitat loss, predation and hunting are the main factors responsible for population declines.The study was conducted in farmlands within a matrix of commercial *Eucalyptus* plantation and remnants of natural habitats of *Cerrado* vegetation (savannah-like) in southeastern Brazil. Rhea groups were located visually in different habitats visited monthly from January 2004 to December 2005. Time spent searching greater rheas in each habitat were equally distributed. Data were collected using scan sampling with instantaneous recording of behaviours every minute. The time-activity budget of greater rheas was influenced by habitat structure, time of the day, season and group size. Rheas spent more time in open areas than in forested areas ($p < 0.001$). Vigilance behaviours were more displayed in forested areas, in the dry season and by solitary and small groups of birds. Resting behaviours occurred more often in openlands and within groups with more than three rheas. Food availability, good visibility, and low human presence are the possible factors for the preference of greater rheas for pasture lands. The results support the resource availability hypothesis where it is expected that habitats with a higher food availability will be more used by the animals, group-size hypothesis, where the scarcity of resources will lead to smaller groups of animals, and that forestry modifies greater rheas habitat use and behaviours.

Key-words: greater rhea, time budget, habitat use, group-size, resource availability.

Introduction

The abundance and availability of resources (e.g.: food) and the risk of predation are considered the main forces that determine how animals use their habitat and if they will live solitarily or socially (Conradt 1998; Neuhaus and Ruckstuhl 2002; Ruckstuhl 1999). It is expected that habitats with a higher risk of predation will be less used by the animals and that the scarcity of resources will lead to smaller groups of animals (Abramsky et al 1996; Hendrie et al 1998; Kenagy et al 1989; Kotler et al 1991; Lacki et al 1984; Vásquez 1996). During dry seasons, when food sources tend to be scarce, strong intragroup competition will favour the formation of smaller groups; in the other hand, during wet seasons, food tended to be abundant, diminishing competition between the animals. In this case, the risk of predation instead of the competition for food will mainly act favouring larger groups (Corp et al 1997; Daly et al 1990; Sommer 2000).

Time-activity budget is important to understand how animals cope with their habitats, energy demands, conspecifics, predators and seasonality (Lafever et al 2008; Lee 1997; Litzow and Piatt 2003; Maxson and Pace III 1992; Muzaffar 2004; Neumann 2001; Palmer et al 2001; Rodway 1998). Evolutionary implications of the activity budget had been suggested since animals that present the best time budget for any circunstances will have more reproductive success and should be selected spreading its genes (Lafever et al 2008; Lee 1997; Orians 1961; White et al 2006).

The aim of this study was to evaluate the behaviour of wild greater rheas in relation to habitat disturbance (*Eucalyptus*-planted forests and pasture lands; the influences of human-disturbed habitats are poorly understood for greater rheas), to know which habitats are used most frequently by greater rheas *Rhea americana* Linnaeus, 1857 (Rheiformes, Rheidae, Aves) in an Brazilian agro-ecosystem; and which behaviours they are expressing

in each habitat, to evaluate the daily activity time-budget in each area, and if group size and seasonality influences the expression of the behaviours. Habitats with mixed cultivated and wild plant species or grasslands are preferred by greater rheas in their occurrence areas (Bazzano et al 2002; Bellis et al 2004; Giordano et al 2008; Martella et al 1996). These studies also showed that large monocultivated grain areas are avoided by the birds due to frequent human contact and intensive agricultural disturbances. Such information will allow better decisions for future conservation efforts for this species.

Greater rheas are threatened flightless birds of South America; populations of these species are vanishing locally in Brazil, Argentina, Paraguay and Uruguay (Codenotti and Alvarez 2000; Di Giacomo and Krapovickas 2005; Giordano et al 2008; Herrera et al 2004). In Brazil, these birds inhabits areas of *Cerrado* vegetation (savannah-like vegetation) (Davies 2002; Sick 1997), one of the most threatened and diverse biomes of Brazil (Machado et al 2004a; Myers et al 2000; Skole et al 1994).

Although greater rheas are not present in the Brazilian National Red List (Silveira and Straube 2008), its populations are declining in several Brazilian States due to habitat loss, hunting, feral dog predation, crop burnings and egg losses caused by agricultural machinery (Dani 1993; Machado et al 1998; Sick 1997). According to IUCN, greater rheas are near threatened (a species that do not fill the requirements for being classified as vulnerable, endangered ou critically endangered, but there are signs that it will be threatened in the near future, IUCN 2008).

Greater rheas are gregarious birds usually found in family groups of three to eight individuals, but groups with more than 90 birds can also be found (Davies 2002). They spent almost all of their time walking and feeding; fights are common, specially between males (Davies 2002). Vigilance behaviours tends to be performed in areas with tall

vegetation and in small groups of birds due to the higher risk of predation (Martella et al. 1995).

They are ground-nesting birds; their mating systems combines harem polygyny and sequential polyandry (Bruning 1974). Adult males fight for harens of females; females lay 20-30 eggs communally in a nest built by the males, which incubate the eggs for approximatelly 40 days (Fernández and Reboreda 2002). Parental care is provided only by the males. Reproduction occurs from July to September, but it can vary throughout their distribution wich encompasses all central South America. Wing reproductive displays (front of the body lowered slightly, neck in a low U-curve and wings extended laterally) and head-bobbing displays (male bobes its neck and head up and down vigorously) are performed by males to females during the mating season (Davies 2002).

The main defensive behaviour that greater rheas uses is a zigzag run controlled by the wings; 60 km/h is the maximun velocity achieved by the birds during these runs. Greater rheas are omnivorous birds that collect their food mainly in the soil; food itens can also be collected in shrubs and trees (Folch 1992). Rheas spent most of their time feeding on vegetables and invertebrates, but small vertebrates are also consumed (Sick 1997).

We predict that greater rheas uses more open areas than forested areas since forestry alters greater rheas behaviour, reduces habitat availability and quality (diminishes food resources) and increases the potential risk for rhea predation due to increased vegetation cover and reduction of visibility. It is also expected that rheas, expresses more vigilance behaviours in the dry season, since they will split larger groups to diminish competition for food; and larger groups expresses more resting behaviours than solitary birds or small groups of rheas.

Materials and Methods

Study area

This study was conducted at the farm named UNISE MG-03 and its surroundings in the Felixlandia Municipality, northwestern Minas Gerais, Southeastern Brazil (79º30'N, 48º23'S) (Fig. 1). The region is largerly dominated by comercial *Eucalyptus* plantation within a matrix of pasture lands and patches of remnant natural vegetation of the Cerrado biome of central South America (a savannah-like vegetation detailed described in Oliveira and Marquis 2002). Annual temperature average of 24ºC (ranging from 16 to 35ºC) and an annual precipitation average of 1235mm, with two distinct seasons: a dry season from May to October (average 41 mm/month rainfall), and a wet season from November to April (average 165 mm/month rainfall) (Carmo et al. 2002).

UNISE MG-03 is a 10568.28ha *Eucalyptus* forest farm owned by 'PLANTAR S.A. Reflorestamentos'. This farm has 66% of its area occupied by *Eucalyptus* trees and 34% destinated for areas of natural vegetation. UNISE MG-03 vicinities are mainly *Brachiaria brizantha* pasture lands for cattle maintenance and it is composed by many small farms with different sizes.

The study area was divided into two distinct habitats, an *Eucalyptus* area (forested area; 6975.06 ha) and a pasture land area (grassland area; 6593.22 ha) (Figure 1). In the *Eucalyptus* and pasture land areas, it were found various predators of eggs, chicks and adults of greater rheas, such as the maned wolf (*Chrysocyon brachyurus*), cougar (*Puma concolor*), crab-eating fox (*Cerdocyon thous*), south american quati (*Nasua nasua*), jaguarundi (*Herpailurus yaguarondi*), ocelot (*Leopardus pardalis*), crab-eating racoon (*Procyon cancrivorus*), domestic dog (*Canis lupus familiaris*) and three species of armadillos (*Cabassous* sp., *Dasypus novencinctus* and *Euphractus sexcinctus* (Sábato et al

2005). *Eucalyptus* spp. was the only species found in the *Eucalyptus* forests ; in the pasture lands, it were identified the monocotyledonous *Brachiaria brizantha*, and the dicotyledonous *Tabebuia aurea*, *Psidium cinereum*, *Albizia lebbeck*, *Duguetia furfuracea*, *Eugenia dysentherica*, *Solanum lycocarpum*, *Solanum palinacanthum*, *Persea americana,* and *Hytis suaveolens* (Azevedo et al 2006a)

Data collection

Both habitats were visited monthly in five-days field trips from January 2004 to December 2005. Time spent searching greater rheas in each habitat were equally distributed (20 hours per habitat per trip; 480 hours in total for each habitat; the minimum observational period was of one hour and the maximum was of four hours a day), but the hour of the day searching changed daily (field searchings range from 6:00 AM to 8:00 PM; but rheas could be searched in different hours; e.x.: from 6:00 AM to 10:00 AM in one day; from 11:30 AM to 15:30 PM in the second day, and so on); in each day roads inside the habitats were run on foot or by car in different hours to equally distributed samples among the periods of time (each period was sampled five hours per field trip; 120 hours in total for each period of the day). Rheas' footprints were followed to facilitate the location of the birds, specially in the *Eucalyptus* forests. Rhea groups were located visually by the three researchers of the team using a 16 X 50 binoculars (Bushmater 1650HWR®) and them followed for the maximum possible time in both habitats. The sighting areas were recorded using a GPS device (eTREX IEC 529IPX7®) and data were collected using scan sampling with instantaneos recording of behaviours every minute (Altmann 1974; Lehner 1996; Martin and Bateson 2007). Number of rheas in the group and the period of the day were recorded. All observations were performed from a distance sufficient to avoid the influence of the researchers on the behaviours expressed by the rheas. The ethogram used was based

on Codenotti et al (1995) and comprised the following behaviours: walking (WAL), alert (AL), foraging (FOR), inactive (IN), preening (PREE), running straight (RS), drinking (DRI), fighting (FIGHT), courting (COUR), jumping (JUM), defending nest (DFN), dust bathing (DUS), running in zigzags (RZ), egg incubation (EGI), intimidating (INT), regurgitating (REG), vocalizing (VOC). Behaviours that not fit in any of these categories were assigned as "other behaviours". Two reproductive periods were sampled in this study (greater rheas reproduction occur from August to October in the study area; Azevedo et al 2006b).

Data analysis

Habitat use was evaluated counting the time spent by the rheas in each area; Fisher's exact test was used to evaluate the significance of the difference between area use (Zar 1999). Activity budget was achieved from two years of rheas observation from 6:00 AM to 8:00 PM; daytime was than splitted in early morning (6:00 to 10:00 AM), late morning (10:00 AM to 12:00 PM), early afternoon (12:00 to 16:00 PM) and late afternoon (16:00 to 20:00 PM). Activity budgets and the time spent in each habitat is given in percentages. Behaviours were quantified and analysed using Friedman non-parametrical test. The Tukey test was used *post-hoc* to compare behavioural responses according to group size (solitary, two, three, four, five or more rheas) and period of the day. Mann-Whitney U-test was used to compare behavioural responses according to the season (dry and wet) and to the habitat (*Eucalyptus* forests and pasture lands). The behavioural responses of the greater rheas according to the season and to group size were only performed for the birds found in the pasture lands due to the larger data recorded in these habitat. The number of samples in each test varied depending to the number of complete hours of behavioural recording (e.g. check sheets with only 15 minutes of data collected

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were excluded from analysis). All statistical analysis were run using MINITAB v.12 and under a confidence level of 95% (= 0.05) (Zar 1999).

Results

Rheas were found mostly using the pasture land areas (90.3% of the time); only in 9.7% of the samplings rheas were found using the *Eucalyptus* forests (Z = -13.11; p < 0.001; DF = 2). The number of birds recorded in the *Eucalyptus* area was 14 and in the pasture land areas was 150, distributed in groups varying from two to nine individuals in the pasture lands and one to three individuals in the *Eucalyptus* forests; solitary rheas were often recorded (86% of the recordings in the *Eucalyptus* forests and less than 10% in pasture lands). Some of the rhea groups (47%) recorded on the pasture lands were associated with cattle.

Rheas' activity in *Eucalyptus* forests was mainly walking (74,3%), running straight (10.5%), alert (8.3%), egg incubation (3.5%) and foraging (3.5%). Walking was the most expressed behaviour in all periods (EM: 96%; LM: 85%; EA: 44%; LA: 72%). Running straight was observed in late morning (15%) and early afternoon (27%), and foraging was observed only once in late afternoon (14%), as well as egg incubation (14%). Alert was more expressed in early afternoon (29%). All behaviours, except walking (F = 6.90, p = 0.07, DF = 3), foraging (F = 5.76, p = 0.12, DF = 3), and egg incubation (F = 5.76, p = 0.12, DF = 3) differed significantly between the periods of the day (AL: F = 8.10, p = 0.04, DF = 3; RS: F = 8.10, p = 0.04, DF = 3). Tukey *post-hoc* test showed that the alert and running straight behaviours were much more expressed during the early afternoon than in the other periods of the day (p < 0.05 for both cases). In *Eucalyptus* areas whith trees in the initial growth stage (less than one meter high), rheas walked and stood alert frequently while

walking and running straight were the most displayed behaviours of the rheas in areas with *Eucalyptus* more than three meters high (egg incubation was also recorded for in these areas).

In the pasture lands, the majority of rheas activity consisted in foraging (35.0%), walking (28.2%), inactive (11.3%), alert (7.3%) and preening (4.1%). All other behaviours occurred less than 1% of the time in this habitat. Foraging was most expressed in early morning (37.0%), early afternoon (36.6%) and late afternoon (36.1%); walking was most expressed in late morning (47.3%). Alert was also more expressed in late afternoon (15.5%); both inactivity and preening behaviours were more expressed in early afternoon (15.2% and 7.6% respectivelly). None of the behaviours differed statistically between the periods of the day (WALK: $F = 1.00$, $p = 0.80$, DF = 3; FOR: $F = 1.00$, $p = 0.80$, DF = 3; AL: $F = 5.70$, $p = 0.13$, DF = 3; IN: $F = 2.0$, $p = 0.57$, DF = 3; PREE: $F = 1.30$, $p = 0.73$, DF = 3).

Walking was more performed in the *Eucalyptus* forests than in pasture lands ($U = 25.0$, $p = 0.05$, DF = 2), but foraging, inactive and preening were more expressed in the pasture lands if compared to the *Eucalyptus* forests ($U = 10.0$, $p = 0.03$, DF = 2; $U = 2.31$, $p = 0.02$, DF = 2; $U = 2.31$, $p = 0.02$, DF = 2, repectivelly). None other behaviours differed between the habitats.

The expression of the behaviours changed from the dry season to the wet season; during the wet season, walking occupied almost 25% of the activity budget of the greater rheas; this percentage doubled during the dry season (51.4%). The same pattern was observed for the behaviour foraging, but this behaviour was more expressed during the wet season (54.9%). Alert and inactive behaviours did not show much variation between the seasons (9.8% for the dry season and 8.8% for the wet season for alert behaviour; 4.8% for the dry season and 6.7% for the wet season for inactive behaviour). Preening was almost

three times more expressed during the wet season if compared to the dry season (3.7% and 0.8% respectivelly). Mann-Whitney U-Test showed that the differences between the expression of the behaviours during the wet and dry seasons were significant for walking, foraging, preening and inactive (Table 1).

The largest group observed was formed by nine rheas. The size of the group also influenced greater rheas behaviours. Only the behaviours walking (F = 10.55, p = 0.03, DF = 4), alert (F = 25.56, p < 0.01, DF = 4) and preening (F = 9.59, p = 0.05, DF = 4) differed statistically between the groups; solitary rheas spent much more time walking and alert than groups with four or more birds. Preening was more common for groups with or larger than five birds. Inactivity was greater for solitary rheas and foraging was greater for groups with five or more birds, but both behaviours did not differed between the group sizes (F = 0.86, p = 0.93, DF = 4; F = 4.16, p = 0.39, DF = 4, respectively); running straight (F = 7.90, p = 0.09, DF = 4), intimidation (F = 4.00, p = 0.41, DF = 4), drinking (F = 4.00, p = 0.41, DF = 4) and other behaviours (F = 3.31, p = 0.51, DF = 4) did not differ between the group sizes.

Discussion

The time-activity budget of greater rheas was influenced by habitat structure, time of the day, season and group size. The results of this study indicates that forestry alters greater rheas habitat use and behaviours; the same results were found for breeding birds in Australia (Recher et al 1985). The results found in this study support the resource availability hypothesis, since smaller greater rhea groups were found mostly during the dry season and that foraging activities were more exhibit in open areas than in the *Eucalyptus* forests (Corp et al 1997; Daly et al 1990; Sommer 2000).

Rheas spent significantly more time in pasture lands than in forested areas, a fact that was also observed by Bazzano et al (2002), Bellis et al (2004), Codenotti and Alvarez (2000), Giordano et al (2008) and Herrera et al (2004). Three factors may be responsible for the preference of greater rheas for pasture lands: (1) food availability, (2) good visibility, and (3) low human disturbance.

The pasture lands studied were covered mainly by *Brachiaria brizantha* (Hochst. Ex A. Rich) Stapf grass and had small patches of native plants such as *cagaita* (*Eugenia dysenterica* D.C.), guava (*Psidium guajava* L.) and Caribbean Trumpet (*Tabebuia aurea* Benth and Hook. f. ex. S. Moore) trees. *Brachiaria* species are normally used for cattle feed due to its high productivity and protein/fiber content (Barnabé et al 2007; Pupo 2002). Native Cerrado plants are also notable for their nutritive and antioxidant values (Costa and Vieira 2004; Roesler et al 2007). Since it was observed greater rheas eating great amounts of *B. brizantha*, fruits/leaves/flowers of different native plant species, arthropods and small vertebrates in the pasture land areas, it is supposed that their nutrient requirements are meet in the pasture land areas (Azevedo et al 2006a). Foraging activities in the *Eucalyptus* area, on the other hand, were seldom recorded and it was directioned mainly to insects and plant species in the edges of the forest; none *Eucalyptus* leaves were found in faeces of greater rheas in a previous study in the area (Azevedo et al 2006a) perhaps due to their low digestability (Cork 1984; Foley et al 1999).

Although food availability was not directly measured in both habitats, it is well known that the management of *Eucalyptus* forests (mechanical and chemical undergrowth weedings), the allelopatic substances produced by the *Eucalyptus* trees that inhibited shrub growth, and the less structured environment of monocultural forests diminishes the richness, abundance and diversity of animals if compared to natural forests (Majer and Recher 1999;

Ramos et al 2004; Souto et al 2001; Viana and Pinheiro 1998). Pasture lands also presents low species richness and diversity indexes (Bernarde and Abe 2006; Bernarde 2007; Macedo et al 2008), but the semi-intensive management, where the pasture do not receive chemical weedings, allows the survival of a larger number of animal species, specially insects and small mammals (mainly rodents and marsupials), items frequently consumed by greater rheas (Azevedo et al 2006a; Martella et al 1996). Thus, it is assumed that in the pasture land area more food items were available for the greater rheas if compared to the *Eucalyptus* forests, a fact that may be responsible for the preference of use of this area by the birds, but further studies that measure sistematicaly the food availability (specially invertebrates and small vertebrates) in both habitats should be conducted to confirm this hypothesis.

The sight of predators at long distances is important for flightless birds because it gives sufficient time for escaping runnings, diminishing the chances of being killed by a surprise attack (Krebs and Davies 1993). In forested areas, vision is diminished by the understory vegetation making these areas less attractive for the rheas (Bellis et al 2006; Codenotti and Alvarez 2000; Giordano et al 2008). On the other hand, pasture lands provide openness that favour vigilance and a rapid scape (Bellis et al 2004). Associations of greater rheas with cattle, recorded many times in this study, are also possible advantage against predation since the excelent eyesight of the formers with the great scent of the latters increases de efficiency in detecting predators at a distance (Folch 1992).

Human disturbance, such as the use of heavy agricultural machinery that destroy nests and eggs, harvesting, pest control, weeding and even the elevation of the risk of predation by the people's accompanying dogs, have been considered the main reason for greater rheas to avoid crop plantations (Bellis et al 2004; Codenotti and Alvarez 2000;

Giordano et al 2008). The management of the silvicultural *Eucalyptus* plantations was intensive and the presence of people was constant (Aracruz 2008; Viana 2004). Besides, the practice of weeding the understory allied with the low digestive and nutritive value of *Eucalyptus* leaves could deter foraging activities in the area (Aracruz 2008; Cork 1984; Foley et al 1999;). In pasture lands, the presence of people is not so frequent due to the extensive cattle management techniques adopted and the possibility of greater rheas to locate humans at long distances is facilitate (Bellis et al 2006; Demaria 1994; Herrera et al 2004).

