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**SOIL BEETLES ASSEMBLAGES AS ECOLOGICAL
INDICATORS IN THE NEOTROPICS: IMPLICATIONS FOR
ECOSYSTEM CONSERVATION AND RESTORATION**

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Lajeado, 2018

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INDICATORS IN THE NEOTROPICS: IMPLICATIONS FOR
ECOSYSTEM CONSERVATION AND RESTORATION**

Thesis submitted as part of the requirements for the degree of Doctor with Environment and Development Postgraduate Program, University of Taquari Valley, Brazil, with area of concentration in Ecology.

Supervisors

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Dedicated to my family and friends.

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It may be argued that to know one kind of beetle is to know them all. But a species is not like a molecule in a cloud of molecules - it is a unique population.

(Edward O. Wilson, 1985).

LIST OF FIGURES

Chapter 1

- Figure 1. Location of the study region in the municipality of Uruará, state of Pará, northern Brazil. Location of the sampling areas: NV, Native vegetation; MS, Mature secondary succession; ES, Early secondary succession; Pa, Pasture; Ag, Agriculture..... 23
- Figure 2. Habitats studied in the municipality of Uruará, Pará. (A) Native Vegetation, (B) Maturing secondary succession, (C) Early Secondary succession, (D) Agriculture - Ag (cocoa plantations), (E) Pasture. 24

Chapter 2

- Figure 1. Location of the Brazilian Amazon (Amazonia) in South America..... 37
- Figure 2. Cumulative rate of deforestation (Km²) between 1988-2013 in the Brazilian Amazon (Inpe, 2014). 38
- Figure 3. "Arc of deforestation" in the Brazilian Amazon - in dotted lines (adapted from Lui and Molina, 2009)..... 38
- Figure 4. Flowchart showing the main causes and effects of Amazon deforestation in Brazil: The causes which lead to deforestation are black boxes; and consequences for human society are indicated by gray boxes (Adapted the Davidson et al., 2012)..... 41
- Figure 5. Types of ecological indicators. 43
- Figure 6. Criteria for selection of ecological indicators..... 45
- Figure 7. Diversity of invertebrate species compared to other indicators groups (Adapted the Calor, 2009)..... 46
- Figure 8. Diversity of beetles compared to the main groups of insects (Adapted the Cajaiba, 2013). 48

Chapter 3

- Fig 1. Location of the study region in the municipality of Uruará, state of Pará, northern Brazil. Location of the sampling areas: NV, Native vegetation; MS, Mature secondary succession; ES, Early secondary succession; Pa, Pasture; Ag, Agriculture..... 81
- Fig. 2. Scarab beetles individual-based rarefaction curves for the studied sites (centre lines). External lines indicate 95% confidence intervals. NV, Vegetation native; MS, Mature Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture; Pa, Pasture. 85
- Fig. 3. Box and Whisker plots expressing the differences in the projected values for (a) Number of species (\pm SE) (b) number of individuals (\pm SE), (c) Shannon diversity (\pm SE) and (d) Berger-Parker dominance (\pm SE) of Scarabaeidae community in the different habitats considered. The values followed by the same letters are not significantly different according to Tukey test. NV, Vegetation native; MS, Mature Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture; Pa, Pasture. 86
- Fig. 4. Canonical Correspondence Analysis (CCA) used to estimate the influence of environmental variables on Scarabaeidae communities. Four axes were considered in the Principal Components Analysis (PCA), including 4 principal components (PC). NV, Native vegetation; MS, Mature secondary succession (15 years of natural regeneration of

vegetation); ES, Early secondary succession (five years of natural regeneration of vegetation); Ag, Agriculture (Cacao plantations); Pa, Pasture. 89

Chapter 4

Fig. 1. Abundance of Scarabaeinae feeding guilds by habitat. NV: Native vegetation; MS: Mature secondary; ES: Early secondary; Ag: Agriculture (Cocoa); Pa: Pasture. 113

Fig. 2. Abundance of Scarabaeinae on the behavioral guild in different habitats studied. NV: Native vegetation; MS: Mature secondary; ES: Early secondary; Ag: Agriculture (Cocoa); Pa: Pasture. 114

Fig. 3. Differences between abundance (A) and species richness (B) of the Scarabaeinae studied with different bait type. Co: Control bait; RB: Rotten banana; RM: Rotten meat; HF: Human faeces. The values followed by the same letters are not different according to Dunn test. 115

Fig. 4. Differences between species richness (a) and abundance (b) of the Scarabaeinae studied in different habitats (NV=Native vegetation, MS=Mature secondary, ES=Early secondary, Ag=Agriculture, Pa=Pasture) for each bait type (Co=Control bait, RB=Rotten banana, RM=Rotten meat, HF=Human feces). 115

Chapter 5

Figure 1. Location of the study sites in the municipality of Uruará, state of Pará, northern Brazil. Abbreviations: Ag, Agriculture; ES, Early secondary succession; MS, Mature secondary succession; NV, Native vegetation; Pa, Pasture. 131

Figure 2. (A) Abundance (number of individuals \pm SE), and (B) richness (number of species \pm SE) of the DB community in the different ecosystems considered. The values labelled with the same letters are not significantly different according to the Tukey post-hoc test. Abbreviations: Ag, agriculture; ES, early secondary succession (vegetation with five years of regeneration); MS, mature secondary succession (vegetation with 15 years of regeneration); NV, native vegetation; Pa, pasture. 134

Figure 3. (A) Abundance (number of individuals \pm SE) and (B) richness (number of species \pm SE) of dung beetles in different periods of sampling in the Brazilian Amazon. The values labelled with the same letters are not significantly different according to the Tukey post-hoc test. Abbreviations: D, dry; I, intermediary; R, rainy. 135

Figure 4. Temporal variation of the abundance and richness of dung beetles by ecosystem. The values followed by the same letters are not significantly different according to the Tukey post-hoc test. Abbreviations: D, dry; I, intermediary; R, rainy. 137

Figure 5. Bray-Curtis similarity index for the different habitats and season sampled (cophenetic correlation coefficient = 0.85). Abbreviations: Ag, agriculture; D, dry; ES, early secondary succession (vegetation with five years of regeneration); I, intermediary; MS, mature secondary succession (vegetation with 15 years of regeneration); NV, native vegetation; Pa, pasture; R, rainy. 138

Chapter 6

Figure 1. Location of the study region in the municipality of Uruará, state of Pará, northern Brazil. Location of the sampling areas: NV, Native vegetation; MS, Mature secondary succession; ES, Early secondary succession; Ag, Agriculture; Pa, Pasture. 159

Figure 2. Differences in the projected values for (a) Richness (\pm SE), and (b) Abundance (\pm SE) of Histerid beetles (HB) community in the different ecosystems considered. The values followed by the same letters are not significantly different

according to Tukey test. NV, Vegetation native; MS, Mature Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture; Pa, Pasture. 162

Figure 3. Non-metric multidimensional scaling (NMDS) showing Histerid beetles grouped in accordance with the ecosystems (using Bray-Curtis similarity). NV, Vegetation native; MS, Mature Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture (Cocoa); Pa, Pasture. 163

Figure 4. Abundance and richness of Histerid beetles (a) and distribution of environmental variables during the study period (b). 164

Chapter 7

Fig. 1. Location of the study region in the municipality of Uruará, state of Pará, northern Brazil. Location of the ecosystems sampling: NV, Native vegetation; MS, Maturing secondary succession; ES, Early secondary succession; Pa, Pasture; Ag, Agriculture. 178

Fig. 2. Rove Beetles individual-based rarefaction curves for the studied sites. NV, Vegetation native; MS, Maturing Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with 5 years of regeneration); Ag, Agriculture; Pa, Pasture. 183

Fig. 3. Barplot expressing the differences in the projected values for (a) Number of morphospecies, (b) number of individuals, (c) Shannon diversity, and (d) Berger-Parker dominance of Staphylinidae community in the different ecosystems considered. Error bars indicate 95% confidence intervals. The values followed by the same letters are not significantly different according to Tukey test. NV, Vegetation native; MS, Maturing Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with 5 years of regeneration); Ag, Agriculture; Pa, Pasture. 185

Fig. 4. Non-metric multidimensional scaling (NMDS) showing RB grouped in accordance with the ecosystems (using Bray-Curtis similarity). NV, Native vegetation; MS, Maturing Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture (Cocoa); Pa, Pasture. 186

Fig. 5. Comparison of dissimilarity values for β_{sor} (overall dissimilarity), β_{sne} (dissimilarity resulting from nestedness), and β_{sim} (turnover) for the different ecosystems. NV, Native vegetation; MS, Maturing Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture (Cocoa); Pa, Pasture. 188

Fig. 6. Canonical Correspondence Analysis (CCA) used to estimate the influence of environmental variables on Rove beetle (Staphylininae) communities. Four axes were considered in the Principal Components Analysis (PCA), including 4 principal components (PC). NV, Native vegetation; MS, Maturing secondary succession (15 years of natural regeneration of vegetation); ES, Early secondary succession (5 years of natural regeneration of vegetation); Ag, Agriculture (Cacao plantations); Pa, Pasture. 189

Chapter 8

Figure 1. Location of the study area in the municipality of Uruará, state of Pará, northern Brazil with sampled habitats: Ag, Agriculture; ES, Early secondary succession; MS, Mature secondary succession; NV, Native vegetation; Pa, Pasture. 224

Figure 2. Box and Whisker plots expressing the differences in the projected values for (a) species richness (\pm SE) and (b) number of individuals (\pm SE) of Scolytinae in different habitats in the municipality of Uruará, state of Pará, northern Brazil. The values followed by the same letters are not significantly different according to Tukey test. Ag, Agriculture; ES, Early secondary succession; MS, Mature secondary succession; NV, Native vegetation; Pa, Pasture. Circles in the figures indicate the outliers. 228

Figure 3. Bray-Curtis similarity index for the different habitats sampled (cophenetic correlation coefficient = 0.90) in the municipality of Uruará, state of Pará, northern Brazil. Ag, Agriculture; ES, Early secondary succession; MS, Mature secondary succession; NV, Native vegetation; Pa, Pasture. 229

Figure 4. Scolytinae community in different periods of sampling in the municipality of Uruará, state of Pará, northern Brazil: (a) number of individuals (\pm SE) and (b) number of species (\pm SE). The values followed by the same letters are not significantly different according to Tukey test. D: Dry; I: Intermediary; R: Rain. Circles in the figures indicate outliers..... 231

Chapter 9

Fig. 1. Location of the study region, (a) Brazil, (b) Pará state, and (c) city of Uruará. Location of the sampling areas: NV, Native vegetation; MS, Mature secondary succession; ES, Early secondary succession; Pa, Pasture; Ag, Agriculture. 241

Fig. 2. Box and Whisker plots expressing the differences in Ground Beetles (A) recognizable taxonomic unit (RTU) richness and (B) RTU abundance for the different ecosystems. The values followed by different letters are significantly different according to the Dunn test. NV: Native vegetation; MS: Mature Secondary succession (vegetation with 15 years of regeneration); ES: Early Secondary succession (vegetation with five years of regeneration); Ag: Agriculture; Pa: Pasture. 247

Fig. 3. Non-metric multidimensional scaling (NMDS) showing Ground beetles (GB) recognizable taxonomic units (RTU) grouped in accordance with the ecosystems (using Bray-Curtis similarity). NV, Native vegetation; MS, Mature Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture (Cocoa); Pa, Pasture. 248

Fig. 4. Comparison of dissimilarity values for β sor (overall dissimilarity), β sne (dissimilarity resulting from nestedness), and β sim (turnover) for the different ecosystems. NV, Native vegetation; MS, Maturing Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture (Cocoa); Pa, Pasture..... 249

Fig. 5. Canonical Correspondence Analysis (CCA) used to estimate the influence of environmental variables on Ground Beetles communities. Four axes were considered in the Principal Components Analysis (PCA), including 4 principal components (PC). NV, Native vegetation; MS, Mature secondary succession (15 years of natural regeneration of vegetation); ES, Early secondary succession (five years of natural regeneration of vegetation); Ag, Agriculture (Cacao plantations); Pa, Pasture. PC1 was positive correlated with the following environmental variables: Circumference at Breast Height (CBH); Circumference at breast ankle (CBA); Canopy Cover (CC); richness of plants (RP); Height of Leaf Litter (HLL); Percentages of Leaf Litter Cover (PLC); Percentage of Green (vegetation) Cover (PGC); Abundance of Plants (AP); Richness of Shrubs (RS), and negative correlated with Temperature (T) and Percentage of Exposed Soil (PES). PC2 was positive correlated with Humidity (H), PC3 positive correlated with Abundance of Shrubs (AS) and PC4 with Precipitation (P), also positively..... 251

Fig. 6. Canonical Correspondence Analysis (CCA) used to estimate the influence of environmental variables on Ground Beetles communities. Four axes were considered in the Principal Components Analysis (PCA), including 4 principal components (PC). NV, Native vegetation; MS, Mature secondary succession (15 years of natural regeneration of vegetation); ES, Early secondary succession (five years of natural regeneration of vegetation); Ag, Agriculture (Cacao plantations); Pa, Pasture. PC1 was positive correlated with the following environmental variables: Circumference at Breast Height (CBH); Circumference at breast ankle (CBA); Canopy Cover (CC); richness of plants (RP); Height of Leaf Litter (HLL); Percentages of Leaf Litter Cover (PLC); Percentage of Green (vegetation) Cover (PGC); Abundance of Plants (AP); Richness of Shrubs (RS), and negative correlated with Temperature (T) and Percentage of Exposed Soil (PES). PC2 was positive correlated with Humidity (H), PC3 positive correlated with Abundance of Shrubs (AS) and PC4 with Precipitation (P), also positively..... 252

Fig. 6. CCA for Carabidae RTU using 14 environmental variables. For a description of the abbreviations of environmental parameters, see Table S2, and for abbreviations of GB RTU see Table S3, Supplementary material. 252

LIST OF TABLES

Chapter 1

Table 1. Dissimilarity percentage among the five studied areas, with significant difference ($p < 0.01$) according to PERMANOVA ($F_{4,207} = 36.59$). NV, Vegetation native; MS, Mature Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture; Pa, Pasture..... 87

Table 2. Analysis of SB' indicator species (IndVal) from NV, Vegetation native; MS, Mature Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture; Pa, Pasture (only taxa with IndVal > 25%)..... 90

Chapter 5

Table 1. Pearson correlation between environmental variables and ecological indices (abundance and richness) of dung beetles collected in Uruará, Pará, northern Brazil. Asterisks indicate statistical significance at levels of: * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$ 136

Chapter 6

Table 1. Total numbers of Histerid beetles species in the environments different in the Brazilian Amazon. NV = Native Vegetation; MS = Mature Secondary succession (15 years of regeneration); ES = Early Secondary succession (5 years of regeneration); Ag = Agriculture (Cocoa); Pa = Pasture. R = Rainy Season; Intermediary Season; D = Dry Season. 161

Table 2. Pearson correlation between environmental variables and ecological indices (abundance and richness) of Histeridae collected in Uruará, Pará, northern Brazil. Asterisks indicate statistical significance at level of * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$ 164

Chapter 7

Table 1. Analysis of RB' indicator species (IndVal) of NV, Vegetation native; MS, Maturing Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with 5 years of regeneration); Ag, Agriculture; Pa, Pasture. A, Mean Abundance; B, Relative frequency of occurrence (for details of A and B, see De Cáceres and Legendre, 2009). 191

Chapter 8

Table 1. Species of Scolytinae collected in different habitats and their abundance in different seasons in the municipality of Uruará, state of Pará, northern Brazil. Ag, Agriculture; ES, Early secondary succession; MS, Mature secondary succession; NV, Native vegetation; Pa, Pasture. The letters R, I and D following the vegetation types correspond to the rainy, intermediary and dry seasons, respectively. 226

Table 2. Multivariate regressions between environmental variables and ecological indices (abundance and richness) of Scolytinae collected in the municipality of Uruará, state of Pará, northern Brazil. H, Humidity; HLL, Height of Leaf Litter; P, Precipitation; PES, Percentage of Exposed Soil; PLC, Percentages of Leaf Litter Cover; T, Temperature. 229

Chapter 9

Table 1. Recognizable taxonomic unit (RTU) selected by the IndVal method. NV, Native vegetation; MS, Mature Secondary succession (vegetation with 15 years of regeneration);

ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture; Pa, Pasture (only taxa with IndVal>25% were considered). 253

CONTENTS

Chapter 1	19
General Introduction / Contextualization	19
Area of study	21
Structure of thesis / specific objectives	25
References	28
Chapter 2	33
Assessing The Potential Role Of Ground Beetles (Coleoptera) As Ecological Indicators In Tropical Ecosystems: A Review	33
Abstract	34
Introduction	35
Causes and Effects of Amazon Deforestation.....	36
Surface Soil Invertebrates as Ecological Indicators of Environmental Changes	42
Soil Surface Coleoptera as Indicators of Environmental Changes.....	46
Soil Surface Beetles as Indicators of Environmental Changes in the Tropics with a Focus on the Brazilian Amazon.....	52
Perspectives and Challenges	54
Final Remarks	56
Appendices.....	58
References	60
Chapter 3	77
Does the composition of Scarabaeidae (Coleoptera) communities reflect the extent of land use changes in the Brazilian Amazon?.....	77
Abstract	78
Material and Methods	80
<i>Study area</i>	80
<i>Scarab beetle sampling</i>	81
<i>Environmental variables</i>	82
<i>Assemblage analysis</i>	83
<i>Association of Scarab beetle assemblages with environmental variables</i>	83
<i>IndVal</i>	84
Results.....	84
<i>Assemblages analysis</i>	84
<i>Association of Scarab beetle assemblages with environmental variables</i>	87

<i>IndVal</i>	89
Discussion	91
<i>Assemblages analysis</i>	91
<i>Association of Scarab beetle assemblages with environmental variables</i>	92
<i>Value indicator - IndVal</i>	93
Conclusion	94
Acknowledgments.....	96
References	97
Appendix A.....	104
Chapter 4	107
Attractiveness of <i>Scarabaeinae</i> (Coleoptera: Scarabaeidae) to different baits in the Brazilian Amazon region.....	107
Abstract	108
Material and methods.....	110
<i>Study sites</i>	110
<i>Sampling method</i>	111
Results.....	112
<i>General results/Functional diversity</i>	112
<i>Bait attractiveness</i>	114
Discussion	116
Resumen.....	119
References	120
Appendix 1	123
Appendix 2.....	126
Chapter 5	127
Can dung beetles (<i>Scarabaeinae</i>) indicate the status of Amazonia's ecosystems? Insights integrating anthropogenic disturbance with seasonal patterns	127
Abstract	128
Introduction.....	129
Material and methods.....	130
<i>Study sites</i>	130
<i>Sampling method</i>	132
<i>Data analysis</i>	132
Results.....	133

Discussion	138
Final remarks.....	141
Acknowledgments.....	142
References.....	143
Supplementary material	148
Chapter 6.....	154
Seasonal patterns in the diversity of histerid beetles (Histeridae) are ecosystem specific? A case in para state, northern Brazil	154
Abstract	155
Introduction	156
Material and methods	158
<i>Study area</i>	158
<i>Histerid beetles sampling</i>	159
<i>Assemblage analysis</i>	160
Results.....	161
<i>Composition of Histeridae</i>	161
<i>Seasonality patterns in Histerid beetles assemblages</i>	163
Discussion	164
<i>Composition</i>	164
<i>Seasonality</i>	166
Acknowledgements	168
References.....	169
Chapter 7.....	172
Are disturbance gradients in neotropical ecosystems detected using rove beetles? A case study in the Brazilian Amazon	172
Abstract	173
Introduction	174
Material and methods	177
<i>Study sites</i>	177
<i>Staphylinidae sampling</i>	178
<i>Environmental variables</i>	179
<i>Assemblage analysis</i>	180
<i>Association of Rove Beetle assemblages with environmental variables</i>	181
<i>Species indicator value - IndVal</i>	182

Results.....	182
<i>Assemblages analysis</i>	182
<i>Composition</i>	185
<i>Effects of environmental variables on Rove Beetle assemblages</i>	188
<i>IndVal method</i>	190
Discussion.....	191
<i>Assemblages analysis</i>	191
<i>Composition of Staphylinidae</i>	193
<i>Effects of environmental variables on Rove Beetle assemblages</i>	195
<i>Value indicator - IndVal</i>	196
<i>Contributions for conservation/ challenges and final remarks</i>	197
Acknowledgments.....	200
References.....	201
Supplementary material.....	211
Chapter 8.....	219
Diversity of Scolytinae (Coleoptera: Curculionidae) in different landscapes in northern Brazil.....	219
Abstract.....	220
Resumo.....	221
Introduction.....	222
Material and Methods.....	223
Results and Discussion.....	226
References.....	233
Chapter 9.....	236
How informative is the response of Ground Beetles' (Coleoptera: Carabidae) assemblages to anthropogenic land use changes? Insights for ecological status assessments from a case study in the Neotropics.....	236
Introduction.....	238
Material and Methods.....	240
<i>Study sites</i>	240
<i>Carabidae sampling</i>	242
<i>Environmental variables</i>	242
<i>Statistical analysis</i>	243
<i>Assemblage analysis</i>	243

<i>Association of Ground Beetle assemblages with environmental variables</i>	245
<i>Species indicator value – IndVal</i>	245
Results.....	245
<i>Assemblages analysis and composition of Ground Beetles</i>	245
<i>Ground Beetles diversity and ecosystems</i>	246
<i>Effects of environmental variables on Ground beetle assemblages</i>	249
<i>IndVal Method</i>	252
Discussion	253
<i>Neotropical Ground Beetles, ecosystems and environmental conditions</i>	253
<i>Assemblages analysis and composition of Ground Beetles</i>	253
<i>Effects of environmental variables on Ground beetle assemblages</i>	255
<i>Neotropical Ground Beetles as ecological indicators of anthropogenic disturbances</i> ...	256
<i>Perspectives and challenges on using Neotropical Ground Beetles metrics for environmental management of forest landscapes</i>	257
Conclusions	259
Acknowledgments.....	261
References	262
Supplementary material	272
Chapter 10	278
Synthesis and recommendations	278
General discussion and key findings.....	278
Challenges and limitations	281
Priorities for future work and recommendations.....	282
References	284
Appendices – Other Outcomes	287

Chapter 1

General Introduction / Contextualization

Due to their diversity, complexity and extension (KIM et al., 2015), neotropical forests contribute disproportionately to local, regional and global ecosystem processes (FOLEY et al., 2007; CARDINALE et al., 2012). These forests are fundamental for global climate regulation (ANDERSON-TEIXEIRA et al., 2012), carbon sequestration and storage (BERENQUER et al., 2014), agricultural pests control (HAHN et al., 2014), and biological diversity conservation (FOLEY et al., 2007). Also have an important economic role, providing timber and non-timber products to countries and communities associated (FRANÇA, 2015).

Neotropical ecosystems and especially pristine forests are biodiversity hotspots fast retreating and/or changing through anthropogenic pressures associated with intense population growth and socio-ecological regime shifts (GARDNER et al., 2009; GIBSON et al., 2011). Human pressures are considered “new” disturbances with which organisms in tropical rainforests may not be evolutionarily adapted (GEERAERT, 2014). Trends show a substantial reduction in closed-canopy forests, converted to other uses (e.g. the logging of a natural forest for agricultural use) and a fragmentation of forest remnants (DIAS et al., 2016). Consequently, most neotropical forests (and in general tropical ecosystems) are now imperilled (GIBSON et al., 2011; KIM et al., 2015; FRANÇA, 2015). These trends are also originating changes in ecosystem structure and functioning with significant impacts on the provision of economic goods, ecosystem services and unknown global environmental consequences (NEWBOLD et al., 2015; LIN et al., 2018).

Particularly in the Brazilian Amazon (Amazonia), the deforestation has been causing a sharp erosion of biodiversity and possibly a disruption of complex global climate phenomena (e.g. HIDASI-NETO et al., 2012; PIRES et al., 2016; SOLAR et al., 2016). Several authors consider that the conservation of the Amazonia's ecosystems represents a core challenge for sustaining earth's functioning and ultimately humankind (e.g. VIEGAS et al., 2014; LEAL et al., 2016; SOLAR et al., 2016). Although the Amazon is considered a biodiversity hotspot, our knowledge of

the diversity and distribution of species in the Amazon biome is still at an early stage of development (FABRÉ et al., 2012; CAJAIBA et al., 2015). Scientists have been studying the Amazonia ecosystems since the beginning of the 18th century and a huge number of books and articles focusing on its biological diversity are available (e.g. PITMAN et al., 2007), anyhow, and considering its vastness, the information available is still fragmented with most ecosystems functioning and diversity unknown: e.g., many of the specimens that have been collected to date have not yet been studied (VERWEIJ et al., 2009; CAJAIBA et al., 2015).

Indicator/surrogate species to monitor/assess environmental changes is a widely used method by researchers (HEINK; KOWARIK, 2010; TIEDE et al., 2017; GHANNEM et al., 2018). Studies showed that knowledge on the homogeneity/heterogeneity of species composition in the ecosystems is essential for understanding and monitoring changes, whether they result from natural and/or anthropogenic pressures (ANDRADE et al., 2014; BARNES et al., 2016; BROSE; HILLEBRAND, 2016).

Soil arthropods are considered indicators of environmental quality due to their diversity and sensitivity to physical and biological characteristics (SANTORUFO et al., 2012; LUZ et al., 2013; SIQUEIRA et al., 2014; ZHU; ZHU, 2015; SPILLER et al., 2018). Among these arthropods, beetles (Insecta, Coleoptera) are assertive indicators, since in addition to their abundance, several ecological niches and diets were identified (MAGURA et al., 2013; BICKNELL et al., 2014; FILGUEIRAS et al., 2015; NAGY et al., 2016; QODRI et al., 2016). Moreover, they have wide geographic distribution, are abundant and active, namely in neotropical systems, throughout the year (KOTZE et al., 2011). However, due to their huge diversity and taxonomic impediment, studies on some Coleoptera species in tropical regions are scarce (BASELGA; NOVOA, 2008; PIMENTA, 2011). Even for groups that have been extensively investigated there is lack of basic ecological information on its natural history, geographic distribution, and their role in ecosystem dynamics (NOVOTNY et al., 2006; MORAES et al., 2013).

Given the rate at which we are changing neotropical ecosystems and landscapes (DIRZO et al., 2014; PIMM et al., 2014) and the importance of tropical forests for

global ecosystem services (e.g. SAATCHI et al., 2007) it is critical to understand the response of ecological indicators, such as beetles to the ongoing trends. This will not only increase our understanding of ecological systems, but will also allow accurate predictions of the consequences of human-driven actions on biodiversity and ecosystem functioning.

Area of study

The study was developed in the municipality of Uruará, southwestern Pará state. The region is located approximately 1000 km away from Belém, the capital of Pará state, and is crossed by the Transamazon Highway (official designation BR-230) (Figure 1). Uruará was part of the Altamira PIC (Integrated Colonization Project), one of the first official settlements in the Amazon (ARIMA et al., 2005), created to resettle families from the south, southeast and northeast of Brazil (PERZ; WALKER, 2002).

Uruará experienced two main large waves of in-migration in its development history due to a combination of pull and push factors (PPERZ; WALKER, 2002; PERZ et al., 2010). The first wave of in-migration appears in the early 1970s (WALKER et al., 2002), where the military government demarcated 100-hectare lots for the old colonists to grow annual crops initially (PERZ; WALKER, 2002). It later diversified into perennials and pastures due to the household life cycles (PERZ; WALKER, 2002). The second wave of large in-migration occurred throughout the whole 1980s due to the stimulation of economic boom and raising prices of perennial crops (PERZ et al., 2010). A third wave of great in-migration is currently occurring, mainly due to the construction of the Belo Monte dam in Vitoria do Xingu and Altamira.

Land cover is characterized by large deforested areas radiating from the main road (TransAmazon) to the feeder roads (travessões), and spreading westward over time from the area of initial settlement in the east. The area has unevenly distributed patches of high-fertility soils known as terra roxa. Extensive livestock production, exploitation of timber at a large scale (mostly illegal) and cacao production are the main agro-pastoral strategies, but are usually complemented by annual crops and

horticulture. In some parts of the study area, sandy soils predominate and pasture and annual crop production are favoured (GODAR et al., 2012).

The municipality of Uruará has fairly similar physiographical conditions, with neighboring municipalities within a radius of approximately 300 km, a predominance of moist humid upland forests and a dry season from July to December (ALDRICH et al., 2006; GODAR et al., 2012).

In our study, we selected different areas that represented the main uses and land cover, namely (Figure 1 and Figure 2a-2e):

i. Native Vegetation – NV: Composed of vegetation whose facies is an upper canopy formed mainly by green trees, with crowns that touch each other, creating a dense and enclosed canopy all year round. In this ecosystem no traces of anthropic action were verified (for example, trails, residues, fires, selective cutting of wood, agricultural activities, among others) (Figure 2a).

ii. Maturing secondary succession - MS (vegetation with 15 years of regeneration): A secondary forest (or second-growth forest) is a forest or woodland area which has re-grown after abandonment of agriculture. Biological diversity gradually increases as long as there are primary remnants to supply seeds. The average height of the trees is over 12 meters and the average diameter is over 14 centimeters (Figure 2b).

iii. Early Secondary succession – ES (vegetation with five years of regeneration): Same as above. This phase typically lasts for up to six years and in some cases up to ten years, depending on soil quality and / or seed bank. The average height of trees is no more than four meters and the diameter of the main trees can reach eight centimeters (Figure 2c).