Walking was the most expressed behaviour in the *Eucalyptus* forest habitat and this was probably due to the use of this area as a passing way from pastoral and *Cerrado* areas adjacent to the *Eucalyptus* forest. The use of the roads inside the *Eucalyptus* forests as corridors between natural areas by the animals was also observed by Machado and Lamas (1996).

Vigilance and defense behaviours (alert and running straight) changed significantly between the period of the days for the *Eucalyptus* forest (both were displayed more in the early afternoon); footprints suggesting predation events were observed inside the *Eucalyptus* area, then, if predation is frequent in this area, more defensive behaviours should be performed; but seasonal and group size effects, as well as the small sample size in these habitat (only 14 birds were observed) could be responsible for such variations and further studies focusing exclusively in the forested areas should be conducted for better understandings of its effects on greater rhea behaviours.

Walking and foraging were the most performed behaviours during all periods of the day in pasture lands; these behaviours, together with inactive and preening were not displayed in dangerous or stress situations. Rheas became more alert in late afternoon, just

before nightfall, period when the location of predators is affected by low light intensities (Rand et al 1997; Richmond et al 2004). The major predators of greater rheas are the jaguar, cougar, maned wolf, domestic dogs (kill adults and chicks), common tegu (eat eggs), bush dog and some birds of prey (kill chicks) (Dani 1993). Small carnivores, like foxes, grisons, and racoons also hunt greater rheas, specially chicks and eggs (Azevedo et al 2006c; Bazzano et al 2002; Bellis et al 2004). Most of the predators listed are present in the study area and are predominantly nocturnal (Azevedo et al 2006b; Vale and Neto 2002, 2005) supporting the hypothesis of a higher risk of predation at night.

Seasonal variation was also observed influencing the behaviour expression in greater rheas. Birds spent significantly more time walking in the dry season (May to October) and foraging, preening and inactive in the wet season (November to April). Alert was more frequently displayed in the dry season, but it does not differed statistically from the wet season. Carro and Fernandéz (2008), studying seasonal variation in the diurnal activity budget of greater rheas in the Argentinean Pampas (Buenos Aires Province), found that behaviours like walking, foraging and alert were more expressed in the wet season (March to April); this result is the contrary found for the rheas studied here. The authors argued that this was due to the breeding season, which occurs from September to January in that region, when rheas walk, forage and stay alert more frequently because of their sexual activities (searching for other groups to copulate, sexual displays, competition for mating, etc.) (Lombardi 1994).

Breeding season in Felixlandia ranged from August to October (Azevedo et al 2006b); sexual displays and matings were recorded during August and nests and egg incubations were recorded in September and October. Sexual behaviours were observed in the end of the dry season and not in the wet season, differing from the results reported by

Carro and Fernandéz (2008), but rheas in the northwestern Minas Gerais behave similar to the rheas of Argentina, walking, foraging and staying alert more during breeding than non-breeding season. This was also reported by Reboreda and Fernandéz (1997) for rheas in a ranch of Entre Rios, Argentina.

Depending of the size of the group, rheas behave differently. Solitary birds spent more time walking and expressing vigilance and defensive behaviours, such as alert and running straight. Resting behaviours, such as foraging and preening were more expressed in groups with five or more rheas. To detect and escape from predators, a solitary rhea needs to spend great amounts of time in vigilance in detriment of other behaviours and, in fact, this has been observed by many researchers (Bellis et al 2006; Carro and Fernandéz 2008; Fernandéz et al 2003; Martella et al 1995; Reboreda and Fernandéz 1997). In larger groups, any individual have the possibility to be less vigilant, since there are many other birds to be alert in its place; a situation known as the many eyes effect (Pulliam 1973). Meanwhile, the value of the vigilance of other member of the group is lower than an individual's own vigilance, who respond quickly to an attack when it is alert (Elgar et al 1984; Lima 1995). This has been proposed to explain why in larger groups rheas increase their vigilance behaviours, a fact smoothly observed in this study.

An alternative explanation for the higher levels of vigilance by solitary rheas is the breeding season; solitary males could be more alert not because of the risk of predation, but because their are searching conspecific competitors or female groups to mate (Carro and Fernandéz 2008; Lombardi 1994; Reboreda and Fernandéz 1997). If so, it is expected that solitary birds walk more than grouped birds, a result found in this study. This is confirmed by the fact that during the breeding season, rheas displayed more alert behaviour than in the

non-breeding season. Breeding season is also suggested to explain the higher levels of vigilance in ostrich groups in Africa (Burger and Gochfeld 1988).

Time-activity budgets of captive ostriches and wild emus showed that these birds changed their behaviours according to the season and time of the day (Csermely et al 2007; Dawson et al 1984; Deeming, 1998; McKeegan and Deeming, 1997); gender and the size of the group also influenced in their behavioural expression, specially for ostriches (Csermely et al 2007; Deeming, 1998; McKeegan and Deeming, 1997). Time-activity budgets for the other ratites are unavailable; they should be run to permite phylogenetic comparisons and the development of better management procedures for captivity populations.

Greater rheas used pasture and open lands more often than forested ones, displayed more vigilance behaviours when solitary or when living in small groups and were more observed living in small groups during the dry season, facts that corroborate the habitat-influence hypothesis (forestry exclusion) and resource availability hypotheses. Time-activity budgets changed seasonally and according to the period of day and number of birds in the flock. Such information is important and necessary to the conservation of threatened species, allowing better decisions during management plans, both in the wild and captivity environments.

All the observations were conducted in accordancy with Brazilian laws.

The authors declare that they have no conflict of interest.

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TABLE

Table 1: Mean number of behavioural records ± standard error and Mann-Witney U-Test results for the comparison of the expression of behaviours by greater rheas during the wet and dry seasons at Felixlandia Municipy, nortwestern Minas Gerais, Brazil, during 2004-2005 (.= 0.05; DF = 2; N = 62)

Behaviour	Dry	Wet	Mann-Whitney	P-value
Walking	18.16 ± 1.63	11.16 ± 1.10	3262.0	< 0.01*
Foraging	10.16 ± 1.42	21.95 ± 1.73	4785.0	< 0.01*
Alert	3.46 ± 0.66	3.48 ± 0.48	4133.0	0,191
Preening	0.33 ± 0.09	2.31 ± 0.36	4818.0	< 0.01*
Inactive	1.85 ± 0.56	4.58 ± 0.87	4568.5	< 0.01*
Running straight	0.55 ± 0.13	0.35 ± 0.09	3777.5	0.536
Fighting	-	0.05 ± 0.03	-	-
Intimidanting	0.02 ± 0.02	0.05 ± 0.03	3937.0	0,315
Courting	0.03 ± 0.02	0.05 ± 0.03	3906.0	0,655
Egg incubation	0.81 ± 0.81	-	-	-
Drinking	0.06 ± 0.05	-	-	-
Defending nest	0.06 ± 0.06	-	-	-
Jumping	0.02 ± 0.02	-	-	-
Dust bathing	-	0.06 ± 0.05	-	-
Regurgitating	-	0.02 ± 0.02	-	-
Other behaviours	0.32 ± 0.16	0.11 ± 0.05	3810.5	0.533
Not visible	25.11 ± 2.80	16.26 ± 2.31	3503.5	0.06

* = behaviours that differed significantly between seasons.

FIGURE LEGENDS

Fig. 1 Study area at Felixlândia Municipality, northwestern Minas Gerais, southeastern Brazil. UNISE-MG-03 farm is represented by the hachured area (*Eucalyptus* forests); dashed lines represents the surroundings of the UNISE MG-03 farm also studied (pasture lands)
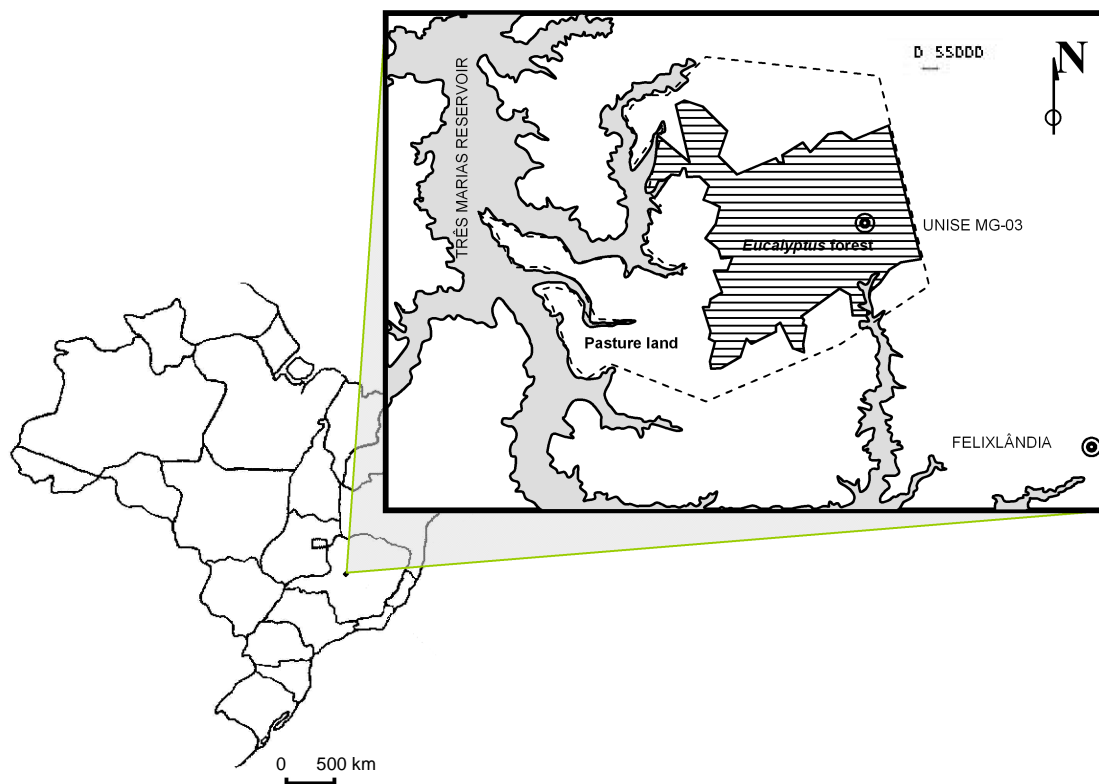
Figure 1

Capítulo 2

Seed Germination In Brazilian *Cerrado*: The Role Of Greater Rheas *Rhea americana*

Artigo a ser submetido para: **Current Zoology.**

Seed germination in the Brazilian c*errado*: the role of greater rheas *Rhea americana*

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Running title: Seed dispersal by greater rheas.

ABSTRACT

Frugivores seed dispersers play an important role in the regeneration of natural populations and communities. One possible seed disperser of *cerrado* plants is the greater rhea, and we studied the potential of seed dispersal by greater rheas through experiments of seed germinability before and after ingestion. The germinability and the mean time of germination of two groups of seeds (treatment: seeds that passed through the birds' digestive system; control: seeds extract direct from the fruits) were evaluated. The tested plant species were guavas (*Psidium guajava*), wolf apple (*Solanum lycocarpum*), *juá* (*Solanum palinacanthum*), passion fruit (*Passiflora edulis*), field *araticum* (*Annona crassiflora*), *mangaba* (*Harconia speciosa*), *jatoba* (*Hymenaea stignocarpa*), *cagaita* (*Eugenia dysenterica*), and souari nut (*Caryocar brasiliense*). Seeds of the wolf apple germinate less in the treatment group than in the control group, and seeds of *juá* germinate more in the treatment group than in the control group. The seeds of guavas germinate more in the control group than in the treatment group, and the seeds of the passion fruit germinate more in the treatment group than in the control group, but these differences were not significant. Only one seed of *mangaba* germinated in the treatment group. None of the seeds of the field *araticum, souari nut* and *cagaita* germinated in either group. Seeds in the treatment groups germinated faster than in control groups for guavas, wolf apple, passion fruit and *jua*, but only for the last species the result was statistically significant. We concluded that greater rheas act as an efficient seed disperser of some *cerrado* plant species; their habit of walking and foraging over great areas increases their role in seed dispersal.

Key-words: *cagaita*; guava; *jatoba*; *jua*; *mangaba*; passion fruit; souari nut; wolf fruit.

INTRODUCTION

Birds and mammals are the main seed dispersers of the Neotropics (Peres and Baider, 1997, Willson and Travaset, 2000), and fruits constitute the primary source of energy for animals, since they are easily found, processed and consumed (Levey, 1994, Galetti et al., 2004). Seed dispersal is an important function carried out by the frugivores (Holbrook et al., 2002); it is responsible for the maintenance of the integrity of the plant communities where they occur (Jordano et al., 2006) and play an important role in the regeneration of natural populations and communities (Willson and Travaset, 2000).

Seed dispersal provides advantages to plants: (1) the escape from high mortality rates of seeds and plantules under or near the adults (escape hypothesis; Janzen, 1970); (2) the possibility of colonization of new sites and environments (colonization hypothesis; Wilson, 1993); and (3) the dispersal to particular favorable microhabitats (directed-dispersal hypothesis; Howe and Smallwood, 1982). Besides, seed scarification during gut passage can alters their permeability to oxygen and water, making the seed more suitable for germination (Vásquez-Yanes and Orozco-Segovia, 1993). One disadvantage is the possibility of seed damage by animals' teeth, beak or stomach acids; in these cases, animals act as seed predators instead of dispersers (Hulme and Benkman, 2002).

Many studies have evaluated the effects of gut passage on seed germination, and the results are diverse: some showed an increase in seed germination (Julliot, 1996, Travaset, 1998, Santos et al., 2003, Varela and Bucher, 2006), and others showed a decrease in seed germination (Uieda and Vasconcelos-Neto, 1985, Verdú and Travaset,2004). Most of the studies, however, showed no effects on seed germination (Barnea et al., 1990, Moll and Jansen, 1995, Julliot, 1996, Knogge et al., 2003, Cáceres and Monteiro-Filho, 2007, Sato et

al., 2008). The evaluation of the germination rates of seeds collected from the faeces is the first step to determine if a species is a seed disperser or predator (Galetti et al., 2004).

Greater rheas (*Rhea americana*, Rheidae, Aves) are omnivorous flightless birds that inhabit grasslands and *cerrado* of South America (Sick, 1997); they are the largest bird species of South America, ranging from 1.34 to 1.70m height and weighting 32-35kg (Sick, 1997). Although greater rheas ingest small vertebrates and invertebrates, the majority of their diet consists of fruits, leaves and seeds of *cerrado* plants (Azevedo et al., 2006, Comparatore and Yagueddú, 2007). Noble (1991) suggest that the greater rheas are possible dispersers of many plant species from the *cerrado* biome, but this hypothesis has never been tested (Calviño-Cancela et al., 2006). Renison et al. (2010) found that greater rheas are good seed dispersers of *Geoffroea decorticans*, *Ziziphus mistol* and *Prosopis nigra* in Argentina; this is the only research that has investigated the role of greater rheas in seed dispersal.

The *cerrado* is considered a biodiversity hotspot (Myers et al., 2000), meanwhile, it is one of the most exploited biomes of Brazil (Klink and Machado, 2005), with more than 54% of its original area destroyed or disturbed in some degree by human activities (Ratter et al., 1997); with the actual destruction rate, it is expected that the *cerrado* biome will disappear by 2030 (less than 1% of the Biome is legally protected in natural reserves; Machado et al., 2004). The loss of habitat leads to species extinction (Brooks et al., 2002), and many animal and plant species of the *cerrado* are now threatened by extinction to some degree (Klink and Machado, 2005); this is the case of the greater rheas, near threatened in many areas and vulnerable to extinction in others (Sick, 1997, Davies, 2002), such as the *cerrado* of Minas Gerais State, southeastern Brazil (Machado et al., 1998). Larger species seems to be more susceptible to extinction than smaller ones (Gaston and Blackburn, 1995,

Cardillo, 2003) and the loss of certain seed dispersers can generate the failure of the whole ecological networks in an area (Memmot et al., 2005), mainly due to the disruption of the mutualistic relations between plants and animals (Cordeiro and Howe, 2003), diminishing the regeneration of disturbed areas (Link and Di Fiore, 2006).

Plants with larger diaspores prevent many animals from swallowing them; thus, large-bodied frugivores have been hypothesized to shape plant communities through their ability to disperse large fruits and seeds (Lord, 2004). According to Guimarães et al. (2008), some plant species of the *cerrado* are now in risk of extinction or facing genetic problems due to the lack of seed dispersers since their seeds are too large to be dispersed by the actual *cerrado* fauna (greater rheas and the tapir – *Tapirus terrestris* Perissodactila – are the largest frugivores of the extant fauna of the *cerrado*) (Galetti et al., 2001).

In the *cerrado* of Brazil, many studies have been conducted with seed dispersal and they have found that the numbers of zoochoric species are enormous (Gottsberger and Silberbauer-Gottsberger, 1983, Oliveira and Moreira, 1992, Batalha and Mantovani, 2000, Vieira et al., 2002), but none evaluated the role of the greater rheas in seed dispersal. In fact there are few studies that have investigated the role of the ratites in seed dispersal, the majority of these studies being with the Australian ratites, the kiwi, emu and cassowary (Noble, 1991, Webber and Woodrow, 2004, Westcott et al., 2005, 2008, Calviño-Cancela et al., 2006). These studies showed variable results; with some seeds germinating more (Noble, 1991, Webber and Woodrow, 2004) or less (Westcott et al., 2008) after gut passage. The aim of this study was to evaluate the potential of seed dispersal of *cerrado* plants by greater rheas and this was evaluated through experiments of seed germinability. We hypothesized that the greater rheas disperse the *cerrado* fruits, especially the bigger ones, playing an important role in the maintenance of the *cerrado* community dynamics.

MATERIALS AND METHODS

Fruits of guavas (*Psidium guajava* L., Myrtaceae), wolf apple (*Solanum lycocarpum* St.-Hill, Solanaceae), *juá* (*Solanum palinacanthum* Dunal, Solanaceae), passion fruit (*Passiflora edulis* Sims., Passifloraceae), field *araticum* (*Annona crassiflora* Mart., Annonaceae), *jatoba* (*Hymenaea stignocarpa* Mart., Caesalpiniaceae), *mangaba* (*Hancornia speciosa* Gom., Apocinaceae), *cagaita* (*Eugenia dysenterica* DC, Myrtaceae), and souari nut (*Caryocar brasiliense* Camb., Caryocaraceae), typical climax plant species of the *cerrado* (except the *juá*, which is an invasive *cerrado* species) (Lorenzi, 2000, 2002a, b), were offered to six captive greater rheas held by the Belo Horizonte Zoo (19º51'44" S; 44º00'40" W), Brazil, in the years of 2008, 2009 and 2010. All plant species were chosen based on a field study of the diet of the greater rheas (Azevedo et al., 2006) and on scientific literature (plant species with possible fruits ingested by greater rheas mainly due to their size; Lorenzi, 2002a, b).

Mature fruits of each species were offered to the rheas in plastic bowls on five consecutive days from 8 30h to 9 30h. None of the fruits were offered simultaneously. Faeces were collected from the enclosure in the following five days, always at 8 00h. Faeces were collected manually using a small shovel and they were washed through a mesh under a constant flow of water to promote seed separation and cleaning. After processing, seeds were blotted dry with a paper towel in a shaded at ambient temperature. Seeds with visible mechanical damage were descarted.

Seeds of each species were separated in two groups: (1) control: seeds extracted directly from the fruits; (2) treatment: seeds that were offered to greater rheas – which had passed through the greater rheas gut and collected from the faeces. Each group comprised of 100 seeds. For the germination tests, the seeds of both groups were placed in 11 cm

gerbox dishes filled with 2 cm of vermiculite as substrate, except for souari nut seeds that were sown individually in 500mL plastic cups filled with a 4 cm layer of sand (in the bottom of the cup) and a 2 cm layer of vermiculite (Pereira et al., 2004). A total of 100 seeds distributed 4 replicates of 25 were used in all treatments. The gerbox dishes and plastics cups were placed in the greenhouse and were watered with tap water three times a week in order to keep continuously moist. Germination was recorded when cotyledons emerged (Borghetti and Ferreira, 2004) and seedlings were counted every two days. Recording continued until 180 days after the seeds were sown for the souari nut and 90 days for the rest of the species tested.

The percentages of germination and mean times of germination (results presented in days to germination) were calculated. The calculation of the mean time of germination (MT) followed the formula proposed by Labouriau (1983): $MT = (n_i . t_i) / n_i.$, where n_i represents the number of seeds that germinate in the time interval t_i. Results from both groups of data were analyzed using the G-test (Zar, 1999) and were conducted using a 95% level of confidence (p 0,05).

RESULTS

The greater rheas individuals consumed the fruits of eight species, but they avoided consuming of jatoba fruit (excluded from the analysis). The percentage of germination varied according to the species evaluated (Table 1). The results differed statistically only for three species: seeds of the wolf apple germinated less in the treatment group than in the control group, seeds of the passion fruit germinated more in the treatment group than in the control group, and seeds of *jua* germinated more in the treatment group than in the control group. The germinability of seeds of guavas between control and treatment groups was not significant (Table 1). Only one seed of *mangaba* of the treatment group germinated and

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none seed of the field *araticum* and souari nut and *cagaita* germinated in either group (Table 1).