*iv. Agriculture - Ag (cocoa plantations, *Theobroma cacao* L.):* Agroecosystem represented by areas planted with cacao crops (*Theobroma cacao* L.) (Figure 2d).

v. Pasture for extensive livestock – Pa: Agroecosystem dominated by herbaceous vegetation (mainly species of the genus *Brachiaria* spp) used for extensive livestock farming (Figure 2e).

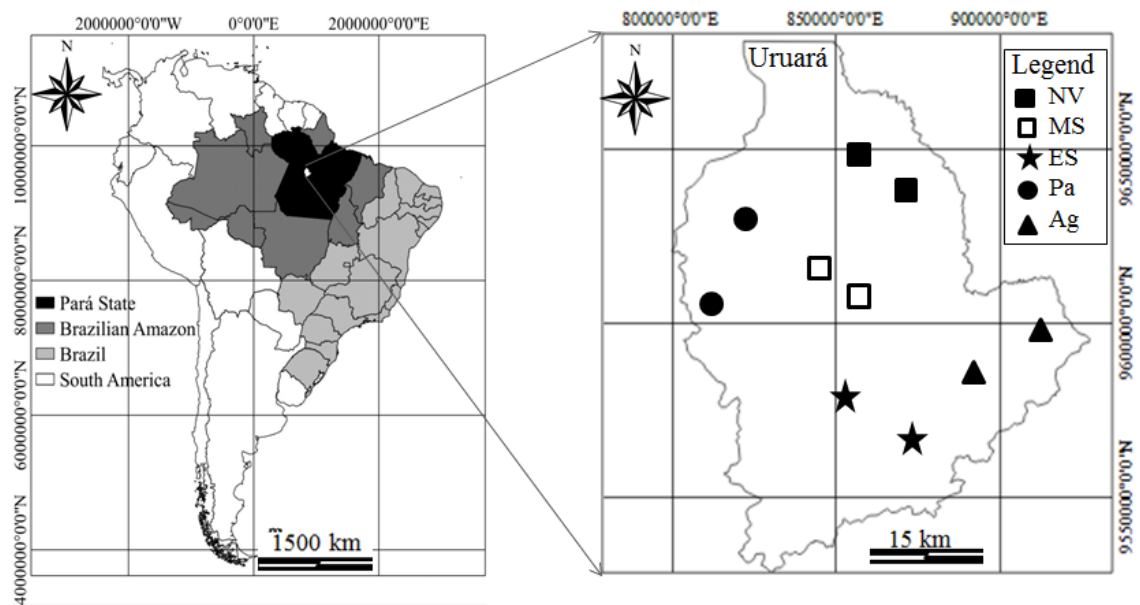


Figure 1. Location of the study region in the municipality of Uruará, state of Pará, northern Brazil. Location of the sampling areas: NV, Native vegetation; MS, Mature secondary succession; ES, Early secondary succession; Pa, Pasture; Ag, Agriculture.

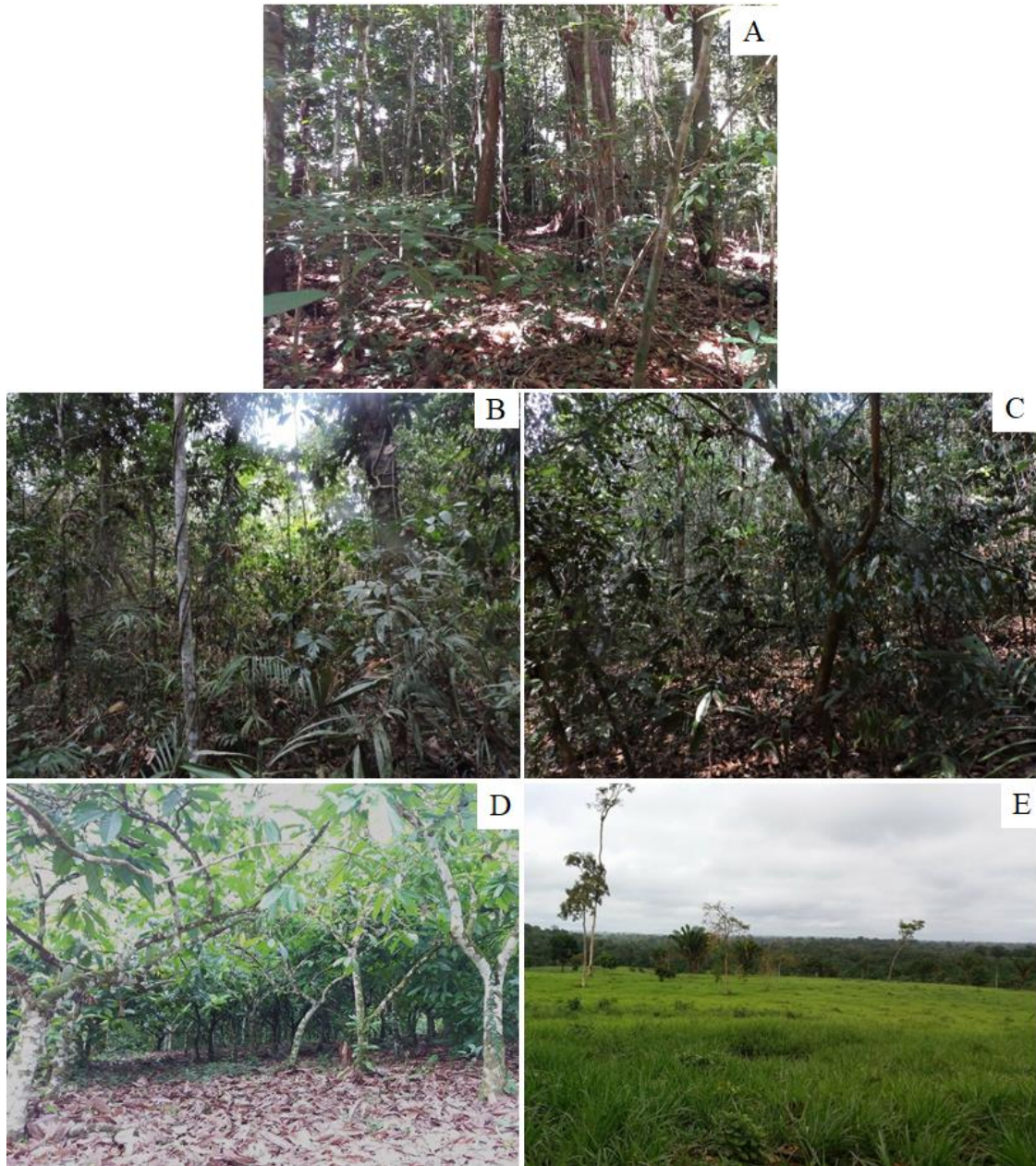


Figure 2. Habitats studied in the municipality of Uruará, Pará. (A) Native Vegetation, (B) Maturing secondary succession, (C) Early Secondary succession, (D) Agriculture - Ag (cocoa plantations), (E) Pasture.

General objective of research

Although each chapter of this thesis presents specific objectives (see *Structure of thesis/ specific objectives*), our main aim was to evaluate the responses of soil beetle assemblages to increasing levels of stress in the Amazonia, in the scope of its usefulness as ecological indicators of anthropogenic disturbance. Therefore, we selected areas with different land use/land cover (agriculture, pasture, native vegetation and secondary vegetation different stages of regeneration, see *Area of study*).

Structure of thesis / specific objectives

The thesis is divided in interrelated but stand-alone chapters. The overlap in the text content between chapters has been kept to a minimum; however, there is some common material presented, particularly in the introductions to the chapters, methods and references. All chapters were published or submitted to scientific journals and books. Although Univates has its own standard format for submission of academic papers, the present thesis, except for chapter one and chapter ten (which obeys the format of Univates), meets the specific standards of each journal.

This **first chapter** presents a general introduction to the topic, in addition to the structuring of the thesis.

The **second chapter**, entitled “**Assessing the potential role of ground beetles (coleoptera) as ecological indicators in tropical ecosystems: A review**”, is the introductory part of this thesis. It describes the ecological role of tropical forests (with emphasis to the Brazilian Amazon), the main causes of deforestation and the impacts caused by these changes. Based on extensive bibliographical review, we examined the use of soil invertebrates as ecological indicators, focusing on soil surface Coleoptera. The advantages and disadvantages of the use of beetles to monitor the effects of anthropogenic changes on the functioning of tropical systems were discussed. We also highlighted the main advances and applications, as well as proposals for future studies with this group.

In the **third chapter**, entitled “**Does the composition of Scarabaeidae (Coleoptera) communities reflect the extent of land use changes in the Brazilian Amazon?**” we examine the effects of anthropogenic impacts on Scarabaeidae

composition. The chapter objectives are to: (i) assess the effects of ecosystem substitution on neotropical Scarabaeidae assemblages and, (ii) demonstrate the use of Scarabaeidae as simple, suitable and intuitive ecological indicators for informing land use managers, namely by their ability of responding to key changes in such ecosystems and detecting levels of stress associated.

The **fourth chapter**, entitled “**Attraction of Scarabaeinae (Coleoptera: Scarabaeidae) to different baits in the Brazilian Amazon region**”, species richness and abundance of **Scarabaeidae** captured using various types of baits (human faeces, rotten banana, rotten meat and a nonbaited trap) was compared, to demonstrate attractiveness dissimilarities for several habitats of the Brazilian Amazon. In this study, species were classified in guilds for a more comprehensive analysis, according to their diet and habitat/nesting resources.

The **fifth chapter**, entitled “**Can dung beetles (Scarabaeinae) indicate the status of Amazonia’s ecosystems? Insights integrating anthropogenic disturbance with seasonal patterns**” focuses its emphasis on evaluating the seasonal patterns of **Scarabaeinae communities** in a gradient of ecosystems/ habitats for to determine their usefulness as ecological indicators.

The **sixth chapter**, entitled “**Seasonal patterns in the diversity of histerid beetles (Histeridae) are ecosystem specific? A case in Pará state, northern Brazil**”. The objective of this work was to characterize the seasonal variation in the community of Histeridae of within ecosystems in the Brazilian Amazon for to demonstrate the patterns of habitat preference and seasonal variations, and to verify their possibility to use as ecological indicators.

The **seventh chapter**, entitled “**Are disturbance gradients in neotropical ecosystems detected using rove beetles? A case study in the Brazilian Amazon**”, the responses of the Rove Beetles (Staphylinidae) to gradients of disturbance in ecosystems of the Brazilian Amazon were evaluated. Specifically, we assessed the possibility of using patterns in richness, abundance and taxonomic diversity of Rove Beetles as anthropogenic disturbance indicators.

The **eighth chapter**, entitled “**Diversity of Scolytinae (Coleoptera: Curculionidae) in different landscapes in northern Brazil**”, aimed to evaluate if the

composition and diversity of species of Scolytinae vary among the different habitats and between seasons with the aimed to support research, add information about these insects, that which are important from the economic point of view.

The **ninth chapter**, entitled “**How informative is the response of Ground Beetles’ (Coleoptera: Carabidae) assemblages to anthropogenic land use changes? Insights for ecological status assessments from a case study in the Neotropics**” the effects of anthropogenic impacts on Carabidae composition are examined. The main objective was to analyse the dissimilarity of GB assemblages by testing the following hypotheses: (1) the GB community composition, using morphospecies, is sensitive to disturbance of reference ecosystems such as primary forest; (2) increasing disturbance can be assessed by an increasing dissimilarity of the selected ecological indicators from pristine forests, namely morphospecies richness, abundance, taxonomic diversity and ecological composition. These hypotheses were tested through representative gradients in order to provide new insights for the GB diversity standard assessments as indicators of disturbance in Neotropical ecosystems.

The **tenth chapter**, final part of the thesis, provides a summary and synthesis of the findings from each of the preceding chapters, and considers their importance for management, conservation and restoration policies on neotropical forests, as well as highlighting future research needs.

Finally, I present at the end of the thesis the appendices that demonstrate the publications that resulted from this doctoral research.

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Chapter 2

ASSESSING THE POTENTIAL ROLE OF GROUND BEETLES (COLEOPTERA) AS ECOLOGICAL INDICATORS IN TROPICAL ECOSYSTEMS: A REVIEW

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ABSTRACT

Emerging concerns with environmental issues has led researchers to look for indicators capable of providing information on the condition of ecosystems. Several animal species have been used for this role. Among these organisms, ground-beetles were found to be appropriate indicators, since they are adapted to all terrestrial habitats, being associated with ecosystem function and sensitive to ongoing changes. Several families of ground beetles, e. g. Carabidae, Cicindelidae, Scarabaeidae, Cerambycidae, Histeridae, Staphylinidae and Curculionidae were used in diverse ecological assessment studies due to its main characteristics, such as habitat fidelity and taxonomic diversity, and because they provide cheap and easy measurements from functionally diverse trophic chains.

Although the growing number of studies on ecological indicators little is known about the ecological patterns in the communities of ground beetles of tropical ecosystems. The present study provides a review on the use of ground beetles as ecological indicators in tropical ecosystems, with focus on the Brazilian Amazon region. We discuss its use for monitoring the effects of anthropogenic induced changes, namely the conversion of forests to agriculture and farming practices, among others on the functioning of tropical systems. We also highlight the major advances and applications employed, as well as proposals for future studies with this group.

Keywords: Coleoptera, ecological indicators, tropical ecosystems

INTRODUCTION

Human action has caused directly or indirectly, the reduction and loss of biodiversity (Halffter, 2005) due to the change of the natural ecological processes and the emergence of new combinations of species, mainly by the habitats transformation, fragmentation and damage (Marques et al., 2002). Several factors have been identified including the suppression of natural habitats, ecosystem fragmentation and overexploitation of natural resources, climate change and the introduction of alien species. However, all these factors have an intimate relation with a primary cause, population growth (Ibama, 2004). This biodiversity loss threatens the sustainability of the entire system, with the consequent reduction of the environmental services, such as pest control, nutrient cycling and the maintenance of soil structure. In many situations the changes are enhanced by the use of pesticides, fertilizers and inadequate soil management that can generate even more negative impacts on the biota (Giller et al., 1997).

Insects have been shown to be appropriate ecological indicators for evaluating changes in the environment, due to several characteristics that they possess (Holloway et al., 1981). An excellent group are beetles (Coleoptera), the largest group of insects, with about four hundred thousand known species, distributed in more than a hundred families occupying practically all ecosystems and feeding on a variety food sources (Audino et al., 2007). Our knowledge of the diversity and distribution of species in the Amazon biome is still at an early stage of development (Silva et al, 2005; Peres, 2005). Although Amazonia has welcomed a steady stream of scientists since the beginning of the 18th century and a huge number of books and articles on its diversity are available (e.g. Pitman et al., 2007), they give a very fragmented picture of the region. Much of the region is still little explored by scientists and many of the specimens that have been collected to date have not yet been studied (Silva et al., 2005; Verweij et al., 2009). Understanding the extent and importance of insect diversity in the humid tropics is one of the biggest challenges in modern ecology (Godfray et al., 1999). The fact that only a fraction of tropical insects (certainly less than 20%) has been properly described is certainly not the least of the problems in this respect (Verweij et al., 2009).

Currently there is much speculation about the effects of human generated disturbance on the biodiversity of tropical forests, in particular, the impacts on species composition and on the ecological services provided (Whitmore and Sayer, 1992; Laurance and Bierregaard, 1997).

This chapter describes the ecological importance of tropical forests (with an emphasis on the Brazilian Amazon), the main causes of deforestation and the impacts caused by such changes. We examine the use of soil invertebrates as ecological indicators, with a focus on the Soil Surface Dwelling Beetles (SSDB), considering their sensitivity to landscape changes and their abundance. We discuss the advantages and disadvantages of its use to monitor the effects of anthropogenic changes on the functioning of tropical systems. We also highlight the major advances and applications employed, as well as proposals for future studies with this group.

CAUSES AND EFFECTS OF AMAZON DEFORESTATION

The Amazonian has circa 40% of the world's remaining tropical rainforests (Laurance et al., 2001) being considered a hotspot in biodiversity but also important for carbon storage and regional/global regulation of the hydrological cycles and climate (Fernandes, 2009).

Located in northern South America, the Amazon rainforest occupies 5.5 million square kilometers, spread over territories of Brazil, Venezuela, Colombia, Peru, Bolivia, Ecuador, Suriname, Guyana and French Guiana (Rivero and Covas, 2013). In Brazil, this type of ecosystem, occurs in the states of Amazonas, Amapa, Rondônia, Acre, Pará, Roraima and part of Mato Grosso, Tocantins e Maranhão (Figure 1).



Figure 1. Location of the Brazilian Amazon (Amazonia) in South America.

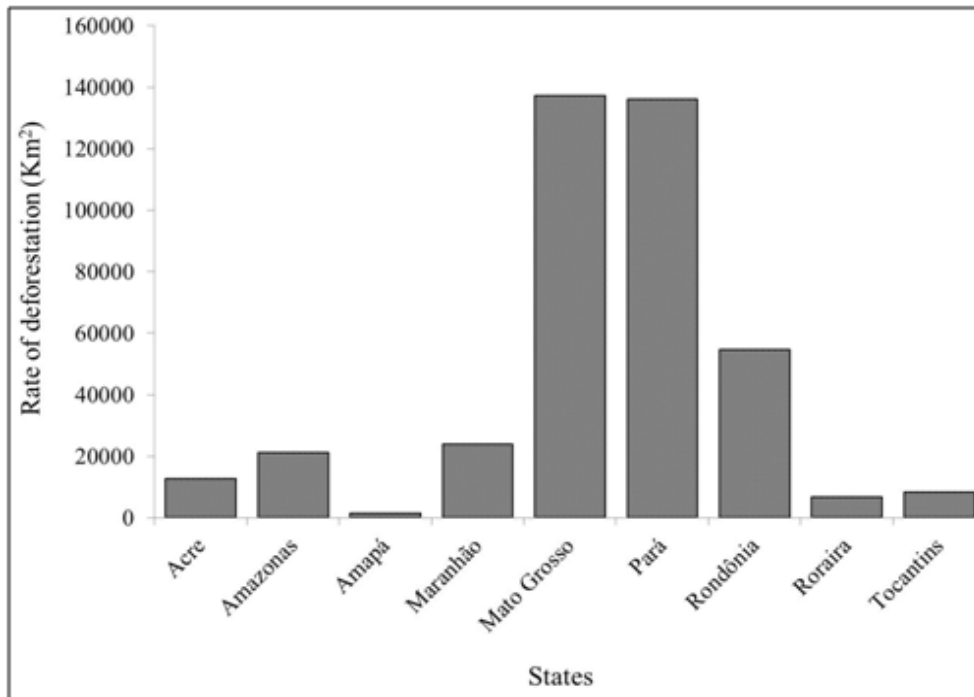


Figure 2. Cumulative rate of deforestation (Km²) between 1988-2013 in the Brazilian Amazon (Inpe, 2014).

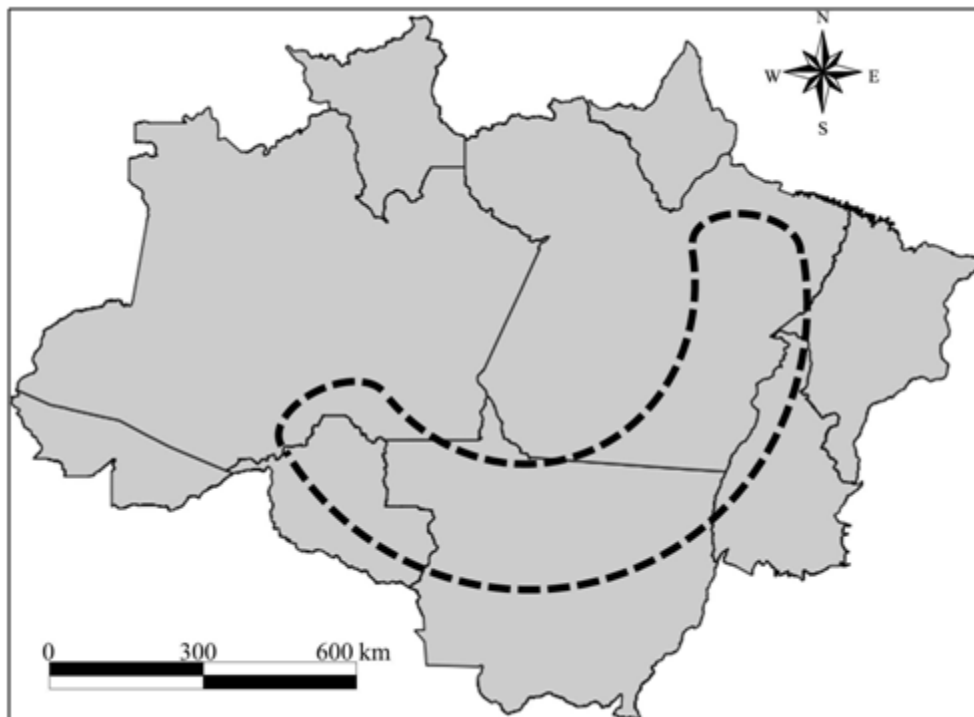


Figure 3. "Arc of deforestation" in the Brazilian Amazon - in dotted lines (adapted from Lui and Molina, 2009).

The deforestation of the Brazilian Amazon has attracted considerable attention over the last two decades (Souza-Rodrigues, 2011). According to satellite images, between 1988 to 2013 the accumulated rate of deforestation was 402.615 Km², divided by the states of Mato Grosso (137,251 Km²), Pará (136,127 Km²) and Rondônia (54,772 Km²) (Inpe, 2014) (Figure 2). Although large areas remain intact, the rate of forest loss is dramatic and future projections simulate its increase (Fearnside 2005a; Soares-Filho et al., 2005).

The current deforestation has focused on the axis set to the "Arc of deforestation" (Becker, 2004) (that goes from the south of Pará, passing to the north of Mato Grosso, Rondônia and southwest of Amazonas until it reaches Acre) (Figure 3), where deforestation has been buoyed by land speculation and, more recently, by increasing the areas occupied by exports such as soy and beef (Fernandes, 2009).

The process of deforestation in the Amazon was molded by settlement patterns of the past and driven by different factors. There are, however, temporal and spatial variations in deforestation (Laurance et al., 2001b) and the causes and dynamics are very distinctive for different locations. In fact it is difficult to generalize what drives or accurately predict the extension of the phenomena (Fernandes, 2009).

The causes of deforestation change constantly, being mostly associated with socio-economic pressures. The main is the rise in the population, previously dominated by economic extractive activities and family farming. Indeed, the population of the Amazon has increased considerably, from 2.5 million in 1960 to more than 20 million in 2000 (Ibge, 2000) further increasing the pressure on forests (Fernandes, 2009). Additionally new economic activities begin to stand out: ranching, logging, mining and intensive agriculture, besides the construction of hydropower plants (Angelo and Sá, 2007).

The advance of soy plantations in the region represents a major threat, encouraged by massive government investments in infrastructures, such as waterways, railroads and highways (Fearnside, 2001). The Brazilian Amazon produces more than a third of the country's soy and the state of Mato Grosso accounts for more than 20% of the national area of soybean cultivation (Ibge, 2006). The expansion of soy in the Amazon states totaled 14.1% per year (1990-2005) with major increases in the last five

years. The expansion of soybean production is causing serious deforestation, through the dynamics encompassed by forest burning/logging, implementation of livestock and subsequent conversion in intensive agriculture. With the increasing mechanization of production, several workers are excluded from these processes and tend to be displaced into marginal areas as contributing to moving forest frontiers or expanding the areas occupied by livestock and agriculture (Domingues and Bermann, 2012). According to Costa (2000) the cultivation of soy is only profitable if practiced in large areas, which demand large amounts of capital and technical expertise, generating a process of land and income concentration.

The logging of natural forests is another activity that plays a growing economic role in most of the Amazon region (Angelo and Sá, 2007). Forest is logged aiming first the trade of wood and possibly funding of economic activities that will be established next. When it comes to the implementation of another activity (Angelo and Sá, 2007), loggers acquire from ranchers the right to exploit logged areas (Veríssimo et al., 1995; Angelo and Sá, 2007). Most of the logging in the Amazon is illegal (Lentini et al., 2005), leading to the depletion of natural resources which could otherwise be used to generate sustainable socioeconomic development.

Due to the availability of cheap land for livestock, their numbers are expanding rapidly in the Amazon. Livestock is also present in small farms and it's the main land use associated with these properties. Increases in income expectations associated with livestock may have impacts on both types of agricultural systems of the Amazonia. That is, the increased expectation of profitability of livestock activity tends to have systemic effects with impacts on the rates of deforestation (Rivero et al., 2009). The expansion of livestock is associated with the increase of internal and external demands for beef, but is also influenced by other factors, such as reduction of transportation costs, increases in productivity and efficiency in management and the insignificant land price in the expansion regions of the Amazon (Rivero et al., 2009).

The construction of new roads and the rehabilitation of existing ones boost access to new areas, encouraging deforestation (Laurance et al., 2001; Nepstad et al., 2001; Soares-Filho et al., 2004) and in general anthropogenic pressures in areas adjacent to roads. The demographic distribution in the Amazon region shows a

tendency of concentration of the rural population next to urbanized areas with higher road density (Angelo and Sá, 2007). Of all deforestation that occurred in the Brazilian Amazon, 85% took place within a radius of 30 km from roads (Verweij et al., 2009). Many of these projects create corridors between densely populated areas and the remote Amazonian frontier (Laurence et al., 2001), facilitating the process of colonization, which subsequently leads to deforestation and other irreversible environmental effects. According to Cattaneo (2002) the reduction of transport costs would imply a significant increase in return on arable land, thus increasing the incentive to deforestation (Verweij et al., 2009).

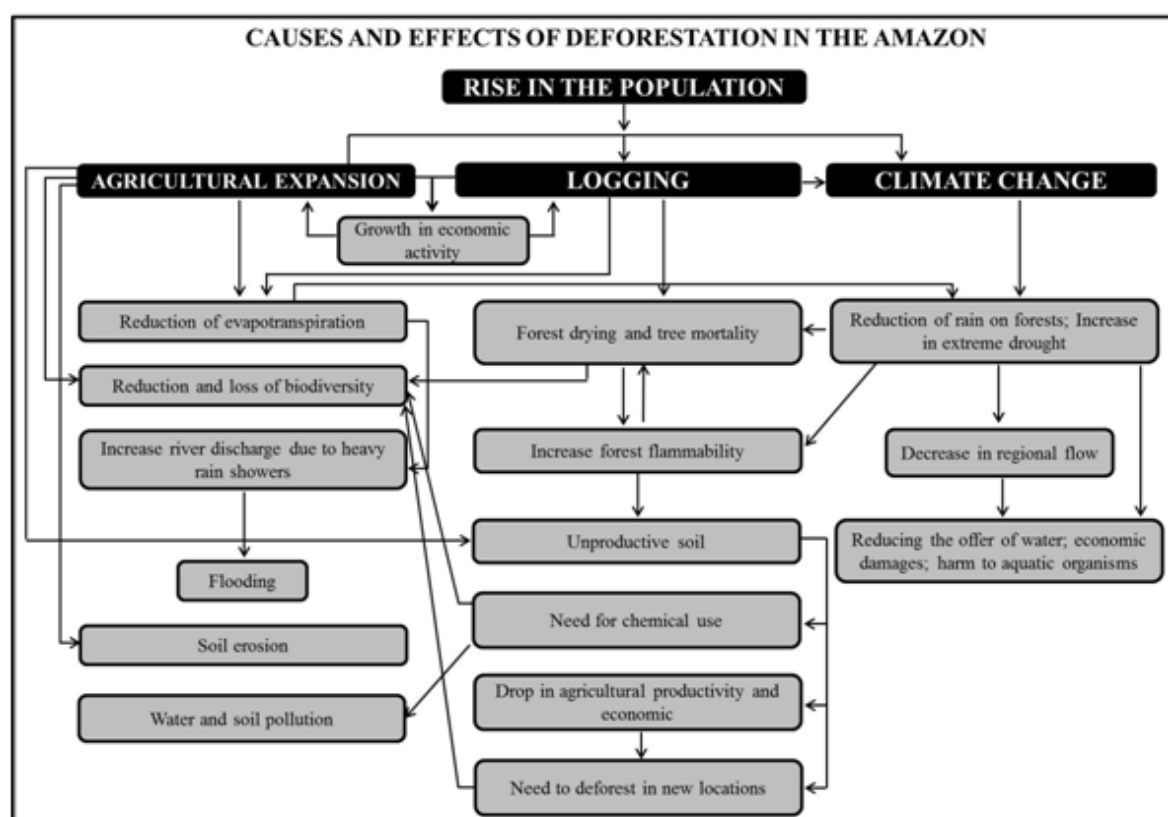


Figure 4. Flowchart showing the main causes and effects of Amazon deforestation in Brazil: The causes which lead to deforestation are black boxes; and consequences for human society are indicated by gray boxes (Adapted the Davidson et al., 2012).