The mean time of germination of the seeds from the *juá* differed statistically between the treatments, being faster for the seeds that had passed through the digestive system of the greater rheas (Table 2); seeds of the control group started germination on the thirtieth day and seeds of the treatment group started germination on the tenth day. Seeds of all others species germinated faster in the treatment group than in the control group, although these differences did not reach statistical significances (Table 2). Seeds of wolf apple of both groups started germination on the twentieth day and thus the comparison was not statistically significant (Table 2). Only one seed of *mangaba* germinated and this was after 44 days from the start of the germination experiment (G-test could not be performed to evaluate the mean time of germination for this species due to the low rate of seed germination; Table 2)

DISCUSSION

We found that the greater rheas are good dispersers of *juá*, guavas and passion fruits, but do not function as dispersers of wolf apples and *jatoba*. The results for the *mangaba*, souari nut, *cagaita* and field *araticum* indicate that greater rheas can disperse their seeds, but don't promote the faster or major seed germination of these species.

Although wolf apple and *juá* belong to the same family (Solanaceae), the results of seed germination were the opposite between them, with the seeds of *juá* increasing in number and in velocity of germination, while seeds of wolf apple diminishing their number and increasing slightly their velocity of germination. We believe that these results were due to differences in gut retention time and the nature of the species evaluated (invasive versus non-invasive species).

The dispersal potential of the wolf apple by the maned wolf (*Chrysocyon brachyurus*, Canidae) is widely recognized (Lombardi and Mota Jr., 1993, Rodrigues, 2002, Santos et al., 2003), but seeds of this species have also been found in the faeces of tapirs (*Tapirus terrestris*, Tapiridae) (Pinto, 1998), crab-eating foxes (*Cerdocyon thous*, Canidae) (Rodrigues, 2002), hoary foxes (*Lycalopex vetulus*, Canidae) (Dalponte and Lima, 1999) and tegu lizards (*Tupinambis merianae*, Teiidae) (Castro and Galetti, 2004). The germinability of the seeds that passed through the guts of these species varied; there were no differences in the germination velocity for the seeds ingested by the tegu lizards and crab-eating foxes, but seeds ingested by the maned wolves and tapirs germinated faster than those not ingested by these animals (Rodrigues, 2002). Seeds of *juá* were found in scats of three bat species in a gallery forest, but no germination test was run (Piccoli et al., 2007).

The differences in the germinability and in the velocity of germination of seeds were suggested to be related to the gut retention time, but the conclusions are inconsistent, with some authors claiming that longer gut retention times improve germination by seed scarification by gut acids (Travaset et al., 2001), and others claiming that shorter gut retention times increase germination by avoiding seed damage by gut acids (Murray et al., 1994). The number of seeds in each scat also influences their germinability (Gosper et al., 2005). In this study, although not systematically measured, gut retention times were similar for both *Solanum* species (seeds appeared in the scats 12 hours after being ingested by the birds), but the number of seeds of *S. palinacanthum* collected in the scats was higher than the seeds of *S. lycocarpum*.

Solanum species contains glycoalkaloids, such as -solanine and -solamargine, in their fruits, and these secondary metabolites influence gut retention time and seed germinability (Ripperger and Schreiber, 1981). Wahaj et al. (1998) demonstrated that great

quantities of glycoalkaloids delay seed germination in *S. americanum*, but did not influence gut retention time. It has been hypothesized that because birds are generally more mobile than mammals of an equivalent size, shorter retention times are beneficial for bird dispersal (to avoid dispersal outside the proper habitat) and longer retention times are beneficial for mammal dispersal (to ensure adequate dispersal distance within the habitat) (Wahaj et al., 1998). Mammal-dispersed *Solanum* species have higher concentrations of glycoalkaloids than bird-dispersed (Cipollini and Levey, 1997). Thus, it would be expected that wolf apple seeds remained in the rheas' digestive system for longer periods than *juá* seeds, but this was not observed in this study (systematic gut retention studies needs to be conducted). Greater rheas ingested the entire *juá* fruits, but only parts of the wolf apple; it is possible that concentrations of the glycoalkaloids in the pieces of wolf apple ingested were low enough to avoid constipation in the birds, but further studies should be conducted to evaluate this hypothesis.

S. palinacanthum is considered an invasive species in the Brazilian *cerrado* (Lorenzi, 2000); invasive species normally presents an r-strategy of reproduction, producing great numbers of seeds with faster growth (Pianka, 1970). Besides, invasive species often produce seeds capable of germinating under a wide range of environmental conditions (Cervera and Parra-Tabla, 2009). The greater germinability and the lower mean time of germination of seeds that passed throught the rheas' gut corroborates this hypothesis.

The guavas did not show any differences in the germinability and mean time of germination between the treatment and control groups. Seeds of guava are impermeable to water and gases, presenting low germinability (Singh and Sonil, 1974). In this study, guava seeds germinated at higher rates (80% in the control group and 73% in the treatment group),

and the non-controlled temperature of the greenhouse and the scarification by digestive acids, which creates small cracks in the seed tegument, could have facilitate the water and gases intake, allowing greater germinability (Bewley and Black, 1982). The few studies that evaluated other Myrtaceae seed dispersal by birds and reptiles did not show any differences between treatment and control groups (Barnea et al., 1991, Castro and Galetti, 2004). Gresseler et al., (2006) suggested that the relationships between the Myrtaceae and its dispersers should be extensively studied to determinate the efficiences of seed dispersal of each vertebrate clade.

Seeds of the passion fruit germinated significantly more in the treatment group than in the control group; the mean time of germination, although faster in the treatment group, did not differed statistically from the control group. Passion fruit seeds germinability was negatively influenced by its aril (Pereira and Dias, 2000), and the complete cleaning of the seeds, which passed through the greater rheas' digestive system, could be responsible for its greater germinability.

According to Heringer (1962), the presence of germination inhibitors in the souari nut could be responsible for the delay of seed germination for about 12 months. Melo and Gonçalves (2001) found germination inhibitors in the endocarp, pulp and spines of the souari nut, but not in the seeds. In natural conditions, the seeds are accompanied by the fruit, and the germination inhibitors inside the fruit can allow the seeds to germinate only when the environmental conditions are adequate (Melo and Gonçalves, 2001). The passage through the digestive system of the greater rheas did not remove the endocarp and the spines of the souari nuts, and this could be responsible for the results observed in this experiment.

The role of greater rheas in field *araticum* dispersal could not be evaluated in this study due to the lack of seed germination in both groups. According to Rizzini (1973), in *A. crassiflora*, the embryo is poorly developed when the fruit is formed, and this fact delays the germination of the seeds by eight months (a time not covered by this study), and this may be the reason for the lack of germination for this species. However, since the seeds recovered from rheas' scats were not mechanically damaged, the embryos inside may also have been intact and the germinability may thus be equal between seeds ingested and non-ingested by rheas. If this was the case, greater rheas may function as a seed disperser of the field *araticum* by increasing the distances of the seeds from their progenitors, but this hypothesis must be verified in a future field-laboratory study. The role of the greater rheas in the dispersal of *cagaita* and *mangaba* fruits could not be determinate in this study due to the lack or low percentuals of seed germination in both groups (control and test); the experiments were run in three consecutive years (2008-2010), but in all trials the seeds were contaminated with fungi (only one seed of *mangaba* germinated in the treatment group).

Endozoochory contributes positively to the colonizing and stablishment of plantules in new environments (Varela and Bucher, 2006). For plant species with large fruits, the loss of large frugivores, such as the greater rheas, could bring problems to their populations, since their seed dispersion would decline (Cramer et al., 2007). Our results indicates that greater rheas act as an efficient seed disperser of some *cerrado* plant species; their habit of walking and foraging over great areas (Davies, 2002), and their capacity to swallow large fruits (Renison et al., 2010) increases their role in seed dispersal and in habitat restoration.

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TABLES

Table 1: Comparison between the percentages of seed germination of the eight plant species offered to the greater rheas of Belo Horizonte Zoo.

Species	Control %	Treatment %	G test	*p*-value
Psidium guajava	80	73	1.37	0.24
Solanum lycocarpum	**46**	16	20.45	< 0.001*
Solanum palinacanthum	17	**46**	21.32	< 0.001*
Passiflora edulis	37	**51**	3.99	0.05*
Eugenia dysenterica	0	0	-	-
Hancornia speciosa	0	1	-	-
Annona crassiflora	0	0	-	-
Caryocar brasiliense	0	0	-	-

*: Results that differed statistically.

Table 2: Comparison between the mean times in days (MT) of seed germination of the eight plant species offered to the greater rheas of Belo Horizonte Zoo.

Species	Control	Treatment	G test	*p*-value
	MT	MT		
Psidium guajava	36.50	23.44	2.09	0.15
Solanum lycocarpum	51.57	49.86	0.02	0.89
Solanum palinacanthum	46.57	**21.26**	6.86	< 0.01*
Passiflora edulis	44.50	33.83	2.86	0.09
Eugenia dysenterica	0	0	-	-
Hancornia speciosa	0	44	-	-
Annona crassiflora	0	0	-	-
Caryocar brasiliense	0	0		

*: Results that differed statistically.

Capítulo 3

Does people's knowledge about an endangered bird species differ between rural and urban communities? The case of the greater rhea (*Rhea americana*, Rheidae) in Minas Gerais, Brazil

Artigo a ser submetido para: **Conservation Biology.**

Does people's knowledge about an endangered bird species differ between rural and urban communities? The case of the greater rhea (*Rhea americana*, Rheidae) in Minas Gerais, Brazil

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Abstract

Greater rheas are threatened South American birds. Populations of this species are now held in captivity for future conservation programmes. Meanwhile, for a conservation program to succeed it is necessary that the human community close to an *in-situ* conservation project be involved. Thus, the perceptions of the human community must be evaluated and incorporated into the conservation activities planned. It is expected that the human communities that are inserted in the environment where the endangered species occurs will give more importance to its conservation, than human communities that live far from the problem. The aim of this study was to evaluate if the perceptions of rural and urban communities about greater rheas is different due to experience and proximity with the problem. The study was conducted in São José do Buriti (rural community) and at BH Zoo (urban community), both localized in Minas Gerais State, southeastern Brazil. One-hundred and thirty-five questionnaires were applied to people in both areas. People's knowledge about the greater rheas' risk of extinction differed in some aspects between rural communities and urban communities, but both communities agreed that the local extinction of greater rheas would bring ecological problems. Rural communities related local environmental problems (e.g., silvicultural activities) as one of the main reasons for greater rheas's declining populations while urban communities related more general reasons (e.g. trafficking and hunting). Rural people were better able to recognize greater rheas than the urban community. Environmental education programmes should be implemented in both areas to maximize conservation actions.

Key-words: conservation, environmental education, greater rhea, people's knowledge, zoo.

Introduction

More than 12% of the world's bird species are threatened by extinction to some degree (IUCN 2008). In Brazil, 25.5% of the threatened species are birds according to the National Red List (Machado et al. 2008); according to the IUCN's red list, 6.7% of the Brazilian bird species are threatened (IUCN 2008). Differences between the two lists are generated mainly due to the criterions used by each Institution for the classification of species in the threat categories (ex. the yellow-nosed albatross *Thalassarche chlororhynchos* is classified as endangered by IUCN and as vulnerable by the National Red List of Brazil) (Rodriguez et al. 2000; Rodrigues et al. 2005; Marini and Garcia 2006; Milner-Gulland 2006).

Greater rheas *Rhea americana* Linnaeus, 1857 (Rheiformes, Rheidae, Aves) are the largest birds of South America, occurring from the north of Brazil to the south of Argentina (Sick 1997; Davies 2002), and it is one of the bird species that differed between the two red lists cited earlier. Although greater rheas are not present in the Brazilian National Red List (Machado et al. 2008), their populations are declining in several Brazilian States due to habitat loss, hunting, feral dog predation, crop burning and egg losses caused by agricultural machinery (Dani 1993; Sick 1997; Machado et al. 1998; Navarro and Martella 1998; Fernandéz and Reboreda 2000; Di Giacomo and Krapovickas 2005). According to the IUCN, greater rheas are classified as 'near threatened' [a species that does not fulfil the requirements for being classified as vulnerable, endangered or critically endangered, but there are signs that it will be threatened in the near future (IUCN 2008)].

Conservation efforts are therefore necessary to reduce the impacts of human activities upon wild populations of greater rheas. One of the most indicated actions to protect any threatened species is the implementation of environmental education activities (Feisinger 2004; Padua et al. 2004). Environmental education can be defined as the joining element of

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educational systems that induce in the society the consciousness for the need of sustainable development of communities (Almado 1996). It serves not only to transmit knowledge, but also to transform human attitudes towards nature, inserting each person in their environment, showing their responsibilities in its management (Arai and Sprules 2001). It is known that there exists a correlation between people's behaviour and how they perceive the environment (Fishbein 1967). An investigation about what people know about threatened species (etnozoology) could affect species conservation (Ruiz-Mallen and Barraza 2008), since it could indicate actions to be implemented using environmental education; thereby, enhancing the effectiveness of these actions through the insertion of the community (Campbell et al. 2010).

Depending on the conflicting demands of local and regional communities, desires about the conservation of a certain endangered species may change (Jobes 1991; Power 1991; Rasker et al. 1992; Reading et al. 1994), but it is expected that human communities, which are inserted in the environment where the endangered species occurs will give more importance to its conservation than human communities that live far from the problem, especially those communities who exploit this species to some degree (Gadgil et al. 1993; Berkes et al. 2000).

The aim of this study was to investigate the level of knowledge and interest about greater rheas of the school children and their parents in a community at the Felixlândia Municipality (north-western Minas Gerais, Brazil) and to compare them to an urban community far from the greater rheas' natural environment. The results of this study could contribute to the development of more efficient conservation measures for this bird species.

Method

Study areas

The study was conducted in two distinct areas, one rural and one urban. The rural area was located at São José do Buriti (local population: 1,390), a District of the Felixlândia Municipality, north-western Minas Gerais State, south-eastern Brazil (18ºS, 45ºW). The region is inserted in the *Cerrado* biome (*latu sensu*) (Veloso and Goés-Filho 1982), and declining wild greater rhea populations was locally observed (Azevedo et al. 2006). The urban area, the zoological garden (BH Zoo), was located at Belo Horizonte Municipality (local population: 2,375,444), capital of the Minas Gerais State (19ºS; 44ºW). This area is the second biggest green area of Belo Horizonte (1,450,000 m^2), with elements of the *Cerrado* and Atlantic Forest vegetation (Silva et al. 2006); there are seven greater rheas on-exhibit to the visitors of the zoo. BH Zoo receives 1.2 million visitors annually. The rural area and the urban area were separated by 225 km.

Data collection and analysis

Two similar semi-structured questionnaires (Appendixes 1 and 2) were applied in both areas; in the rural area, it was applied to the local population of the São José do Buriti. In the urban area, the questionnaire was applied to the visitors at the greater rheas' enclosure at the Belo Horizonte Zoo. In total, it we randomly applied 270 questionnaires, being 135 in each area; however, our initial objective was to interview 200 people but we were limited in the rural community by the number of people available or willing to respond; however, our sample size was 10% of the rural population. Some questions of the questionnaires were not answered by the interviewee, which explains different samples numbers in the result section (e.g., only 124 people in the local community answered their gender; 11 people did not answer this question).

The questionnaires intended to evaluate the knowledge and the perceptions of the populations about the conservation of the greater rheas. We collected general demographic information about the interviewed public such as gender, age, schooling (level), profession and birthplace, and specific information, like the role of introduced predators (e.g., feral dogs) and zoos in the conservation of greater rheas. The schooling categories followed the Brazilian Educational System: fundamental level (seven to 14 years old), secondary level (15 to 18 years old), technical school (15 to 18 years old school), and university degree (19 to 23 years old). Data were summarized and analyzed statistically using the Chi-square Test. All tests were run using MINITAB v.12 and with a confidence level of 95% (= 0.05) (Zar 1999).

Results

Rural Community (São José do Buriti)

We interviewed 84 men (62.22%) and 40 women (29.63%). The majority of people aged between 10 and 19 years old (45.19%; N = 61), the next highest category was of 20-29 years old (17.04%; N = 23) (Table 1). Most of the interviewees had incomplete primary school (seven to 14 years old) (33.34%; N = 45) or incomplete secondary school (15 to 18 years old) (31.85%; N = 43) (Table 2). Students (N = 26), farm-hands (N = 14) and forest helpers (N = 11) were the most encountered professions.

Most of the interviewees recognized the greater rhea photograph (N = 120; 88.89%). One-hundred and two people (75.56%) said that they had seen greater rheas in the study region, and 10 people (7.41%) had never seen this bird. Most of the interviewees (N = 96; 71.11%) reported seeing greater rheas only in opened habitats, like grasslands or open *Cerrado*; some (N = 3; 2.22%) reported seeing greater rheas in forests and 8.89% (N = 12) saw greater rheas inside *Eucalyptus* forests; the same percentage saw rheas in both opened and

forest areas. One interviewee (0.74%) told that he saw a greater rhea locked inside a pen, and one interviewee (0.74%) saw greater rheas in all habitats.

More than 85% of the interviewees (N = 115) believed that greater rheas' local populations were vanishing, and 12% (N = 16) did not believed that greater rhea populations were reducing. When questioned about the possible causes of the diminution in the local populations, people answered: habitat loss (22%; N = 30), poisoning from pesticides (19%; N = 26), hunting (7%; N = 10) and other reasons not listed in the questionnaire, such as the increased transit of people and cars in the region, drying of water resources and the establishment of *Eucalyptus* plantations in the area, which were cited by 17% (N = 24) of the interviewees (Figure 1).

Only three interviewees (2.22%) had the habit of collecting greater rheas' eggs to eat, but when asked if this habit could cause any risks to the birds, only one individual (33.33%) answered yes; one answered no and the other did not answer this question. Three questions evaluated the degree of knowledge of the role of feral dogs in the greater rhea extinctions: more than a half of the interviewees had a dog (N = 79; 58%), 28% (N = 38) allowed their dogs to run free through the vicinities of their houses, and 37% (N = 49) thought that this habit did not bring risk to greater rhea populations (Figure 2).

Ninety-six (71.11%) interviewees thought that it was important to conserve greater rheas locally, and 87% (N = 117) would like to participate in a conservation program if implemented in the region. When asked why we should conserve greater rheas, many of the interviewees (53.33%, N = 72) answered that if the rhea disapear, the biological equilibrium would be at risk; 26.67% (N = 36) answered that the greater rhea should be conserved due to its beauty, and 8.89% (N = 12) due to its economic value (Figure 3).

Urban community (zoo visitors)

We interviewed 62 men (45.93%) and 73 women (54.07%). The majority of people had an age varying between 20 and 29 years old (N = 48; 35.56%), the next most prominent category being of 30-39 years old (N = 37; 27.41%) (Table 1). Schooling varied from a complete fundamental level (seven to 14 years old) (17.78%; N = 24) to complete secondary school (15 to 18 years old) (44.44%; N = 60) (Table 1). Students (N = 13; 9.63%), drivers (N = 9; 6.67%) and teachers (N = 6; 4.44%) were the most encountered professions.

Many of the interviewees recognized the greater rhea (67.41%; N = 91), and 44 (32.59%) individuals confused greater rheas with ostriches or did not recognize the bird. One-hundred and fourteen (84.44%) persons said that they had not seen greater rheas in the wild; 19 (14.07%) people had seen wild greater rheas at least once in their lifetimes.

More than 71% of the interviewees (N = 97) believed that greater rheas populations are in risk of extinction, and almost 26% (N = 35) believed that greater rhea populations were not in risk of extinction. When questioned about the possible causes of the diminution in the greater rheas' populations, people answered that habitat loss (28.15%; N = 38), hunting (14.81%; N= 20) and these two causes acting together (45.93%; N = 62) were the main reasons (Figure 1).

One hundred and thirty-two (97.78%) interviewees thought that it is important to conserve greater rheas, and 83.70% (N = 112) thought that the maintenance of greater rheas in zoos could be a good strategy to conserve this species. When asked why we should conserve greater rheas, many of the interviewees (40%; N = 54) answered that greater rheas have as much right to live as any other living creature, 22.96% (N = 30) answered that ecological

problems due to the extinction of greater rheas could be difficult to manage, and 13.33% (N = 18) thought that it is important to conserve greater rheas due to their aesthetics; since they are beautiful birds to watch (Figure 3). The main reason given by the interviewees about the role of zoos in a conservation strategy for greater rheas were protection (53.91%; N = 61), educational opportunities (20%; N = 23) and reintroduction stocks (19.13%; N = 22); 5% (N = 11) did not answer this question and 1.74% (N = 2) said that zoos should be the last alternative to conservation of greater rheas or any other species.

People were asked if their actions could stress greater rheas' and if they do, what would be the consequences of this. More than 70% (N = 95) of the interviewees answered that the visitors do indeed stress the birds and 69.62% (N = 94) of the persons believed that this is not a good thing for the birds. The major problems cited as a consequence of keeping stressed birds in zoos were: reproductive failure (17.31%; N = 18), death (16.35%; N = 17) and increase in the aggressiveness (13.46%; N = 14). Educational problems (6.73%; N = 7), the expression of abnormal behaviours (4.81%; N = 5), decreasing of feeding (2.88%; N = 3) and low psychological welfare (1.92%; N = 2) were also suggested by the interviewees as problems of keeping stressed greater rheas in captivity.

Rural data versus urban data

A chi-squared test showed that the interviewees opinions differed significantly between rural and urban populations for the sightings of greater rheas in the wild, with rural communities seeing more birds than urban ones ($X^2 = 143.41$; DF = 1; p < 0.01). In terms of recognition of the greater rheas, rural communities identified more correctly than urban communities ($X^2 = 18.24$; DF = 1; p < 0.01). In terms of opinion about greater rheas' risk of extinction, the rural community believed that greater rheas are in greater risk of

extinction than urban communities ($X^2 = 8.60$; DF = 1; p < 0.05), and as for the causes of greater rheas' population decrease, urban people suggested more causes than the rural community ($X^2 = 37.09$; DF = 1; p < 0.01). There were also a statistical diference between the rural and urban communities for the reasons that greater rheas should not disapear ($X^2 = 59.56$, p < 0.01) with the urban community suggesting more reasons than the rural community.

Discussion

Rural Community (São José do Buriti)

The rural community had some knowledge about greater rheas' biology and their conservation status; although a few aspects of the birds' biology and population threats were apparently unknown to them (e.g., most of the interviewees did not recognized that their dogs could kill greater rheas). The predominant age and schooling level were 10-19 years old with incomplete fundamental or secondary school, due to the majority of the data set being collected at the São José do Buriti State School. According to Padua et al. (2004), the choice of the interviewee public is extremely important and depends on the identified problem/theme. In this study, the problem was the extinction of the local greater rheas' population, and the choice of the São José do Buriti State School as a data collection site was due to the importance of the children in the transmitting the information and experience in environmental education received in the school to their relatives. It is easier to implement environmental education programs to change habits and concepts about the whole of the humankind in the protection of the planet and its wildlife for children (Strong

1998). It has been shown that children also have the power to change their parent's habits, increasing the net of nature's conservancy (Vaughan et al. 2003).

Most people already knew or had seen wild greater rheas in the study area; most of them made sightings in opened areas, which was expected since greater rheas inhabit grasslands and opened *Cerrados* (Del Royo et al. 1992; Sick 1997; Davies 2002). Few interviewees made sightings of greater rheas in forests or inside the *Eucalyptus* plantations. Although uncommon, greater rheas can use such habitats (Martella et al. 1996; Bazzano et al. 2002; Bellis et al. 2004; Giordano et al. 2008), but the low detectability of birds inside forests (Martella and Navarro 1992; Donatelli et al. 2004) may influenced this results.

The great majority of the interviewees believed that the number of local greater rheas had diminished, and that the habitat loss and the poisoning by pesticides were the main reasons. Both reasons were linked by the population to the silvicultural activities in the study area (production of *Eucalyptus* forests); the use of pesticides inside the plantations was pointed out as one of the greatest problems of such silviculture. Few interviewees believed that greater rheas migrated from *Eucalyptus* forests to the opened areas in the vicinity.

Although the local community had reported great mortality of rheas after the implementation of the silvicultural farms in the region, especially due to poisoning by pesticides, this hypothesis are not supported by a previous study of environmental impacts conducted in the area (Del Rey Engenharia 2000). According to this study, although *Eucalyptus* forests had been applied with pesticides to eliminated weeds, which could cause the death of rheas due to bioaccumulation, only two birds were found dead in the plantations and none of them died by poisoning, as toxicological exams confirmed later. According to the Monsanto do Brasil Company, the manufacturer of the pesticide used in

the *Eucalyptus* forests (Scout[®]), the possible bioaccumulation of the pesticide is virtually inexistent, since it acts only on weeds and is soon degradated in the enviroment (Monsanto 2004). This idea was passed to the local population by local environmental activists and even by some teachers in the schools. This could explain the great number of interviewees suggesting this reason. If so, it demonstrates the great power of schools in teaching notions of environmental impacts and education (Vaughan et al. 2003) and, obviously, the need to do this with correct information.

The hypothesis of the migration of greater rheas to the vicinity of the *Eucalyptus* forests seemed more plausible than the hypothesis of the poisoning. The *Eucalyptus* silviculture was planted over human-altered areas, covered by exotic grasses used for cattle production (Del Rey Engenharia 2000). Thus, it did not destroyed native forests or *Cerrados*, but it eliminated grasslands, areas that are frequently used by greater rheas, forcing the birds to migrate to nearby areas. During this migration, many birds could have died, since they become more susceptible to hunting, capture, predation and accidents with cars. According to Lank et al. (2003), predation of individuals during migration events is high since animals become more conspicuous to predators. But, since studies were not run in the region at that time, the migration hypothesis and the higher levels of predation events are only speculation.

It is important to emphasize that the local community believes that the conservation of the greater rheas' population would help in the maintenance of some ecological equilibrium in the region, and that they want to participate in conservation programs for the species.

The desire of the local community to participate in the conservation of local greater rheas populations is extremely positive. Pádua et al. (2004) suggested that a key-element for an

environmental education program is to develop mechanisms to promote the participation of the local communities in conservation actions. The individual's strength and her sense of identity are essential for environmental education, and need to emerge from inside each person (Glazer 1999). The community's participation in all solutions and decisions in a conservation program should be guaranteed; in this way, the feelings of "to conserve what matters to me" could blossom and dominate their actions.

The majority of the interviewees had dogs, and more than a half allowed their dogs free access to explore the surrounding areas (grasslands, *Cerrados*, forests, etc.). The opinions about the possible problems of this habit were equally divided, with half of the persons thinking that dogs bring problems to greater rheas and half of the persons thinking that this was not a problem to greater rheas.

Predation of wild animals by domestic animals is a common problem around the globe (Ruxton et al. 2002; Butler et al. 2004; Cavalcanti 2004; Kays and DeWan 2004). The domestic dog, is one of the most problematic animals in terms of wildlife predation; it causes great impacts not only in the prey populations but also in the native predator populations, since it competes with them for prey (Yanes and Suárez 1996; Manor and Saltz 2004).

Carcasses of greater rheas predated by dogs normally show a great number of injures on the legs, and this is due to their bites during the attack and also preys are often not ingested by the dogs (Cavalcanti 2004). Two of three greater rhea carcasses found in the study area by the researchers showed these characteristics, and one video recording of a persecution and killing of a greater rhea by a dog proved that this was a real problem in this area.

One of the biggest problems faced by conservation researchers is the lack of knowledge that the owners of domestic animals have about their pets and the danger they can cause to native fauna if left to roam freely (Galetti and Sazima 2006). In the study area this problem existed as well as the problem of domestic dog predation on greater rheas. Besides, the owners that left their dogs free thought that their dogs did not hunt rheas, not believing that their dogs represented a real problem for greater rhea's conservation. Environmental education programs should be implemented in the region approaching the problem of dogs to wildlife conservation, especially for the greater rheas.

Many reports of people capturing greater rhea adults and chicks for consumption or trafficking were collected in this study, although 97% (N = 131) of the interviewees said that this practice was not done. The local population should understand that they play an important role in the conservation of greater rheas by teaching their children to respect this species or even participating in the vigilance of the area, protecting the birds from activities of persons from outside of their community. Again, environmental education is a valuable tool to achieve these goals (Baral et al. 2007).

Finally, it is important to note that the interviewees' data were in general biologically accurate and therefore we can be confident that their responses were in general genuine/honest.

Urban community (zoo visitors)

The urban community had less knowledge about greater rheas and their conservation status, but had more general knowledge about conservation problems than the local community. The predominant age categories were 20-29 and 30-39 years old, but all classes of ages were represented. The same could be observed when the schooling was analyzed. All

classes were representative, showing a great variety of public knowledge. Tunnicliffe (2006) stated that the most important work that biology education researchers can do is to identify the starting points for teaching from and then stimulate the manner in which new biological knowledge is built upon the learner's existing knowledge. This creates a challenge in the educational activities at zoos, since it needs to be able to reach people practically from illiterate to post-graduated educational levels. But, it is important to remember that people who visit zoos acquire information about the animals in TV shows, books and from parents, relatives and friends, so, they have some degree of knowledge, even this knowledge being far from scientific terms or not necessarily correct (Boulter et al. 2003).

Most people recognized greater rheas, but a large sample did not know the bird or confused it with the ostrich (*Struthio camelus*). According to Bruner et al. (1966), people have a need to name the animals and they allocate a name of nearest fit according to their knowledge. In this study, some people thought that the greater rheas were ostriches; ostriches are the closest genetic relative to greater rheas depending on the methods used to infer phylogenies (Sibley and Alquist 1990; Del Hoyo et al. 1992; van Tuinem 1998; Davies 2002). The ostrich is the African ratite and, as the other large animals of African savannas, it appears more frequently on TV documentaries, cartoons and even in children books. This fact could be responsible for the confusion of the zoo visitors; since they rarely go to the zoo to see native fauna, but often to see the African fauna, a phenomenon that occurs in most zoos of the world (Auricchio 1999; Achutti 2003).

Most of the interviewees had not seen greater rheas in the wild; from the interviewees that had already seen greater rheas in the wild, most were from other Brazilian States, such as

Mato Grosso and Maranhão; these states are in concordance with greater rheas' known distribution (Sick 1997). Some of the interviewees reported that although they had seen greater rheas in the wild, there had been a while since their last sightings, which is in accordance to the studies that showed that greater rheas have declining populations all over their distribution area (Dani 1993; Sick 1997; Machado et al. 1998; Navarro and Martella 1998; Fernandéz and Reboreda 2000; Di Giacomo and Krapovickas 2005).

The great majority of the interviewees believed that the number of greater rheas is diminishing, and that the habitat loss, hunting and trafficking were the main reasons. The increase of human population was cited by many as the motive for habitat loss. Hunting and trafficking may have been cited as reasons due to the recent explosion of reports about these themes on Brazilian TV, but no programs talking specifically about greater rheas have been made.

Zoo visitors believed that it was important to conserve greater rhea populations due to their intrinsic right to live and their contribution in the maintenance of some ecological equilibrium in nature. More than 87% (N = 115) of the interviewees thought that zoos play an important role in conservation efforts for greater rheas, and for their protection from hunters and predators. It is important to highlight the educational opportunities that captive environments offer to the public, so long as the animals are experiencing good levels of animal welfare (Fernandes and Timberlake 2008). Since, for example, animals expressing a lot of abnormal behavior tend to generate pity rather than interest in the species (Swaisgood 2007).

According to the public interviewed, zoo animals have the chance to become stressed due to visitor disturbances, and this can cause troubles for conservation efforts. Low levels of

welfare can reduce feeding and reproduction activities, turning the animals more susceptible to diseases and, in the last case, shorten the animals' lifespan. These problems have been observed by many zoo researchers (Davey and Henzi 2004; Hosey 2005; Davey 2007; Farrand 2007), thus, low levels of animal welfare should be avoided by using environmental enrichment, and the visitors should receive special training or participate in educational programs to know how to behave properly when visiting the zoo (Fernadez et al. 2009).

Rural community versus urban community

The communities differed in some important aspects: (1) people from rural community had more chance to observe the birds in their natural habitat than people living in the city, an expected result; since most of the city's citizens did not have any opportunity to watch wildlife unless they went to a zoo (Young 2003); (2) people from rural community recognized more frequently greater rheas than city people. TV documentaries are more consumed by city people than rural people, but on the other hand, TV docummentaries often show exotic fauna and that should be the reason of so many answers confusing greater rheas with ostriches by zoo visitors; and (3) habitat loss was the main reason cited as being responsible for the decline of greater rheas by both communities, but the other reasons differed significantly between them, with urban people relating trafficking and hunting, and rural people relating poisoning by pesticides as important factors. The knowledge about animal extinction and wildlife trafficking increased with schooling level. Urban people had more formal knowledge than rural people and this could be decisive in this difference; the rural community related local problems as the main reason for the diminution of greater rheas' population while urban community related general reasons

(e.g., rural people suggested that the implementation of an *Eucalyptus* forest in the area could be responsible for the diminution of the greater rheas, since greater rheas do not live in forested areas; urban people suggested that habitat loss could be responsible for the extinction of the greater rheas, but no further information were given by them).

People fight for what they care about, and what they care about is strongly linked to what they know (Brewer, 2006). The affective domain seems to be crucial in the development of attitudes towards nature conservancy (Moyer 1975; Ruiz-Mallen and Barraza 2008). It is important to evaluate the feelings of rural and city people about greater rheas, and, based on the results of such an evaluation, create an environmental education program. A citizen-science strategy is proposed by Brewer (2006) and it is strongly suggested for the case of the greater rheas; it consists in connecting conservation biologists with members of the public, who will help with data collection and research. This would increase their knowledge about nature, the role of humans in shaping the environment, demystifying and increasing appreciation of science (Brewer 2006).

Appendix 1: Questionnaire applied to the rural community of São José do Buriti, Minas Gerais State, southeastern Brazil, to determine their knowledge about the biology and conservation of the greater rhea (*Rhea americana*, Rheidae, Aves).

Date:_____ Local: _____ Gender: M (   ) F (   )

Age:_____ Schooling: _____ Profession: _____

1 – Mark the greater rhea photograph.

(   )   (   )   (   )

2 – Have you seen greater rheas in your region? (   ) Yes   (   ) No

3 – If yes, in what habitat?

Open (grasslands and *Cerrados*) (   )

Forest (   )

Eucalyptus forest (   )

4 – Do you think that the greater rheas' populations are declining in your region? Yes (   )
No (   )

5 – If yes, what are the causes?

(   ) Habitat loss   (   ) Hunting   (   ) Trafficking

(   ) Poisoning (   ) Other

6 – Do you have the habit of eating greater rheas' eggs or hunting greater rheas for food?

(   ) Yes   (   ) No

7 – If yes, do you think that these activities could represent a risk for the greater rhea's population in the region? Yes (   ) No (   )

8 – Do you think that it is important to conserve greater rheas in the region?

Yes (   ) No (   )

9 – Why?

10 – Would you like to participate in a conservation program for the greater rheas in the region? Yes ( ) No ( )

11 – Do you have dogs? Yes ( ) No ( )

12 – Do you leave your dog to run free in natural areas? Yes ( ) No ( )

13 – Do you think that allowing your dog to run freely through such areas represents a risk to the greater rheas of the region? Yes ( ) No ( )

14 – Why?

Appendix 2: Questionnaire applied to the urban community of Belo Horizonte, Minas Gerais State, southeastern Brazil, to determine their knowledge about the biology and conservation of the greater rhea (*Rhea americana*, Rheidae, Aves).

Date:_____ Local: _____ Gender: M ( ) F ( )

Age:_____ Schooling: _____ Profession: _____

1 – What is the bird you are looking at?

2 – Have you seen this bird in your region? ( ) Yes ( ) No

3 – Do you think that this species is in risk of extinction? ( ) Yes ( ) No

4 – If yes, what are the causes?

( ) Habitat loss ( ) Hunting ( ) Trafficking ( ) Poisoning

( ) Other

5 – Do you think that it is important to conserve greater rheas in the region?

Yes ( ) No ( )

6 – Why?

7 – Do you believe that is a good conservation measure to maintain greater rheas in captivity? ( ) Yes ( ) No

8 – Why?

9 – Do you believe that the zoo visitors can stress the greater rheas? ( ) Yes ( ) No

10 – Do you see any problems in keeping stressed greater rheas in captivity? ( ) Yes ( ) No

11 – If yes, what problems?

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Table 1: Age and schooling of the rural and urban interviewees about greater rheas (N = absolute number; % = relative number).

Rural Community (São José do Buriti)

Schooling	N	%	Age	N	%
Incompleted fundamental	45	33.34	10-19	61	45.19
Completed fundamental	9	6.67	20-29	23	17.04
Incompleted secondary	43	31.85	30-39	8	5.93
Completed secondary	13	9.63	40-49	14	10.37
Incompleted university	0	0.00	50-59	9	6.67
Completed university	2	1.48	60-69	6	4.44
Technical	4	2.96	70-79	5	3.70
Not answered	19	14.07	Not answered	9	6.67
Total	**135**	**100**	**Total**	**135**	**100**

Urban community (Zoo visitors)

Schooling	N	%	Age	N	%
Incompleted fundamental	1	0.74	10-19	10	7.41
Completed fundamental	24	17.78	20-29	48	35.56
Incompleted secondary	20	14.81	30-39	37	27.41
Completed secondary	60	44.44	40-49	22	16.30

Incompleted university	7	5.19	50-59	15	11.11
Completed university	15	11.11	60-69	1	0.74
Technical	0	0.00	70-79	2	1.48
Not answered	6	4.44	Not answered	0	0.00
Total	**135**	**100**	**Total**	**135**	**100**

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Figure 1: Reasons pointed out by the interviewees from rural and urban communities for the decline of greater rheas' populations (results presented in percentages).

Figure 2: Perceptions of the rural community (São José do Buriti, Minas Gerais, Brazil) about the role of domestic dogs in the risk of extinction faced by greater rheas (results presented in percentages).

Figure 3: Reasons pointed out by the rural and urban people communities (zoo visitors) for the conservation of greater rheas (results presented in percentages).

Figure 1
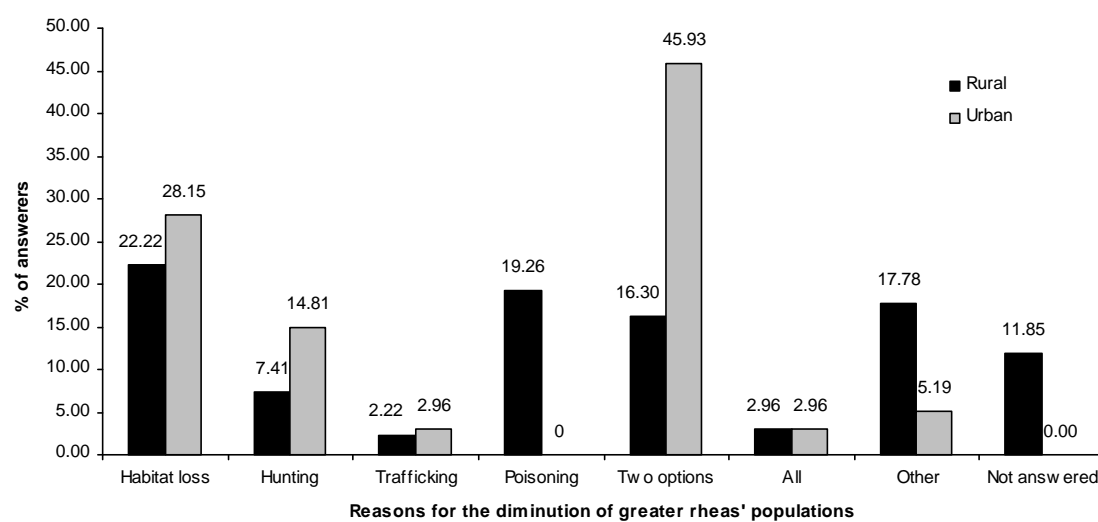
Figure 2
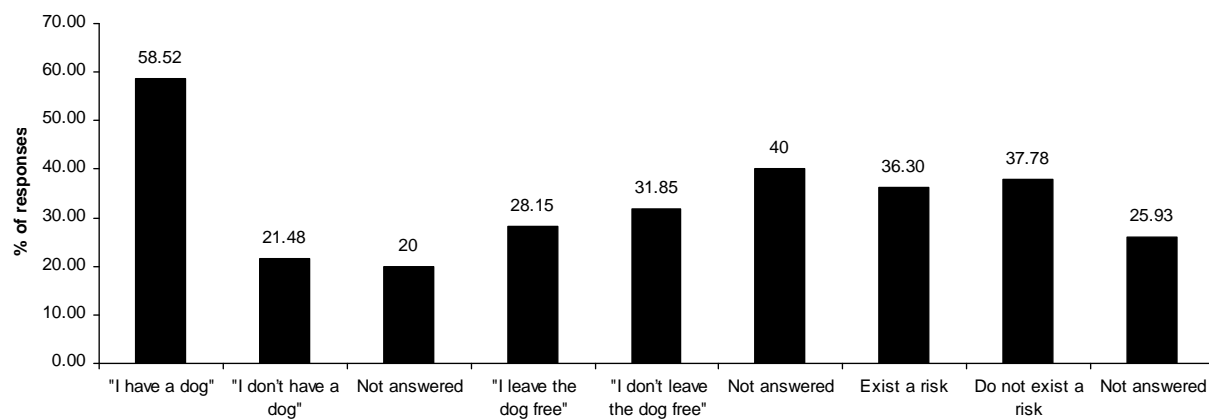
Figure 3
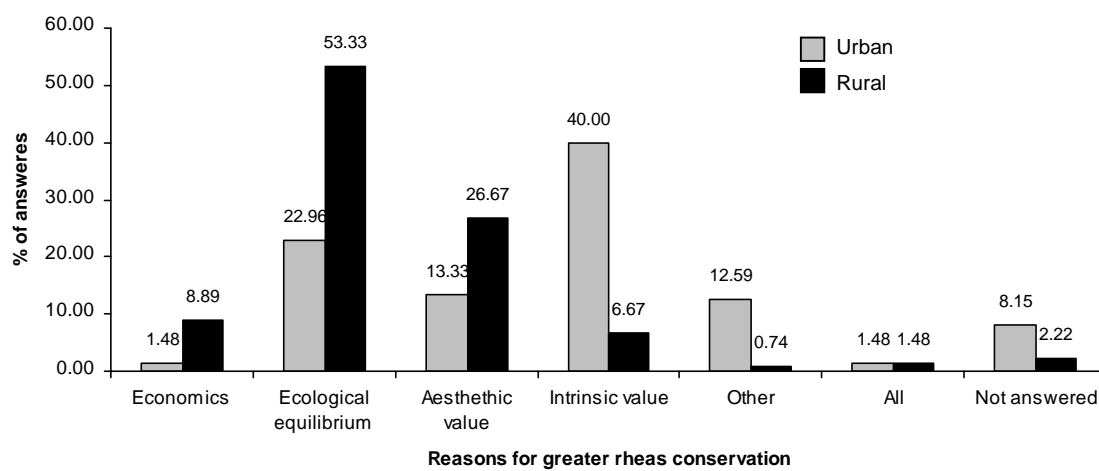
Capítulo 4