Forests also are lost due to flooding from reservoirs associated with hydroelectric dams that usually tend to occupy vast areas (Fearnside, 1995, 2008). Besides the destruction of forests and riparian systems, hydroelectric projects have

much wider impacts in encouraging road building, new migration, boosting deforestation (Fearnside 2005b, 2006, 2008). There are plans, in the near future, for more than 70 new dams that will submerge and destroy a total area of forest of more than 100,000 km² (Verweij et al., 2009). Figure 4 illustrates the main causes and effects of deforestation in the Amazon.

SURFACE SOIL INVERTEBRATES AS ECOLOGICAL INDICATORS OF ENVIRONMENTAL CHANGES

To assess the state of degradation or conservation of an ecosystem, biologists use ecological indicators (Fränzle, 2006; Heink and Kowarik, 2010). These indicators must be highly sensitive to changes in the structure of an ecosystem, if possible providing information concerning the alteration in the structure and functioning on the system according to their presence, absence and/or abundance (Majer, 1981).

In this context, Lindenmayer et al. (2000) defines seven types of attributes with relevance as ecological indicators at the level of species, guilds and communities: (1) species whose presence indicates the presence of a set of other species, (2) key species whose addition to or loss from an ecosystem leads to major changes, (3) species whose presence indicates a biotic conditions of human change as air pollution or water pollution, (4) dominant species that provide much of the biomass or number of individuals in an area, (5), species that indicate particular environmental conditions, such as soil or certain types of rocks; (6) species considered sensitive and thus serving as an early warning of environmental change (also called bioindicators) and (7) a management indicator species, reflecting the effects of a disturbance regime or the effectiveness of efforts to mitigate particular disturbance. These seven types of attributes can be effectively classified in three classes of ecological indicators in function of their main assessment vocation: (1) biodiversity, (2), environment or (3) ecosystem integrity (McGeoch, 1998).

Biodiversity indicators provides descriptive data with ecological relevance, such as species richness, native vs. exotic composition and/or degree of endemism in a given habitat (Noss, 1990, Gaston and Blackburn, 1995; Cano and Schuster, 2008). Species or groups of species that exhibit predictable responses to environmental

degradation or to other changes in the environmental status, providing cheap and easy measurements if standard methodologies are applied, are termed environmental indicators (Venier and Pearce, 2004). Finally, ecological integrity indicators that demonstrate the effects of ecosystem changes, such as changes to habitats, fragmentation, climate change, pollution and other factors that generate impact on biota (McGeoch, 1998; Cano and Schuster, 2008). All three categories of ecological indicators cannot necessarily be exclusive and the most promising approach depends on their suitability for the investigation of key cause-effect relationships, as well as for the quality of the monitoring data and final diagnosis in supporting local management decisions (Van Straalen and Krivolutsky, 1996; Gerlach et al., 2013) (Figure 5).

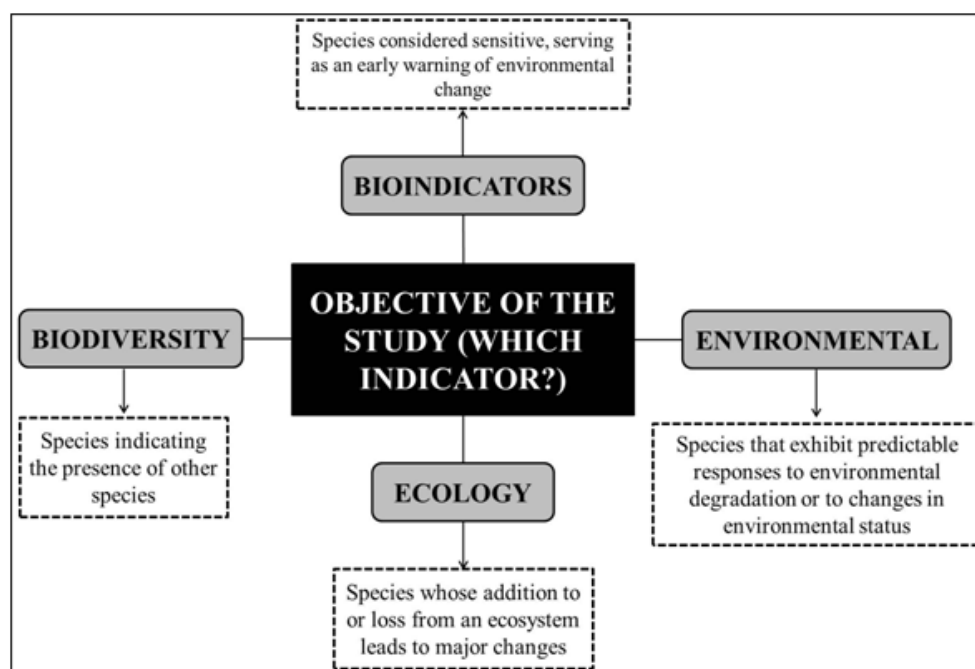


Figure 5. Types of ecological indicators.

The concept of bioindication was initially developed for aquatic environments while its application to terrestrial ecosystems gained momentum from 1980 (McGeoch, 1998). Subsequently, several criteria were proposed for the choice of suitable more generic ecological indicators (Figure 6) to be used also at the level of terrestrial systems (Pearson, 1994; Andreasen et al., 2001; Dale and Beyelerb, 2001; Freitas et al., 2006):

- i. Taxonomy relatively well provided – groups with taxonomic problems provide low quality information, because of the difficulty or impossibility of defining the boundaries between the species;
- ii. Knowledge about natural history, genetics, chemistry, and other aspects of biology – must have a recognized response to environmental change as well as responding clearly to the disturbance. In this way, must inform about the structure, functioning and composition of the ecological system and should be monitored in environmental disturbances of short- and long-term;
- iii. Convenient diversity - tremendous diversity can derail the process of sorting and analysis, while a very low diversity may compromise the quality of the data;
- iv. Short life cycle - in theory, the lower the time of generation, the more rapidly the effects of environmental changes are perceived.
- v. Ecological diversity - a group that has different species with ecological associations provides information on different compartments of the habitat.
- vi. Fidelity of habitat - species with little faithfulness of habitat do not provide specific information.
- vii. Close association to resources or other species – is not essential, but species closely related to each other are more informative (represent another taxonomic group or resource).
- viii. "Sedentary" relative - at one end, migratory species or very dispersive may be present in an environment without any relationship with the environmental conditions of the same.
- ix. Ease in sampling, sorting and identification, including for non-specialists.
- x. Little human use – groups whose species have a high economic value (for example, hunted for consumption or ornamentation) may disappear from the system regardless of their conservation status.

We highlight the surface soil invertebrates as good environmental and ecological indicators as their small size makes them sensitive to local conditions, while their mobility enables them to rapidly move in response to changing conditions (Samways et al., 2010), both at the habitat level and according to the gradients of environmental impact intensity (Oliver et al., 1998; Lewinsohn et al., 2005).

Additionally as insects they tend to be very diverse and abundant when compared with the groups of vertebrates and plants (Figure 7). Although historically neglected in conservation and monitoring strategies, they are now becoming an important asset in the disciplines of landscape ecology and conservation (McGeoch et al., 2011; Gerlach et al., 2013).

Studies have been developed to evaluate the behavior of soil invertebrates in degraded environments and how these organisms respond to different restoration measures serving as a tool for monitoring (e.g. Correia, 2002; Rovedder et al., 2009; Cajaiba and Silva, 2014). Soil invertebrates composition reflects the functioning of the ecosystem due to its intimate association with litter fall-soil system processes and its great sensitivity to environmental changes (Ruf et al., 2003; Yi et al., 2006; Hoffmann et al., 2009; Oliveira and Souto, 2011).

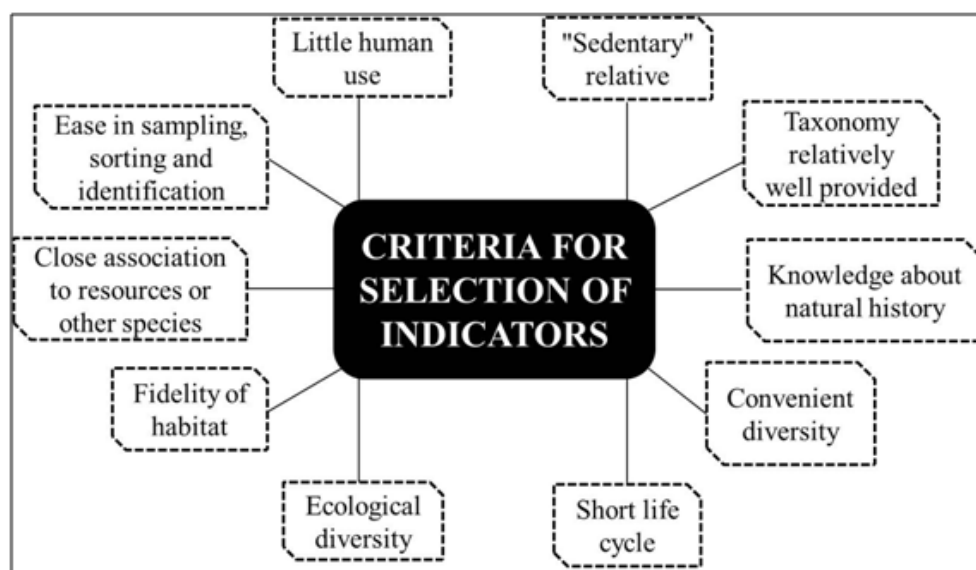


Figure 6. Criteria for selection of ecological indicators.

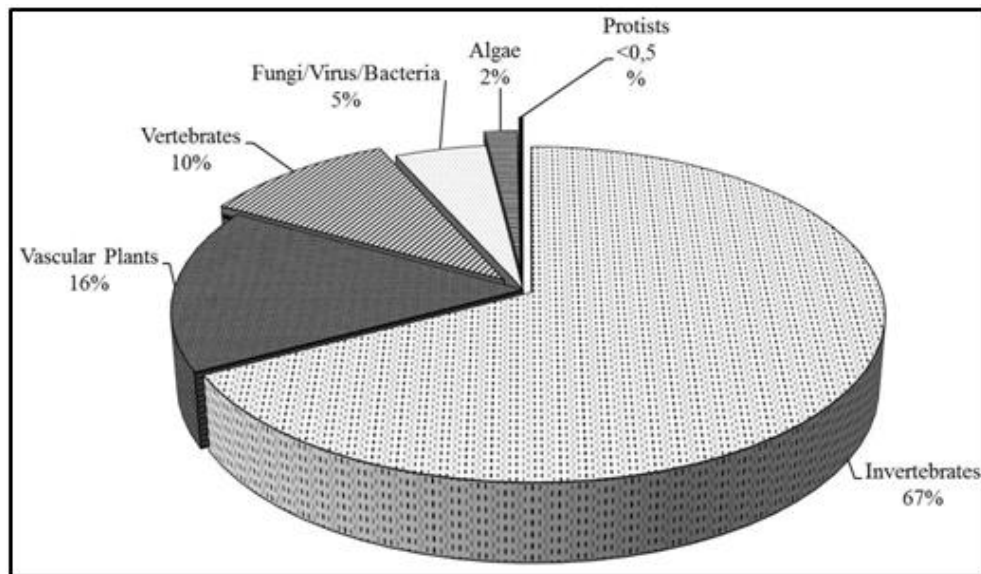


Figure 7. Diversity of invertebrate species compared to other indicators groups (Adapted the Calor, 2009).

Functional groups of soil fauna may disappear or be reduced (in their abundance and richness) as a result of soil degradation processes or be displaced by few opportunist organisms, highly adapted to disturbances (Lavelle, 1996; Loranger et al., 1998). Thus, environmental indicators must be highly sensitive organisms to changes in the structure of an ecosystem, featuring the integrity of the soil (Maluche et al., 2003; Lima et al., 2006).

SOIL SURFACE COLEOPTERA AS INDICATORS OF ENVIRONMENTAL CHANGES

The insects of the order Coleoptera, known popularly as beetles represent one of the most diverse animal life groups on Earth, with about 4000 thousand species described (Lara, 1992; Costa, 1999; Cajaiba, 2013), representing more than a third of all known insect species (Figure 8). They occupy most of the Earth's environments, with the exception of the open seas, exhibiting diverse diets and lifestyles (Grebennikov and Leschen, 2010).

Beetles have characteristics that qualify them as appropriate for ecological studies (Gaston et al., 1992; Marinoni and Dutra, 1997; Carlton and Robinson, 1998; Didham et al., 1998) monitoring different compartments of the system (Marinoni,

2001). Several studies suggest that beetles are strongly associated with the structure of the habitat and factors associated with microclimates (Halfpter and Mathews, 1966; Thiele, 1977; Schwarzkopf and Rylands, 1989; Bishop et al., 2009) that can be altered by anthropic activities. For example, beetles respond to environmental disorder (Koivula et al., 1999; Niemelä et al., 2000; Pearce et al., 2003; Pearson and Cassola, 2005, 2007; Kaiser et al., 2009; Negro et al., 2010; Vasquez-Velez et al., 2010), soil depth (Bultman and Uetz, 1984), soil disturbance (Bergeron, 1991; Pearce et al., 2003), local humidity (Epstein and Kulman, 1990; Rykken et al., 1997), temperature (Li and Jackson, 1996), soil pH (Paje and Mossakowski, 1984) and pollution (Garcia et al., 2010). Beetles were also used for indicating success in managing and restoration of habitats (Jacobs et al., 2010; Paoletti et al., 2010; Gomez, 2010).

Carabidae, Scarabaeidae, Cerambycidae, Staphylinidae and Coccinellidae are groups that have species with high ecological fidelity, diversified taxonomically and ecologically, easily collectable in large samples, easily identifiable and functionally diverse (Oliver and Beattie, 1996; Langor and Spence, 2006; Pearce and Venier, 2006). These families also form groups relatively well known with other species and resources (Brown, 1991; Vulinec, 2000; Nichols et al., 2008).

Monitoring changes has been investigated with the use of the family Scarabaeidae, subfamily Scarabaeinae mainly (Spector, 2006) due to its relation with the forest environment and mammals that inhabit it (Gill, 1991; Halfpter, 1991; Davis et al., 2001; Nichols et al., 2008). Scarabaeidae were also used as ecological indicator (McGeoch et al., 2002; Spector, 2006; Nichols et al., 2007, 2008), considering this group close relationship and dependency on the environment in which they live and of the resources available to the community (Halfpter and Arellano, 2002; McGeoch et al., 2002; Andresen, 2003; Spector and Ayzama, 2003; Larsen and Forsyth, 2005; Scheffler, 2005; Nichols et al., 2007; Almeida et al., 2011). Several studies demonstrate that the composition and structure of Scarabaeinae communities are affected in modified areas by human action (Klein, 1989; Didham et al., 1998; Halfpter and Arellano, 2002; Scheffler, 2005; Nichols et al., 2007), namely by habitat fragmentation and isolation (Klein, 1989; Andresen, 2003; Nichols et al., 2007; Escobar et al., 2008). In the tropical belt, their abundance and richness is heavily

influenced by the climate, namely by precipitation that sets the advent of most species (Halffter and Matthews, 1966; Gill, 1991; Hernández, 2007; Andresen, 2008).

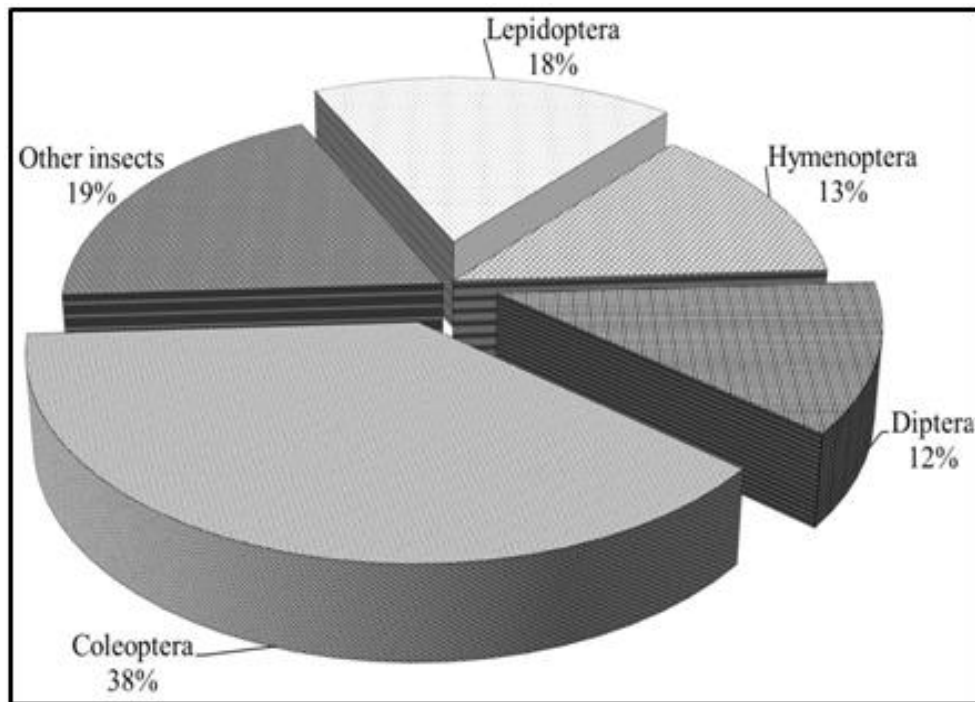


Figure 8. Diversity of beetles compared to the main groups of insects (Adapted the Cajaiba, 2013).

Kimberling et al. (2001), Escobar et al. (2007) highlight the Scarabaeidae sedentary nature and vulnerability to environmental changes (e.g. vegetation cover, vegetation type, fragmentation, habitat loss, physical structure, altitude of ecosystem, among others). The scavenger habit of these species may also indicate abundance/diversity of vertebrates in forest environments (Halffter, 1991). Size and biomass of Scarabaeidae are related to the presence of groups of vertebrates due to food scraps associated (Klein, 1989; Estrada et al., 1993; Scheffler, 2005).

The family Carabidae has been suggested to be used as indicator in studies with biodiversity and for evaluating habitat changes, for their diversity in morphological, behavioral, ecological and taxonomic traits, abundance and sensitivity to environmental changes (Heliovaara and Vaisanen, 1993; Niemelä et al., 2000; Koivula et al., 2002; Rainio and Niemelä, 2003; Niemelä and Kotze, 2009). This group is also used in assessing the sustainability of agricultural systems, being negatively affected by intensive agriculture, mechanized control of weeds and fire (Stork and Eggleton,

1992), urbanization, crop and forest management, overgrazing by domestic livestock, tourist flow in natural landscapes and soil pollution (Avgın and Luff, 2010). Their dominance is altered with the rhythm and phenology of the crop, as well as by the microclimate (Kromp, 1999). The group has also been used as indicator of metal contamination in soils (Paoletti et al., 1991; Avgın and Luff, 2010). Pearce and Venier (2006) demonstrate in his review that typical forest species tend to prefer lower temperatures and higher humidity, while open-habitat species are associated with for higher temperatures and lower humidity. Thus, large fluctuations in temperature and moisture levels can disrupt the assemblages of Carabidae (Pearce and Venier, 2006). Several disadvantages were also discussed, including seasonal variations, the uneven distribution and high number of generalist species. Also in tropical regions their taxonomy and ecology are little known (Rainio, 2009).

The family Staphylinidae, have been proposed as bioindicators or surrogates (Bohac, 1999; Anderson and Ashe, 2000; Pohl et al., 2007) given that they are abundant and diverse; occupy a great variety of habitats worldwide; are easy to collect, given that the majority of the species are found among leaf litter and fallen trunks, forming one of the commonest groups of insects and ecologically the most important of the soil-related fauna (Bohac, 1999; Navarrete-Heredia et al., 2002; Newton et al., 2005; Vasquez-Velez et al., 2010). Their abundance and diversity are dependent on the moisture of the soil foliage. For example, in studies conducted by García-Cárdenas et al. (2001) in fragments of forest, a positive relation was observed between richness of staphylinids and leaf litter moisture, as well as between richness and area of the fragments (García-Cárdenas and Ulloa, 2005). Preliminary evaluations suggest that richness and diversity depend, to a great extent, on the habitats with greater plant heterogeneity such as forests and riparian corridors (Bermúdez et al., 2006; Vasquez-Velez et al., 2010). This family is many times associated with locations containing important concentrations of potassium and phosphorus, which are frequent in soils containing organic material (Dunxião et al., 1999). Many species of Staphylinidae are also associated with the soil of agricultural crops (Sotherton, 1984), namely direct planting systems (Stinner and House, 1990). These organisms have already been cited in studies of population surveys on crops such as soybeans, cacao, guava and eucalyptus forest (Zanuncio et al., 1993). The biggest problem in the use of

Staphylinidae as ecological indicators lies in the difficulties of identifying the species based on the observation of internal structures (Hammond, 1997; Freitas et al., 2006).

Another family of Coleoptera which presents good characteristics to be used as ecological indicators of environmental integrity/disturbance is the Coccinellidae. The microclimatic conditions and dietary factors, accordingly with Iperti (1999), are important in determining the habitat of beetles of this family, because each species shows preferences to the type of vegetation stratification and its seasonal variations. The specie *Semiadalia undecimnotata* was considered very sensible to the increase in chemical and physical pollution, increased cultivation of cereal (which create problems on trophic chain) and the impact of climatic fluctuations (Wink et al., 2005). Thomazini and Thomazini (2002) noted that the number of species as well as individuals was considerably less on pasture than in other areas, when compared with primary woods and secondary vegetation. Despite the already mentioned features, there are few studies developed with this group in tropical regions.

The Cerambycidae are important part of forest ecosystems, occupying from the ground up to the treetops and, in the process of co-evolution between the communities of insects and flora, have also played the important role of pollinators (Hequet, 1996). These insects have been used for planning of conservation and monitoring, because they maintain a close relationship with the natural resources they use, being sensitive to environmental changes (Brown, 1997; Lewinsohn et al., 2005). According Marinoni and Ganho (2003), the population dynamics of some species of Cerambycidae (e.g. *Nyssodrysinia lignaria*) is influenced by high temperatures, humidity and daylight. This group is easily collectable by several types of traps (Monné and Hovore, 2005) and participates in the process of decomposition of dead wood, incorporating nutrients and allowing the renewal of forests through the opening of spaces and consequent seed germination. In Brazil, some areas such as the South and Southeast regions have important studies on the fauna of Cerambycidae, but in the Caatinga, Pantanal and especially the Amazon forest little is known about their natural history, behavior and ecology (Martins, 1999; Silva et al., 2005; Souza and Silva, 2012).

The beetles of the family Tenebrionidae are common, easily captured in pitfall traps, and straightforwardly identifiable when compared with other groups.

Communities of these beetles are dependent on the availability of specific resources such as debris, vegetation cover and various environmental characteristics of soil such as humidity, hardness and granulometric composition (Henschel et al., 2010). They have also been considered sensitive biodiversity indicators due to its sensitivity to habitat loss and degradation. Their abundance and diversity is affected to forest fragmentation, fires, climatic factors, among others (Parenzee, 2001; Cheli, 2009; Fattorini et al., 2011; Thakare et al., 2012). The family also responds to vegetation post-fire recovery (Fattorini, 2010). Despite these advantages, deeper studies are needed to determine whether changes in diversity of Tenebrionidae are purely seasonal and bioecological results or whether they are influenced by land use changes (Henschel et al., 2010; Saji and Al Dhaheri, 2011).

Another group of beetles widely used in studies of conservation/degradation is the Histeridae family. These beetles are functional predators, both in larval stage and adulthood, using wide-ranging environments for their food, and debris attract the scavengers that comprise important portion of its diet (Summerlin et al., 1991). Theoretically the community of predatory insects that occurs in decomposed matter maintains is high diversity in environments with complex vegetation structure possibility by the existence of superior biomass of vertebrates (suppliers of resources) in these locations (Holloway and Schnell, 1997; Smith and Merrick, 2001). Lopes et al. (2005) studying restinga vegetation in the State of Espírito Santo, Brazil, found that species richness of Histeridae was practically equal in each of the four areas studied (Restinga arboreal, Restinga open from *Clusia* spp, Restinga burned and Pasture), but the pattern of diversity was very different, with great diversity in the pristine restinga arboreal environment. The same authors observed a tendency of decrease of diversity (H') and equity (J') with a reduction of the complexity of the vegetation. Summerlin (1989) mentions the trend of reduced abundance and high richness of Histeridae in forest environments, as opposed to greater abundance and reduced diversity of Histeridae in open habitats of grassland. For better understanding, studies with this family are necessary, because, as pointed out by Degallier et al. (2011) the number of known species in tropical areas is diminutive.

The Table 1 (the appendices) presents a summary of the main families of beetles used as indicators, as well as their advantages and disadvantages.

SOIL SURFACE BEETLES AS INDICATORS OF ENVIRONMENTAL CHANGES IN THE TROPICS WITH A FOCUS ON THE BRAZILIAN AMAZON

Despite the recognized importance of using ground beetles as ecological indicators, its application in Brazil and more precisely in the Amazon, has been extremely limited. The effects of human actions (e.g. effects of deforestation, fire, grazing, logging and mining) on the functionality and structuring of ground beetles communities of the Amazonian forests are still poorly understood (Blanche et al., 2001; Dawes-Gromadzki, 2007; Andrade et al., 2011; Samu et al., 2011). Often, researcher's results are not comparable because of the lack of standardization in the sampling methodologies, hampering a direct comparison with other studies. In addition, there are few studies that examine long-term effects (Debinski and Holt, 2000; McGarigal and Cushman, 2002) owing practical difficulties and logistics of sampling and repetition in the same location and using the same methodology (Quintero and Halffter, 2009). As the number of forest areas to be searched or the total area under management increases, the number of researchers involved in surveys and the time required also increases, making surveys unfeasible (Aguilar-Amuchastegui and Henebry, 2007).

Studies with some families of Coleoptera such as Scarabaeidae, Carabidae, Staphylinidae have grown tremendously in the recent years, while for the other families most of the work are incipient. Amazonian Scarabaeidae beetles were studied in states Rondônia (Vulinec, 2000, 2002), Pará (Andresen, 2002, 2003; Scheffler, 2005; Gardner et al., 2008; Matavelli and Louzada, 2008; Vulinec et al., 2008; Barlow et al., 2010) and Amazonas (Vulinec, 2002; Quintero and Roslin, 2005; Radtke et al., 2007; Quintero and Halffter, 2009; Vulinec et al., 2009).

These studies highlight the effects of forest logging in the Amazon showing that abundance, species richness and composition of ground beetles is severely affected by forest fragmentation (Klein, 1989). According to the results of Imbrósio

(2012), that performed studies in different soil uses (eucalyptus plantations, native forest and selective cutting of wood) and Gardner et al. (2008) in eucalyptus plantations, native and secondary forest, the system with original forest cover sustains a community with a different functional structure namely by the reduction in the variability in the more humanized systems. According to these studies, the fauna of dung beetles are severely depleted in eucalyptus plantations and secondary vegetation compared to areas of primary forest. According to the authors, dung beetle communities that are closely associated with tropical forest habitats are greatly influenced by differences in vegetation structure (Halfpter & Arellano, 2002; Gardner et al., 2008), with individual species often having specific affinities for certain structural habitat properties (Davis et al., 2001; Gardner et al., 2008). Furthermore, secondary forests and Eucalyptus plantations, are typically characterized by low, relatively open canopies with hot and dry understory environments (Gardner et al., 2008). These microclimatic differences could help explain the observed impoverishment of dung beetle communities through a physiological intolerance to high temperatures (Chown, 2001; Campos and Hernández, 2013), the influence of solar radiation on adult activity patterns (Lobo et al., 1998; Moreira et al., 2009; Korasaki et al., 2013) or the influence of decreased soil moisture content on larval survival (Sowig, 1995; Gardner et al., 2008).

Teixeira (2006) analyzed the effect of isolation of forest islands and the effect of roads on the Scarabaeidae community noted that there was reduction of species in areas close to roads and in forest fragments when compared with intact and continuous regions. Vulinec (2002) in studies conducted in the States of Pará, Amazonas and Rondônia, noted that the diversity of most species of Scarabaeinae decreases in areas with high levels of impact (e.g. forest clearing and cutting), concluding that larger forest gaps affect the dynamics of this group. Scheffler (2005) examined the distribution and diversity of dung beetles (Coleoptera: Scarabaeidae) in intact forest and under three disturbance regimes (selective logging, clear-cutting and pasture), as a result, the author observed that Principal Components Analyses and the Sorensen's Index indicated that the intact and selectively logged areas were similar in terms of species composition but that the pasture areas and clear-cut areas had substantially different groups of species. Beetle size and diversity (as measured by species richness,

Shannon index and Simpson's index) were lower in clear-cuts and pasture than in intact forest. While the fact that selective logging has a relatively minor impact on the dung beetle community is heartening, the projected increase in the amount of highly disturbed landscape in Amazonia is expected to have a severe impact on dung beetle biodiversity in the region (Scheffler, 2005).

PERSPECTIVES AND CHALLENGES

Despite already having a reasonable number of researchers and publications, the use of ecological indicators in Brazil is in its initial stage. However, these studies were restricted mostly to the South and Southeast of the country. The Amazon has still few studies with ecological indicators/ground beetles, and can be considered insignificant when compared with its biodiversity and environmental problems. Systematics, general inventories, and testing the effects of habitat disturbance should be considered a priority by research groups engaged in biology and natural resources managing (Freitas et al., 2006).