Failure of captive-born greater rheas (*Rhea americana*, Rheidae, Aves) to discriminate between predator and non-predator models

Artigo a ser submetido para: **Behavioural Processes.**

Failure of captive-born greater rheas (*Rhea americana,* Rheidae, Aves) to discriminate between predator and non-predator models

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Abstract

The capacity to recognize and respond to predators can be lost by captive animals. Habituation to humans provoked by the captive environment can be transferred to other stimuli, such as predators, a situation that conservation biologists wish to avoid. Greater rheas are threatened South American birds, for which there are plans for reintroduction programmes in Brazil. The goal of this study was to evaluate the behavioural responses of greater rheas to different models of predators and non-predators. Seventeen captive-born greater rheas from the Belo Horizonte zoo, Brazil were studied. Three predator and three non-predator models were presented to the birds and their behavioural responses recorded. Predators versus non-predator models and baseline (standard behaviours of the rheas) versus models versus post-models treatments were analysed. Captive-born greater rheas modified their behaviours (but not significantly) in the presence of both predator/non-predator models, showing some degree of antipredator behaviour persistence, but they were not able to discriminate between predator and non-predator models. Habituation to humans was not transferred to other stimuli, such as the predators, showing that this did not prevent antipredator responses. In conclusion, antipredator training should be implemented to develop the expression of this behaviour before reintroductions are attempted.

Key-words: greater rhea, predator model, generalization, habituation, conservation.

1. Introduction

Greater rheas (*Rhea americana*, Rheidae, Aves) are the largest flightless birds of South America; they live on the grasslands and opened *Cerrados* of Brazil, Argentina, Paraguay, Uruguay and in the southern Bolivia (Davies, 2002). In Brazil, the *Cerrado* biome has been over-exploited by humans and more than 50% of its original area is now altered to some degree (Silva, 1995). In these areas, greater rheas' populations are decreasing, making this species threatened with extinction (Del Hoyo et al., 1992; Machado et al., 1998). Conservation measures are, therefore, necessary.

Reintroduction programmes for greater rheas may be a good solution to their *in-situ* conservation problems since they are easily produced in captivity (Brandt and Neto, 1999). However, the negative effects of captivity, such as stress caused by small enclosures, the lack of predatory stimulation/contact and negative animal-visitor interactions need to be avoided (Carlstead and Shepherdson, 2000, Young, 2003).

The capacity of predator recognition can be lost when an animal is born in captivity because animals can fail to the develop recognition skills or lose their skills; it can occur within a few generations or over evolutionary time (McPhee, 2003; Adams et al., 2006; Blumstein et al., 2006). These recognition abilities can be evaluated using predator and non-predator models (Griffin et al., 2001; 2002); if behavioural responses do not differ between model types, then antipredator training session should be considered. Thus, the evaluation of the response of captive breed animals to predators before reintroduction is important to define if the antipredator training sessions are necessary or not.

The major predators of greater rheas in the wild are the jaguar (*Panthera onca*), maned-wolf (*Chrysocyon brachyurus*), domestic and feral dogs (*Canis lupus familiaris*) (these three pradators kills adults and chicks of greater rheas), common tegu (*Tupinambis teguixin*) (it consumes greater rheas' eggs), bush dog (*Speothos venaticus*), and some species of birds of prey (they kill chicks of greater rheas) (Dani 1993). The common antipredator behaviour exhibited by the greater rheas is running in zigzags, using movements of the wings to maintain balance (Codenotti et al. 1995).

Habituation to humans, a characteristic often observed in captive environments, may have detrimental effects on the survival of the animals when reintroduced to the wild, since human-reduced fearfulness can be transferred to natural predators (van Heezik et al. 1999; Coleman et al. 2008). It is important, therefore, to evaluate if the habituated response is being generalized by the animals before reintroduction programmes.

The aims of this study were to evaluate the behavioural responses of captive-born greater rheas to different models of predators and non-predators (including a human model to evaluate tameness), and to evaluate if predator and non-predator models changes the behavioural expression of the birds.

2. Materials and Methods

2.1. Study place, animals, housing and maintenance

The study was conducted at Belo Horizonte Zoo (BH Zoo, Minas Gerais, Brazil - S 19º51'44.8"; W 44º00'40.1") facilities from March to September 2008. Our subjects were seventeen captive-born greater rheas divided into four groups (Table 1). Birds were housed in wire-fenced enclosures of different sizes and with different

levels of human-contact (Table 1). They were fed twice a day with a mixture of ratite ration (Socil®, 1.2 kg) and vegetables (carrot and cabbage; 1.3 Kg). Water was provided *ad libitum* from a water hole.

2.2. Experimental protocol

The study was divided into two experiments: (1) the influence of animal models on the behaviour of greater rheas; and (2) a predator versus non-predator recognition test.

To evaluate the models' influence on the behaviour of the greater rheas, this part of the study was divided into three sequential treatments: baseline, models and post-models. Baseline treatment was composed of the behaviours expressed by the rheas before the exposition to the predators or non-predators models; models treatment was composed of the behaviours expressed by the rheas during the exposure to predator or non-predator models and the post-models treatment was composed of the behaviours expressed by the rheas after the remove of the predator or non-predator models, when the conditions returned to that of the baseline treatment. A one week of interval was left between treatments. Thirty hours of behavioural data were collected in each treatment, totalizing 90 hours of behavioural observation. All data were collected using scan sampling with instantaneous recording of behaviours every minute (Altmann, 1974). All behavioural data collection sessions were conducted between 1500h and 1600h daily (this time period was chosen as pilot observations had shown rheas to be most active at this time of day).

An ethogram for greater rheas was constructed based on scientific literature (Codenotti et al., 1995; Brandt and Neto, 1999) and on 12 hours of *ad libitum* pilot observations (Martin and Bateson, 2007) (Table 2).

Predators consisted of taxidermized-models of a puma (*Puma concolor*), a crab-eating zorro (*Cerdocyon thous*), and a roadside hawk (*Buteo magnirostris*); non-predators consisted of a human (*Homo sapiens;* considered here as a non-predator due to captive contact and management), a plastic chair and a lesser anteater stuffed-model (*Tamandua tetradactyla*). The models were presented randomly to the greater rheas, being that each model was presented five times to the birds; each greater rhea group received only one model per day and they were never exposed to the same model on consecutive days. The models were put inside the enclosure, in the centre of a circle previously delimited on the enclosure's floor. Each model was visible to the rheas for one hour daily (1500h to 1600h), and was presented to the four groups of rheas (Table 1).

To evaluate if the greater rheas distinguish between predator and non-predator models, the behavioural data were divided in two treatments: (1) with predator models; and (2) with non-predator models. Fifteen hours of behavioural data were collected in each treatment, using the same behavioural collection methods described previously, totalizing 30 hours of behavioural observations. To evaluate the rheas' habituation to the predator and non-predator models, we compared the first 10 minutes of exposure to the models to the last 10 minutes of exposure to the models.

2.3. Statistical analysis

Data were tested, using an Anderson-Darling test, to see if they met the requirement for parametric statistics, which they did not, therefore nonparametric statistical tests were used throughout. The behavioural responses of the greater rheas were compared between the predators and non-predators models and between the first and the last 10 minutes of exposure to the models using the Mann-Whitney U-Test. The behavioural responses of the greater rheas to the models in the three treatments (baseline, models and post-models) were tested using Friedman non-parametrical ANOVA test. The Tukey test was used *post-hoc* to compare the behavioural responses between treatments and between groups. For all statistical analyses, the confidence level was 95% (= 0.05) (Zar, 1999).

3. Results

3.1. Models influence on the behavioural expression of greater rheas

Greater rheas modified their behaviours during the presentation of the predator/non-predator models. "Alert" ($p < 0.01$; N = 120; DF = 2) and "pacing" ($p < 0.01$; N = 120, DF = 2) were significantly more expressed during the models treatment; "foraging" ($p < 0.01$; N = 120; DF = 2) and "not visible" were significantly more recorded during baseline; and "other behaviours" were significantly more recorded in the post-models treatment ($p < 0.01$; N = 120; DF = 2). "Walking" ($p = 0.73$; N = 120; DF = 2) and "inactive" ($p = 0.17$; N = 120; DF = 2) showed no differences between the treatments.

The behaviours expressed by the greater rheas of group 1 were significantly modified by the predator and non-predator models. "Walking" was more expressed during treatment 2 (models) than during the baseline (F = 14.52, $p < 0.01$, DF = 2)

and "foraging" was less expressed during the same treatment in relation to baseline (F = 8.87, p < 0.01, DF = 2). "Alert" and "inactive" were less expressed during post-model treatment and more expressed during baseline (F = 7.27, p = 0.03, DF = 2; F = 19.52, p < 0.01, DF = 2), and "pacing" was more expressed during the same treatment in relation to baseline (F = 14.07, p < 0.01, DF = 2).

Greater rheas from group 2 expressed less the behaviours "walking", "foraging" and "other behaviours" in the models treatment than in baseline (F = 21.12, p < 0.01, DF = 2; F = 16.07, p < 0.01, DF = 2; F = 9.27, p < 0.01, DF = 2, respectively), and most expressed "pacing" in this same treatment in relation to baseline (F = 37.80, p < 0.01, DF = 2). Rheas became more visible during the post-models treatment than in baseline (F = 9.87, p < 0.01, DF = 2).

For group 3, the behaviour "foraging" was the only one less expressed during the models treatment in comparison to the post-model treatment (F = 10.22, p < 0.01, DF = 2), with "pacing" being equally expressed during treatments 2 and 3, when compared to baseline rates (F = 21.12, p < 0.01, DF = 2). "Observing" was most expressed during the models treatment (it was not expressed during baseline or post-model treatments; F = 45.00, P < 0.01, DF = 2) and "walking" was most expressed during the post-models treatment than in the baseline (F = 20.85, p < 0.01, DF = 2).

"Alert", "pacing", and "observing" were the behaviours most expressed by the greater rheas of group 4 during the models treatment when compared to baseline and post-models treatments (F = 45.07, p < 0.01, DF = 2; F = 13.27, p < 0.01, DF = 2; F = 14.45, p < 0.01, DF = 2, respectively); "inactive" and "other behaviours", on the other hand, were less expressed during this phase when

compared to baseline and post-models treatments, respectively (IN: F = 8.27, p <

0.02, DF = 2; OTH: F = 33.87, p < 0.01, DF = 2). Greater rheas were more visible

during the post-models treatment and less visible during baseline (F = 23.82, p <

0.01, DF = 2).

Only the behaviours "pecking" and "running" did not differed statistically

between the rhea groups during the experiments (Table 3). All other behaviours

differed significantly in some degree between the groups in the three treatments

(baseline, models and post-models) (Table 3).

3.2. Predators versus non-predators

Greater rheas from all groups did not respond differently to predators or non-

predators models (Alert: U = 1719.0, p = 0.67; Walking: U = 1699.0, p = 0.59;

Foraging: U = 1656.5, p = 0.45; Inactive: U = 1630.5, p = 0.37; Pecking: U =

1770.0, p = 0.87; Observing: U = 1656.5, p = 0.45; Running: U = 1738.0, p = 0.74;

Pacing: U = 1728.0, p = 0.71; Other behaviours: U = 1502.0, p = 0.12; Not visible:

U = 1681.0, p = 0.53; N = 60, DF = 1 for all behaviours), and the behaviours

"foraging" (F = 18.31; p < 0.01; DF = 5; N = 20), "running" (F = 39.76; p < 0.01; DF

= 5; N = 20), and "other behaviours" (F = 19.71; p < 0.01; DF = 5; N = 20) differed

between the models. Rheas forage less during the puma model if compared to the

chair and people models (p < 0.05), run more when exposed to the puma and bush

dog models in relation to the people (p < 0.05), and exhibited more "other

behaviours" when exposed to the hawk, chair and people, if compared to the puma

model (p < 0.05).

For group 1, only the behaviour "preening" differed statistically between treatments, being more expressed when predators were presented (U = 287.5; N = 15; p = 0.02), and for group 4, only "other behaviours" differed statistically between treatments, being most expressed when the non-predators models were presented (U = 183.5; N = 15; p = 0.04). For groups 2 and 3, none of the behaviours differed between treatments.

Behavioural responses of greater rheas differed, meanwhile, between groups for almost all behaviours (Table 4). The most expressed behaviours of all four groups were "pacing" (34.82% of the behaviours with predator models and 32.55% with non-predator models), "alert" (21.86% with predator models and 18.24% with non-predator models) and "walking" (14.91% with predator models and 15.57% with non-predator models).

"Pacing", in the predator model treatment, was expressed more by rheas of group 2, followed by the rheas of group 4; greater rheas of the group 3 expressed this behaviour less followed by the rheas of group 1; the differences between the groups 1 and 2 with groups 3 and 4 were statistically significant (Table 4). Greater rheas' responses for "pacing" with non-predator models showed the same pattern (Table 4).

"Alert" was more expressed by greater rheas of group 4 followed by group 3, and less expressed by rheas of group 1 in the predator models treatment. The differences between these groups were statistically significant (Table 4). The results for the non-predator models treatment were similar to the predator models, but there was an inversion in the groups that more expressed this behaviour; group 3 expressed "alert" the most and was followed by group 4 (Table 4).

The results found for the behaviour "walking" were similar for both treatments, with greater rheas of the group 4 expressing more this behaviour than all the other groups; group 1 was followed by group 4. Rheas of the group 2 expressed the least this behaviour and the differences between these three groups were statistically significant (Table 4).

The responses of the greater rheas to the predator and non-predator models differed statistically across time only for the behaviours "observing" ($W = 15894.0$, $p < 0.01$, $N = 120$, $DF = 1$) and "other behaviours" ($W = 11788.5$, $p < 0.01$, $N = 120$, $DF = 1$), being the former behaviour more expressed during the 10 first minutes of test and the latter behaviour more expressed during the 10 last minutes of test. The behaviours "alert", "walking", "foraging", "inactive", "running" and "not visible" diminished from the beginning to the end of the test, but not significantly ($W = 14849.5$, $p = 0.47$; $W = 14498.0$, $p = 0.94$; $W = 13938.0$, $p = 0.29$; $W = 14730.0$, $p = 0.61$; $W = 6840.0$, $p = 0.50$; $W = 14719.5$, $p = 0.44$, respectively; $N = 120$, $DF = 1$ for all behaviours). "Pacing" did not varied across time ($W = 14595.5$, $p = 0.80$, $N = 120$, $DF = 1$).

4. Discussion

Although greater rheas responded to the predator/non-predators models during the experiments, they did not distinguish between them, showing the same behavioural responses to both types of models. These results are similar to that found by Johnsson et al. (1996) and Alvarez and Nicieza (2003) when studying brown trout (*Salmo trutta*), and Johnsson et al. (2001) when studying Atlantic salmon (*Salmo salar*). In all of these studies, authors suggested that domestication diminished the antipredator responses of the fishes, but how exactly domestication modifies

antipredator behaviours was only speculative. The absence of predation events or predator encounters or the counterselection of avoidance reactions mediated by selection on correlated traits were suggested by Alvarez and Nicieza (2003) as possible reasons for this. Domestication could be affecting the greater rheas' behavioural responses performed in this study.

Greater rheas of all groups responded similarly to the models: they became alert, began to walk and pace, and diminished the time allocated to foraging and eating activities, although some of the groups showed stronger responses than others. These responses suggest that the presence of the models was uncomfortable to the birds, including the non-predator models. Encounters with predators in nature can be classified as acute stressful events (Teixeira et al., 2007), and the animals should respond properly to predators upon their first encounter; if they do not display the correct repertoire of antipredator behaviours, they could be killed by the predators (Griffin et al., 2000).

Animals should discriminate, through perception systems, predators and non-predators or prey species (Griffin et al., 2002); this would prevent the loss of energy and time as animals do not need to respond to species, which are not harmful or dangerous to them (Helfman, 1989; Ferrari, 2009). Some evidence exists that animals use frontally-placed eyes, apparent size, speed and body shape to differentiate predators from non-predator animals (Curio, 1993; Coss and Goldthwaite, 1995; Blumstein et al., 2000; Griffin and Evans, 2003; Stankowich and Blumstein, 2005). The predator models used in this study varied in shape and size, but all had front-placed eyes; of the non-predators models, only the humans had front-placed eyes; shape and size also differed among them. Thus, greater

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rheas showed fear responses to the novel stimuli presented, but the results did not support recognition of predators by the birds.

In nature, reintroduced rheas that respond in a neophobic manner to all animals that they encounter would probably die soon after their release, since they could suffer from long-term or chronic stress (Moberg and Mench, 2000; Teixeira et al., 2007). Cavigelli and McClintock (2003) showed that neophobic rats were 60% more likely to die at any point in time than neophilic ones and this was related to higher levels of blood glucocorticoids. Antipredator and discriminate learning training sessions are, thus, important to be applied to birds destined for reintroduction, especially if linked to direct measurements of stress, such as behaviour tests (e.g. open-field) or physiological tests (blood cortisol levels).

The groups of greater rheas differed in the strength of their responses to the models, with groups 1, 2 and 4 responding stronger to the models than group 3. Groups 1, 2 and 4 showed more alert and antipredator behaviours, such as "alert", "pacing", and "running". Group 3 also showed stress behaviours such as "pacing", but in general, they were more relaxed than greater rheas of groups 1, 2 and 4.

Group 4 was formed by rheas habituated to humans (this was the group exposed to the zoo visitors) (Azevedo; unpublished data), and group 3 was formed by rheas that received the least amount of human contact (being restricted to the keepers of the zoo). Groups 1 and 2 received an intermediate level of contact with humans: zoo keepers and staff being their main visitors (rheas of groups 1, 2 and 3 were not exposed to zoo visitors). It has been proposed that human habituated fauna could transfer their habituation from one stimulus to another [i.e., generalization; humans to other animals (Jones and Waddington, 1992; van

Heezik et al., 1999)]. The results found in this research do not support the habituation transference hypotheses since the rhea groups 1, 2 and 4 increased their wariness to the models despite being the most habituated birds. These results were similar to those found by Coleman et al. (2008) when studying Gunther's dik-diks (*Madoqua guentheri,* Bovidae). The different composition of the groups (number of birds and sex ratio) was not an important factor in determining greater rheas' behavioural responses to models since all birds were adults.

It is important to remember that there are advantages of using stuffed models in the predator recognition tests (no possibility of attack or disease transmitting and no ethical or logistical problems – Griffin et al. 2000), but these advantages can be overcome by the fact that the animals could rapidly habituate to them. Live models have the advantages of natural movements and sounds (Shalter, 1984). In this study, greater rheas showed signs of habituation to the models since the majority of behaviours diminished between the first and the last 10 minutes of exposure to the models, especially the behaviours "observing" and "running".

In conclusion, captive-born greater rheas modified their behaviours appropriately in the presence of the predator and non-predator models, showing some degree of antipredator behaviour persistence, but they were not able to discriminate between predator and non-predator models. Habituation to humans was not transferred to other stimuli, such as the predators, showing that this did not inhibit antipredator responses. Antipredator training should be implemented to develop the rheas' antipredator and discriminating skills before reintroduction.