While recognizing the qualities of Coleoptera for ecological integrity studies, several works should also try to solve the highlighted difficulties/disadvantages identified. The key problem is the taxonomic challenge, considering that a low proportion of species are known or described (Samways et al., 2010; Cardoso et al., 2011; Gerlach et al., 2013). The rates of knowledge differ markedly, hindering the identification by non-specialists, and some of the taxa proposed for indicators can only be reliably identified by taxonomic specialists (Behan-Pelletier, 1993; Báldi, 2003). Thus, a group of invertebrates can show excellent characteristics to be used as ecological indicators, but their widespread practice can be impeded by taxonomic difficulties (Hodkinson and Jackson, 2005). A solution to overcome these challenges is the new advances in molecular identification techniques (particularly DNA barcoding) (Janzen et al., 2005). DNA barcoding (Hebert et al., 2003) has been particularly successful in the identification and delimitation of new species from various groups (Hebert et al., 2004; Ward et al., 2005; Cywinska et al., 2006; Hajibabaei et al., 2006a, b; Smith et al., 2007; Borisenko et al., 2008; Kerr et al., 2009). This method has

received increased acceptance because it is simple and affordable (Padial and De La Riva, 2007; Pires and Marinoni, 2010). The advantages of the method would be the possibility of identifying individuals at any stage of development and the prospect of discriminating between morphologically identical species (Pires and Marinoni, 2010).

Another criterion that could "attenuate" this disadvantage would be sorting by morphospecies. This approach involves, above all, the creation of a reference collection of all new species found throughout the study. The fundamental key to this approach is that individuals of a species found in different samples are assigned the same species name (morphospecie). This allows the researcher to study the diversity and composition changes through a study, without having to wait for the species to be correctly identified (or described) (Gerlach et al., 2013), speeding the process until more accurate results are completed (Oliver and Beattie, 1996). Therefore, the approach by morphospecies is no substitute for taxonomy, just accelerate the studies and seems to provide good results concerning structuring and functioning of systems (Samways et al., 2010; Gerlach et al., 2013). Added to these factors, some impediments to the use of ground beetles as indicators (Cardoso et al., 2011):

- i. Public dilemma – invertebrates and their ecological services are mostly unknown to the public;
- ii. Political dilemma – policymakers and stakeholders are unaware the problems of invertebrate conservation;
- iii. Scientific dilemma – the study of soil invertebrates is scarce and underfunded.
- iv. Linnean shortfall – most species are not described;
- v. Wallacean shortfall – the distribution of species described is virtually unknown;
- vi. Prestonian shortfall – the abundance of species and their changes in time and space are unknown;
- vii. Hutchinsonian shortfall – the ways of life of the species and their sensitivity to changes in habitat are largely unknown.

Besides the above-mentioned factors, many of the challenges of research applied to soil beetles could be significantly improved through a greater effort to

collect standardized data sets on the species and patterns of functional response to disturbance. This could include the creation of a wide range of public available resources, including a comprehensive online catalog, new keys to genera and species and standardized survey protocols (Nichols and Gardner, 2011). Additionally, greater financial support to researchers and incentives for conducting research in more remote regions would help in solving these challenges.

FINAL REMARKS

In the last decade, there has been a growing recognition of the value of using ecological indicators for monitoring changes in terrestrial environments. Ground beetles are good indicators of biodiversity and environmental quality in a variety of spatial and temporal scales. This is because they are sensitive to climatic and ecological conditions, as well as to edaphic, physiognomic, trophic and microclimatic factors (Davis et al., 2004).

However, before choosing suitable ecological indicators, certain steps must be followed. It is necessary to define clearly the goals and endpoints of the study (McGeoch, 1998). It must be determined what to measure, how it will be measured, and why. Account must be taken in accordance with the nature of the problem, even if it is a response to a single pollutant in a restricted location or an attempt to compare the biodiversity of a wider area. The lack of accuracy at this stage will result in inadequate indication and results of little reliability (Hodkinson and Jackson, 2005). Only after establishing the groundwork it's possible to select the indicator group appropriate to support the rigorous protocols required (Hodkinson and Jackson, 2005).

Sometimes taxa that are good indicators at a spatial scale could lose their reliability at higher or lower scales (Allen et al., 1999). Instead of focusing on a few indicator species, more reliable information can be obtained from studies of a set of species/groups or at the community level, with measurements made not at the level of presence/absence but as numbers, biomass and dominance. The use of trophic guilds as detritivores, predators, parasitoids, decomposers, saprophagous, among others, may also reveal interesting differences in the landscape (Paoletti, 1999).

Selecting the appropriate indicator taxon is a compromise between advantages and drawbacks. The success of using ground beetles is based on cost-effective data collection and sensitivity to different environmental factors (Rainio and Niemelä, 2003). Despite the advantages and features already mentioned and the ecological importance of beetles, species identification is still very difficult for some groups, which added to the lack of systematic studies and natural history causes some beetles to be unsuitable for indicator studies (Gaston et al., 1992; Marinoni and Dutra, 1997; Freitas et al., 2006). However, these features are common to most invertebrate groups.

Finally, many existing studies on the change of habitat suffer from methodological limitations that may interfere with the interpretation of results (Gardner et al., 2007a) and such sampling problems are increasingly recognized in studies of biodiversity in general (Hamer and Hill, 2000; Hill and Hamer, 2004; Barlow et al., 2007a, b, c; Gardner et al., 2007b; Koh, 2007; Gardner et al., 2008). We stress here the importance of inventories well conducted, suggesting further discussion about general protocols and standardization of methods, taking into account the particularities of each group. The differences of method and the absence of general protocols, scientifically tested and feasible, can simplify complex realities, leading to wrong decisions (Silveira et al., 2010). However, more consistent studies it is necessary for understand the relationship between beetles and environment, and further explore the technique of beetles as ecological indicators, which might be applied to monitoring and evaluation of environment.

APPENDICES

Table 1. Advantages and limitations of ground beetles used with potential use as ecological indicators

Family	Main studies	Advantages	Limitations	Reference
Carabidae	Agriculture; Forest fire; Pollution by metals; Urban ecology; Insecticides; Climate change.	Each type of habitat has given set of species with generalist and specialist species. Thus, species of Carabidae individual or sets of species can be used as bioindicators.	Due to the high number of carabidae generalist species have been criticized as bioindicators.	Basedow (1990); Eyre and Luff (1990); Paoletti et al. (1991); Stork and Eggleton, (1992); Halme and Niemelä (1993); Butterfield (1996); Rykken et al. (1997); Venn (2000); Niemelä et al. (2000); Niemelä (2001); Maryański et al. (2002); Mozdzer et al. (2003); Ermakov (2004), Gongalsky et al. (2004); Schwerk et al. (2006); Belskaya and Zinoviev (2007); Cárdenas and Hidalgo (2007).
Scarabaeidae	Deforestation; Fragmentation; Logging; Agriculture; Reduction of rainfall; Livestock; Reduction of mammals.	Can represent to other animals (vertebrates); Easily collectable for several types of traps; Close relationship and dependency on the environment in which they live.	-	Howden and Nealis (1975); Wolda (1987); Klein (1989); Nummelin and Hanski (1989); Estrada et al. (1993); Hill et al (1995); Intachat et al. (1999); Nummelin (1996); Andresen (2003); Scheffler (2005); Nichols et al. (2007); Hernández (2007); Escobar et al. (2008); Andresen (2008).
Staphylinidae	Fragmentation; Agriculture;	Easily collectable for several types of	The biggest problem in the	Sotherton (1984); Stinner

Family	Main studies	Advantages	Limitations	Reference
	Fertilization of soil; Seasonality; Insecticides; Herbicides.	traps; Close relationship and dependency on the environment in which they live.	use of Staphylinidae as biological indicators lies in the difficulties of identifying the species, since this is often subject to the observation of internal structures.	and House (1990); Zanuncio et al. (1993); Dunxião et al. (1999); Bohac (1999); Hunter (2002); Büchs (2003).
Coccinellidae	Climatic conditions; Food factors; Urbanization; Use of pesticides; Deforestation; Physical and chemical pollution.	Each species shows preferences to the type of vegetable strata and different seasonality.	Few studies developed with this group in tropical regions.	Iperti (1999); Thomazini and Thomazini (2002); Wink et al. (2005).
Cerambycidae	Planning of conservation and monitoring; Influence of temperatures and humidity	Easily collectable for several types of traps.	Little is known about their ecology, mainly in tropical forests.	Hequet, 1996; Brown, 1997; Marinoni and Ganho (2003)
Tenebrionidae	Climatic factors; Effects of fire; Influence of land use.	Are relatively plentiful, easily captured in pitfall traps and readily identifiable when compared with other groups. Sensitive to the change of biodiversity	Few studies developed with this group in tropical regions.	Parenzee (2001); Cheli (2009); Fattorini et al (2011); Fattorini (2010); Henschel et al. (2010); Saji and Al Dhaheri (2011); Thakare et al. (2012).
Histeridae	Effect of fire; Pasture; Replacement of natural landscape by agricultural activities.	Close relationship and dependency on the environment in which they live.	Few studies developed with this group in tropical regions.	Holloway and Schnell (1997); Smith and Merrick (2001); Lopes et al. (2005)

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Chapter 3

Does the composition of Scarabaeidae (Coleoptera) communities reflect the extent of land use changes in the Brazilian Amazon?

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Abstract

Scarab beetles (Coleoptera: Scarabaeidae) have been used to investigate the effects of environmental disturbances on forest structure and diversity. This group is recognized as sensitive to habitat perturbations and ecosystem changes. Here we examine the effects of anthropogenic impacts on Scarabaeidae composition, testing the following hypotheses: (1) Scarab beetle communities react to land use disturbances with predictable trends, (2) disturbed habitats are able to retain only a part of the Scarab beetle community of native forests or late secondary forests; (3) habitats largely differ in terms of species richness, taxonomic diversity and ecological composition, supporting exclusive and indicator species. We selected areas of native forest, agriculture, pasture for extensive livestock and secondary forests in different stages of regeneration. Our results show that the Scarabaeidae species were not indifferent to the gradient of structural changes represented by the studied areas. In fact, their patterns of habitat preference reveals communities more abundant and diverse in pristine habitats. In contrast, disturbed habitats, dominated by agricultural activities and pasture, indicated clear detrimental effects on the abundance of all forest Scarab beetle specialists. On the other hand, the generalist species, mainly associated with open environments, seemed to be favoured by the prevailing conditions induced by agricultural activities. Overall, the composition of the Scarab beetle communities is variable and sensitive to those structural gradients and, therefore, capable of responding as useful ecological indicators for assessing the extent of land use change or degradation.

Keywords: Land use change; Indicator species; Landscape heterogeneity; Scarabaeidae; Brazilian Amazon.

Introduction

There is enormous concern and speculation about the effects of human disturbance in the biodiversity of tropical ecosystems, namely the impacts on species composition and the modifications in the ecological services provided (e.g. Morris, 2010; Aerts and Honnay, 2011; Cajariba et al., 2015a). The deforestation in the neotropics, namely in the Brazilian Amazon (Amazonia), has been causing a sharp erosion of biodiversity and the disruption of complex global climate phenomena (e.g. Hassan et al., 2005; Morris, 2010). Therefore, the conservation of the Amazonia' ecosystems represents a growing core challenge for sustaining earth's functioning and ultimately humankind (e.g. Viana and Pinheiro, 1988; Viegas et al., 2014). Even though a steady stream of scientists and a large number of publications and projects have been investigating this problem, they provide a very fragmented picture concerning the environmental changes and its ecological consequences. Furthermore, considering its vastness, most of the Amazonia' ecosystems are still marginally studied (Verweij et al., 2009; Teston et al., 2012).

Terrestrial invertebrates and especially insects play a crucial role in most ecological processes and are key components of ecosystems' structure and functioning (Bicknell et al., 2014; Viegas et al., 2014; Cajariba and Silva, 2015; Cajariba et al., 2015b; Campos and Hernández, 2015). In this context, insects' abundance and richness are related with other taxa, climate and soil characteristics, thus representing potential target indicators of environmental changes (e.g. Nichols et al., 2008; Cajariba et al., 2015a). Therefore, the understanding of the ecological relevance of insects in the humid tropics could even support decision-making and robust management/recovery of imperilled ecosystems in the scope of the need for rapid, standardized and cost-saving assessment methodologies (Godfray et al., 1999). Among insects, ground beetles of the family Scarabaeidae (Scarab beetles) share several features that make them highly appropriate for ecological studies, namely because they usually occur in high densities, they are functionally diverse in the tropical food webs, they are sensitive to landscape and habitat changes, and they provide cheap and easy measurements if standard methodologies are applied (Nichols et al., 2013; Bicknell et al., 2014; Viegas et al., 2014; Cajariba et al., 2015b; Filgueiras et al., 2015).

The diversity of Scarab beetles (SB) is deeply interrelated with the structure of the habitats, with most species being highly sensitive to microclimate variations (Nichols et al., 2008; Lopes et al., 2011). In fact, SB communities respond quickly to changes in the vegetation canopy and vegetation diversity, fragmentation and/or isolation of tropical forests remnants (e.g. Nichols et al., 2007; Viegas et al., 2014; Campos and Hernández, 2015). Light intensity and humidity (Nichols et al., 2008) determined mostly by vegetation cover, soil type (Gardner et al., 2008) and the availability of faeces as a food source (Davis and Philips, 2009) are considered determinants for SB distribution. Their dependence on the faeces of vertebrates makes them also useful to monitor mammal communities (Estrada et al., 1999; Nichols et al., 2007). Additionally they are a low cost trap with relative straightforward identification (Gardner et al., 2008). Few studies relate the effects of pristine forest substitution by other anthropogenic ecosystems, such as pastures or agriculture, in the composition of SB communities in the Amazonia (Scheffler, 2005; Quintero and Halffter, 2009; Silva et al., 2014). We studied SB assemblage through habitat gradients ranging from primary forest to secondary forests, cocoa plantations and pastures in the Amazonia context. We based our analyses on three main hypotheses: (1) SB communities react to disturbances with predictable trends, (2) disturbed habitats are able to retain only a part of the SB community of native forests or late secondary forests, (3) habitats largely differ in terms of species richness, taxonomic diversity and ecological composition, supporting exclusive and indicator species compositions. Specifically, in this study we intended to: (i) assess the effects of ecosystem substitution on a neotropical SB assemblage and, (ii) demonstrate the use of SB as simple, suitable and intuitive ecological indicators for informing land use managers, namely by their capability of responding with rigour to key changes in such ecosystems, namely by detecting different levels of impacts.

Material and Methods

Study area

This study was performed near the city of Uruará, state of Pará, northern Brazil (Fig. 1). The territorial extension of the municipality is 10796 km² and its population encompasses circa 44789 inhabitants. The dominant land use/ land cover (LU/LC) is

natural forest (69% of the area) and deforestation is concentrated mainly in the south-central part of the territory and near the main roads. Extensive livestock production and the exploitation of timber at a large scale (mostly illegal) are currently considered the most serious environmental threats (Cajaiba et al., 2015b). The studied areas contain the pertinent gradients, in terms of biophysical and ecological characteristics, for testing the response of Scarab beetle (SB) communities (Cajaiba et al., 2015b) facing the main anthropogenic drivers. These gradients encompass: Native Vegetation – NV, Early Secondary succession – ES (vegetation with five years of regeneration), Mature Secondary succession - MS (vegetation with 15 years of regeneration), Agriculture - Ag (cocoa plantations, *Theobroma cacao* L.) and Pasture for extensive livestock - Pa. The climate of the study area is classified as Aw (Köppen), hot and humid and the average annual rainfall is 2000 mm (Cajaiba, 2014).

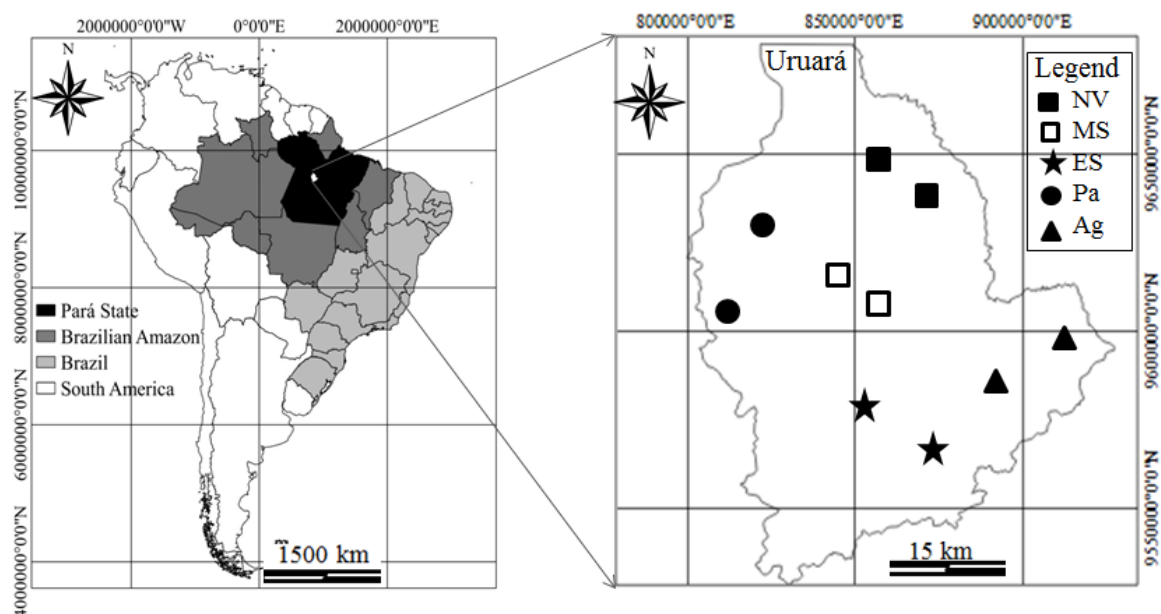


Fig 1. Location of the study region in the municipality of Uruará, state of Pará, northern Brazil. Location of the sampling areas: NV, Native vegetation; MS, Mature secondary succession; ES, Early secondary succession; Pa, Pasture; Ag, Agriculture.

Scarab beetle sampling

Sampling was carried out during the year 2015, in the months of February/March (rainy season), June (final of rainy season and early dry season) and September/

October (dry season). This allowed integration of annual seasonal differences in the activity of SB (Kasule, 1968). The sample points were placed at a minimum distance of 100 meters from ecotones, to ensure that most beetles captured were associated to the ecosystem in study (Buse and Good, 1993). Pitfall traps with 75 mm diameter and 110 mm deep were filled with preservative liquid consisting of formalin, alcohol, water and a few drops of detergent to break the surface tension. A roof was attached to each trap to prevent rainwater from entering the trap, remaining installed for 48 h prior to collection. Each pitfall contained different baits such as human faeces, meat and banana in order to attract different species according to their feeding habits (non-baited pitfalls were used as control).

In each study site (NV, MS, ES, Ag and Pa) seven sample points were placed 100 m apart. Each sample point contained four pitfall traps including the different baits (faeces, carrion, banana and non-baited), separated by 5 meters. The distance between pitfall traps allowed individuals to choose their preferential food resource (Almeida and Louzada 2009, Silva et al 2012). This protocol was applied to all areas and periods of collections, totalizing a sampling effort of 840 traps. The SB specimens collected were conserved in 70% ethanol and subsequently preserved in entomological blankets and identified in the laboratory to the species level where possible or assigned to morphospecies. In order to complement the pitfall derived information (Cajaiba et al., 2014), specific methods were used to collect leaf litter invertebrates. Ten random sampling points of 1 m² were selected in each ecosystem and sampling period. At each collection point ('litter-only'), only the loose soil was gently scraped with a metal trowel, to include those SB into the samples that fell out of the leaf litter during this collection procedure. We refrained from simply separating leaf litter and soil, considering that when we remove the litter, SB might flee the litter and hide in the loose topsoil.

Environmental variables

Fourteen microclimate and habitat variables were measured by location (Felton et al., 2006): Temperature-T, Humidity-H, Precipitation-P, Circumference at Breast Height-CBH, Circumference at Ankle Height-CAH, Canopy Cover-CC, Number of Plant Species-NPS, Number of Plants-NP, Number of Species of Shrubs-NSS, Number

of Shrubs-NS, Percentage of Exposed Soil-PES, Percentage of Green (vegetation) Cover-PGC, Percentages of Leaf Litter Cover-PLC, Height of Leaf Litter-HLL. The T, H and P of each point were measured during the traps installation and removal with a portable weather station (model Oregon Scientific WMR200A). To assess the environmental complexity of each sampling site, the quadrat-section method was adopted (Campos and Hernández, 2015). Using a cross as a reference, four quadrants (northeast, northwest, southeast, southwest) were marked and, in each quadrant the following variables were measured: the distances to the centre of the cross, height, crown diameter and trunk diameter of all trees with circumference at breast height (CBH) greater than 15 cm and all shrubs with CAH less than 15 cm and height greater than 1 m. Trunk diameter was taken at breast height (1.3 m) for the trees and ankle height (CAH = 0.1 m) for the shrubs. In each quadrant, the HLL in 1 m × 1 m marked square (using PVC pipe) was measured with a ruler, and the percentages of PLC, PGC and PES were measured by visual estimation using the following classes, 0–5%, 6–25%, 26–50%, 51–75%, 76–95% and 96–100% (Campos and Hernández, 2015). Using these same classes, the percentage of CC in the four quadrats was calculated with a convex spherical densiometer (D) Lemmon (Lemmon, 1954).

Assemblage analysis

Species richness, Abundance, Shannon-Weiner diversity index and dominance Berger Parker index of each sampling site were measured, and differences among sites were gauged using One-Way-Analysis-of-Variance (ANOVA) and subsequent Tukey post hoc tests to check for specific differences. Before applying ANOVA, the normality of the data was verified by the Shapiro-Wilk test. The taxonomic composition of SB communities between sites was compared using Permutational Multivariate Analysis of Variance (PERMANOVA).

Association of Scarab beetle assemblages with environmental variables

Canonical Correspondence Analysis (CCA) was used to estimate the influence of the fourteen environmental variables on SB communities. Principal Components Analysis (PCA) was used to reduce variables to 4 principal components (PC). The CCA was performed with the Bray-Curtis dissimilarity index on two axes.

IndVal

To determine possible indicator species, single value indicator (IndVal) developed by Dufrêne and Legendre (1997) was calculated, combining specificity (patterns of relative abundance) of a given species in a given environment with its fidelity within that environment (patterns of incidence). Species with a high specificity and high fidelity within a habitat are considered to achieve the highest indicator value. Only taxa with IndVal>25% were saved in the final lists (Dufrene and Legendre, 1997). This analysis was performed in R 3.2.4 program (R Core Team, 2016) using the *indicspecies* package 1.7.5 (De Cáceres and Jansen, 2015) with 9999 permutations, using data for beetle abundance.

Results

Assemblages analysis

A total of 15109 Scarab beetles were captured, distributed by nine genera and 112 species. From these, 86 species (8804 specimens) were identified in the Natural Vegetation (NV), 69 species (3941 individuals) in the Mature Secondary succession (MS), 33 species (707 individuals) in Early Secondary succession (ES), 38 species (1219 individuals) in Pasture (Pa) and 33 species (438 individuals) in Agriculture (Ag). The genus *Ateuchus* (15 species) and *Canthon* (19 species) were prevalent in all communities, with 43 percent of all specimens captured (Appendix A). Rarefaction curves for all sites reached their asymptotes values, supporting the sampling effort undertaken (Fig. 2).

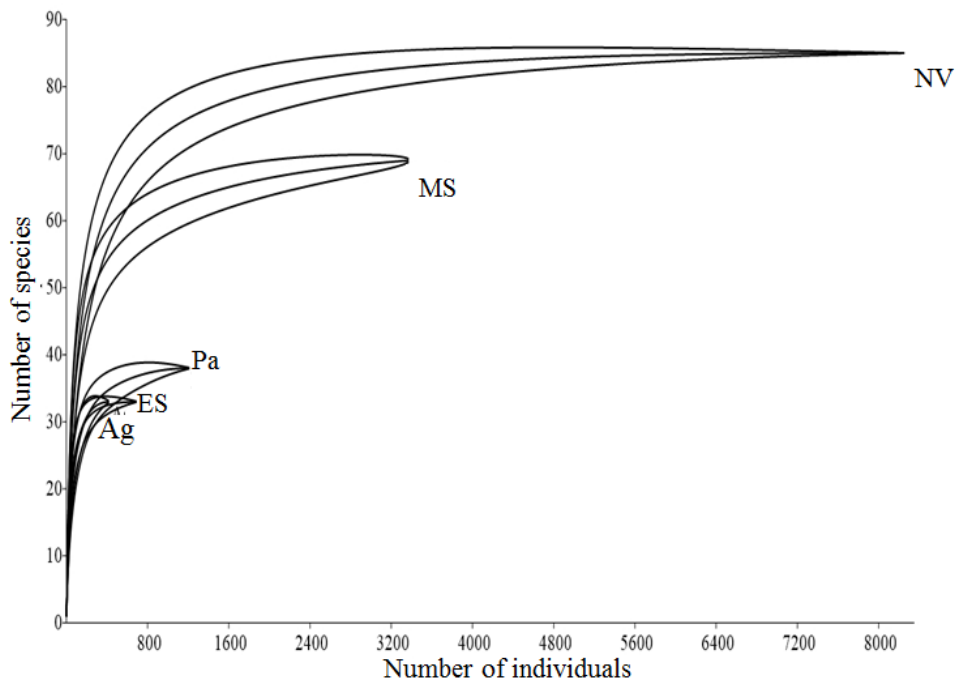


Fig. 2. Scarab beetles individual-based rarefaction curves for the studied sites (centre lines). External lines indicate 95% confidence intervals. NV, Vegetation native; MS, Mature Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture; Pa, Pasture.

The species richness varied significantly between the study sites ($F_{4,205} = 106.74$, $p < 0.01$). The average Scarab beetle (SB) richness was significantly higher in NV ($p < 0.01$), differing from the other habitats considered. The second largest species richness was observed in MS, although less than NV but higher than the other sites categories ($p < 0.05$). There was no difference in richness between ES, Pa and Ag ($p > 0.05$) (Fig. 3a). The abundance of SB varied significantly between the study sites ($F_{4,205} = 65.72$, $p < 0.001$). Mean abundance was significantly higher in NV ($p < 0.01$), differing from the other sites. The second largest abundance was observed in MS, being less than NV and higher than the others ($p < 0.05$) (Fig. 3b).

Shannon diversity reached higher values in NV ($H' = 3.73$), followed by MS ($H' = 3.64$), while Ag and Pa showed greater dominance of specific species ($D = 0.24$ and $D = 0.18$, respectively) (Appendix A). The Shannon diversity varied significantly between the study sites ($F_{4,205} = 88.32$, $p = 0.0001$). Shannon was significantly higher

in NV ($p < 0.01$) differing from other areas. MS presented the second highest index, being different from the other sites ($p < 0.01$). On the other side, Pa and Ag were similar each other and different from NV ($p < 0.001$), MS ($p < 0.01$) and ES ($p < 0.05$) (Fig. 3c). There was a significant difference in the Berger-Parker dominance index of the five sites studied ($F_{4,205}=34.35$, $p < 0.01$), with NV differing from Pa ($p < 0.05$), Ag ($p < 0.05$) and ES ($p < 0.01$) ((Fig. 3d).

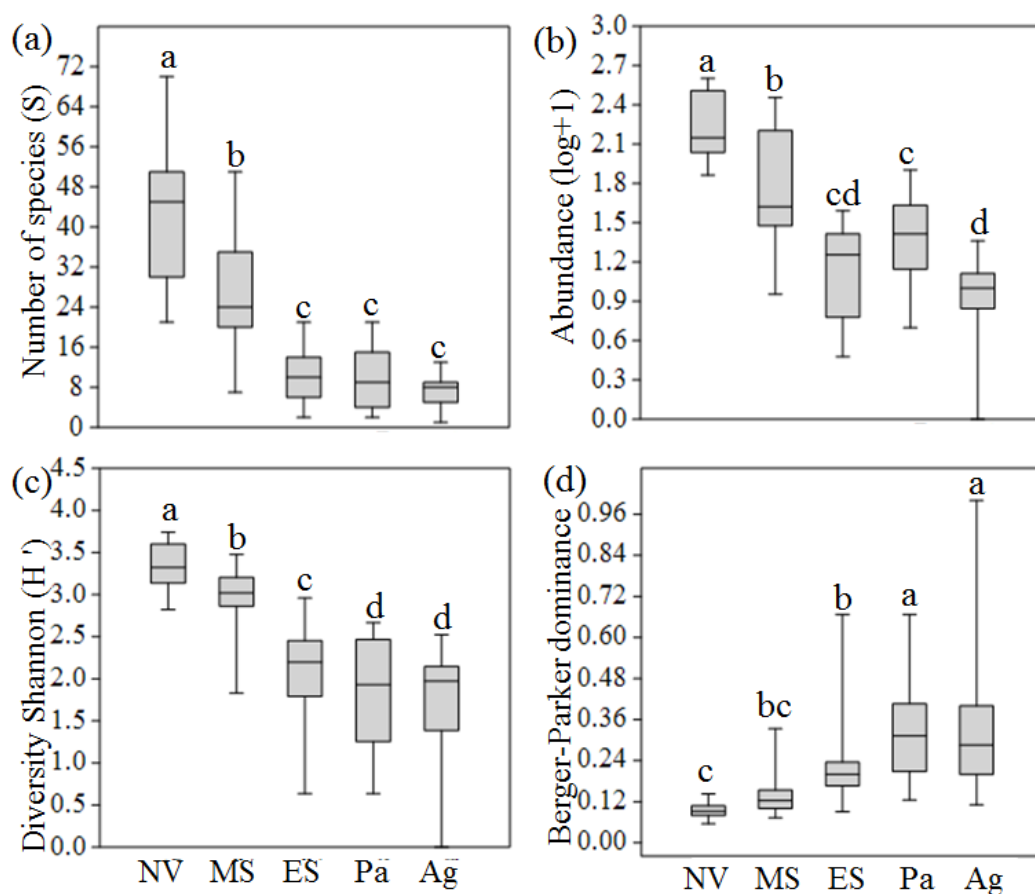


Fig. 3. Box and Whisker plots expressing the differences in the projected values for (a) Number of species (\pm SE) (b) number of individuals (\pm SE), (c) Shannon diversity (\pm SE) and (d) Berger-Parker dominance (\pm SE) of Scarabaeidae community in the different habitats considered. The values followed by the same letters are not significantly different according to Tukey test. NV, Vegetation native; MS, Mature Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture; Pa, Pasture.

Of the 112 species sampled, 16 species were collected exclusively on native vegetation, followed by pastures with eight unique species and late secondary vegetation with five (Appendix A). There was a greater sharing of species between native vegetation and late secondary vegetation (64 species, ~57%). Native vegetation showed greater dissimilarity with cocoa, secondary vegetation and pasture (Table 1). However, a Permutational Multivariate Analysis of Variance (PERMANOVA) showed that SB taxonomic composition varied significantly among the five sites studied ($F_{4,207} = 36.59, p < 0.001$).

Table 1. Dissimilarity percentage among the five studied areas, with significant difference ($p < 0.01$) according to PERMANOVA ($F_{4,207} = 36.59$). NV, Vegetation native; MS, Mature Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture; Pa, Pasture.

Habitat	NV	MS	ES	Ag	Pa
NV	0	-	-	-	-
MS	70.68	0	-	-	-
ES	92.77	81.94	0	-	-
Ag	93.54	85.22	87.04	0	-
Pa	92.74	88.24	83.30	84.35	0

Association of Scarab beetle assemblages with environmental variables

PC1 was positive correlated with the following environmental variables: Circumference at Breast Height (CBH); Circumference at breast ankle (CBA); Canopy Cover (CC); Number of Plant Species (NPS); Height of Leaf Litter (HLL); Percentages of Leaf Litter Cover (PLC); Percentage of Green (vegetation) Cover (PGC); Number of Plants (NP); Number of Species of Shrubs (NSS), and negative correlated with Temperature (T) and Percentage of Exposed Soil (PES). PC2 was positive correlated with Humidity (H), PC3 positive correlated with Number of Shrubs (NS) and PC4 with Precipitation (P), also positively.

The first two axes generated by CCA explained 74.3% (50.1% in axis 1 and 24.2% in axis 2) of the total variation in the SB composition. Among the PC's, environmental variables represented by PC1 ($R = -0.8313$) were the most important

factors affecting the first axis, and the Number of shrubs (NS) (represented by PC3) ($R=-0.7431$) had a great correlation with the second axis (Fig. 4).

The SB communities in Mature Secondary succession (MS) was mainly determined by Number of shrubs (NS) (PC3), among which Pasture was correlated with Humidity (H) (PC2) and Precipitation (P) (PC4); Agriculture (Ag) was correlated with H (PC2) and P (PC4), as well as environmental variables represented by PC1; Habitats Native Vegetation (NV) and Early Secondary succession (ES) were correlated with environmental variables represented by PC1.

At the species level, *T. externepunctatum* was associated to sites with the highest percentage of exposed soils – PES; *C. mutabilis* and *C. lituratus* were associated with precipitation – P; *A. platensis*, *Aphodius* sp2, *Blackburneus* sp1 and *Canthidium* sp2 were associated with temperature – T; *E. parallelus* was associated Height of Leaf Litter – HLL; *Eurysternus* sp2 was associated with humidity – H; and *O. carinifrons* was associated with Canopy Cover – CC and Percentages of Leaf Litter Cover – PLC.

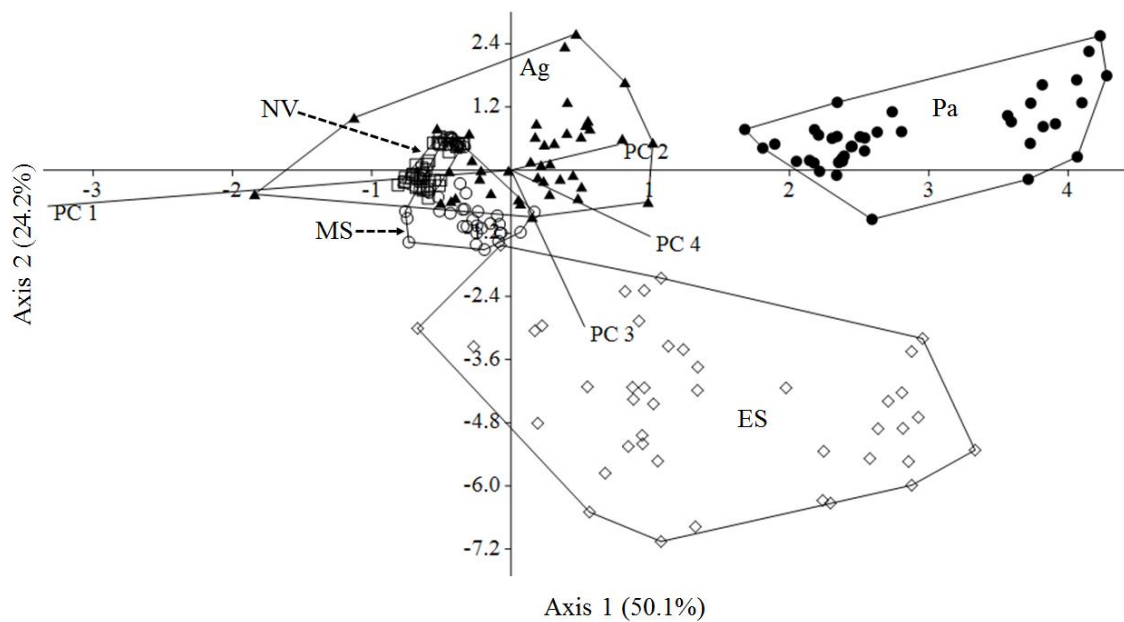


Fig. 4. Canonical Correspondence Analysis (CCA) used to estimate the influence of environmental variables on Scarabaeidae communities. Four axes were considered in the Principal Components Analysis (PCA), including 4 principal components (PC). NV, Native vegetation; MS, Mature secondary succession (15 years of natural regeneration of vegetation); ES, Early secondary succession (five years of natural regeneration of vegetation); Ag, Agriculture (Cacao plantations); Pa, Pasture.

IndVal

According to IndVal, 23 SB species were significantly associated with NV, 8 SB species were associated with PA, and 6 with ES. Ag and MS have only 1 associated species. The genus that presented the highest number of indicator species (5 species) was *Onthophagus*, 4 for native vegetation and 1 for pasture. *Canthon* also features 5 species, all associated to altered habitats, including 3 to ES and 1 for Ag and Pa. In general, 14 genera have exclusive species for sites (NV: *Uroxys*, *Oxysternon*, *Coprophanaeus*, *Eurysternus*, *Dichotomius*, *Scybalocanthon*, *Deltochilum*, *Phanaeus*; ES: *Diabroctis*, *Ataenius*, *Cryptocanthon*; Pa: *Aphodius*, *Gromphas e Sulcophanaeus*) (Table 2).

Table 2. Analysis of SB' indicator species (IndVal) from NV, Vegetation native; MS, Mature Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture; Pa, Pasture (only taxa with IndVal>25%).

Species	Indicator value	<i>p</i>	Site
<i>Ateuchus connexus</i> (Harold 1868)	1.000	0.005	NV
<i>Uroxys</i> sp1	0.976	0.005	NV
<i>Uroxys</i> sp2	0.964	0.005	NV
<i>Onthophagus haemathopus</i> (Harold 1875)	0.864	0.005	NV
<i>Onthophagus bidentatus</i> (Drapiez 1819)	0.843	0.005	NV
<i>Ateuchus murrayi</i> (Harold 1868)	0.837	0.005	NV
<i>Ontherus carinifrons</i> (Luederwaldt 1930)	0.829	0.005	NV
<i>Ateuchus pygidialis</i> (Harold 1868)	0.825	0.005	NV
<i>Oxysternon durantoni</i> (Arnaud 1984)	0.816	0.005	NV
<i>Coprophanaeus telamon</i> (Erichson 1847)	0.756	0.005	NV
<i>Onthophagus clypeatus</i> (Blanchard 1846)	0.690	0.005	NV
<i>Eurysternus strigilatus</i> (Génier 2009)	0.667	0.005	NV
<i>Dichotomius boreus</i> (Olivier 1789)	0.595	0.005	NV
<i>Onthophagus</i> sp2	0.563	0.005	NV
<i>Dichotomius subaeneus</i> (Castelnau 1840)	0.535	0.005	NV
<i>Oxysternon</i> sp1	0.512	0.005	NV
<i>Eurysternus ventricosus</i> (Gill 1990)	0.448	0.005	NV
<i>Scybalocanthon sexpilotus</i> (Guérin 1855)	0.444	0.005	NV
<i>Coprophanaeus</i> sp1	0.352	0.005	NV
<i>Canthidium</i> sp3	0.342	0.005	NV
<i>Deltochilum carinatum</i> (Westwood 1848)	0.309	0.010	NV
<i>Dichotomius melzeri</i> (Luederwaldt 1922)	0.309	0.010	NV
<i>Phanaeus bispinus</i> (Bates 1868)	0.309	0.010	NV
<i>Ateuchus</i> sp8	0.674	0.005	MS
<i>Canthon</i> sp6	0.772	0.005	ES
<i>Diabroctis mimas</i> (Linné 1758)	0.577	0.005	ES
<i>Canthon chalybaeus</i> (Blanchard 1846)	0.535	0.005	ES
<i>Canthon</i> sp7	0.408	0.005	ES
<i>Ataenius</i> sp4	0.282	0.005	ES
<i>Cryptocanthon peckorum</i> (Howden 1973)	0.267	0.025	ES
<i>Canthon</i> sp1	0.674	0.005	Ag
<i>Onthophagus hirculus</i> (Mannerheim 1829)	0.716	0.005	Pa
<i>Canthon histrio</i> (LePeletier e Serville 1828)	0.469	0.005	Pa
<i>Ontherus pubens</i> (Genier 1966)	0.469	0.005	Pa

<i>Aphodius</i> sp4	0.442	0.005	Pa
<i>Canthidium aff. ardens</i> (Bates 1887)	0.413	0.005	Pa
<i>Gromphas aeruginosa</i> (Perty 1830)	0.413	0.005	Pa
<i>Aphodius</i> sp3	0.383	0.005	Pa
<i>Sulcophanaeus</i> sp1	0.271	0.020	Pa

Discussion

Although considerable effort has been made to identify the most appropriate ecological indicators for assessing the status of disturbed habitats, this exercise have revealed widely differing views on why and what to measure and quantify. Since the conventional measures regarding biodiversity studies require multiple specialization levels and are difficult to implement due to resource constraints, we proposed alternative indicators capable of responding with comparable rigour to key changes in such habitats by using the species composition of Scarab beetles (SB) assemblages as surrogates. This proposal can only be properly addressed when the sampling effort associated with the heterogeneity of the habitats is considered in the scope of the need for rapid, standardized and cost-saving assessment methodologies (Scott and Anderson, 2003). In this context, the rarefaction curves of our work, depicted their asymptotes values, indicate that the sampling effort was enough to sample most SB fauna in heterogeneous habitats. The species richness (112 species) is comparatively higher when compared to the values reported from Malaysia (Davis, 2000), Peru (Valencia, 2001), French Guyana (Feer, 2000), African rain forests (Cambefort and Walter, 1991) rain forests of Mexico (Estrada and Coates-Estrada, 2002), Colombia (Escobar, 2000), Australia (Howden et al., 1991) and even to the Brazilian rain forests (Korasaki et al., 2013; Silva et al., 2014; França et al., 2016).

Assemblages analysis

The complexity of the habitat appears to influence species richness, diversity and the abundance of Scarab beetles, corroborating the results of Tews et al. (2004) and Navarrete and Halffter (2008). The applied indexes attained higher values for native vegetation and/or mature secondary vegetation, as a result of greater environmental complexity observed in relation to areas of anthropic influence (as

pastures, cocoa and vegetation in initial stage of regeneration). In fact, environmental complexity allows the coexistence of a larger set of species in pristine areas, due to the wider variety of resources offered (Blake and Kar, 1987; Casas et al., 2016). This also justifies the extraordinary abundance of a few species in pastures and agriculture (for details see Appendix A), mostly adapted to homogeneous microclimatic and structural factors (Quintero and Halfpter, 2009; Silva et al., 2014). According to Harper and Hawksworth (1994), Vinod and Sabu (2007), a taxonomic composition index is a measure of spread of biodiversity, indicating differences among habitats. The PERMANOVA analysis corroborated this finding showing that the most pristine habitats are similar, while the most disturbed environments form the farthest group. The observed differences in the composition of the beetles in this study might also be explained by the diversity of mammals (primarily primates) in native vegetation and late secondary vegetation, leading to superior availability of food resources for the Scarab beetles (SB). Furthermore, these areas have a higher fruit production, which serve as a secondary food resource for several species of SB (Culot et al., 2013; Silva and Hernández, 2016). Disturbed habitats, such as pastures and cocoa, are typically characterized by a relatively open canopy with hot and dry understory environments (Gardner et al., 2008), which might explain the impoverishment observed in the communities of SB. In fact, SB are intolerant to high temperatures (Chown, 2001; Campos and Hernandez, 2013), to solar radiation on adult activity patterns (Moreira et al., 2009; Korasaki et al., 2013) and to the decrease of soil moisture on survival of larvae, all considered limiting factors for several SB species (Gardner et al., 2008; Cajaiba et al., 2015a).

Association of Scarab beetle assemblages with environmental variables

The PCA/CCA analysis demonstrated that SB assemblages of anthropogenic habitats (Pasture and Agriculture) are mainly influenced by patterns of humidity and precipitation. In contrast, the communities of primary forest (native vegetation) are positively influenced by the structural vegetation characteristics, soil, and temperature, which are also important determinants of dung beetle communities in other regions of Brazil (Campos and Hernández, 2013; Silva and Hernández, 2016).

The lower temperatures that occur in pristine forests affect positively larger SB, such as *C. telamon* and *O. durantoni*, found exclusively on primary forest. These low temperatures increase their ability to build nests in underground galleries, increasing the survival rate of larvae in response to moist soils (Anduaga, 2004). These results are similar to the findings on the Amazon by Gardner et al. (2008) who observed an increased abundance of larger SB species in more pristine areas, suggesting that the differences in SB communities can be explained by changes in the structure of the vegetation.

The species richness observed in forest areas and/or mature succession could be associated with the availability of niches and food resources. On the other hand, disturbed areas support mostly generalist and tolerant SB species. In fact, the distribution of dung beetles along different environmental characteristics seems to demonstrate discrete associations with particular biotypes within the landscape (Viegas et al., 2014; Silva and Hernández, 2016). Furthermore, the species richness, abundance and biomass seem to be negatively affected in disturbed habitats (Gardner et al., 2008; Silva and Hernández, 2016). These environmental characteristics are also expected to affect the distribution of some mammalian species (Culot et al., 2013) and therefore, the intake of food resources for dung beetles (Silva and Hernández, 2016). Our results also substantiate the meta-analysis carried out by Rey Benayas et al. (2009), which concluded that restoration efforts tend to increase species richness, diversity and abundance of SB. Nichols et al. (2007) concluded that the land use systems with a high degree of forest cover harbour SB assemblages, similar to the results found in the pristine forests of our study area.

Value indicator - IndVal

The IndVal has been used in several works (Barlow et al., 2010; Viegas et al., 2014; Bicknell et al., 2014), due to its efficiency in identifying habitat bioindicators (Shahabuddin et al., 2005; Korasaki, 2010). In this study, *O. hirculus*, *Canthon* sp6 and *Canthon* sp1, appear as good surrogates of disturbed environments, namely in pastures, early secondary vegetation and cacao, respectively. It is interesting to highlight the ecological function of the *O. hirculus* (pasture indicator), because it is an excavator, providing valuable services in removing faecal masses on pasture area and contributing

to the biological control of fly and helminth parasites of veterinary importance (Silva et al., 2012). On the other hand, the species *A. connexus*, *Uroxys* sp1 and sp2, *O. haemathopus*, *O. bidentatus*, *A. murrayi*, *O. carinifrons*, *A. pygidialis*, *O. durantoni* and *C. telamon* are indicators of native vegetation and *Ateuchus* sp8 mature secondary vegetation. These species are commonly collected in primary forests and secondary late vegetation (Korasaki, 2010; Korasaki et al., 2012).

The results of this analysis, signals indicator species for all habitats might help with the management of neotropical systems highly affected by several anthropogenic pressures (Simões, 2013), allowing environmental managers and decision-makers, to anticipate impacts and support the development of measures to minimize the problems identified, in addition to support the management strategies and restoration of the forest ecosystems for a certain region of interest. Our work should be complemented by ecological and behavioural studies to understand and assess the state of conservation of the ecosystems considered (Brown, 1997), because ecological assessment and monitoring are important procedures to address effective management of ecosystems and natural resources, in which the use of essential indicators of ecosystem integrity is considered crucial to measure and evaluate the status and trends of target environmental systems (Kandziora et al. 2013).

Conclusion

Our results suggest a decrease in species richness and abundance of Scarabaeidae throughout gradients of habitat disturbance of the native tropical forests in Amazonia, with main finding supporting to the hypotheses that: (1) habitats largely differ in terms of species richness, taxonomic and ecological composition, with each habitat supporting exclusive and indicator species; (2) disturbed habitats (i.e. replacement of native forests by pasture or crops) are able to retain less species of Scarabaeidae than native forests or late secondary forests; (3) forest generalist species with more preserved habitat requirements may disappear after the destruction or depletion of their habitat. Our findings highlight the role of Scarabaeidae as a suitable key taxonomic group to evaluate the ecological status of changing neotropical ecosystems. Nevertheless, since the ecological status of the studied habitats can be only partly assessed by the Scarab beetles (SB) composition, this approach also

provides a useful starting point, allowing the precise development of more complete standardized data sets and information on the patterns of the species' functional response to changing ecosystems.

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Appendix A

Total numbers of Scarabaeidae species in the environments different in the Brazilian Amazon. NV = Native Vegetation; MS = Mature secondary succession (15 years of regeneration); ES = Early secondary succession (5 years of regeneration); Ag = Agriculture (Cocoa); Pa = Pasture.

Species	NV	MS	ES	Ag	Pa
<i>Coprophanaeus lancifer</i>	48	39	0	0	0
<i>Coprophanaeus telamon</i>	41	0	0	0	0
<i>Coprophanaeus degallieri</i>	0	0	0	0	2
<i>Coprophanaeus</i> sp1	7	1	0	0	0
<i>Oxysternon durantoni</i>	105	0	0	0	0
<i>Oxysternon conspicillatum</i>	23	0	43	0	0
<i>Oxysternon</i> sp1	19	0	0	0	0
<i>Sulcophanaeus faunus</i>	4	2	0	0	0
<i>Sulcophanaeus</i> sp1	0	0	0	0	3
<i>Eurysternus atrosericus</i>	81	30	0	8	0
<i>Eurysternus parallelus</i>	133	54	6	24	0
<i>Eurysternus caribaeus</i>	174	76	11	9	0
<i>Eurysternus wittmerorum</i>	71	54	9	3	0
<i>Eurysternus strigilatus</i>	37	10	0	0	0
<i>Eurysternus ventricosus</i>	17	3	0	0	0
<i>Eurysternus foedus</i>	21	20	8	5	0
<i>Eurysternus hamaticollis</i>	14	12	12	0	0
<i>Eurysternus</i> sp1	75	32	9	16	0
<i>Eurysternus</i> sp2	46	46	21	0	0
<i>Eurysternus</i> sp3	90	118	44	0	0
<i>Uroxys</i> sp1	320	0	0	0	0
<i>Uroxys</i> sp2	90	0	0	0	0
<i>Ateuchus pygidiali</i>	101	6	0	0	0
<i>Ateuchus murrayi</i>	470	98	0	0	98
<i>Ateuchus connexus</i>	304	0	0	0	0
<i>Ateuchus pygidialis</i>	123	51	0	0	0
<i>Ateuchus candezei</i>	133	44	0	0	0
<i>Ateuchus simplex</i>	59	13	0	0	0
<i>Ateuchus</i> sp1	29	33	11	11	24
<i>Ateuchus</i> sp2	51	15	0	0	36
<i>Ateuchus</i> sp3	296	98	26	4	0
<i>Ateuchus</i> sp4	181	55	4	0	12
<i>Ateuchus</i> sp5	320	88	31	2	0
<i>Ateuchus</i> sp6	68	37	0	0	0
<i>Ateuchus</i> sp7	28	47	0	0	0
<i>Ateuchus</i> sp8	0	43	0	0	2
<i>Ateuchus</i> sp9	323	109	6	0	0

<i>Anomiopus</i> sp1	2	0	0	0	0
<i>Canthon luteicollis</i>	136	107	31	9	6
<i>Canthon simulans</i>	11	25	66	15	232
<i>Canthon chalybaeus</i>	0	0	15	0	0
<i>Canthon aequinoctialis</i>	81	36	0	12	0
<i>Canthon fulgidus</i>	68	84	48	12	106
<i>Canthon quadriguttatus</i>	21	9	0	3	11
<i>Canthon triangularis</i>	165	89	0	0	0
<i>Canthon subhyalinus</i>	26	27	0	2	3
<i>Canthon lituratus</i>	68	15	77	0	98
<i>Canthon mutabilis</i>	0	0	33	0	16
<i>Canthon histrio</i>	0	0	0	0	12
<i>Canthon</i> sp1	0	2	0	21	0
<i>Canthon</i> sp2	312	90	11	106	77
<i>Canthon</i> sp3	24	5	0	27	0
<i>Canthon</i> sp4	0	0	0	29	14
<i>Canthon</i> sp5	36	53	32	25	111
<i>Canthon</i> sp6	21	0	16	9	0
<i>Canthon</i> sp7	0	0	30	0	0
<i>Canthon</i> sp8	0	0	7	0	0
<i>Canthonella</i> sp1	26	0	0	0	16
<i>Deltochilum orbiculare</i>	61	28	0	0	2
<i>Deltochilum submetallicum</i>	337	173	0	0	0
<i>Deltochilum carinatum</i>	5	0	0	0	0
<i>Deltochilum icarus</i>	4	2	0	0	0
<i>Deltochilum</i> sp1	0	1	0	0	0
<i>Sylvicanthon</i> sp1	15	10	0	0	2
<i>Scybalocanthon sexpilotus</i>	12	1	0	0	0
<i>Canthidium deyrollei</i>	548	236	0	0	0
<i>Canthidium ardens</i>	0	0	0	0	12
<i>Canthidium</i> sp1	6	23	0	20	10
<i>Canthidium</i> sp2	9	3	0	7	20
<i>Canthidium</i> sp3	54	14	0	0	0
<i>Canthidium</i> sp4	7	0	0	3	0
<i>Canthidium</i> sp5	487	196	0	0	0
<i>Dichotomius apicalis</i>	43	21	0	0	0
<i>Dichotomius boreus</i>	28	6	0	0	0
<i>Dichotomius subaeneus</i>	80	82	0	0	0
<i>Dichotomius lucasi</i>	78	70	0	0	0
<i>Dichotomius subaeneus</i>	21	0	0	0	0
<i>Dichotomius</i> sp1	404	297	0	0	0
<i>Dichotomius</i> sp2	0	3	0	0	0
<i>Dichotomius worontzowi</i>	10	6	0	0	0
<i>Dichotomius melzeri</i>	4	0	0	0	0

<i>Deltochilum carinatum</i>	3	0	0	0	0
<i>Ontherus carinifrons</i>	463	201	0	0	0
<i>Ontherus pubens</i>	0	0	0	0	23
<i>Ontherus sp1</i>	2	0	0	0	0
<i>Onthophagus bidentatus</i>	302	98	0	0	0
<i>Onthophagus haemathopus</i>	213	69	0	0	0
<i>Onthophagus clypeatus</i>	37	0	0	0	0
<i>Onthophagus hirculus</i>	0	0	0	0	54
<i>Onthophagus sp1</i>	5	0	0	0	3
<i>Onthophagus sp2</i>	26	5	0	0	0
<i>Onthophagus sp3</i>	3	0	0	0	0
<i>Ataenius platensis</i>	27	3	0	6	26
<i>Ataenius sp1</i>	24	19	27	17	43
<i>Ataenius sp2</i>	3	0	0	4	8
<i>Ataenius sp3</i>	0	13	2	4	12
<i>Ataenius sp4</i>	0	0	6	3	0
<i>Phanaeus chalcomelas</i>	9	5	0	0	0
<i>Phanaeus bispinus</i>	4	0	0	0	0
<i>Phanaeus sp1</i>	0	2	0	0	0
<i>Aphodius sp1</i>	13	19	2	10	12
<i>Aphodius sp2</i>	7	1	0	0	13
<i>Aphodius sp3</i>	0	0	0	0	12
<i>Aphodius sp4</i>	1	0	0	0	0
<i>Blackburneus sp1</i>	12	0	0	3	12
<i>Diabroctis mimas</i>	0	0	25	0	0
<i>Cryptocanthon peckorum</i>	0	0	3	0	0
<i>Trichillum externepunctatum</i>	0	0	29	0	65
Scarabaeidae 1	0	0	4	0	2
<i>Digitonthophagus gazela</i>	0	0	0	2	0
<i>Gromphas aeruginos</i>	0	0	0	0	18
<i>Macraspis sp1</i>	0	0	0	2	0
Abundance	8.804	3.941	707	438	1.219
Richness (S)	86	69	44	33	38
Shannon-Weiner diversity index (H')	3.73	3.64	3.16	2.95	2.96
Dominance Berger Parker (D)	0.06	0.08	0.10	0.24	0.18

Chapter 4

Attractiveness of *Scarabaeinae* (Coleoptera: Scarabaeidae) to different baits in the Brazilian Amazon region

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Abstract

Dung beetles (Coleoptera: Scarabaeidae: Scarabaeinae) are considered essential for enhancing the physicochemical characteristics of soils, principally by changing organic materials (e.g. dead animals, faeces, fruits e leaf litter). This study compared the species richness and abundance of dung beetles captured using various types of baits, to demonstrate attractiveness differences in variable habitats of the Brazilian Amazon. Samplings were carried out with pitfall traps baited with human faeces, rotten banana, rotten meat and a non-baited trap, in February, March, June, September and October 2015. Habitats included native forests, agriculture areas, pastures and disturbed forests in different regeneration stages. A total of 13 736 Scarabaeinae beetles were captured, distributed over 98 species. Most individuals were captured using traps baited with faeces (76.7 % of individuals), followed by rotten meat baited traps (17.8 % of individuals), fermented banana baited traps (3.9 % of individuals) and finally by non-baited traps (1.6 % of individuals). A significant difference in attractiveness of the different baits used and habitats types was observed. Most of the captured assemblages were composed by coprophagous (42 %), generalist species (32 %), necrophagous (15 %) and none was classified as saprophagous. Approximately 54 % of the specimens were tunnelers, 25 % were rollers and 12 % were dwellers. The species of Scarabaeinae sampled in the region revealed qualitative and quantitative differences among their assemblages and the phytophysiognomies. The forest environments housed the greatest species richness observed, and a fraction of these is exclusive of those areas. We concluded that some species of Scarabaeinae have an important potential as disturbance indicators in the Amazonian ecosystem.

Key words: dung beetles, dung attractiveness, food preference, trophic guild, tropical forest.

The subfamily Scarabaeinae (Coleoptera: Scarabaeidae) includes dung beetles (DB), a globally distributed group of detritus-feeding insects, determinant of ecological functions such as nutrient recycling, secondary seed dispersal, bioturbation, and natural control of cattle parasites (Nichols et al., 2008; Simmons & Ridsdill-Smith, 2011). Their contribution to the improvement of the physico-chemical properties of the soil and plants is associated with increased edaphic aeration and hydration (Andresen, 2002; Nichols et al., 2008).

Mammalian dung is one of the most important food resources for dung beetle communities, being also the dominant substrate for oviposition (Filgueiras, Liberal, Aguiar, Hernández, & Iannuzzi, 2009). Anyhow, in the Neotropics, fungi, plants and carcasses are also used by several species as food resource (Halffter & Matthews, 1966). This seems related to the local availability of the ecosystem that provides food for the maintenance of diverse DB strategies. When the food source is not available due to various limiting factors, several DB can use other similar state resources (in the process of decomposition) as food (Da Silva & Audino, 2011). This feeding diversification observed in the Neotropics is believed to have occurred during the extinction of large mammals in the late Pleistocene (Halffter, 1991; Larsen, Lopera, & Forsyth, 2006).