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Table 1: Description of the greater rhea groups at BH Zoo and their enclosures.

Group number	Enclosure number	Number of birds	Sex ratio	Human contact	Enclosure size (m^2)
1	ASE 009	4	1 ; 3	Medium	288
2	ASE 010	3	1 ; 2	Medium	252
3	ASE 015	3	3	Low	386
4	Birds' square	7	3 ; 4	High	1021

ASE = *Aves setor extra* (translation: Bird's Extra Sector); Human contact: low = only zoo keepers; medium: zoo keepers and zoo staff; high = zoo keepers, zoo staff and zoo visitors (the scale of human contact is qualitative and subjective).

Table 2: Ethogram of the greater rheas (*Rhea americana*, Rheidae, Aves) at BH Zoo.

Behaviour	Description
Alert	*Rheas stood still or walked with its neck up high.*
Observing	*Rheas observes the models but not with an alert posture.*
Walking	*When rheas walked through the enclosure.*
Running	*When rheas run straight or in zigzags inside the enclosure.*
Pecking	*When rheas peck objects such as the fence or the models.*
Inactive	*When rheas stood still, sitted or when they were sleeping.*
Foraging	*When rheas walked pecking and ingesting items from the ground.*
Preening	*When rheas preen their feathers with their beaks.*
Pacing	*When rheas walked from one side of the enclosure to the other, repeatedly.*
Not visible	*When greater rheas were out of sight.*
Other behaviours	*Behaviours not present in the above list.*

Table 3: Comparison between the behaviours exhibited by the greater rhea groups of BH Zoo during the baseline ($_b$), models ($_m$) and post-models ($_{pm}$) treatments (mean ± standard error; DF = 2; N = 30; = 0.05; Behav. = behaviours; F = Friedman).

Behav.	Group 1	Group 2	Group 3	Group 4	F	p-value
AL$_b$	22.23 ± 1.80^{ab}	49.17 ± 3.88^{acd}	68.63 ± 3.20^{bc}	57.33 ± 3.25^{d}	57.99	< 0.01*
AL$_m$	22.13 ± 1.92^{ab}	43.17 ± 3.36^{acd}	68.57 ± 4.65^{bc}	74.40 ± 10.40^{d}	52.39	< 0.01*
AL$_{pm}$	17.33 ± 1.47^{abc}	38.27 ± 2.66^{ad}	79.50 ± 4.37^{bde}	15.77 ± 1.19^{ce}	67.23	< 0.01*
WAL$_b$	43.60 ± 4.39^{ab}	53.45 ± 7.59^{cd}	18.63 ± 1.77^{ace}	79.83 ± 4.91^{bde}	51.37	< 0.01*
WAL$_m$	55.57 ± 4.12^{abc}	4.37 ± 2.72^{a}	32.13 ± 3.29^{bd}	66.23 ± 4.79^{cd}	52.05	< 0.01*
WAL$_{pm}$	29.83 ± 3.18^{a}	17.13 ± 3.49^{b}	35.03 ± 1.82^{c}	74.73 ± 4.04^{abc}	53.92	< 0.01*
FOR$_b$	21.73 ± 2.46^{ab}	6.38 ± 1.29^{acd}	6.27 ± 1.14^{ce}	51.60 ± 5.08^{bde}	67.09	< 0.01*
FOR$_m$	11.23 ± 1.39^{ab}	1.10 ± 0.50^{acd}	3.93 ± 1.66^{ce}	53.10 ± 5.85^{bde}	59.81	< 0.01*
FOR$_{pm}$	16.77 ± 2.35^{ab}	3.00 ± 0.66^{ac}	7.43 ± 1.50^{d}	47.77 ± 5.88^{bcd}	54.51	< 0.01*
IN$_b$	30.07 ± 3.86^{ab}	28.34 ± 4.34^{cd}	0.37 ± 0.37^{ace}	84.37 ± 5.78^{bde}	77.15	< 0.01*
IN$_m$	22.70 ± 3.49^{a}	32.40 ± 4.16^{b}	0.00 ± 0.00^{abc}	49.87 ± 6.41^{c}	59.33	< 0.01*
IN$_{pm}$	10.57 ± 1.73^{abc}	33.00 ± 4.30^{ade}	0.20 ± 0.17^{bdf}	79.03 ± 6.19^{cef}	83.63	< 0.01*
PAC$_b$	28.83 ± 6.56	51.00 ± 8.62^{ab}	65.07 ± 5.79^{a}	94.70 ± 11.40^{b}	21.19	< 0.01*
PAC$_m$	45.13 ± 7.06^{ab}	143.67 ± 6.47^{ac}	35.83 ± 6.91^{bd}	125.20 ± 12.10^{cd}	49.77	< 0.01*
PAC$_{pm}$	61.70 ± 6.97^{a}	66.63 ± 6.06	35.83 ± 5.35^{ab}	70.50 ± 10.60^{d}	13.05	< 0.01*
OBS$_m$	0.07 ± 0.07^{a}	0.05 ± 0.03^{bc}	24.90 ± 5.37^{abd}	3.57 ± 1.63^{cd}	52.47	< 0.01*
PEC$_m$	0.00 ± 0.00	0.03 ± 0.03	0.00 ± 0.00	0.00 ± 0.00	0.06	1.00
RUN$_m$	0.03 ± 0.03	0.00 ± 0.00	0.60 ± 0.60	2.23 ± 1.14	0.72	0.87
OTH$_b$	35.07 ± 3.70^{a}	46.34 ± 5.87	24.03 ± 3.59^{ab}	47.70 ± 3.70^{b}	19.69	< 0.01*
OTH$_m$	25.60 ± 2.93^{a}	19.57 ± 3.00	16.97 ± 4.24^{b}	47.33 ± 6.99^{ab}	22.81	< 0.01*
OTH$_{pm}$	46.23 ± 4.08^{a}	24.93 ± 3.92^{b}	25.00 ± 4.31^{c}	133.53 ± 6.02^{abc}	54.73	< 0.01*
NV$_b$	1.10 ± 0.71^{ab}	3.48 ± 1.09^{c}	0.00 ± 0.00^{ad}	11.30 ± 1.06^{bcd}	48.05	< 0.01*

NV_m	0.53 ± 0.31^a	0.53 ± 0.32^b	0.13 ± 0.13^c	5.27 ± 0.74^{abc}	32.41	< 0.01*
NV_{pm}	0.50 ± 0.21^a	0.13 ± 0.10^b	0.00 ± 0.00^c	4.03 ± 0.65^{abc}	38.03	< 0.01*

* = behaviours that differed statistically between treatments; groups marked with same superscript letters differed statistically according to Tukey's *post-hoc* test (P <0.05). (AL = alert; WAL = walking; FOR = foraging; IN = inactive; PAC = pacing; OBS = observing; PEC = pecking; RUN = running; OTH = other behaviours; NV = not visible).

Table 4: Comparison between the behaviours exhibited by the greater rhea groups from the BH Zoo when presented with predator and non-predator taxidermized-models (mean ± standard error of behavioural expression - absolute numbers; DF = 3; N = 15;   = 0.05; Behav. = behaviours; Treat. = treatment; F = Friedman statistic).

Behav.	Treat.	Group 1	Group 2	Group 3	Group 4	F	p-value
Alert	Pred.	21.87 ± 2.45^{ab}	44.33 ± 5.30	69.60 ± 6.81^a	90.90 ± 18.40^b	28.84	< 0.01*
	Non-pred.	22.40 ± 3.03^{ab}	42.00 ± 4.30	67.53 ± 6.57^a	57.80 ± 8.53^b	24.42	< 0.01*
Walking	Pred.	53.67 ± 6.40^a	2.87 ± 2.66^{abc}	35.53 ± 4.68^b	62.60 ± 8.16^c	24.54	< 0.01*
	Non-pred.	57.47 ± 5.37^a	5.87 ± 4.82^{ab}	28.73 ± 4.62^c	69.87 ± 5.18^{bc}	28.60	< 0.01*
Foraging	Pred.	10.40 ± 2.08^a	0.47 ± 0.34^{ab}	4.60 ± 2.97^c	45.00 ± 8.43^{bc}	30.50	< 0.01*
	Non-pred.	12.07 ± 1.88	1.73 ± 0.93^a	3.27 ± 1.60^b	61.20 ± 7.85^{ab}	29.58	< 0.01*
Inactive	Pred.	19.87 ± 4.26^b	31.40 ± 5.26^a	0.00 ± 0.00^{abc}	43.60 ± 9.92^c	29.16	< 0.01*
	Non-pred.	25.53 ± 5.58^b	33.40 ± 6.62^a	0.00 ± 0.00^{abc}	56.13 ± 8.13^c	30.70	< 0.01*
Observing	Pred.	0.13 ± 0.13^b	0.27 ± 0.15^a	0.00 ± 0.00^{abc}	6.07 ± 3.17^c	26.76	< 0.01*
	Non-pred.	0.00 ± 0.00^b	0.80 ± 0.62^a	0.13 ± 0.13^{ab}	1.07 ± 0.36	25.74	< 0.01*
Pecking	Pred.	0.00 ± 0.00	0.07 ± 0.07	0.00 ± 0.00	0.00 ± 0.00	0.12	0.99
	Non-pred.	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	-	-
Running	Pred.	0.07 ±	0.00 ± 0.00	0.00 ± 0.00	4.47 ±	1.92	0.59

		0.07			2.16		
	Non-pred.	0.00 ± 0.00	0.00 ± 0.00	1.20 ± 1.20	0.00 ± 0.00	0.16	0.98
Pacing	Pred.	49.67 ± 9.63[ac]	145.60 ± 5.98[ab]	32.27 ± 9.37[bd]	133.50 ± 17.00[cd]	31.62	< 0.01*
	Non-pred.	40.60 ± 10.50[a]	141.70 ± 11.70[ab]	39.40 ± 10.40[b]	116.90 ± 17.70	19.72	< 0.01*
Other behaviours	Pred.	27.13 ± 3.65	18.27 ± 4.41	10.27 ± 3.88[a]	35.20 ± 5.68[a]	12.14	< 0.01*
	Non-pred.	24.07 ± 4.67	20.87 ± 4.20[a]	23.67 ± 7.28[b]	59.50 ± 12.20[ab]	12.22	< 0.01*
Not visible	Pred.	0.27 ± 0.21[a]	0.73 ± 0.56	0.00 ± 0.00[b]	5.60 ± 1.32[ab]	12.42	< 0.01*
	Non-pred.	0.80 ± 0.60[b]	0.33 ± 0.33[a]	0.27 ± 0.27[c]	4.93 ± 0.73[abc]	20.84	< 0.01*

* = behaviours that differed statistically between treatments; groups marked with same superscript letters differed statistically according to Tukey's *post-hoc* test (P<0.05).

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Capítulo 5

Feeding enrichment decreases abnormal behaviors in greater rheas (*Rhea americana*, Rheidae, Aves): conservation implications

Artigo a ser submetido para: **Zoo Biology.**

Feeding enrichment decreases abnormal behaviors in greater rheas (*Rhea americana*, Rheidae, Aves): conservation implications

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Running title: Feeding enrichment decreases abnormal behaviors in rheas.

Abstract

Animals kept by zoos may express abnormal behaviors, which are indicators of low welfare status. To maintain the whole behavioral repertoire of animals is a challenge for zoos, but it is important for the use of the animals in conservation programs, such as reintroduction. Environmental enrichment is a technique of animal management (husbandry), which may stimulate the expression of normal behaviors. The aim of this study was to evaluate the effects of a simple feeding enrichment method in the reduction of the expression of abnormal behavior by captive-born greater rheas at Belo Horizonte Zoo, southeastern Brazil. As enrichment, we offered to a group of seven birds, scattered vegetables from April to September 2009, and the behavior of the birds was recorded using scan sampling with instantaneous recording of behavior every one minute for one hour daily (90 hours of observation). The study was divided into three treatments: baseline, enrichment and post-enrichment. The abnormal behaviors "pacing", "eating faeces", "drinking water" and "escaping behavior" diminished during the enrichment treatment, but only the first three behaviors differed significantly between the treatments; both "walking" and "foraging" increased during the enrichment treatment. These results showed that the implementation of simple feeding enrichment can stimulate greater rheas to exhibit more natural behaviors and thereby increase their welfare, making captive bred animals more suitable for reintroduction programs.

Introduction

The maintenance of animals in captivity may lead to the expression of quantitative and qualitative abnormal behaviors; the lack of sufficient and adequate stimuli provided by this unnatural environment is thought to be the main reason for this (Young, 2003). The expression of abnormal behaviors is considered a good measure of welfare (Broom and Johnston, 1993; Moberg and Mench, 2000; Hötzel et al., 2005; Bracke and Hopster, 2006): the more the animal expresses abnormal behaviors, the worse the welfare it experiences (Olsson and Dahlborn, 2002). Self-mutilation, pacing and 'tongue playing' are examples of abnormal behaviors (Young, 2003; Mason and Rushen, 2006).

According to Shepherdson (1994) and Young (2003), environmental enrichment is a concept, which describes how the environments of captive animals can be changed for the benefit of their inhabitants by the implementation of enrichment devices that simulate natural situations (i.e., increasing behavioral opportunities for animals). In most cases, environmental enrichment also increases the welfare of the animals (Shepherdson et al., 1998; Young, 2003; Adams, 2007).

Environmental enrichment is used in the prevention and diminution of abnormal behaviors, and it is becoming a tool for species conservation, since it allows the maintenance of the natural behavioral repertoire of a species and may reduce stress caused by the captive environment (Shepherdson et al., 1998). A recent review of translocation and reintroduction programs noted the importance of considering and eliminating distress for animals that will participate in such programs (Teixeira et al., 2007). Increased adaptability to changing environments caused by the environmental enrichment contributes positively to reintroduction programs, since animals with such experiences have greater chances of

survival after released into the wild (Miller et al., 1998; Castro et al., 1998; Hare et al., 2003; Salvanes et al., 2007).

Greater rheas (*Rhea americana*, Rheidae, Aves) are native of the *Cerrados* and grasslands of South America (Sick, 1997; Davies, 2002). It is the largest flightless bird of Brazil and it is threatened with extinction in most of its distribution (Codenotti and Alvarez, 2000; Herrera et al., 2004; Di Giacomo and Krapovickas, 2005; Giordano et al., 2008). Habitat loss, crop burning, feral dog predation and egg losses caused by agricultural machinery are the main factors responsible for the decline of greater rhea populations (Dani, 1993; Sick, 1997; Machado et al., 1998; Navarro and Martella, 1998; Fernandéz and Reboreda, 2000; Di Giacomo and Krapovickas, 2005).

Greater rheas lives in familiar groups normally varying from three to eight individuals, but groups with more than 90 birds have been observed (Davies 2002). Their main activities are walking and feeding; they walk great distances (1.8 km in average per day; Bellis et al. 2004), eating vegetables and invertebrates, although small vertebrates can also be consumed (Sick 1997). They collected their food mainly in the ground, but food items can also be collected from shrubs and trees (Del Hoyo et al. 1992).

Fights are common, especially between males during the formation of harems in the breeding season (Davies 2002), which occurs from July to September (although it can vary throughout their distribution). Their mating systems combines harem polygyny and sequential polyandry (Bruning 1974); females lay 20-30 eggs communally in a nest built by the males on the ground (Fernández and Reboreda 1998). Egg incubation takes 40 days on average, and parental care is provided exclusively by the males (Sick 1997). Wing displays (front of the body lowered slightly, neck in a low U-curve and wings extended laterally)

and head-bobbing displays (male bobes its neck and head up and down vigorously) are performed by males to females during courtship (Davies 2002).

Vigilance behaviors tends to be performed in areas with tall vegetation and in small groups of birds (Martella et al. 1995), and the main defensive behavior that greater rheas use is a zigzag run controlled by the wings; they can achieve a speed of 60 km/h during this behavior (Sick 1997).

A reintroduction program is planned to be undertaken by the Belo Horizonte Zoo, Brazil (BH-Zoo). During an on-going study (Azevedo et al; unpublished data), it were detected some abnormal behaviors [pacing, eating faeces, drinking water (quantitatively abnormal), escaping behavior (considered abnormal since the birds expressed it with no apparent reason and in a stereotyped way; see Table 1)] being expressed by captive-born greater rheas, and consequentially environmental enrichment activities were implemented for these birds. This study intended to evaluate the efficiency of the feeding enrichment in the diminution of abnormal behaviors of greater rheas.

Methods

Study area, housing and maintenance

The study was conducted in the "Bird Square" of the Belo Horizonte Zoo (BH-Zoo), Minas Gerais, southeastern Brazil (S 19º51'44.8"; W 44º00'40.1"). Seven greater rheas (three males and four females), all bred at BH Zoo, were studied. Birds varied from four to eight years old.

Birds were housed in a wire-fenced enclosure 28m in length and 16m wide (the same enclosure and situation where the abnormal behaviors were previously recorded). They were fed twice a day with a mixture of ratite ration (Socil®, 1.2 kg) and vegetables (carrot and cabbage; 1.3 Kg) at 0900 and 1400 hours. Birds were allowed to eat fruits and flowers

from the trees planted inside the enclosure, such as guavas (*Psidium guajava*, Myrtacea) and licuri fruit (*Syagrus coronata*, Arecaceae). Water was provided *ad-libitum* in a water hole. The enclosure was cleaned three times a week.

Experimental protocol

The study was divided into three sequential treatments: baseline, enrichment and post-enrichment. Baseline treatment was the standard husbandry regime of the rheas (i.e., the behaviors expressed by the rheas before the implementation of the enrichments); enrichment treatment consisted of offering scattered fruit and vegetables for the rheas (i.e., the behaviors expressed by the rheas during the enrichment); and the post-enrichment treatment was a return to baseline conditions (i.e., behaviors expressed by the rheas after the remove of the enrichment). Enrichment used consisted of chopped fruit and vegetables (apple, banana, strawberry, papaya, pineapple, pear, guava, cucumber, tomato, and beetroot; 2.5 kg in total daily) scattered through the enclosure and/or hung on the trees inside the enclosure. The enrichment was offered to the birds five minutes before data collection began, once per day at 1455h (rheas consumed all the enrichment food offered before observations began on the following day). The enrichment foods were offered concomitantly with the normal diet of the birds to test its power in the elicitation of foraging activities.

Thirty hours of behavioral data were collected in each treatment, totalizing 90 hours of behavioral observations. All data were collected using scan sampling with instantaneous recording of behaviors every minute (Altmann, 1974). All behavioral data collection sessions were conducted between 1500h and 1600h daily, from April to September 2009. This time period was chosen as pilot observations had shown rheas to be active at this time of day. Each treatment lasted two months.

An ethogram for greater rheas (Table 1) was constructed based on scientific literature (Codenotti et al., 1995; Brandt and Neto, 1999) and on 12 hours of *ad libitum* pilot observations (Martin and Bateson, 2007).

Statistical analysis

We used an Anderson-Darling test to determine if our data met the requirements for parametric statistics, which they did not, therefore non-parametric tests were used throughout. Behaviors were quantified (number of behavioral recordings were summed and means and percentages calculated) and statistically analyzed using Friedman non-parametrical ANOVA test. The Tukey test was used *post-hoc* to compare the behavioral responses between treatments. Environmental factors (weather, husbandry, visitors, etc.) were registered and showed no significant variations between the treatments, which permit the data analysis without the necessity of randomization of the treatment days. For all statistical analyses, the confidence level was 95% (= 0.05) (Zar, 1999).

Results

The expression of the abnormal behaviors "pacing", "eating faeces" and "drinking water" were significantly reduced in the enrichment treatment (Table 2). "Pacing" was more expressed in the baseline treatment (13.92%), decreased in the enrichment (8.11%) and increased in the post-enrichment treatment (8.82%). The expression of pacing differed statistically between the baseline and the post-enrichment treatments (Table 2). "Eating faeces" was most expressed in the baseline (7.09%) and decreased abruptly in the enrichment treatment (0.89%); a slight increase in the expression of this behavior during the post-enrichment treatment occurred (2.81%); whereas, baseline differed from the enrichment and post-enrichment treatments statistically (Table 2). "Drinking water" was most expressed during the baseline treatment (11.32%) and almost disappeared during the

enrichment treatment (0.25%), increasing again during the post-enrichment treatment (1.81%) (Table 2). "Escaping behavior" was not statistically different between the three treatments although it decreased from baseline (0.04%) to post-enrichment treatments (0.02% in the enrichment treatment and 0.01% during post-enrichment treatment).

The category "not visible" and the behaviors "alert", "walking", "foraging", "eating", "vocalization", "courting", "incubating eggs", and "other behaviors" also differed statistically between the treatments (Table 2). "Alert" was most expressed during the baseline treatment (5.97%), less during the enrichment treatment (5.38%) and reached their lowest expression during the post-enrichment treatment (4.00%) (Table 2). "Walking" was most expressed during the enrichment treatment (25.03%) and less expressed during the post-enrichment treatment (14.63%) (Table 2); "eating" was more exhibited during the baseline treatment (7.48%), decreased during the enrichment treatment (4.00%) and increased during the post-enrichment treatment (5.23%) (Table 2).