In tropical regions, the main types of baits used for monitoring DB are cattle dung (Andresen, 2008; Ueda, Dwibadra, Noerdjito, Kon, & Fukuyama, 2015) and human faeces (Vieira, Louzada, & Spector, 2008). Alternative baits have included the dung of other mammals, rotting fruits (Vieira et al., 2008; Ueda et al., 2015) and carrion (Ueda et al., 2015). Different food preferences reduce inter specific competition, allowing the coexistence of diverse DB species, in particular neotropical ecosystems. Indeed the occupation of diverse ecological niches increases diversity for specific locations (Da Silva & Audino, 2011). The presence/ abundance and type of food types, associated with other factors such as climate, animal migrations and life cycles, in addition to altitude and landscape features, might affect the spatial and temporal distribution of Scarabaeinae (Filgueiras et al., 2009).

The use of DB in the Brazilian Amazon is in its initial stage, despite already having a reasonable number of publications associated, mostly restricted to the South

and Southeast part of the country. Furthermore, the DB research and applications could be significantly improved through using standardized methods and data sets, supported on the species preferences and patterns of functional response to disturbance (Cajaiba, Perico, Cabral, & Santos, 2015a). In fact, understating food preferences of DB assemblages is fundamental to enlighten managers on the effects of changes occurring in tropical regions. The use of trophic guilds/functional guilds (necrophagous, saprophagous, coprophagous, tunnelers, dwellers and rollers) may also reveal interesting differences in the structure and functioning of ecosystems and landscape (Paoletti, 1999).

The present study compared the DB richness, abundance and functional diversity when captured using different types of baits in variable habitats of the Brazilian Amazon, in order to: a) assess the general attractiveness of baits, and b) gauge the relation between specific baits' diversity and the dominant habitats. The presented method and results could be used to support a standardization of field work for effective ecological monitoring of this group studies in Neotropical areas.

MATERIAL AND METHODS

Study sites

The study was performed in the municipality of Uruará, state of Pará, Northern Brazil (-03°43'27"S; -53°44'8"W). The dominant land use/land cover of the study area was a natural forest (69 % of the area), where deforestation is mainly concentrated in the South-central part of the territory, near the main roads. Extensive livestock production and the exploitation of timber at a large scale (mostly illegal) are currently considered the most serious environmental threats (Cajaiba, Cabral, & Santos, 2015b). The studied areas encompass habitats that, in terms of physical characteristics and anthropogenic disturbances, are representative of the regional habitats: Native Vegetation-NV, Mature Secondary succession-MS (vegetation with 15 years of regeneration), Early Secondary succession-ES (vegetation with five years of regeneration), Agriculture-Ag (cocoa plantations, *Theobroma cacao* L.) and Pasture for extensive livestock-Pa. This habitats gradient was considered fundamental to

analyse the response of DB communities (Cajaiba et al., 2015b). The climate of the study area is classified as Aw (Köppen), hot and humid and the average annual rainfall is 2 000 mm (Peel, Finlayson, & McMahon, 2007).

Sampling method

Sampling was carried out during the year 2015, in the months of February, March, June, September and October. The sample points were placed at a minimum distance of 100 meters from ecotones, to guarantee that most DB captured in the pitfalls were associated to the specific LU/LC monitored. Pitfall traps (75 mm diameter and 110 mm deep) were filled with preservative liquid consisting of formalin, alcohol, water and a few drops of detergent to break the surface tension (Cajaiba et al., 2015b). A roof was attached to each pitfall to prevent rainwater from entering, remaining installed for 48 h prior to collection. Each pitfall contained different baits: Human faeces-HF; Rotten meat-RM; Rotten banana-RB in order to attract different species according to their feeding habits and non-baited pitfalls were used as control-Co.

In each study site (NV, ES, MS, Ag and Pa) seven sample points were placed 100 m apart. Each sample point contained four pitfall traps separated by five meters and included the different baits (HF, RM, RB, Co). The distance between pitfall traps allowed individuals to choose their preferential food resource (Almeida & Louzada, 2009; Da Silva, Vaz-De-Mello, & Di Mare, 2012). The Scarabaeinae collected were conserved in 70 % ethanol and taken into the laboratory and identified to the species level when possible. The identification was based on the keys proposed by Vaz-de-Mello, Edmonds, Campo, and Schoolmeesters (2011) and Pacheco and Vaz-de-Mello (2015).

Species were classified within guilds, according to their use of food and nesting resources. Nesting guild included rollers (those that roll balls of food on the surface of soil to some distance from the source of resource, where they bury them); tunnelers (those that carry food resource into the soil, making tunnels on the side or below the resource); and dwellers (which do not reallocate food, using it directly in the source) (Simmons & Ridsdill-Smith, 2011; Da Silva & Di Mare, 2012). Concerning the feeding guild, species were classified as coprophagous (≥ 80 % of captures occurred in

traps baited with faeces), necrophagous (≥ 80 % of captures occurred in traps baited with rotten meat), saprophages (≥ 80 % of captures occurred in traps baited with rotten banana) and generalists (species not covered in the previous groups) (Almeida & Louzada, 2009; Da Silva & Di Mare, 2012).

Two way analysis of variance (ANOVA)/Kruskal-Wallis tests, and associated multiple comparisons tests, Tuckey and Dunn were used for testing: a) possible differences of nesting guild selection (tunnelers, dweller, rollers) and feeding guild selection (coprophagous, necrophagous, saprophages and generalist) within the different habitats; b) possible differences in richness and abundance of Scarabaeinae by bait, and c) differences in abundance and richness of the Scarabaeinae by baits within the habitats studied. The normality of the data was verified by the Shapiro-Wilk test. In order to homogenize the variances and normalize the residues, both the abundance and richness changed to logarithmic values ($\log+1$). All analyses were performed using PAST software version 3.14 (Hammer, Harper, & Ryan, 2001).

RESULTS

General results/Functional diversity

A total of 98 species and 13 736 scarabaeine beetles were captured. The most abundant species were *Canthidium deyrollei* Harold 1867 (985 individuals), *Canthidium* sp. 4 (890 individuals), *Dichotomius* sp.1 (701 individuals), *Ateuchus murrayi* Harold 1868 (666 individuals), *Ontherus carinifrons* Luederwaldt 1930 (664 individuals) and *Deltochilum submetallicum* Castelnau 1840 (612 individuals). Of the total number of collected individuals, 76.7 % were captured in faeces baited traps, 17.8 % in rotten meat traps, 3.9 % in rotten banana traps, and only 1.6 % in non-baited traps. Eight species were captured in all traps.

Considering the feeding guild, coprophagous comprised 42% of the collected specimens, 32% were considered generalists, and only 15% were determined as necrophagous (Appendix 1). The coprophagous were more abundant in NV when compared to other habitats ($F_{4,205} = 12.35$, $P < 0.001$), namely between NV vs MS ($Q 5.69$, $P < 0.01$), NV vs ES ($Q 8.12$, $P < 0.001$), NV vs Pa ($Q 7.83$, $P < 0.001$) and NV

vs Ag (Q 8.33, $P < 0.0001$). Generalists were also more abundant in NV when compared to other habitats ($F_{4,155} = 12.75$, $P < 0.05$), specifically between NV vs MS (Q 5.52, $P < 0.05$), NV vs ES (Q 8.32, $P < 0.001$), NV vs Pa (Q 7.53, $P < 0.001$) and NV vs Ag (Q 8.65, $P < 0.0001$). In relation to the necrophagous abundance, no statistical differences were detected ($F_{4,70} = 2.40$, $P = 0.06$) (Fig. 1).

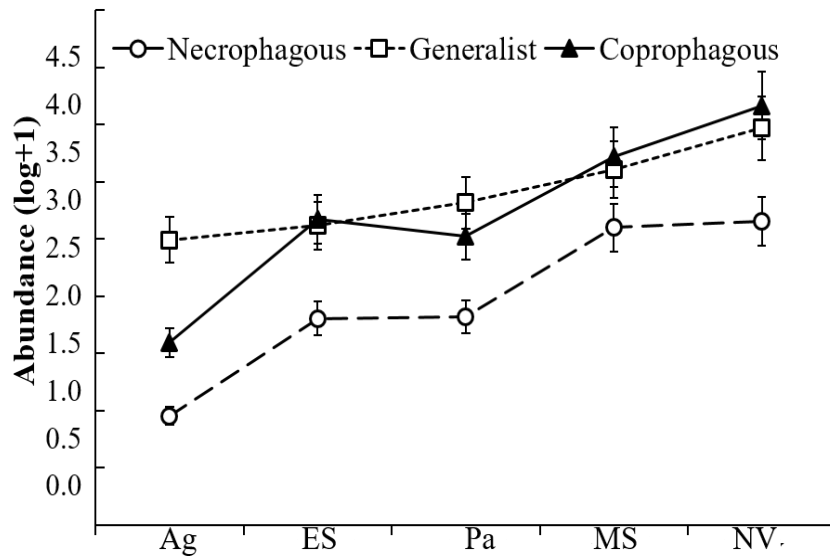


Fig. 1. Abundance of Scarabaeinae feeding guilds by habitat. NV: Native vegetation; MS: Mature secondary; ES: Early secondary; Ag: Agriculture (Cocoa); Pa: Pasture.

Concerning the nesting guild, approximately 54 % of the specimens (53 species) were tunnelers, 25 % were rollers (25 species) and 12 % were dwellers (12 species). The tunnelers were more abundant in NV when compared to other habitats ($F_{4,255} = 18.04$, $P < 0.05$), namely between NV vs MS (Q 6.61, $P < 0.05$), NV vs ES (Q 9.87, $P < 0.05$), NV vs Pa (Q 9.52, $P < 0.05$) and NV vs Ag (Q 9.92, $P < 0.001$). The number of rollers was significantly higher in NV ($F_{4,124} = 2.82$, $P = 0.02$), with significant differences between NV vs ES (Q 3.93, $P = 0.04$) and NV vs Ag (Q 4.26, $P = 0.02$). Due to the low abundance of dweller species (12 species) it was not possible to perform statistical analysis (Fig. 2).

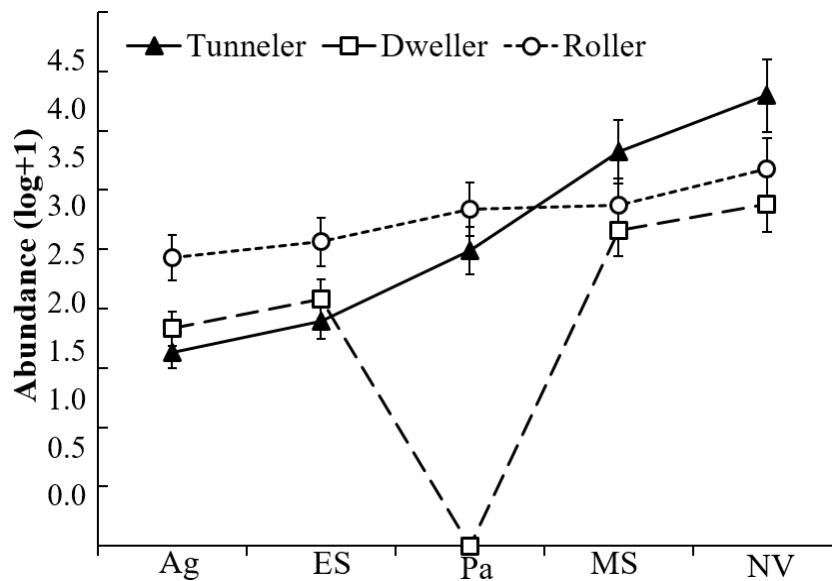


Fig. 2. Abundance of Scarabaeinae on the behavioral guild in different habitats studied. NV: Native vegetation; MS: Mature secondary; ES: Early secondary; Ag: Agriculture (Cocoa); Pa: Pasture.

Bait attractiveness

Kruskal-Wallis tests showed significant statistical differences in the medians of species richness by bait types (Kruskal-Wallis = 252.8, d.f. = 3, $P < 0.01$). According to an a posteriori Dunn's test, significant differences were found between medians of HF vs RM (71.59, $P < 0.05$), HF vs RB (146.59, $P < 0.01$), HF vs Co (160.79, $P < 0.0001$), RM vs RB (75.01, $P < 0.01$), RM vs Co (89.20, $P < 0.01$). There was no difference between RB and Co (14.20, $P > 0.05$) (Fig. 3A).

Kruskal-Wallis tests also showed significant statistical differences in the medians of species abundance by bait types (Kruskal-Wallis = 252.8, d.f. = 3, $P < 0.01$). Significant differences were found between medians of HF vs RM (52.22, $P < 0.05$), HF vs RB (193.87, $P < 0.001$), HF vs Co (225.05, $P < 0.0001$), RM vs RB (141.64, $P < 0.01$), RM vs Co (172.84, $P < 0.05$). There was no difference between the average RB vs Co (31.20, $P > 0.05$) (Fig. 3B).

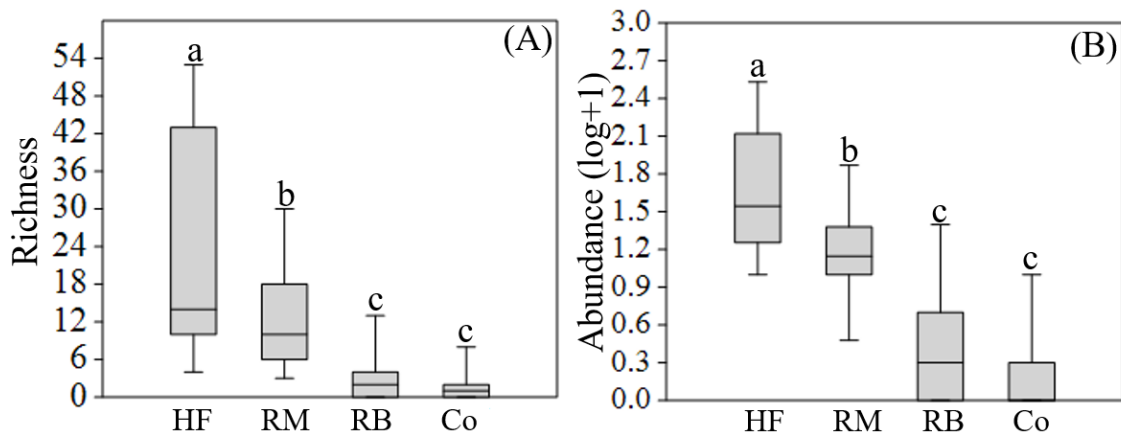


Fig. 3. Differences between abundance (A) and species richness (B) of the Scarabaeinae studied with different bait type. Co: Control bait; RB: Rotten banana; RM: Rotten meat; HF: Human faeces. The values followed by the same letters are not different according to Dunn test.

When the richness of the Scarabaeinae was tested for the different kinds of baits among habitats, significant differences among bait types ($F = 51.71$, d.f. = 3, $P < 0.001$) and habitats ($F = 29.1$, d.f. = 4, $P < 0.001$) were found (Fig. 4A). When the abundance of the Scarabaeinae was tested for the different kinds of bait among habitats, significant differences among bait types ($F = 61.2$, d.f. = 3, $P < 0.01$) and habitats ($F = 47.18$, d.f. = 4, $P = 0.001$) were found (Fig. 4B).

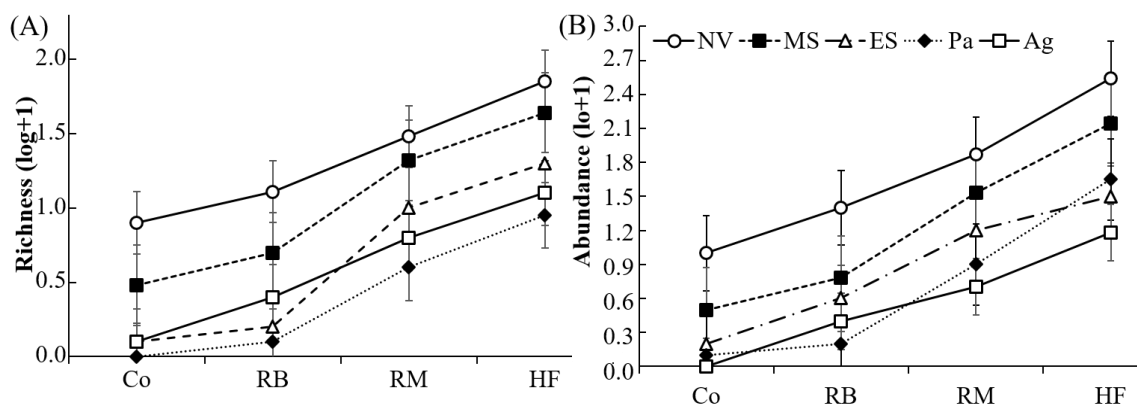


Fig. 4. Differences between species richness (a) and abundance (b) of the Scarabaeinae studied in different habitats (NV=Native vegetation, MS=Mature secondary, ES=Early secondary, Ag=Agriculture, Pa=Pasture) for each bait type (Co=Control bait, RB=Rotten banana, RM=Rotten meat, HF=Human feces).

DISCUSSION

Considering the functional diversity, this work revealed that conserved environments such as NV and MS in this region, hold a larger proportion of Dung Beetle (DB) tunnelers, rollers and dwellers. Anthropogenic disturbances may alter the composition of scarabaeines, changing the primary “services” of dung beetles such as the reduction and decomposition of organic materials through burial and removal. These actions are fundamental for the conversion of biomasses, conserving energy and recycling nutrients in the ecosystem (Arellano, 2016). This group is also involved in other functions which include reducing compaction and improving soil fertility, dispersal of seeds, control of vectors of diseases (e.g. flies) and protection of agricultural/wild seeds against pests (Nichols et al., 2008).

Human actions that reduce specific mammal groups may have direct effects on DB fauna, which in turn may alter nutrient cycling processes and secondary dispersion of seeds (Andresen, 2002). In addition, the biological effects of a reduction in fauna may impact processes (e.g. behavioural / physiological, ecological, and evolutionary) at different environmental scales (e.g., local, regional, ecosystemic, and global) (Galetti & Dirzo, 2013; Bogoni & Hernández, 2014). The high coprophagy specialization in the Scarabaeinae seems to be related to the regular availability of mammal dung in the ecosystem (Halfpter & Matthews, 1966) contrary to the rotting fruits and carcasses of dead animals that might be seasonally and spatially limited (Louzada & Lopes, 1997). Considering that an important proportion of the nutrients consumed by vertebrates remain in their faeces and leftovers (Steinfeld et al., 2006), the ecological function performed by DB is fundamental for nutrient cycles and ecosystem productivity (Huerta, Martínez, Montes, & Favila, 2013; Arellano, 2016). For example if fresh dung is not rapidly incorporated into the soil, most nitrogen is lost through ammonia volatilization (West & Nelson, 2003).

Although generalists, several species present feeding preferences for certain types of resource (Da Silva et al., 2012). In fact, tunnelers were dominant in relation to the other guilds, a common pattern in the Neotropics. The distribution of behavioral guilds seems to be the result of local diversity of DB species in the Neotropics (Louzada & Lopes, 1997; Lima, Silva, Bianchi, Da Silva, & Di Mare, 2015).

When considering bait attractiveness, Halffter and Edmonds (1982) suggested that DB have physiological needs at different times of the year due to their life cycle, which may also explain the use of different food resources (Da Silva & Di Mare, 2012). Anyway, a clear distinction of attractiveness of the different types of baits to Scarabaeinae within habitats was detected. HF presented the highest abundance and richness in all habitats. In fact, the majority of collected individuals were coprophagous, i.e. preferred stool baits. The results obtained are in agreement with other studies in tropical forests and in other types of landscapes in tropical regions (Halffter & Matthews, 1966; Filgueiras et al., 2009; Da Silva et al., 2012). The preference for faeces of omnivorous mammals for nesting and feeding by adults and larvae seems to have resulted from evolutionary processes (Simmons & Ridsdill-Smith, 2011; Da Silva et al., 2012; Da Silva & Di Mare, 2012). This perspective suggests that loss of mammals (i.e., and their feces as a food resource) may alter competitive interactions between dung beetle species and may even cause local extinction of highly specialized species (Bogoni & Hernández, 2014). RM baits were also quite attractive highlighting the importance of this group in nutrient cycling processes in the environments where they live: according to Halffter and Matthews (1966) and Da Silva and Di Mare (2012) species of DB that feed on dead animal evolved with scarcity of large mammals and lower supply of excrement in the Neotropics (Halffter, 1991).

Traps with bait tend to be selective: if the aim of a specific work is an assessment of the overall community, different baits should be used to collect most diversity and estimate relative abundances (Rafael, 2002); if the goal of the research is collecting specific species and/or groups (e.g. trophic/functional guilds), particular baits should be used (Marchiori, 2016). Collection methodologies described here provide an approach to improve the detection and description of the responses of DB to disturbance and anthropogenic pressures, and facilitate a greater integration of ecological data collection efforts in tropical regions. Although spatially and temporally restricted, the results of this study highlight the attractiveness of several baits and the feeding preference of some species of DB.

Although highly specialized in mammalian excrement due to evolutionary processes, many Neotropical species of Scarabaeinae show plasticity in their diet (Larsen et al., 2006). This process seems related to the local availability of the ecosystem to provide food, i.e... when the preferred food is not available, due to different limiting factors, many Scarabaeinae may use other resources in similar state (in decomposition process).

The DB sampled in the Amazon region reveal qualitative and quantitative differences among their assemblages and the phytophysiognomies sampled. The forest environments housed the greatest species richness observed and a fraction of these is exclusive of this environment and hardly occurs in other types of ecosystems (Cajaiba, Périco, Dalzochio, Silva, Bastos, Cabral, & Santos, 2017). However, another part of this fauna is adapted to the open environment, being largely represented by coprophagy species. In this way, the landscape context is very important to the DB, because complementarity of habitats can present a particular diversity that increases the diversity of the landscape (Almeida & Louzada, 2009). Studies like this, focused in the knowledge of the biology and distribution of Scarabaeinae, are essential to support any future initiative for biodiversity and ecosystems conservation (Da Silva et al., 2012; Lima et al., 2015).

RESUMEN

Atracción de *Scarabaeinae* (Coleoptera: Scarabaeidae) por diferentes cebos en la región Amazónica Brasileña. Los escarabajos peloteros (Coleoptera: Scarabaeidae: Scarabaeinae) son considerados fundamentales para la mejora de las características físico-químicas de los suelos, es decir, para la descomposición de materiales orgánicos (p. ej., animales muertos, heces, frutas y hojarasca). Este estudio compara la riqueza de especies y la abundancia de escarabajos peloteros, capturados utilizando diversos tipos de cebos, para demostrar diferencias en la atracción en hábitats de la Amazonía Brasileña. Se realizaron muestreos con trampas cebadas con heces humanas, plátano podrido, carne podrida y una trampa sin cebo. Los hábitats incluyen bosques nativos, zonas de agricultura, pastos y bosques alterados en diferentes fases de regeneración. Se capturaron un total de 13 736 escarabajos Scarabaeinae, distribuidos en 98 especies. La mayoría de los individuos fueron capturados en trampas cebadas con heces (76.7 % de los individuos), seguido por las trampas con cebo de carne podrida (17.8 % de los individuos), trampas con cebo de plátano fermentada (3.9 % de los individuos) y finalmente por las trampas sin cebo (1.6 % de los individuos). Se observó una diferencia significativa en la atracción de los diferentes cebos y hábitats. La mayoría de la comunidad capturada estuvo compuesta de escarabajos coprófagos (42 %), especies generalistas (32 %) y necrófagos (15 %) y ninguno fue clasificado como saprófago. Aproximadamente, el 54 % de las muestras fueron excavadores, 25 % eran rodadores y el 12 % eran residentes. Las especies de scarabaeinae muestreadas en la región revelaron diferencias cualitativas y cuantitativas entre sus ensamblajes y las fitofisiognomías muestreadas. Los ambientes forestales albergaron la mayor riqueza de especies observada y una fracción de ellas es exclusiva de este entorno. Concluimos que algunas especies de Scarabaeinae tienen un potencial importante como indicadores de perturbación en el ecosistema amazónico.

Palabras clave: Escarabajos peloteros, atracción del estiércol, preferencia de alimentación, agrupación trófica, bosque tropical.

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Appendix 1

Numbers of individuals of species of the Scarabaeinae captured with pitfall traps baited with Human Feces (HF), Rotten Meat (RM) Rotten Banana (RB) and Control (Co), in the municipality of Uruará, state of Para, northern Brazil. TH: Trophic Habit of the species (C, coprophagous; N, necrophagous; G, generalist); BG: Behavioral Guild (T, tunneler; R, roller; D, dweller). *insufficient number of individuals or undefined.

Species	Bait				Total	TH	BG
	HF	RM	RB	Co			
<i>Coprophanaeus lancifer</i> (Linnaeus, 1767)	3	83	-	-	86	N	T
<i>Coprophanaeus telamon</i> (Erichson 1847)	-	41	-	-	41	N	T
<i>Coprophanaeus degallieri</i> (Arnaud 1997)	2	1	-	-	3	*	T
<i>Coprophanaeus</i> sp	7	1	-	-	8	C	T
<i>Oxysternon durantoni</i> (Arnaud 1984)	105	-	-	-	105	C	*
<i>Oxysternon conspicillatum</i> (Weber 1801)	43	23	-	-	66	G	*
<i>Oxysternon</i> sp	19	-	-	-	19	C	*
Scarabaeinae 1	6	-	-	-	6	C	T
Scarabaeinae 2	3	-	-	-	3	*	T
<i>Eurysternus atrosericus</i> (Génier 2009)	87	24	-	-	111	G	D
<i>Eurysternus parallelus</i> (Castelnau 1840)	157	60	-	-	217	G	D
<i>Eurysternus caribaeus</i> (Herbst 1789)	156	108	6	-	270	G	D
<i>Eurysternus wittmerorum</i> (Martinez 1988)	98	39	-	-	137	G	D
<i>Eurysternus strigilatus</i> (Génier 2009)	46	9	-	-	55	C	D
<i>Eurysternus ventricosus</i> (Gill 1990)	12	8	-	-	20	G	D
<i>Eurysternus foedus</i> (Guérin-Méneville 1844)	54	-	-	-	54	C	D
<i>Eurysternus hamaticollis</i> (Balthasar, 1939)	38	-	-	-	38	C	D
<i>Eurysternus</i> sp1	62	73	-	-	135	G	D
<i>Eurysternus</i> sp2	59	86	-	-	145	G	D
<i>Eurysternus</i> sp3	179	42	-	-	221	C	D
<i>Uroxys</i> sp1	316	-	-	4	320	C	T
<i>Uroxys</i> sp2	90	-	-	-	90	C	T
<i>Ateuchus murrayi</i> (Harold 1868)	627	-	39	-	666	C	T
<i>Ateuchus connexus</i> (Harold 1868)	304	-	-	-	304	C	T
<i>Ateuchus pygidialis</i> (Harold 1868)	17	95	-	10	122	N	T
<i>Ateuchus candezei</i> (Harold 1868)	37	84	53	2	176	G	T
<i>Ateuchus simplex</i> (Serville 1828)	-	58	-	-	58	N	T
<i>Ateuchus</i> sp1	57	24	27	9	117	G	T
<i>Ateuchus</i> sp2	17	36	3	-	56	G	T
<i>Ateuchus</i> sp3	164	141	80	32	417	G	T
<i>Ateuchus</i> sp4	151	50	36	11	248	G	T
<i>Ateuchus</i> sp5	253	129	81	26	489	G	T
<i>Ateuchus</i> sp6	67	4	25	-	96	G	T
<i>Ateuchus</i> sp7	83	-	10	-	93	C	T
<i>Ateuchus</i> sp8	43	-	0	-	43	C	T
<i>Anomiopus</i> sp	2	-0	0	-	2	*	*
<i>Canthon luteicollis</i> (Erichson 1847)	213	57	11	-	281	G	R
<i>Canthon simulans</i> (Martínez 1950)	258	82	7	-	347	G	R
<i>Canthon chalybaeus</i> (Blanchard 1846)	3	12	-	-	15	N	R
<i>Canthon aequinoctialis</i> (Harold 1868)	58	61	-	10	129	G	R
<i>Canthon fulgidus</i> (Redtenbacher 1867)	204	60	37	16	317	G	R

<i>Canthon quadriguttatus</i> (Olivier 1789)	41	-	3	-	44	C	R
<i>Canthon triangularis</i> (Drury 1770)	159	95	-	-	254	G	R
<i>Canthon subhyalinus</i> (Harold 1867)	58	-	-	-	58	C	R
<i>Canthon lituratus</i> (Germar 1813)	234	24	-	-	258	C	R
<i>Canthon mutabilis</i> (Lucas 1857)	14	35	-	-	49	G	R
<i>Canthon histrio</i> (LePeletier e Serville 1828)	-	12	-	-	12	N	R
<i>Canthon (Glaphyrocanthon) sp1</i>	14	9	-	-	23	G	R
<i>Canthon sp1</i>	380	137	49	30	596	G	R
<i>Canthon sp2</i>	65	44	-	-	109	G	R
<i>Canthon sp3</i>	38	5	-	-	43	C	R
<i>Canthon sp4</i>	170	19	16	25	230	G	R
<i>Canthon sp5</i>	7	33	-	2	42	N	R
<i>Canthon sp6</i>	-	30	-	-	30	N	R
<i>Canthon sp7</i>	-	7	-	-	7	N	R
<i>Canthonella sp</i>	33	16	-	-	49	G	*
<i>Deltochilum orbiculare</i> (van Lansberge 1874)	98	14	2	-	114	C	R
<i>Deltochilum submetallicum</i> (Castelnau 1840)	612	-	-	-	612	C	R
<i>Deltochilum carinatum</i> (Westwood 1848)	5	-	-	-	5	C	R
<i>Deltochilum icarus</i> (Olivier 1789)	6	-	-	-	6	C	R
<i>Deltochilum sp</i>	1	-	-	-	1	*	R
<i>Scybalocanthon sexpilotus</i> (Guérin, 1855)	13	-	-	-	13	C	*
<i>Sylvicanthon sp</i>	14	13	-	-	27	G	*
<i>Canthidium deyrollei</i> (Harold 1867)	950	34	-	-	984	C	T
<i>Canthidium ardens</i> (Bates 1887)	12	0	-	-	12	C	T
<i>Canthidium sp1</i>	42	11	-	-	53	C	T
<i>Canthidium sp2</i>	17	19	-	-	36	G	T
<i>Canthidium sp3</i>	68	-	-	-	68	C	T
<i>Canthidium sp4</i>	3	-	-	7	10	G	T
<i>Canthidium sp5</i>	878	-	12	-	890	C	T
<i>Dichotomius apicalis</i> (Luederwaldt 1931)	64	-	-	-	64	C	T
<i>Dichotomius boreus</i> (Olivier 1789)	34	-	-	-	34	C	T
<i>Dichotomius subaeneus</i> (Castelnau 1840)	183	-	-	-	183	C	T
<i>Dichotomius lucasi</i> (Harold 1869)	24	124	-	-	148	N	T
<i>Dichotomius worontzowi</i> (Pereira 1942)	-	16	-	-	16	N	T
<i>Dichotomius melzeri</i> (Luederwaldt 1922)	4	-	-	-	4	C	T
<i>Deltochilum carinatum</i> (Westwood 1848)	3	-	-	-	3	*	R
<i>Dichotomius sp1</i>	701	-	-	-	701	C	T
<i>Dichotomius sp2</i>	-	3	-	-	3	*	T
<i>Ontherus carinifrons</i> (Luederwaldt 1930)	664	-	-	-	664	C	T
<i>Ontherus pubens</i> (Genier 1966)	23	-	-	-	23	C	T
<i>Ontherus sp</i>	-	-	2	-	2	*	T
<i>Onthophagus bidentatus</i> (Drapiez 1819)	400	-	-	-	400	C	T
<i>Onthophagus haemathopus</i> (Harold 1875)	282	-	-	-	282	C	T
<i>Onthophagus clypeatus</i> (Blanchard 1846)	-	37	-	-	37	N	T
<i>Onthophagus hirculus</i> (Mannerheim 1829)	9	45	-	-	54	N	T
<i>Onthophagus sp1</i>	8	1	2	-	11	G	T
<i>Onthophagus sp2</i>	-	30	-	-	30	N	T
<i>Onthophagus sp3</i>	-	3	-	-	3	*	T
<i>Phanaeus chalcomelas</i> (Perty 1830)	-	14	-	-	14	N	T
<i>Phanaeus bispinus</i> (Bates 1868)	4	-	-	-	4	C	T
<i>Phanaeus sp</i>	-	2	-	-	2	*	T
<i>Diabroctis mimas</i> (Linné 1758)	25	-	-	-	25	C	*

<i>Cryptocanthon peckorum</i> (Howden 1973)	3	-	-	-	3	*	*
<i>Trichillum externepunctatum</i> (Preudhomme de Borre 1880)	94	-	-	-	94	C	D
<i>Digitonthophagus gazella</i> (Fabricius 1787)	1	-	-	-	1	*	*
<i>Gromphas aeruginosa</i> (Perty 1830)	18	-	-	-	18	C	T

Appendix 2

Differences between species richness and abundance of the Scarabaeinae studied in different habitats (NV=Native vegetation, MS=Mature secondary, ES=Early secondary, Ag=Agriculture, Pa=Pasture) for each bait type (Co=Control bait, RB=Rotten banana, RM=Rotten meat, HF=Human feces).