"Foraging" was most expressed during the enrichment treatment (23.91%) than during baseline (19.39%) or post-enrichment (15.31%) treatments (Table 2). "Other behaviors" were more expressed during the enrichment treatment (0.86%) and post-enrichment, and rheas were more visible to the observers during the post-enrichment phase (0.07%) (Table 2).

Reproductive behaviors, such as "vocalization", "courting" and "incubating eggs" also differed statistically between the treatments. "Vocalization" was more expressed during the baseline (0.59%) and decreased during the subsequent treatments until it ceased to be expressed in the post-enrichment treatment (0.27% during enrichment treatment) (Table 2). "Courting" increased in expression from baseline treatment (1.14%) to the enrichment treatment (2.69%), when it reached its peak; "incubating eggs" started to be exhibited

during the enrichment phase (eggs were laid on the tenth day of the enrichment phase; 10.25%) and increased in post-enrichment treatment (30.10%) (Table 2).

Discussion

The abnormal behaviors expressed by the greater rheas ("pacing", "drinking water", "escaping behavior" and "eating faeces") were reduced by the introduction of a cheap and simple feeding enrichment method. An animal that exhibits more of its natural behavioral repertoire is more likely to survive in a reintroduction program, for instance, than an animal expressing abnormal behaviors (Tudge, 1992; Seidensticker and Doherty, 1996; Vickery and Mason, 2003). Thus, environmental enrichment was shown to have potential as a management tool for captive greater rheas destined for reintroduction.

Greater rheas have never been observed ingesting faeces in the wild, but in captivity this behavior is frequently observed (Dani, 1993; Silva, 2001; Hosken and Silveira, 2003); this behavior was considered qualitatively abnormal due to its absence in wild conspecifics and the high proportion of time that it consumed in the baseline time-budget of the birds (7.2%). In the baseline treatment, greater rheas were fed with a mixture of vegetables and ratite ration; they rapidly ate at the feeder and began to search for faeces to eat; once they found a scat they spent a long time ingesting it. During the enrichment treatment, when vegetables were scattered through the enclosure, greater rheas avoided the feeder and spent large amounts of time foraging and walking from one food patch to another. The "eating faeces" behavior almost disappeared in the enrichment treatment, and remained low in the post-enrichment treatment. Many studies have found similar results, with the enriched animals preferring to search for food than to eat the same food at a feeder (Reinhardt, 1994; Azevedo and Faggioli, 2001; Jones and Pillay, 2004; Maloney and Meiers, 2006; Rowntree,

2007), and the carry-over effects of enrichment being detected in the post-enrichment treatment (Marashi et al., 2003).

"Pacing" was also influenced by the enrichment, since it diminished during the enrichment phase. "Pacing" is one of the most expressed abnormal behaviors by captive animals (Carlstead and Seidensticker, 1991; Shepherdson et al, 1998; Mallapur and Choudhury, 2003; Young, 2003; Mason and Rushen, 2006). It is linked to low levels of animal welfare (Mason, 1991; Jordan, 2005; Owen et al., 2005) being expressed by under- or over-stimulated animals (Pitsko, 1993; Mallapur et al., 2002; Dawkins, 2008). Studies of wild greater rheas' time budgets show that these birds spend almost the entire day walking and foraging (Reboreda and Fernandéz, 1997; Bellis et al., 2006; Carro and Fernandéz, 2008). In captivity, the expression of such behaviors is obviously limited by the size of the enclosures, which is often much smaller than the home range size of the species (Shepherdson et al., 1998; Young, 2003). However, enrichment devices can increase the psychological space available to captive animals (Chamove, 1989). The scattered vegetables in this study stimulated the birds to walk and forage for long periods of time instead of pacing, reproducing in part their natural behavior. This, probably, increased welfare and reduced stress, along with ameliorating the fitness of the birds (Bortolotti et al., 2002). Studies with felids (Moreira et al. 2007), monkeys (Boinski et al. 1999; Doyle et al. 2008), rodents (Iwata et al. 2007), rabbits (Verga et al. 2007), dogs (Lefebvre et al. 2009) and pigs (Jong et al. 2000) demonstrated that the use of environmental enrichment decrease stress and increase the welfare of the animals [for a review of the behavioral and physiological evidence see Young (2003), Tables 3.1 and 3.2]. Studies of stress hormones conducted simultaneously with behavioral research could be used to validate this hypothesis.

An uncommon behavior, named "escaping behavior" (ESCB) was observed mostly in the baseline treatment, although it was expressed by the rheas in all treatments. This behavior was characterized by the rheas jumping in the air and then running in zigzags as soon as they touched the ground, with no apparent reason (just like they are trying to escape from something). This behavior was not previously recorded in literature, and it was considered here as qualitatively abnormal. The distribution of environmental enrichment also diminished the occurrence of this behavior, but not significantly, probably, due to its low expression. Sometimes this behavior seemed to be expressed as a form of play, as if the rheas had "too much energy to spend" and used the escaping runs to waste it. Play is a behavior often considered good and beneficial by animal welfare researchers (Fagen and Fagen, 1990; Dybkjaer, 1992; Goodwin and Hughes, 2005; Oliveira et al., 2010), suggesting that animals displaying such behavior experience a good level of welfare (Young, 2003; Odendaal, 2005; Bexell et al., 2007). Play is often expressed by young vertebrates (Oliveira et al., 2010), but all the rheas studied were adults. Energetic and physiological studies need to be conducted to evaluate if this behavior really means good welfare; if this hypothesis is confirmed, then this behavior could be interpreted as a 'luxury' for the birds since it stopped being performed when rheas had the opportunity to express foraging activities (Young, 2003).

"Drinking water" strongly diminished during the enrichment phase. This behavior could be considered quantitatively abnormal in this study since the birds spent long periods at the water hole during baseline (13.06% of the time budget). Excessive water intake, called polydipsia, is a known abnormal behavior in a number of animal species, such as rats (Mumby and Beck, 1988), dogs (van Vonderen et al., 1999), sparrows (Kuenzel and Helms, 1970) and hens (Ralph, 1960). Polydipsia is related to physiological problems, for example,

lesions in the supraotic region of hypothalamus (Ralph, 1960), genetic influence on heat tolerance (Obeidah et al., 1977), or due to the lack of environmental stimulation (van Loo et al., 2004). Perhaps, the enrichment items and the breeding season re-directed drinking time to foraging (provided environmental stimulus), and reproductive behaviors since the rheas studied did not show any apparent brain damage or genetic problems (non-studied genetically-related groups of rheas held by BH Zoo did not showed any signs of polydipsia during the study period; genetic studies of the captive greater rheas' populations are planned to be run in the near future and it would helpful in addressing the polydipsia-genetic related problem).

The abnormal behaviors decreased during the enrichment treatment showing that they were not important to the animals; that is, not behaviorally resilient (Young, 2003). Behavioral resilience; that is, how an animal responds to having less time to perform its daily time budget, is a technique used for the assessment of animal welfare (Young, 2003). It predicts that the animal will maintain only the important behaviors, which ensure its homoeostasis and, consequently, its survivorship and welfare. The reduction in the expression of abnormal behaviors by greater rheas suggests that they were expressed because of a lack of stimulation (i.e., frustration caused by not being able to express consummatory behaviors) and that this abnormal behavior did not became emancipated from their original causation; once the proper stimulus was provided to them, those behaviors decreased in frequency. Abnormal behaviors normally occur to fill a behavioral void (Kuhn 1991), and the decrease in its performance showed that the feeding enrichment provided the right stimuli to fill these voids. Physiological and neurological evidence would permit us a better understanding of the role of the environmental enrichment on the amelioration of greater rheas' welfare (Young, 2003).

"Alert", "intimidating" and "fighting" behaviors also diminished during the enrichment treatment; at the same time the reproductive behaviors began to be recorded. Courting was most exhibited in the enrichment phase and this behavior influenced decisively the expression of inactivity. When female greater rheas were "courted" by males, they stood still in front of the males for long periods of time (Codenotti and Alvarez, 2000; Davies, 2002).

"Walking alert" was most performed by two of the three males who delayed their nest building and egg incubation; each male followed females through the entire enclosure "walking alert" to monitor and avoid the other one; fights often occurred when a male approached another male. In the wild male greater rheas compete aggressively for the formation of their harems with other males during breeding season (Raikon, 1969; Del Royo et al., 1992; Codenotti et al., 1995; Sick, 1997; Codenotti and Alvarez, 2001; Fernandez et al., 2003).

Conclusions

Environmental enrichment diminished captive-born greater rheas' abnormal behavior, probably, increasing their welfare. Environmental enrichment should be implemented for this species with the goal of maintaining more their normal behavioral repertoire. For species that are threatened with extinction, as in the case of the greater rheas, the performance of natural behaviors is important for their survivorship when reintroduced to the wild. Never-the-less, to exhibit greater rheas displaying normal and diversified behaviors have more educational and conservation value (Swaisgood, 2007).

Greater rheas expressed more natural behaviors, not in the sense of behavioral diversity but their use of time was more similar to that of wild conspecifics (Reboreda and Fernandez,

1997; Carro and Fernandez, 2008), when the enrichment was present showing that the method functioned well in the provision of stimuli for the birds.

Acknowledgements

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Table 1: Ethogram of the greater rheas (*Rhea americana*, Rheidae, Aves) at BH Zoo.

Behavioral category	Abbreviation	Behavior	Description
1 - Reproduction	V	*Vocalization*	The male gives a deep-toned hissing, rendered "nan-du".
	MAT	*Mating*	A male mounts a female and copulate with her.
	INC	*Incubating eggs*	A male incubate the eggs in the nest.
	COU	*Courting*	A male exhibits the wing or head-bobbing displays.
2 – Vigilance	AL	*Alert*	The rhea extends its neck up in the air and watches the environment.
	WAAL	*Walking alert*	The rhea extends its neck up in the air and watches the environment while walking.
3 – Activity	WAL	*Walking*	The rhea walks through its enclosure.
	RUN	*Running*	The rhea runs in zigzags or straight through its enclosure.
	PEC	*Pecking*	The rhea picks up objects or pecks other individuals.
4 – Inactivity	IN	*Inactive*	The rhea assumes a standing, crouching, sitting or sleeping posture, with no movements.

5 – Nourishment	FOR	*Foraging*	The rhea searches for food on the ground or on the trees, but not on its feeder.
	EAT	*Eating*	The rhea eats food from its feeder.
6 – Maintenance	PRE	*Preening*	The rhea uses its bill to preen its feathers.
	DUS	*Dust bathing*	The rhea lies down and throws dust over its body using its bill.
	DFU	*Defecating/urinating*	The rhea defecates or urinates.
7 – Abnormal behaviors	EF	*Eating faeces*	The rhea eats its own faeces or other greater rheas' faeces.
	PAC	*Pacing*	The rhea walks from one side of the enclosure to the other, using the same route and with no apparent reason.
	ESCB	*Escaping behavior*	The rhea jumps in the air and then runs in zigzags as soon as they touched the ground, with no apparent reason.
	DRI	*Drinking water*	The rhea extends its neck and dips its bill into the water, in the water hole.
8 – Aggression	FIG	*Fighting*	The rhea fights with other bird, pushing the opponent with its chest, kicking and pecking it.

	INT	*Intimidating*	The rhea charges with his head and neck in an S-shaped curve and tries to strike the other rhea with thrusts and bill snapping.
9 – Not visible	NV	*Not visible*	The rhea is not visible to the observer.
10 – Other behaviors	OTH	*Other behaviors*	The rhea exhibits a behavior not previously recorded.

Table 2 – Results for the comparison of the behaviors expressed by the BH-Zoo's greater rheas (*Rhea americana*, Rheidae, Aves) between the three treatments of the study (baseline, enrichment and post-enrichment) (mean ± standard error; DF = 2; N = 30; α = 0.05 in all cases).

Behavior	Baseline	Enrichment	Post-enrichment	Friedman	*p*-value
V	2.50 ± 0.48^{a}	1.03 ± 0.32	0.00 ± 0.00^{a}	17.62	< 0.01*
MAT	0.00 ± 0.00	0.60 ± 0.26	0.00 ± 0.00	1.25	0.53
INC	0.00 ± 0.00^{abc}	38.57 ± 8.04^{abc}	91.80 ± 5.11^{abc}	45.22	< 0.01*
COU	4.83 ± 0.78^{ab}	10.13 ± 2.03^{a}	0.00 ± 0.00^{b}	22.07	< 0.01*
AL	25.40 ± 2.89^{a}	20.23 ± 1.84^{b}	12.20 ± 1.64^{ab}	10.62	< 0.01*
WAAL	0.60 ± 0.20	2.17 ± 0.86	0.33 ± 0.14	5.22	0.07
WAL	80.73 ± 4.81^{a}	94.17 ± 5.28^{b}	44.60 ± 2.89^{ab}	31.67	< 0.01*
RUN	0.57 ± 0.18	0.40 ± 0.12	0.17 ± 0.10	2.15	0.34
PEC	0.63 ± 0.18	0.07 ± 0.05	0.30 ± 0.10	4.55	0.10
IN	47.00 ± 3.96	44.77 ± 4.95	42.27 ± 5.51	1.27	0.53
FOR	82.47 ± 6.32^{a}	89.97 ± 4.04^{b}	46.70 ± 5.18^{ab}	26.52	< 0.01*
EAT	31.80 ± 4.48^{ab}	15.07 ± 2.21^{a}	15.93 ± 4.03^{b}	10.05	< 0.01*
PRE	8.50 ± 1.37	4.83 ± 1.07	5.83 ± 0.89	1.85	0.40
DUS	2.90 ± 0.70	2.20 ± 0.81	1.73 ± 0.45	1.22	0.54
DFU	0.70 ± 0.14	0.63 ± 0.15	0.37 ± 0.13	3.15	0.21
EF	35.97 ± 5.19^{ab}	3.23 ± 0.88^{a}	8.57 ± 2.20^{b}	31.22	< 0.01*

PAC	60.13 ± 6.99^{a}	29.57 ± 4.53	26.90 ± 4.58^{a}	11.27	$< 0.01^{*}$
ESCB	0.17 ± 0.08	0.07 ± 0.05	0.03 ± 0.03	0.35	0.84
DRI	48.17 ± 7.82^{ab}	0.93 ± 0.28^{a}	5.53 ± 1.70^{b}	31.20	$< 0.01^{*}$
FIG	0.37 ± 0.16	0.30 ± 0.20	0.00 ± 0.00	1.02	0.60
INT	0.27 ± 0.11	0.13 ± 0.06	0.03 ± 0.03	1.07	0.59
NV	2.97 ± 0.51^{ab}	2.07 ± 0.54^{a}	0.20 ± 0.09^{b}	23.45	$< 0.01^{*}$
OTH	0.73 ± 0.26^{a}	3.23 ± 0.88^{a}	1.43 ± 0.46	9.02	0.01^{*}

* = results that differed statistically between the treatments. Letters indicates the Tukey Test results; same letters represents treatments that differed between each other. V = vocalization; MAT = mating; INC = incubating eggs; COU = courting; AL = alert; WAAL = walking alert; WAL = walking; RUN = running; PEC = pecking; IN = inactive; FOR = foraging; EAT = eating; PRE = preening; DUS = dust bathing; DFU = defecating/urinating; EF = eating faeces; PAC = pacing; ESCB = escaping behavior; DRI = drinking water; FIG = fighting; INT = intimidation; NV = not visible; OTH = other behaviors.

Capítulo 6

Visitor influence on the behaviour of captive greater rheas (*Rhea americana*, Rheidae Aves)

Artigo a ser submetido para: **Animal Welfare.**

Visitor influence on the behaviour of captive greater rheas (*Rhea americana*, Rheidae Aves)

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Running title: Visitor effect on captive greater rheas.

Abstract

The effect of visitors can be one of the factors responsible for reducing animal welfare (e.g., increasing the expression of abnormal behaviour). The Belo Horizonte Zoo (BH Zoo), southeastern Brazil, had a group of greater rheas that were intended for reintroduction to the wild. Since this group received public visitation, it was important to evaluate the effect of the public on the behaviour and welfare of these birds. Sixty hours of behavioural observations were conducted, being 30-hours with the public in front of the greater rheas' enclosure and 30-hours in the absence of the public. Data were collected using scan sampling with instantaneous recording of behaviour every minute. Public behaviour, visitor density and time spent observing the birds were also collected. The study was conducted from April to December 2009. More than 4,000 people visited the greater rheas' enclosure, and the average time spent in front of the exhibit was 9.86 ($\pm$ 1.49) seconds. The behaviours most expressed by the public were "walking-watching-talking" and "stopped-watching-talking". The behaviours most expressed by the greater rheas in the presence of visitors were "foraging" and "walking"; the most expressed behaviours by the greater rheas without the presence of the public were also "walking" and "foraging". Mann-Whitney's U-test showed that only the behaviours "walking alert", "defecating/urinating" and "other behaviours" differed statistically between the treatments, being "walking alert" most expressed in the presence of the visitors, and "defecating/urinating" and "other behaviours" being more expressed in the absence of visitors. Greater rheas appeared to be habituated to the visitor presence since the majority of their behaviours did not differ in the presence or absence of public in front of their enclosure.

Key-words: captivity, conservation, greater rheas, habituation, public effect, zoo.

Introduction

Public influence on the behaviour of captive animals, such as inducing behavioural changes, has been widely documented (Hosey, 2000; Davey, 2007; Farrand, 2007; Sekar et al., 2008). This influence is known as the "public effect", often results in stress for captive animals and some studies have shown a link between the increase of people in front of exhibits and a greater expression of aggressive behaviour (Hosey, 2000; Sekar et al., 2008). Thus, captive animals intended to be part of conservation programs (e.g., reintroduction) need to receive special attention in relation to their exposure to visitation. It is predicted that animals, which are not exhibited to visitors would experience less stress than animals exposed to visitors (Glatston et al., 1984; Burrel and Altman, 2006; Davey, 2006) and thus, these animals should be preferred for conservation projects such as reintroduction (McDougall et al., 2006). However, many zoos do not have off-exhibit enclosures.

On the other hand, for captive management, visitor and caretaker habituated animals would suffer less with stress than non-habituated animals (Jezierski et al., 1999). Some authors even suggested that the daily contact with visitors could function as environmental enrichment, providing sources of novelty in the environment and, thus, increasing the welfare of the captive animals (Hosey, 2000; Margulis et al., 2003). Consequently, zoo visitation needs to be evaluated carefully depending on the species in question and on the goals of the zoo; thus, conservation versus animal welfare is a conflict faced by zoo managers.

Greater rheas (*Rhea americana*, Rheidae, Aves) are the largest South American bird (Del Hoyo et al., 1992; Sick, 1997); they are flightless omnivorous birds that occur from Brazil to southern Argentina, habiting grasslands and *Cerrados* also in Paraguay, Uruguay and Bolivia (Davies, 2002). Greater rheas' populations are diminishing throughout their

distribution due to habitat loss, hunting, crop burnings and egg losses caused by the use of heavy agricultural machinery (Dani, 1993; Sick, 1997). In Minas Gerais State, southeastern Brazil, greater rheas are classified as vulnerable on the red list of species threatened with extinction (Machado et al., 1998). Considering this scenario, conservation measures need to be implemented to avoid the greater rheas' local extinction.

The evaluation of the public effect on the behaviour of captive greater rheas is important for the planning of conservation programs, since zoos and other Institutions, which hold these birds constitute the main sources of individuals for potential reintroduction projects. This is the case of the Belo Horizonte Zoo (BH Zoo) in southeastern Brazil who intends to reintroduce greater rheas in the near future. If negative effects of visitors' presence on their behaviour or welfare are detected, then new management strategies could be implemented to conserve greater rheas' natural behaviour and to avoid health problems. The aim of this study was to detect the public effect on captive greater rheas by comparing their behavioural expression on days with and without public visitation; it was expected that greater rheas would exhibit more abnormal behaviours, such as pacing, in the days with visitation, and that larger numbers of visitors would elicit more abnormal behaviour, and that the intensity of visitation (i.e., public behaviour) would influence greater rheas' behaviour.

Material and Methods

Five greater rheas were studied (two male and three female), which were held in the "Bird's Square" of the Belo Horizonte Zoo (BH Zoo), Minas Gerais, Brazil (S 19º51'44.8"; W 44º00'40.1"). The greater rheas' exhibit was open to public visitation from Tuesday to Sunday every week, from 9:00AM to 5:00PM. Birds were housed in a wire-fenced enclosure 28m in length and 16m wide (the front of the enclosure was the visitors' viewing

area - 28m length); a female casuar (*Casuarius casuarius*) and two ostriches (*Struthio camelus*) were housed in neighbouring enclosures. Rheas were fed twice a day with a mixture of ratite ration (Socil®, 1.2 kg in total) and vegetables (carrot and cabbage; 1.3 Kg in total). Water was provided *ad libitum* in a water hole. The enclosure was cleaned three times a week. Birds varied from four to eight years old (5.6 ± 0.8 years) and had been exposed to the public for between three and seven years (birds were exposed only after they were one year old) (4.6 ± 0.7 years).