Site/ Bait	Abundance (log+1)	Richness
NV-HF	2.50 ^a	49.33 ^a
NV-RM	1.82 ^b	27.09 ^b
NV-RB	1.30 ^c	11.42 ^c
NV-Co	0.86 ^d	6.38 ^d
	$F_{3,165}=5245^{***}$	$F_{3,165}=1771^*$
MS-HF	2.11 ^a	40.04 ^a
MS -RM	1.38 ^b	16.76 ^b
MS -RB	0.57 ^c	3.52 ^c
MS -Co	0.20 ^d	1.76 ^d
	$F_{3,165}=1047^{***}$	$F_{3,165}=625.1^{**}$
ES-HF	1.27 ^a	13.28 ^a
ES -RM	0.95 ^b	7.61 ^b
ES -RB	0.31 ^c	0.38 ^c
ES -Co	0.1 ^d	0.20 ^c
	$F_{3,165}=571.7^{***}$	$F_{3,165}=964.8^{**}$
Ag-HF	1.01 ^a	7.28 ^a
Ag-RM	0.76 ^b	4.24 ^b
Ag-RB	0.51 ^b	0.01 ^c
Ag-Co	0.10 ^b	0.04 ^c
	$F_{3,65}=266^*$	$F_{3,65}=61.2^{**}$
Pa-HF	1.54 ^a	11.67 ^a
Pa -RM	1.13 ^b	9.14 ^b
Pa -RB	0.24 ^c	1.71 ^c
Pa -Co	0.11 ^d	1.38 ^c
	$F_{3,65}=1202^{***}$	$F_{3,65}=29.1^*$

Asterisks indicate statistical significance at level of * $p<0.05$; ** $p<0.01$; *** $p<0.001$

Chapter 5

Can dung beetles (Scarabaeinae) indicate the status of Amazonia's ecosystems? Insights integrating anthropogenic disturbance with seasonal patterns

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Abstract

Temporal and spatial variation in dung beetles abundances is a pattern observed in many tropical forests. The present study evaluated the seasonal patterns of dung beetles in a range of increasingly disturbed ecosystems of the state of Pará, northern Brazil, to identify valuable disturbance indicators. The areas included native forest, agriculture, pasture for extensive livestock grazing and secondary forests. Fieldwork was carried out encompassing the complete range of environmental conditions encountered during the year. In total, 13,649 individuals were captured within 23 genera and 99 species but with pronounced differences among ecosystems and seasons. The obtained results seem to demonstrate that dung beetles can be used to help identify ecosystems under very complex and variable environmental conditions. The ecological drift observed also demonstrates the possibility of using dung beetles as ecological indicators of disturbance in Amazonia.

Keywords

Amazon rainforest; biodiversity; beetles; scarab; seasonality; tropical forest

Introduction

The landscape of the Brazilian Amazon (Amazonia) is being transformed by vast investments in roads, leading to the growth of urban areas, extensive livestock rearing and intensive farming (Mertens et al., 2002; Tabarelli et al., 2004). These activities are closely associated with forest logging but also with hydroelectric power and mining (Soares-Filho et al., 2005). The new land uses might generate strong ecological impacts that were not fully anticipated (Cajaiba & Silva, 2017), resulting in isolation of animal populations and local extinctions (Aizen et al., 2012; Valiente-Banuet et al., 2015).

From a conservation perspective, the condition of an ecosystem, the status or difference from reference conditions, might be assessed using informative surrogates named ecological indicators (Heink & Kowarik, 2010; Costanza, 2012; Heath, 2013). Indicator species should be highly sensitive to changes in the structure and functioning of an ecosystem and easily monitored, providing valuable information on the system's qualitative status (Rapport & Hildén, 2013). Dung beetles (DB; Coleoptera: Scarabaeinae) are considered particularly informative (Gardner et al., 2008; Da Silva et al., 2013), because of their sensitivity to abiotic and biotic factors. In fact, soil depth, soil structure and porosity, humidity, temperature, soil pH, and pollution (Nichols et al., 2008; Viegas et al., 2014; Campos & Hernández, 2015; Cajaiba et al., 2017) and the composition of the vertebrate community are strongly correlated with DB communities (Spector, 2006). Their roles as herbivores, carnivores, omnivores, scavengers (Vandewalle et al., 2010) as well as pollinators, seed dispersers, and decomposers highlight their importance in the ecosystems (Nichols et al., 2008; Vandewalle et al., 2010; Bicknell et al., 2014). The suitability of DB for monitoring

the effects of subtle changes in the ecosystems was demonstrated by Scheffler (2005) and Nichols & Gardner (2011).

Diverse methods developed for Amazonia predict huge landscape changes in the future, with warmer and dryer climates, while the status of ecosystems remains without realistic projections (Marengo, 2015; Marengo et al., 2016). This preliminary work examines differences in DB neotropical communities in different seasons and/or associated with ecosystems with increasing anthropogenic disturbances to determine their usefulness as ecological indicators.

This information might guide the construction of more robust ecological assessments aimed at envisaging possible changes in the status of Amazonia's pristine ecosystems, integrating the multiple factors associated with DB dynamics and highlighting the most effective management practices through quantitative metrics (Santos et al., 2016a, b).

Material and methods

Study sites

The study was performed in the municipality of Uruará, state of Pará, Northern Brazil (-03°43'27"S, -53°44'8"W). The dominant land use/land cover of the study area was natural forests (69% of the area). Extensive livestock production and the exploitation of timber at a large scale (mostly illegal) are currently considered the most serious environmental threats (Cajaiba et al., 2016). The climate of the study area is classified as Aw (Köppen), i.e. hot and humid; the average annual rainfall is 2000 mm (Peel et al., 2007). The studied areas encompass ecosystems that, in terms of physical characteristics and anthropogenic disturbances, are representative of the region: Native Vegetation (NV), Mature Secondary succession (MS: vegetation with 15 years of

regeneration), Early Secondary succession (ES: vegetation with five years of regeneration), Agriculture (Ag: cocoa plantations, *Theobroma cacao* L.) and Pasture for extensive livestock (Pa) (fig. 1). This gradient in disturbance was considered fundamental to analyse the response of the DB communities (Cajaiba et al., 2017).

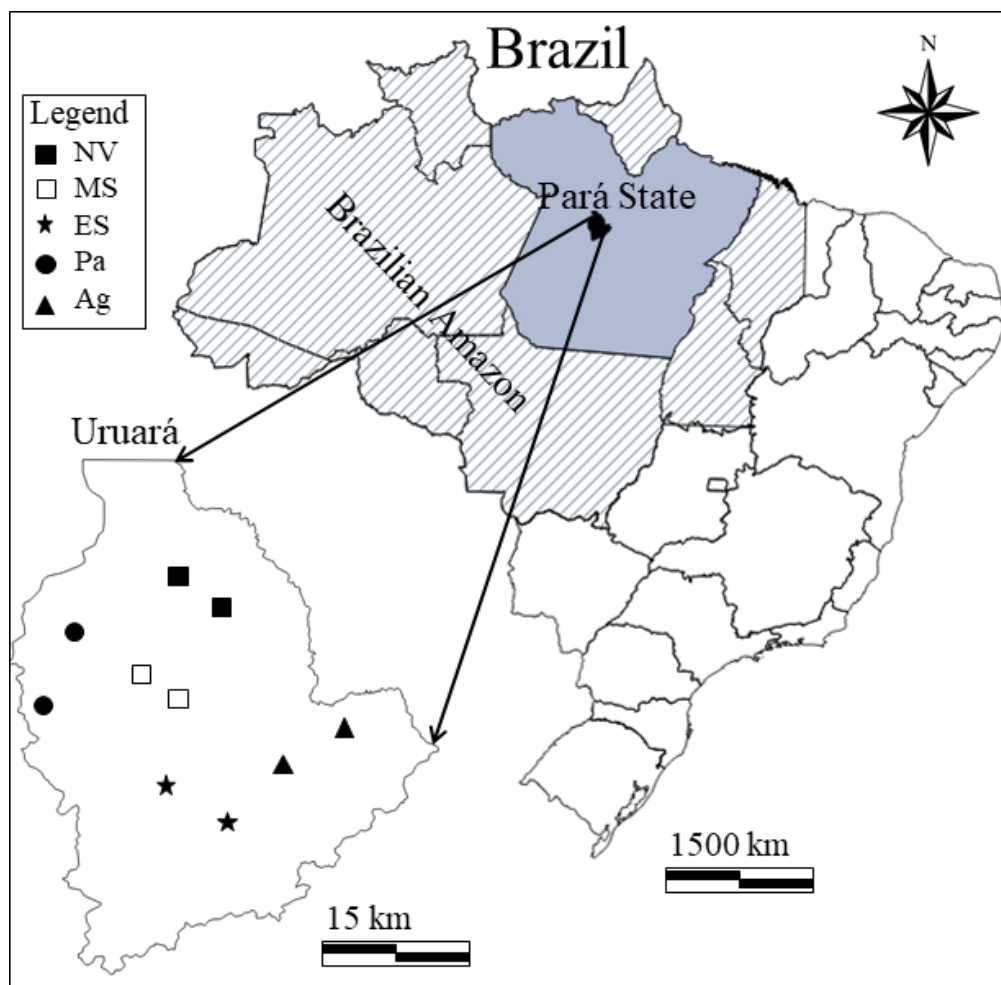


Figure 1. Location of the study sites in the municipality of Uruará, state of Pará, northern Brazil. Abbreviations: Ag, Agriculture; ES, Early secondary succession; MS, Mature secondary succession; NV, Native vegetation; Pa, Pasture.

Sampling method

Sampling was carried out in February/March (rainy season), June (intermediary season), and September/October (dry season) of the year 2015. This allowed checking for seasonal differences in the activity and structure of DB communities. The sampling points were placed at a minimum distance of 100 m from ecotones to guarantee that most DB captured in the pitfalls were associated with the monitored ecosystem. Pitfall traps (75 mm diameter and 110 mm deep) were filled with formalin, alcohol, water, and a few drops of detergent. Each pitfall was covered by a roof to prevent rainwater from entering, and each trap remained installed for 48 h prior to collection. Each pitfall contained different types of bait: HF, human feces; RM, rotten meat; and RB, rotten banana, in order to attract different species according to their feeding habits. Non-baited pitfalls were used as control (Co).

Seven sampling points were placed at each study site at distances of 100 m from each other. Each sample point contained four pitfall traps separated by a distance of 5 m and including the different baits (HF, RM, RB, Co). This protocol was applied to all ecosystems and monitoring periods, creating a total sampling effort of 840 traps. The DB collected were conserved in 70% ethanol, taken to the laboratory and identified to the species level when possible or assigned to morphospecies. The identification was based on the keys proposed by Vaz-de-Mello et al. (2011) and Pacheco & Vaz-de-Mello (2015).

Data analysis

One-way analysis of variance (ANOVA) was used followed by the Tukey test to test for: a) possible differences in the abundance and richness of dung beetles among

ecosystems; b) possible differences in abundance and richness in different sampling periods (seasonal variation) and within each ecosystem (Zar, 1984). Bray-Curtis cluster analysis was applied to verify the similarity between different ecosystems and seasons, and the UPGMA algorithm was used to depict the distance based on the Bray-Curtis index. This index ranges between 0 (indicating no similarity in community composition between sites) and 1 (indicating complete overlap), and it is considered one of the most robust measures of community similarity (Magurran, 2004). The cophenetic correlation coefficient was used to verify that the result of the cluster analysis was significant. In order to check for environmental variables that influence the DB communities at particular periods of the year, a correlation analysis (Pearson correlation) was applied between DB abundance and richness and meteorological data – temperature, humidity and precipitation. The normality of the data was verified by the Shapiro-Wilk test. In order to homogenize the variances and normalize the residues the abundance was transformed by $\log(x + 1)$. All analyses were performed using PAST software version 3.14 (Hammer et al., 2001).

Results

A total of 13 649 dung beetles were captured within 23 genera and 99 species. The genera with the greatest abundances were *Ateuchus* and *Canthon*, which represented about 24% and 20% of the total number of individuals, respectively. The highest number of exclusive species was associated with the NV (11 species), followed by Pa (five species) and ES (four species), while MS and Ag presented only one exclusive species each (Appendix, table A1). DB abundances were dissimilar: the ecosystem with the higher abundances was the Native Vegetation (NV, 8071 individuals),

followed by Mature Sucession (MS, with 3278 individuals), Pasture (Pa with 1100 individuals), Early succession (ES, with 665 individuals), and finally Agriculture (Ag, with 389 individuals). According to the ANOVA, there was a statistical difference in abundance between ecosystems ($F_{4,205} = 84.59$, $P < 0.001$; fig. 2A; see Appendix, table A2, for details of the associated differences and Tukey post-hoc values).

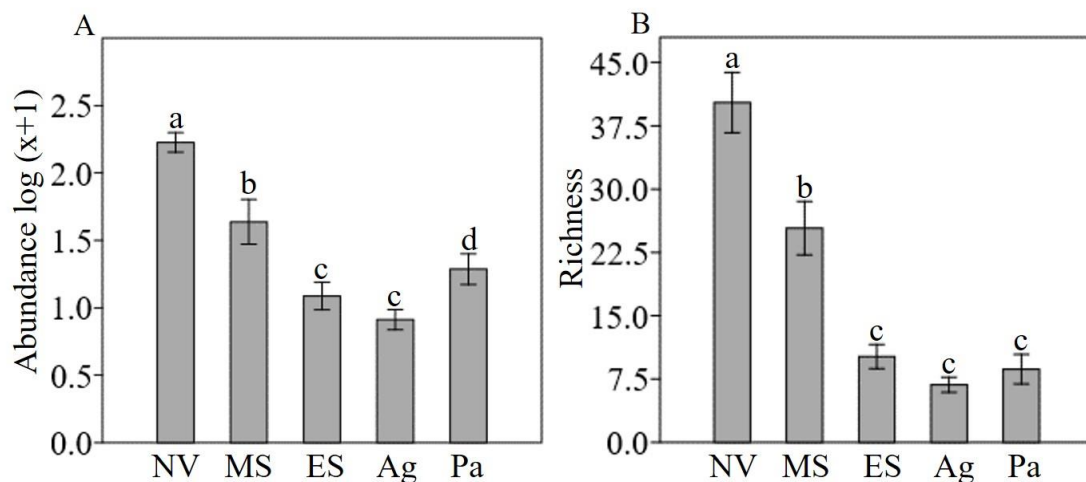


Figure 2. (A) Abundance (number of individuals \pm SE), and (B) richness (number of species \pm SE) of the DB community in the different ecosystems considered. The values labelled with the same letters are not significantly different according to the Tukey post-hoc test. Abbreviations: Ag, agriculture; ES, early secondary succession (vegetation with five years of regeneration); MS, mature secondary succession (vegetation with 15 years of regeneration); NV, native vegetation; Pa, pasture.

Regarding species richness, there were large variations between habitats. The greatest richness was found in the NV (63 species), while 50 species were collected from the MS. A total of 20 species were obtained from the ES and only 12 species from the Ag and 21 for Pa (Appendix, table A1). ANOVA showed statistically significant differences between the studied habitats ($F_{4,205} = 137.3$, $P < 0.001$; fig. 2B;

see Appendix, table A2, for details of the associated differences and Tukey post-hoc values).

Regarding seasonal differences, 8356 individuals were collected in the rainy season, followed by the intermediate season with 2856 individuals and the dry season with 2437 individuals (Appendix, table A1). According to the ANOVA, differences between the sampling periods are statistically significant ($F_{2,207} = 20.25$, $P < 0.05$; fig. 3A; see Appendix, table A3, for details of the associated differences and Tukey post-hoc values).

With respect to species richness, of the 99 species sampled, 54 (54.5%) occurred during all seasons. The highest number of exclusive species was associated with the rainy season (17 species = 17.2%), followed by the intermediary and the dry seasons with three and two exclusive species, respectively. ANOVA showed that there were differences in richness among periods ($F_{2,207} = 11.59$, $P < 0.0001$; fig. 3B) (see Appendix, table A3, for details of the associated differences and Tukey post-hoc values).

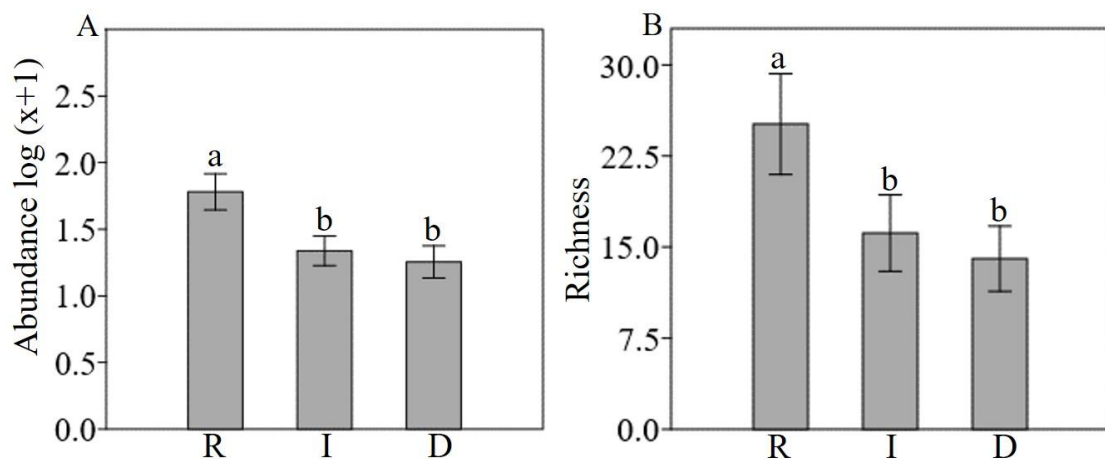


Figure 3. (A) Abundance (number of individuals \pm SE) and (B) richness (number of species \pm SE) of dung beetles in different periods of sampling in the Brazilian Amazon. The values labelled with the same letters are not significantly different according to the Tukey post-hoc test. Abbreviations: D, dry; I, intermediary; R, rainy.

Among the climatic variables evaluated, humidity and precipitation were positively correlated with the abundance and richness of DB. In contrast, the air temperature influenced these indexes negatively (table 1).

Table 1. Pearson correlation between environmental variables and ecological indices (abundance and richness) of dung beetles collected in Uruará, Pará, northern Brazil. Asterisks indicate statistical significance at levels of: * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.

Environmental variables	Abundance log (x + 1)	Richness
Humidity	0.54**	0.60**
Temperature	-0.51*	-0.57**
Precipitation	0.44*	0.32***

Even though abundance and richness attained generally higher values in the rainy season, this seasonal trend was not the same for all ecosystems. In Ag, abundance and richness were higher in the intermediary period and Pa also presented greater richness in the intermediate season (fig. 4; see Appendix, table A4, for details of the associated differences and Tukey posthoc values).

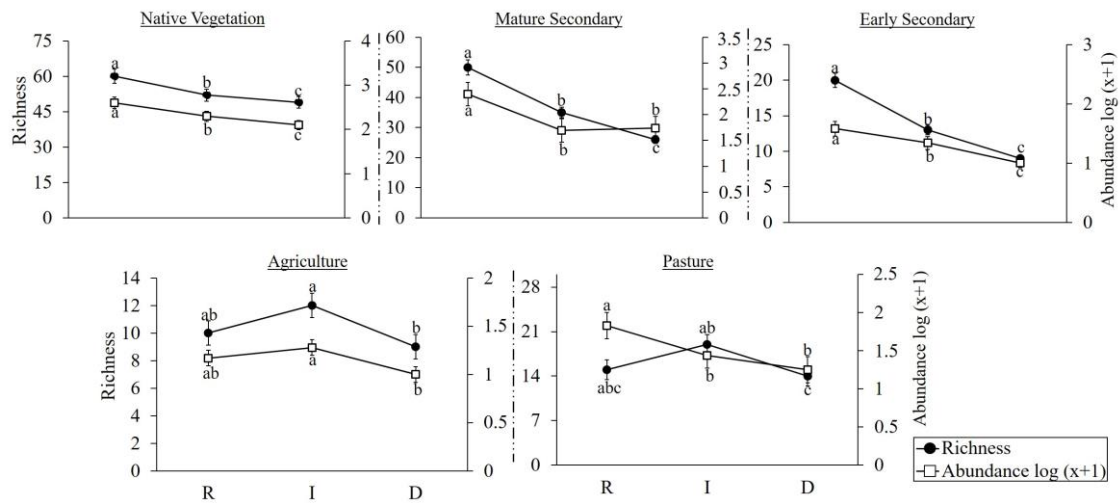


Figure 4. Temporal variation of the abundance and richness of dung beetles by ecosystem. The values followed by the same letters are not significantly different according to the Tukey post-hoc test. Abbreviations: D, dry; I, intermediary; R, rainy.

The community structure, based on individual abundance per species and per ecosystem/sampling period, showed the high similarity of NV in the rainy season (NV_R) and MS in the rainy season (MS_R) (Bray-Curtis similarity index 0.63), followed by NV in the intermediate season (NV_I) and NV in the rainy season (NV_R) (Bray-Curtis similarity index 0.62), which were quite different from the other assemblages. The lowest similarity observed was between NV in the rainy season (NV_R) and ES in the dry season (ES_D) (Bray-Curtis similarity index 0.02). In general terms the more pristine ecosystems are quite apart from the more disturbed ones, even though climatic conditions tend to make the associated DB communities more or less similar. The cophenetic correlation coefficient for the dendrogram was 0.85 (fig. 5).

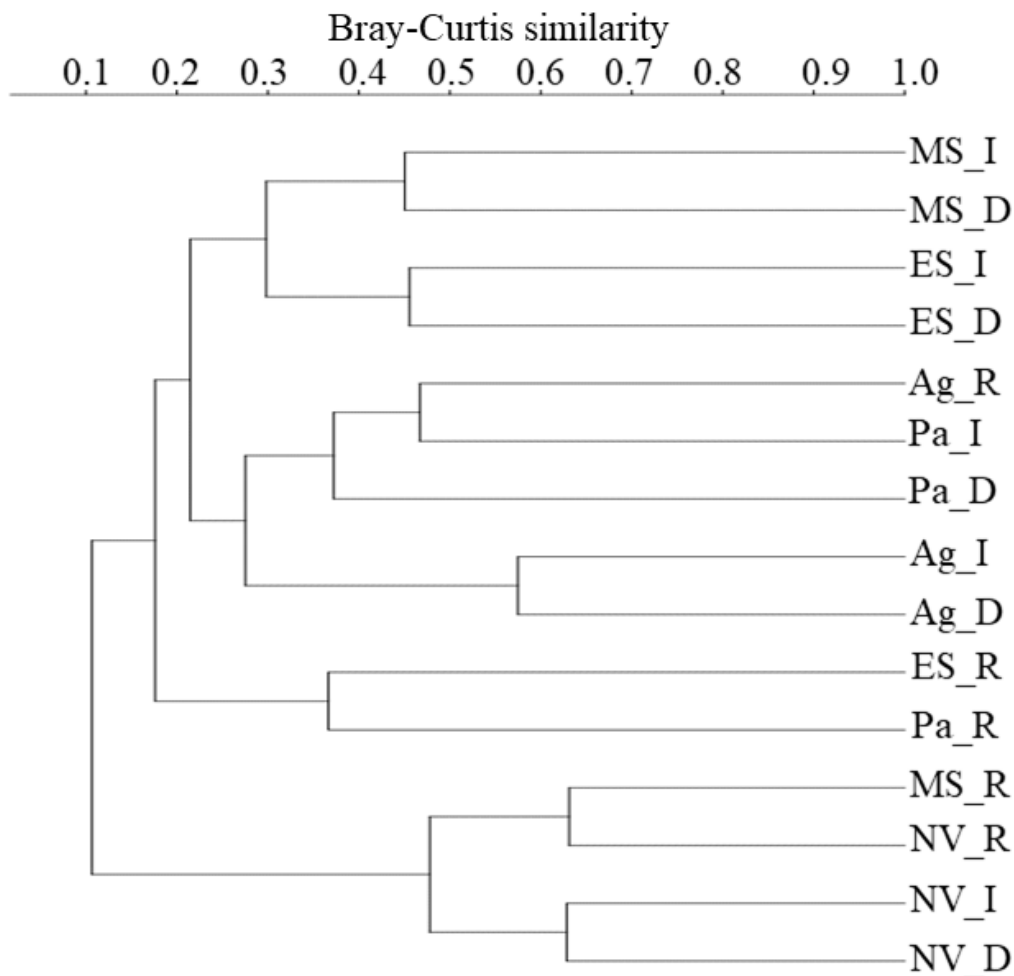


Figure 5. Bray-Curtis similarity index for the different habitats and season sampled (cophenetic correlation coefficient = 0.85). Abbreviations: Ag, agriculture; D, dry; ES, early secondary succession (vegetation with five years of regeneration); I, intermediary; MS, mature secondary succession (vegetation with 15 years of regeneration); NV, native vegetation; Pa, pasture; R, rainy.

Discussion

Results show that the DB community in the study area is very diverse and, like other megadiverse ecosystems in the neotropics, understudied (Lucky et al., 2002). The DB richness (99 species) recorded at Uruará was even higher than the values reported for Malaysia (Davis, 2000), Peru (Valencia, 2001), French Guyana (Feer, 2000), African rain forests (Cambefort & Walter, 1991), rain forests of Mexico (Estrada & Coates-

Estrada, 2002), Colombia (Escobar, 2000), Australia (Howden et al., 1991) and even for the Brazilian rain forests (Korasaki et al., 2013; França et al., 2016).