The study was divided in two treatments: (1) the presence of visitors in front of the greater rheas' enclosure and; (2) the absence of visitors in front of the greater rheas' enclosure. Thirty hours of behavioural data were collected in each treatment. All data were collected using scan sampling with instantaneous recording of behaviours every minute (Altmann, 1974). All behavioural data collection sessions were conducted between 1500h and 1600h daily, from April to December 2009. This time period was chosen as pilot observations had shown rheas to be most active at this time of day.

An ethogram for greater rheas (Table 1) was constructed based on scientific literature (Codenotti et al., 1995; Brandt and Neto, 1999) and on 12 hours of *ad libitum* pilot observations (Martin and Bateson, 2007).

Data on public behaviour were also collected at the same period; data were collected using scan or focal sampling (i.e., when only one visitor was present) with instantaneous recording of behaviours every minute (Altmann, 1974); an ethogram of the public behaviour (Table 2) was based on 10 hours of *ad libitum* pilot observations (Martin and Bateson, 2007). Data concerning visitor number (the number of visitors per hour in front of the greater rheas' enclosure) and time spent in front of the greater rheas' enclosure (the

period that each visitor or group of visitors stayed in front of the greater rheas' enclosure) were also collected.

We tested our data with an Anderson-Darling test to determine if they met the requirements for parametric statistics, which they did not and therefore nonparametric statistical tests were used throughout. Greater rhea behaviours and visitor's behaviours were quantified and statistically analysed using Mann-Whitney U-test. Spearman's rank correlations between behaviours expressed by the greater rheas and by the visitors, and between behaviours expressed by the greater rheas and the number of visitors were undertaken. For all statistical analyses, the confidence level was 95% (= 0.05) (Zar, 1999).

Results

The number of the public that visited the greater rheas' enclosure during the study period was 4,182, with a mean of 139.4 ($\pm$ 15.3) persons per hour. The minimum number of visitors at the same time was one and the maximum number of visitors at the same time was 103. The mean time spent by the visitors in front of the greater rheas' enclosure was 9.86 ($\pm$ 1.49) seconds; the minimum time spent by the visitors in front of the greater rheas' enclosure was nine seconds and the maximum time was 319 seconds. The percentages of behaviours expressed by the visitors are shown in Figure 1. The most performed visitor behaviours were "stopped-watching-talking" (56.00 $\pm$ 8.33; 40.08%) and "walking-watching-talking" (42.27 $\pm$ 5.92; 30.25%). The least exhibited behaviours were "walking-watching-shouting" (0.23 $\pm$ 0.20; 0.17%) and "interacting-with-the-birds" (0.47 $\pm$ 0.20; 0.33%); the behaviour "walking-not-watching-shouting" was not performed by the visitors during the study.

The most expressed behaviours by the greater rheas in the presence of the public were "foraging" (N = 2007 records; 19.12%) followed by "walking" (N = 1954 records; 18.62%);

the least exhibited behaviours by the greater rheas in the presence of the public were

"escaping behaviour" (N = 3 records; 0.03%) and "running" (N = 4 records; 0.04%). The

most expressed behaviours by the greater rheas without the presence of the visitors were

"walking" (N = 2279 records; 21.71%) and "foraging" (N = 2046 records; 19.49%); the

least exhibited behaviours by the greater rheas without the presence of the visitors were

"escaping behaviour" and "intimidating" both with only three records (0.03% each).

Only the behaviours "walking alert", "defecating/urinating" and "other behaviours"

differed statistically between the treatments (see Table 1), "walking alert" being more

expressed in the presence of visitors and "defecating/urinating" and "other behaviours"

being more expressed in the absence of the visitors (Table 3).

A negative significant correlation was found between the behaviours "alert" and "walking-

watching-talking" (r_s = -0.64; N = 30; p < 0.01), "alert" and "running (public)" (r_s = -0.37;

N = 30; p < 0.05), "alert" and "stopped-watching-talking" (r_s = -0.47; N = 30; p < 0.01),

"preening" and "stopped-watching-talking" (r_s = -0.49; N = 30; p < 0.01), and "pacing" and

"other behaviours (public)" (r_s = -0.58; N = 30; p < 0.01); a positive correlation was found

between the behaviours "foraging" and "stopped watching quietly" (r_s = 0.54; N = 30; p <

0.01) (first behaviours in the correlation referred to greater rheas' behaviours; second

behaviours in the correlation referred to the public's behaviours).

A negative significant correlation was found between the greater rheas' behaviours "alert",

"pacing" and "not visible" and the number of visitors in front of the exhibition (r_s = -0.59,

N = 30, p < 0.01; r_s = -0.38, N = 30, p < 0.05; r_s = -0.42, N = 30, p < 0.05).

Discussion

Greater rheas seemed to be habituated to visitor presence since the majority of their

behaviours did not differed in the presence or absence of visitors in front of their enclosure.

Habituation is good for greater rheas' captive management since it, probably, reduces stress (Waitt et al., 2002; Owen et al., 2004), but it should be avoided for conservation programs, since rheas could approach humans after release, which could cause them problems (Kloppers et al., 2005; Lamarque et al., 2008).

Only the behaviours "walking alert", "defecating/urinating" and "other behaviours" differed between treatments. "Walking alert" was more expressed when the visitors were present in front of the greater rheas' enclosure, and this could be first interpreted as a vigilance (anti-predator) response; that is, with the presence of people, greater rheas became more stressed. Stress due to visitors presence has been observed for primates in many studies (Chamove et al., 1988; Wood, 1998; Wells, 2005), Indian leopards (*Panthera pardus*) (Mallapur and Cheilant, 2002), black rhinos (*Diceros bicornis*) (Carlstead and Brown, 2005), birds (Bennet and Zuelke, 1999) and Indian gaurs (*Bos gaurus*) (Sekar et al., 2008).

"Defecating/urinating" was most exhibited when the public was absent from the visitor's area; this behaviour when frequently expressed may be associated with fear (Candland and Campbell, 1962; Munksgaard et al., 1997; Vargas et al., 2000), which was not the case in this study since it only occupied 0.2% (0.53 ± 0.13) of the activity budget of the greater rheas; thus, the recording of this behaviour was disproportionately higher in the absence of visitors, probably, due to chance.

"Other behaviours", such as "head scratching", "stretching" and "building nests" were also more expressed when the public was absent from the visitor's area. These were behaviours which could indicate a "relaxed" behavioural state, and their low occurrence in the presence of visitors could be interpreted as the public functioning as a source of stress to the birds, since without the presence of the public, greater rheas expressed significantly more of these

'relaxed' behaviours. It is important to emphasize that "other behaviours" were expressed at very low levels (less than 1% of the activity time budget; 0.2% in the presence and 0.7% in the absence of visitors).

All abnormal behaviours, such as "pacing", "eating faeces" and "drinking water" (in this case considered abnormal due to its high expression; 8.9% (42.83 ± 8.62 recordings of the activity budget) were more performed when visitors were present, supporting the hypothesis that some stress was caused by human visitation. It is necessary to conduct physiological studies to confirm this hypothesis.

Most of the behaviours expressed by the visitors were not considered as threats to the welfare of the greater rheas: all the correlations found between visitor and rhea behaviour supported this statement. Shouting and rhea-visitor interactions were rare and the time spent by the visitors in front of the greater rheas' enclosure was minimal (9.8 seconds on average). Public flux was high, and this could act as a source of novelty for the birds, even functioning as environmental enrichment (Morris, 1964; Pazol and Bloomsmith, 1993). Thus, audience activities and noisiness, the most common variables inferred as sources of animal stress (Hosey and Druck, 1987; Mitchell et al., 1992a; Birke, 2002), did not appear to show great negative effects on greater rheas' welfare in this study.

Conclusion

The results found in this study showed that the greater rheas of BH Zoo appeared to be habituated to the presence of the public. Some behavioural changes when public were present, such as the greater expression of pacing and lower expression of some relaxed behaviours ("head scratching", "stretching" and "building nests"), indicated that the presence of the public stressed the birds to some degree (this conclusion needs further physiological studies).

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Animal welfare implications

The habituation showed by the greater rheas to the public is indicative of the fact that visitors were, probably, not a significant source of stimuli causing animal welfare problems (i.e., stress). In order to avoid changes in this scenario, it is important that the public, which visits the greater rhea's enclosure, receives educational support from the zoo's staff to avoid shouting or the performance of stress inducing behaviours for the birds (the greater rheas in this study did not showed signs of stress problems, but density and intensity of visitors have been shown to affect the behaviour of captive animals causing stress (Hosey and Drunk, 1987; Sellinger and Ha, 2005). Large crowds producing loud sounds should be avoided in front of its exhibit, since this behaviour could stress the animals (Davey, 2006; 2007).

For animals intended to be reintroduced to the wild, the results found in this study are worrying. Habituated animals have a greater probability of dying after release than non-habituated animals (Kasereka et al., 2006). On the other hand, the maintenance of non-habituated animals in the zoo could generate welfare problems due to acute or chronic stress resulting from daily human contact (Moberg and Mench, 2000).

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Table 1: Ethogram of the greater rheas (*Rhea americana*, Rheidae, Aves) at BH Zoo.

Behaviour	Description
Alert	Rhea stretches its neck, elevating its head, and moves its head from side to side, watching their vicinity.
Walking	Rhea walks through its enclosure.
Foraging	Rhea searches for food on the ground or on the vegetation.
Inactive	Rhea stands or sits inactive.
Eating	Rhea eats the food provided by its keeper.
Drinking water	Rhea drinks water from the water hole.
Running	Rhea runs straight or in zigzags through its enclosure.
Fighting	Two males stretch their necks, striking each other with thrusts and bill snapping.
Preening	Rhea preens its feathers with its beak.
Threatening	Rhea opens its bill and gapes, drawing its head back slightly and turns it so as to face the other rhea continually.
Eating faeces	Rhea eats their own or other rheas' faeces.
Pacing	Rhea walks continually in an eight-shaped route, with no apparent reason.
Vocalization	Rhea produces a deep-toned two note call ("nan-du").
Mating	The female lies flat on the ground as the male grasps the feathers on the back of her neck with his bill; he rests on his legs while copulating.
Incubating eggs	The male incubates the eggs in the nest.
Courting	The male slowly approaches the female, lower his neck in a low loop and walks beside her, sometimes bobbing his head slightly.
Walking alert	Rhea walks with an alert posture.
Escaping behaviour	Rhea jumps in the air and then run in zigzags as soon as it touches the ground, with no apparent reason.
Pecking	Rhea pecks objects in the ground or the enclosure's fence.
Dust bathing	Rhea extends its neck and picks up some soil or dust with its bill, throwing the dust onto its wings and body.
Defecating/urinating	Rhea defecates or urinates.

Other behaviours	Behaviours not previously recorded.
Not visible	Birds not visible to the observer.

Table 2: Ethogram of the visitors' behaviour at the greater rhea exhibit at BH Zoo.

Behaviour	Description
Reading label	Visitor reads the label of the rhea's exhibit.
Walking-watching-quietly	Visitor walks quietly through the visitor's viewing area when watching the rheas.
Walking-watching-talking	Visitor walks talking through the visitor's viewing area when watching the rheas.
Walking-watching-shouting	Visitor walks shouting through the visitor's viewing area when watching the rheas.
Walking-not-watching-quietly	Visitor walks quietly through the visitor's viewing area but not watching the rheas.
Walking-not-watching-talking	Visitor walks talking through the visitor's viewing area but not watching the rheas.
Walking-not-watching-shouting	Visitor walks shouting through the visitor's viewing area but not watching the rheas.
Running	Visitor runs through the visitor's viewing area of the greater rheas' exhibit.
Stopped-watching-quietly	Visitor stops in the visitor's viewing area, and quietly watches the rheas.
Stopped-watching-talking	Visitor stops in the visitor's viewing area, and talks while watching the rheas.
Stopped-watching-shoulting	Visitor stops in the visitor's viewing area, and shouts while watching the rheas.
Stopped-not-	Visitor stops quietly in the visitor's viewing area, but not watching the rheas.

|	
watching-quietly	
Stopped-not-watching-talking	Visitor stops in the visitor's viewing area and talks, but not watching the rheas.
Stopped-not-watching-shoulting	Visitor stops in the visitor's viewing area and shouts, but not watching the rheas.
Interacting with the rheas	Visitor interacts with the rheas, throwing foods to the birds or calling the birds to approaches them.
Other behaviours	Any other behaviour expressed that is not in this ethogram.
Not visible	Visitors not visible to the observer.

Table 3: Behaviours expressed by the greater rheas of BH Zoo in the presence and absence of visitors in front of their enclosure (mean ± standard error; DF = 1; N = 30 in all cases) and Mann-Whitney U-test results.

Behaviour	With public	Without public	Mann-Whitney	p-value
Alert	16.37 ± 2.28	13.23 ± 2.36	838.5	0.26
Walking	64.67 ± 4.50	76.27 ± 6.04	1000.5	0.21
Foraging	66.13 ± 6.14	68.97 ± 6.00	930.0	0.83
Inactive	45.73 ± 5.32	53.10 ± 6.67	948.5	0.63
Eating	16.43 ± 2.77	26.67 ± 4.57	1027.5	0.10
Drinking water	31.23 ± 7.85	21.33 ± 5.66	833.5	0.23
Running	0.13 ± 0.07	0.33 ± 0.15	938.0	0.59
Fighting	0.27 ± 0.20	0.13 ± 0.13	885.5	0.32
Preening	9.73 ± 2.01	8.33 ± 1.48	921.5	0.93
Threatening	0.17 ± 0.08	0.10 ± 0.06	898.5	0.67
Eating faeces	13.10 ± 3.14	13.17 ± 2.97	926.0	0.88
Pacing	42.83 ± 8.62	21.17 ± 3.98	831.0	0.22
Vocalization	0.70 ± 0.30	0.98 ± 0.39	923.5	0.88
Mating	0.20 ± 0.20	4.00 ± 4.00	915.5	1.00
Incubating eggs	32.30 ± 7.77	29.00 ± 7.50	890.0	0.69
Courting	3.53 ± 1.18	3.23 ± 1.12	899.0	0.78
Walking alert	1.03 ± 0.37	0.90 ± 0.80	796.0	0.02*
Escaping behaviour	0.10 ± 0.06	0.10 ± 0.06	915.0	1.00
Pecking	0.23 ± 0.11	0.17 ± 0.08	900.0	0.73

Dust bathing	2.00 ± 0.71	2.40 ± 0.67	942.0	0.66
Defecating/urinating	0.53 ± 0.13	1.00 ± 0.16	1058.0	0.02*
Other behaviours	0.63 ± 0.19	2.47 ± 0.81	1093.0	< 0.01*
Not visible	1.80 ± 0.59	1.33 ± 0.50	853.0	0.30

*: behaviours that differed statistically between treatments.
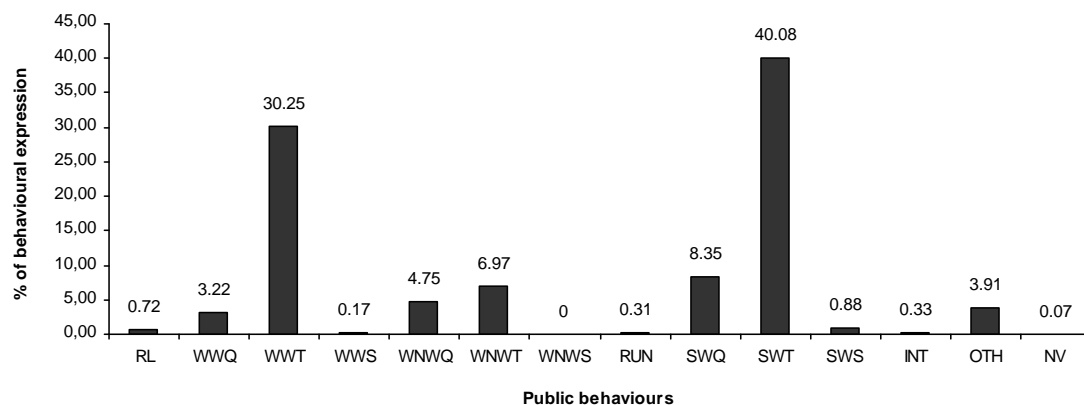
Figure 1: Percentage of visitor behaviours in front of the greater rheas' enclosure at BH Zoo. RL = reading label; WWQ = walking-watching-quietly; WWT = walking-watching-talking; WWS = walking-watching-shouting; WNWQ = walking-not-watching-quietly; WNWT = walking-not-watching-talking; WNWS = walking-not-watching-shouting; RUN = running; SWQ = stopped-watching-quietly; SWT = stopped-watching-talking; SWS = stopped-watching-shouting; INT = interacting with the rheas; OTH = other behaviours; NV = not visible.

Conclusões finais

O comportamento das emas, bem como o uso do habitat, se modifica quando o ambiente é alterado pelo homem. A destruição do Cerrado para a implantação de monoculturas é fator importante na exclusão das emas da área, especialmente se as monoculturas forem de plantas arbóreas (florestas). Pastagens, ao contrário, são bastante utilizadas pelas emas, que usam o capim como fonte principal de alimento.

As emas se mostraram boas dispersoras de sementes de algumas espécies de plantas do Cerrado, e o seu desaparecimento pode comprometer a dinâmica e o equilíbrio das áreas onde ocorre.

O conhecimento da população humana acerca do papel das emas na conservação do Cerrado varia entre pessoas que vivem no campo (na área de ocorrência da ave) e na cidade (fora da área de ocorrência da ave), mas, em geral, as pessoas vêem relação entre a destruição do Cerrado e o desaparecimento das emas. Ações de educação ambiental devem ser realizadas tanto para as populações rurais quanto para as populações urbanas.

A ema é uma espécie mantida pelos zoológicos do Brasil; sua reprodução em cativeiro é facilmente conseguida. Estas Instituições podem, então, servir como centros de reprodução para fornecimento de emas para programas de reintrodução. Entretanto, antes de serem soltas na natureza, vários aspectos de seu comportamento antipredação devem ser avaliados, afim de que aumente as chances de sobrevivência dos indivíduos pós-soltura. A capacidade de reconhecer predadores deve ser avaliada, e caso constatado o não-reconhecimento, sessões de treinamento anti-predação devem ser conduzidas (como observado neste estudo).

O uso do enriquecimento ambiental deve ser estimulado; itens bastante simples, como frutas espalhadas pelo chão, além de estimularem a exibição de comportamentos naturais, diminuem a chance de que comportamentos anormais se desenvolvam. A diminuição do estresse causado pelo ambiente de cativeiro é também um benefício do enriquecimento ambiental. Então, como medida de manejo, um programa de enriquecimento ambiental para emas deve ser conduzido.

O estresse causado pela visitação contínua do público dos zoológicos é fato já demonstrado para uma grande variedade de espécies. Neste estudo, o público visitante não pareceu estressar as emas, já que seu comportamento não foi diferente quando o público estava presente ou ausente. Entretanto, emas destinadas à programas de reintrodução devem ser mantidas em recintos adequados (grandes, naturalísticos e estimulantes) e fora da área de visitação, para evitar estresse ou habituação aos seres humanos.

Para que as emas sobrevivam aos dias atuais, seu ambiente natural precisa ser conservado; a criação de áreas de preservação bem como o manejo adequado das áreas já existentes são medidas urgentes de conservação *in-situ*. O uso dos jardins zoológicos como áreas de conservação *ex-situ* de emas também é fortemente recomendada. Nestes locais, medidas de manejo que evitem o estresse causado pelo ambiente de cativeiro devem ser implantadas, como o uso contínuo do enriquecimento ambiental (desde que itens sejam elaborados periodicamente para se evitar a habituação) e a evitação do contato direto com o público (pelo menos das aves destinadas a programas de conservação). Dietas mais próximas ao natural, recintos mistos, com vegetação natural, grandes e com áreas de escape do público são recomendados para estas aves. A implementação de projetos de reintrodução deve ser conduzida após testes comportamentais que permitam identificar a qualidade do plantel disponível, como testes de estresse, reconhecimento de predadores e de alimento, e

treinamento anti-predação. Tanto as ações *in-situ* quanto as *ex-situ* devem ser conduzidas juntamente com projetos de educação ambiental. Estudos genéticos (estrutura genética da população de emas cativas mantidas pelos zoológicos brasileiros), de doenças (tipos de doenças que mais acometem os plantéis brasileiros) e dos tipos de manejo (intensivo, semi-intensivo ou extensivo, dietas, etc.) das emas no Brasil podem ser conduzidos futuramente para aumentar os subsídios para a elaboração de planos de manejo, que em última instância, geram informações para a idealização e desenvolvimento de programas de conservação de espécies no país.