Regarding the types of ecosystems studied, abundance and richness were higher in Native Vegetation (NV) and Mature Secondary succession (MS) than in the other systems. This trend might be associated with the complex structure of NV and, in particular for MS (Nichols et al., 2009; Bicknell et al., 2014), linked with a range of environmental characteristics that are more favorable for dung beetles' survival and reproduction (deeper and softer soils, higher soil moisture content, more stable air and soil temperature, lower insolation, and higher concentration of food resources; Andresen, 2005). We also observed differences in the species composition: the most disturbed ecosystems (Ag and Pa) are dominated mainly by generalist beetles that occur both in forests and anthropogenic areas (Cajaiba et al., 2017).

The distribution of dung beetles along different environmental gradients, represented by the monitored ecosystems, seems to demonstrate discrete associations with particular biotypes within the landscape (Viegas et al., 2014; Da Silva & Hernández, 2016). The decrease in richness and abundance seem to be negatively correlated with disturbance (Gardner et al., 2008; Da Silva & Hernández, 2016; Cajaiba et al., 2017), which is expected to reduce the populations of several mammalian species (Culot et al., 2013) and, therefore, the food resources for dung beetles (Da Silva & Hernández, 2016).

Climatic conditions also seem to determine the DB communities, with general decreases in diversity during the dry season. Although more studies are needed to describe accurately the seasonal patterns detected, most species were observed to have their abundance reduced or even to be completely absent during the driest season, in

accordance with other work (e.g. Hanski & Cambefort, 1991; Milhomem et al., 2003, in tropical forests; Seymour & Jones, 2000; Nyeko, 2009, for other regions of the world). Silva et al. (2010) suggested two hypotheses that may explain the lower abundance and richness of dung beetles in the dry season: 1) adults in open habitats are sensitive to the effects of drought and remain underground during this period; or 2) the adults die in the dry season and only the immature beetles survive in brood chambers, reaching the adult stage at the beginning of the rainy season. However, according to Cajaiba & Silva (2017), biotic responses to climate changes are not easily understood or predictable because the responses differ between species, suggesting that disturbance history may be an important determinant in the occurrence of changes. Prior knowledge and temporal monitoring of dung beetles communities are required to recognize the disturbances caused by and consequent responses to possible new climatic conditions associated with global change.

Dung beetle species collected in this study demonstrated ecosystem preferences as well as seasonal/climatic trends. The rainy season and native vegetation presented the highest index overall (e.g. abundance and richness). Therefore, information on dung beetles will provide support to future actions for selecting priority areas for preservation in the region, where human activity increasingly threatens the maintenance of natural ecosystems (Da Silva et al., 2013). Although recent studies have increasingly addressed seasonal effects on the ecology of dung beetles in Amazonia, for the majority of species these questions remain to be solved, thus requiring specific studies to understand the relation between dung beetles and environmental conditions and for predicting their usefulness as ecological indicators in changing conditions. Despite this limitation, which is inherent to a demonstrative study

case in progress, the work presented here is of value for studying ecosystems characterized by a high degree of heterogeneity in space and time, influenced by many interacting factors and feedback mechanisms and where ecological indicators are particularly helpful to capture these multi-factor influences.

Final remarks

Current principles used to guide conservation and management consider that protection of indicator species is an efficient way to conserve overall biodiversity and sustain key ecological processes. By using these indicators it is possible to predict how anthropogenic and natural environmental changes affect the species and the communities in disturbed ecosystems. For conservation and management purposes, the use of the correct ecological indicators may reveal what are the consequences of changing environmental factors for the integrity of ecosystems. Although the observed discrepancies between ecosystems could be also associated to seasonal differences, our preliminary work depicts the possibility of using dung beetle communities as ecological indicators, presenting an important source of information to policy makers and helping to guide decision-making.

Acknowledgments

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Supplementary material

Table S1.

Scarabaeinae species (Dung beetles, Coleoptera: Scarabaeidae) collected in different ecosystems and different seasons in the municipality of Uruará, Pará State in northern Brazil. NV: Native Vegetation; MS: Mature secondary succession; ES: Early secondary succession; Ag: Agriculture (Cacao plantations); Pa: Pasture. The letters R, I and D following the vegetation types correspond the rainy season, intermediary and dry, respectively.

Species	NV- R	NV- I	NV- D	MS- R	MS- I	MS- D	ES- R	ES- I	ES- D	Ag- R	Ag- I	Ag- D	Pa- R	Pa- I	Pa- D
<i>Coprophanaeus lancifer</i>	29	18	1	16	23	-	-	-	-	-	-	-	-	-	-
<i>Coprophanaeus telamon</i>	19	21	1	-	-	-	-	-	-	-	-	-	-	-	-
<i>Coprophanaeus degallieri</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	2	-
<i>Coprophanaeus</i> sp1	5	2	-	-	-	1	-	-	-	-	-	-	-	-	-
<i>Oxysternon durantoni</i>	76	29	-	-	-	-	-	-	-	-	-	-	-	-	-
<i>Oxysternon conspicillatum</i>	12	10	1	-	-	-	41	2	-	-	-	-	-	-	-
<i>Oxysternon</i> sp1	19	-	-	-	-	-	-	-	-	-	-	-	-	-	-
<i>Sulcophanaeus faunus</i>	4	-	-	-	2	-	-	-	-	-	-	-	-	-	-
<i>Sulcophanaeus</i> sp1	-	-	-	-	-	-	-	-	-	-	-	-	3	-	-
<i>Eurysternus atrosericus</i>	27	41	13	17	1	12	-	-	-	3	5	-	-	-	-
<i>Eurysternus parallelus</i>	36	61	36	18	6	30	-	6	-	5	12	7	-	-	-
<i>Eurysternus caribaeus</i>	81	51	42	44	10	22	-	11	-	2	-	7	-	-	-
<i>Eurysternus wittmerorum</i>	35	24	12	15	6	33	-	9	-	-	3	-	-	-	-
<i>Eurysternus strigilatus</i>	15	22	-	5	5	-	-	-	-	-	-	-	-	-	-
<i>Eurysternus ventricosus</i>	17	-	-	3	-	-	-	-	-	-	-	-	-	-	-
<i>Eurysternus foedus</i>	-	15	6	-	-	20	-	8	-	-	-	5	-	-	-
<i>Eurysternus hamaticollis</i>	-	9	5	-	-	12	-	12	-	-	-	-	-	-	-
<i>Eurysternus</i> sp1	18	22	35	19	2	11	-	-	9	-	11	5	-	-	-
<i>Eurysternus</i> sp2	17	29	-	17	3	26	9	12	-	-	-	-	-	-	-
<i>Eurysternus</i> sp3	-	41	49	14	35	69	18	15	11	-	-	-	-	-	-
<i>Uroxys</i> sp1	198	90	32	-	-	-	-	-	-	-	-	-	-	-	-
<i>Uroxys</i> sp2	37	29	24	-	-	-	-	-	-	-	-	-	-	-	-
<i>Ateuchus pygidiali</i>	142	40	42	51	6	-	-	-	-	-	-	-	-	-	-
<i>Ateuchus murrayi</i>	212	160	98	98	-	-	-	-	-	-	-	-	98	-	-
<i>Ateuchus connexus</i>	143	78	83	-	-	-	-	-	-	-	-	-	-	-	-
<i>Ateuchus candezei</i>	51	65	17	33	11	-	-	-	-	-	-	-	-	-	-
<i>Ateuchus robustus</i>	154	96	73	49	23	37	-	6	-	-	-	-	-	-	-
<i>Ateuchus simplex</i>	34	25	-	9	4	-	-	-	-	-	-	-	-	-	-
<i>Ateuchus</i> sp1	29	-	-	-	6	27	11	-	-	3	-	8	15	7	2
<i>Ateuchus</i> sp2	51	-	-	-	-	15	-	-	-	-	-	-	21	12	3
<i>Ateuchus</i> sp3	141	57	98	37	34	27	-	14	12	4	-	-	-	-	-
<i>Ateuchus</i> sp4	118	41	22	34	3	18	-	-	4	-	-	-	12	-	-
<i>Ateuchus</i> sp5	96	131	93	48	12	28	24	4	3	2	-	-	-	-	-

<i>Ateuchus</i> sp6	30	20	18	3	8	26	-	-	-	-	-	-	-	-	-
<i>Ateuchus</i> sp7	2	17	9	39	8	-	-	-	-	-	-	-	-	-	-
<i>Ateuchus</i> sp8	-	-	-	35	8	-	-	-	-	-	-	-	-	2	-
<i>Anomiopus</i> sp1	2	-	-	-	-	-	-	-	-	-	-	-	-	-	-
<i>Canthon luteicollis</i>	73	43	20	64	24	19	10	8	13	7	2	-	-	6	-
<i>Canthon simulans</i>	11	-	-	-	5	20	40	14	12	-	9	6	173	50	9
<i>Canthon chalybaeus</i>	-	-	-	-	-	-	9	6	-	-	-	-	-	-	-
<i>Canthon aequinoctialis</i>	43	20	18	27	9	-	-	-	-	7	-	5	-	-	-
<i>Canthon fulgidus</i>	49	19	-	59	4	21	22	20	6	12	-	.	12	29	65
<i>Canthon quadriguttatus</i>	12	9	-	6	3	-	-	-	-	3	-	-	-	3	8
<i>Canthon triangularis</i>	116	27	22	71	18	-	-	-	-	-	-	-	-	-	-
<i>Canthon subhyalinus</i>	18	8	-	9	-	18	-	-	-	2	-	-	-	-	3
<i>Canthon lituratus</i>	24	21	23	15	-	-	68	9	-	-	-	-	90	2	6
<i>Canthon mutabilis</i>	-	-	-	-	-	-	25	8	-	-	-	-	11	4	1
<i>Canthon histrio</i>	-	-	-	-	-	-	-	-	-	-	-	-	8	4	-
<i>Canthon</i> sp1	-	-	-	-	2	-	-	-	-	9	12	-	-	-	-
<i>Canthon</i> sp2	87	125	100	39	24	27	-	3	8	43	33	30	13	61	3
<i>Canthon</i> sp3	11	12	1	-	5	-	-	-	-	-	14	13	-	-	-
<i>Canthon</i> sp4	-	-	-	-	-	-	-	-	-	-	24	5	-	14	-
<i>Canthon</i> sp5	32	4	-	28	9	16	32	-	-	20	5	-	27	36	48
<i>Canthon</i> sp6	-	-	21	-	-	-	-	11	5	2	7	-	-	-	-
<i>Canthon</i> sp7	-	-	-	-	-	-	12	4	14	-	-	-	-	-	-
<i>Canthon</i> sp8	-	-	-	-	-	-	7	-	-	-	-	-	-	-	-
<i>Canthonella</i> sp	23	-	3	-	-	-	-	-	-	-	-	-	13	3	-
<i>Deltochilum orbiculare</i>	46	12	3	19	1	8	-	-	-	-	-	-	-	-	2
<i>Deltochilum submetallicum</i>	225	-	112	173	-	-	-	-	-	-	-	-	-	-	-
<i>Deltochilum carinatum</i>	8	-	-	-	-	-	-	-	-	-	-	-	-	-	-
<i>Deltochilum icarus</i>	4	-	-	2	-	-	-	-	-	-	-	-	-	-	-
<i>Deltochilum</i> sp	-	-	-	-	1	-	-	-	-	-	-	-	-	-	-
<i>Scybalocanthon sexpilotus</i>	12	-	-	1	-	-	-	-	-	-	-	-	-	-	-
<i>Sylvicanthon</i> sp1	4	8	3	1	-	9	-	-	-	-	-	-	2	-	-
<i>Canthidium deyrollei</i>	401	25	122	224	7	5	-	-	-	-	-	-	-	-	-
<i>Canthidium ardens</i>	-	-	-	-	-	-	-	-	-	-	-	-	12	-	-
<i>Canthidium</i> sp1	6	-	-	5	-	18	-	-	-	9	6	5	10	-	-
<i>Canthidium</i> sp2	9	-	-	3	-	-	-	-	-	7	-	-	15	3	2
<i>Canthidium</i> sp3	54	-	-	14	-	-	-	-	-	-	-	-	-	-	-
<i>Canthidium</i> sp4	-	-	7	-	-	-	-	-	-	-	-	3	-	-	-
<i>Canthidium</i> sp5	388	-	99	196	-	-	-	-	-	-	-	-	-	-	-
<i>Dichotomius apicalis</i>	26	10	7	21	-	-	-	-	-	-	-	-	-	-	-
<i>Dichotomius boreus</i>	17	8	3	6	-	-	-	-	-	-	-	-	-	-	-
<i>Dichotomius subaeneus</i>	59	12	9	82	-	-	-	-	-	-	-	-	-	-	-
<i>Dichotomius lucasi</i>	74	3	1	52	18	-	-	-	-	-	-	-	-	-	-
<i>Dichotomius worontzowi</i>	10	-	-	6	-	-	-	-	-	-	-	-	-	-	-
<i>Dichotomius melzeri</i>	-	3	1	-	-	-	-	-	-	-	-	-	-	-	-

<i>Dichotomius</i> sp1	404	-	-	297	-	-	-	-	-	-	-	-	-	-	-
<i>Dichotomius</i> sp2	-	-	-	-	-	3	-	-	-	-	-	-	-	-	-
<i>Ontherus carinifrons</i>	321	98	44	201	-	-	-	-	-	-	-	-	-	-	-
<i>Ontherus pubens</i>	-	-	-	-	-	-	-	-	-	-	-	-	23	-	-
<i>Ontherus</i> sp	-	-	2	-	-	-	-	-	-	-	-	-	-	-	-
<i>Onthophagus bidentatus</i>	191	78	33	98	-	-	-	-	-	-	-	-	-	-	-
<i>Onthophagus haemathopus</i>	82	94	37	69	-	-	-	-	-	-	-	-	-	-	-
<i>Onthophagus clypeatus</i>	21	15	1	-	-	-	-	-	-	-	-	-	-	-	-
<i>Onthophagus hirculus</i>	-	-	-	-	-	-	-	-	-	-	-	-	40	14	-
<i>Onthophagus</i> sp1	3	2	-	-	-	-	-	-	-	-	-	-	3	-	-
<i>Onthophagus</i> sp2	12	13	1	5	-	-	-	-	-	-	-	-	-	-	-
<i>Onthophagus</i> sp3	-	2	1	-	-	-	-	-	-	-	-	-	-	-	-
<i>Phanaeus chalcomelas</i>	9	-	-	4	1	-	-	-	-	-	-	-	-	-	-
<i>Phanaeus bispinus</i>	-	3	1	-	-	-	-	-	-	-	-	-	-	-	-
<i>Phanaeus</i> sp	-	-	-	-	-	2	-	-	-	-	-	-	-	-	-
<i>Blackburneus</i> sp	-	8	4	-	-	-	-	-	-	3	-	12	-	-	-
<i>Diabroctis mimas</i>	-	-	-	-	-	25	-	-	-	-	-	-	-	-	-
<i>Cryptocanthon peckorum</i>	-	-	-	-	-	2	1	-	-	-	-	-	-	-	-
<i>Trichillum externepunctatum</i>	-	-	-	-	-	29	-	-	-	-	-	65	-	-	-
<i>Digitonthophagus gazella</i>	-	-	-	-	-	-	-	-	-	2	-	-	-	-	-
<i>Gromphas aeruginosa</i>	-	-	-	-	-	-	-	-	-	-	-	18	-	-	-

Table S2.

Tukey values for number of individuals (abundance) and number of species (richness) of the DB community in the different ecosystems considered. Tukey's Q the diagonal, p (same) above the diagonal. Not-significant comparisons are gray. NV, Native Vegetation; MS, Mature secondary succession (15 years regeneration); ES, Early secondary succession (5 years regeneration); Ag, Agriculture (Cacao plantations), and Pa, Pasture.

Site	Abundance log (x+1)					Richness				
	$F_{4,205} = 84.59, P < 0.001$					$F_{4,205} = 137.3, P < 0.001$				
	NV	MS	ES	Ag	Pa	NV	MS	ES	Ag	Pa
NV		P<0.001	P<0.001	P<0.001	P<0.001	-	P<0.001	P<0.001	P<0.001	P<0.001
MS	Q=10.41	-	P<0.001	P<0.001	P<0.001	Q=12.15	-	P<0.001	P<0.001	P<0.001
ES	Q=20.13	Q=9.72	-	P=0.183	P=0.092	Q=24.58	Q=12.43	-	P=0.296	P=0.909
Ag	Q=23.23	Q=12.82	Q=3.09	-	P<0.001	Q=27.32	Q=15.17	Q=2.74	-	P=0.820
Pa	Q=16.60	Q=6.19	Q=3.52	Q=6.62	-	Q=25.80	Q=13.65	Q=1.22	Q=1.51	-

Table S3.

Tukey values for number of individuals (abundance) and number of species (richness) of the DB community in the different periods of sampling. Tukey's Q the diagonal, p (same) above the diagonal. Not-significant comparisons are gray. R: Rainy; I: Intermediary; D: Dry.

Periods	Abundance log (x+1)			Richness		
	$F_{2,207} = 20.25, P < 0.05$			$F_{2,207} = 11.59, P < 0.0001$		
	R	I	D	R	I	D
R	-	P<0.001	P<0.001	-	P<0.001	P<0.001
I	Q=7.05	-	P=0.62	Q=5.19	-	P=0.66
D	Q=8.36	Q=1.31	-	Q=6.41	Q=1.22	-

Table S4.

Tukey values for number of individuals (abundance) and number of species (richness) of the DB community in the different periods of sampling in each habitat. NV: Native Vegetation; MS: Mature secondary succession; ES: Early secondary succession; Ag: Agriculture (Cacao plantations); Pa: Pasture. The letters R, I and D following the vegetation types correspond the rainy season, intermediary and dry, respectively.

Site/ Sample	Abundance log (x+1)			Richness		
	R	I	D	R	I	D
	$F_{2,82} = 142.7, P < 0.05$			$F_{2,82} = 20.2, P < 0.05$		
NV_R	-	P<0.0001	P<0.0001	-	P<0.001	P<0.01
NV_I	Q=17.51	-	P<0.001	Q=5.64	-	P=0.06
NV_D	Q=22.84	Q=5.33	-	Q=3.23	Q=3.23	-
	$F_{2,82} = 151.1, P < 0.01$			$F_{2,82} = 51.49, P < 0.001$		
MS_R	-	P=<0.001	P=<0.001	-	P<0.001	P<0.001
MS_I	Q=24.2	-	P=0.071	Q=13.75	-	P=<0.001
MS_D	Q=17.21	Q=6.98	-	Q=10.44	Q=3.31	-
	$F_{2,82} = 29.02, P < 0.01$			$F_{2,82} = 12.7, P < 0.01$		
ES_R	-	P<0.01	P<0.01	-	P<0.05	P<0.001
ES_I	Q=7.15	-	P=0.05	Q=3.59	-	P<0.05
ES_D	Q=10.55	Q=3.40	-	Q=7.12	Q=3.52	-
	$F_{2,82} = 15.97, P < 0.05$			$F_{2,82} = 10.11, P < 0.05$		
Ag_R	-	P=0.99	P=0.06	-	P=0.96	P=0.12
Ag_I	Q=0.07	-	P<0.05	Q=0.39	-	P<0.05
Ag_D	Q=3.41	Q=3.49	-	Q=2.84	Q=3.22	-
	$F_{2,82} = 48.9, P < 0.001$			$F_{2,82} = 22.64, P < 0.05$		
Pa_R	-	P<0.001	P<0.001	-	P=0.07	P=0.06
Pa_I	Q=10.1	-	P=0.07	Q=11.89	-	P<0.05
Pa_D	Q=13.28	Q=3.18	-	Q=7.12	Q=2.23	-

Chapter 6

SEASONAL PATTERNS IN THE DIVERSITY OF HISTERID BEETLES (HISTERIDAE) ARE ECOSYSTEM SPECIFIC? A CASE IN PARA STATE, NORTHERN BRAZIL

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Abstract

The objective of this work was to characterize the seasonal variation in the community of Histerid beetles (Histeridae) of different ecosystems in the Brazilian Amazon, ranging from primary or secondary forests with different stages of regeneration to cocoa farms and pastures. Pitfall traps were distributed within the monitored ecosystems during different periods of the year. A total of 1945 Histerid beetles, belonging to five genera and 14 species were captured. Higher diversities were observed during the rainy season, although with differences among ecosystems. The genera *Phelister* and *Hister* were ubiquitous in all ecosystems, constituting 71% of all the specimens captured. Histerid beetle communities, discriminated by ordination methods, change gradually from the most conserved ecosystems to more disturbed ones. Moreover, the results showed that disturbed ecosystems, namely cocoa farms and pastures, have detrimental effects on the occurrence of Histerid beetles, which are considered forest specialists, while enhancing generalist species.

Keywords: *Amazon rainforest, biodiversity, soil insect, fluctuation, tropical forest*

Introduction

Currently, there is enormous concern and speculation about the effects of anthropogenic disturbance on the biodiversity of tropical ecosystems, including the effects on species composition and the modifications in the ecological services provided (e.g. Morris, 2010; Aerts and Honnay, 2011; Cajariba et al., 2017a). Deforestation in the Neotropics, particularly in the Brazilian Amazon (Amazonia), has been causing a sharp erosion of biodiversity and disruption of the complex global climate phenomena (e.g. Hassan et al., 2005; Morris, 2010). Therefore, the conservation of the Amazonian ecosystems represents a growing core challenge for sustaining earth's functioning and ultimately that of mankind (e.g. Viana and Pinheiro, 1988; Viegas et al., 2014).

Predicting the ecological consequences of land use/land cover (LU/LC) changes is therefore a subject of scientific and political interest in order to support strategic options for sustainable development, land use planning and natural resources management (Turner et al., 2007). In this context, ecological assessment and monitoring are important tools to support effective management of ecosystems and natural resources, in which the use of pertinent indicators is crucial for measuring and evaluating the status and trends of target environmental systems (Cajariba and Silva, 2017).

Ecological integrity is a key concept in natural resource management, and researchers have been trying to improve public understanding of this concept by using simple scientific measures known as ecological indicators (Andreasen et al., 2001). In fact, ecological integrity can be measured and interpreted by changes in abundance, diversity, and composition of groups of indicator organisms that ultimately depend on system resources and conditions. Correlates of integrity in terrestrial landscapes have been proposed, along with many specific indicators that measure various aspects of the ecosystems (Cajariba and Silva, 2017). Among the various indicators, terrestrial invertebrates, and particularly insects, play a crucial role in most ecological processes, and they are key components of ecosystem structure and functioning (Bicknell et al., 2014; Viegas et al., 2014; Cajariba and Silva, 2015; Cajariba et al., 2015; Campos and

Hernández, 2015). Insect abundance and richness are related with other taxa, climate, and soil characteristics, thus representing potential target indicators of environmental changes (e.g. Nichols et al., 2008; Cajaiba et al., 2015). Therefore, a better understanding of the ecological relevance of insects in the humid tropics could support decision-making and robust management/recovery of imperilled ecosystems in the scope of the need for rapid, standardised, and cost-effective assessment methodologies (Godfray et al., 1999).

Beetles have characteristics that make them appropriate for ecological studies (Vasquez-Velez et al., 2010; Paoletti et al., 2010; Cajaiba and Silva, 2015) monitoring different compartments of the system (Marinoni, 2001). Among beetles, those of the family Histeridae (Histerid beetles - HB) are known as generalist predators, with wide range of habitats, which may occur in faeces, fungi, tree trunks, decomposing fruit, roots of trees, bird nests, burrows of mammals or reptiles, and in decaying vegetation. In addition, they are important predators of eggs and larvae, particularly of Diptera (Cyclorrhapha). Some groups are related to other animals, particularly social insects such ants and termites (Leivas et al., 2013). Despite its ecological importance, few studies have been conducted in Brazil using this group, and the published ones focused on the systematics (Degallier et al., 2011; Corrêa et al., 2012; Leivas et al., 2012a, b; Leivas et al., 2015).

In the present study, we aimed to characterize the seasonal communities of HB of different ecosystems in the Brazilian Amazon, ranging from primary or secondary forests with different stages of regeneration, to cocoa farms and pastures. These ecosystems, characterized by dissimilar structure and complexity, could be associated with divergent seasonal patterns in richness and diversity. Specifically, we addressed the following questions: 1) does the composition of Histerid beetle communities vary among the different ecosystems and 2) does the diversity of Histeridae assemblies in the Amazon biome differ among seasons?

Material and methods

Study area

This study was performed near the city of Uruará, state of Pará, northern Brazil (*Fig. 1*). The territorial extension of the municipality is 10796 km² and its population encompasses circa 44789 inhabitants. The dominant land use/ land cover (LU/LC) is natural forest (69% of the area) and deforestation is concentrated mainly in the south-central part of territory and near the main roads. Extensive livestock production and the exploitation of timber at a large scale (mostly illegal) are currently considered the most serious environmental threats (Cajaiba and Silva, 2017). The studied areas contain the pertinent gradients, in terms of biophysical and ecological characteristics, for testing the response of Histerid beetle communities (Cajaiba and Silva, 2017) facing the main anthropogenic drivers. These gradients encompass: Native Vegetation - NV, Early Secondary succession – ES (vegetation with five years of regeneration), Mature Secondary succession - MS (vegetation with 15 years of regeneration), Agriculture - Ag (cocoa plantations, *Theobroma cacao* L.) and Pasture for extensive livestock - Pa. The climate of the study area is classified as Aw (Köppen), hot and humid and the average annual rainfall is 2000 mm (Cajaiba et al., 2017b).

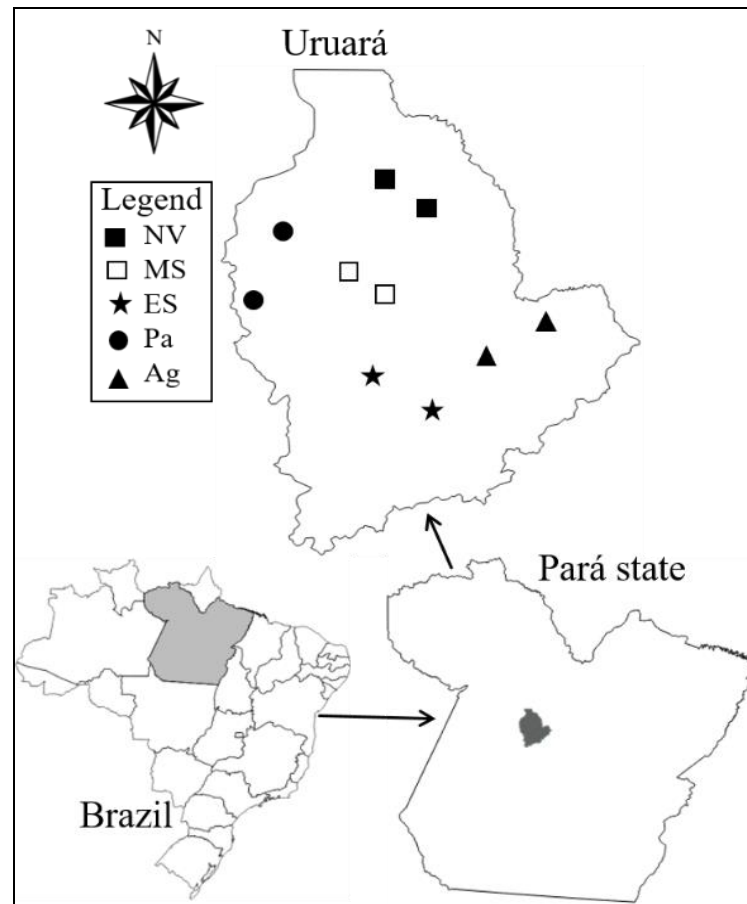


Figure 1. Location of the study region in the municipality of Uruará, state of Pará, northern Brazil. Location of the sampling areas: NV, Native vegetation; MS, Mature secondary succession; ES, Early secondary succession; Ag, Agriculture; Pa, Pasture.

Histerid beetles sampling

Sampling was carried out during the year 2015, in the months of February/March (rainy season), June (final of rainy season and early dry season) and September/October (dry season). This allowed integrating eventual seasonal differences in the diversity of Histerid beetles (HB). The sample points were placed at a minimum distance of 100 meters from ecotones, to ensure that most HB captured were associated to the ecosystem in study. Pitfall traps with 75 mm diameter and 110 mm deep were filled with preservative liquid consisting of formalin, alcohol, water and a few drops of detergent to break the surface tension. A roof was attached to each trap to prevent rainwater from entering the trap, remaining installed for 48 h prior to collection. Each pitfall contained different baits such as human faeces, meat and banana in order to

attract different species according to their feeding habits (non-baited pitfalls were used as control).

In each studied ecosystem (NV, MS, ES, Ag and Pa) seven sample points were placed 100 m apart. Each sample point contained four pitfall traps including the different baits (faeces, carrion, banana and non-baited), separated by 5 meters. The distance between pitfall traps allowed individuals to choose their preferential food resource (Almeida and Louzada, 2009). This protocol was applied to all areas and periods of collections, totalizing a sampling effort of 840 traps. In order to complement the pitfall derived information (Cajaiba et al., 2014), specific methods were used to collect leaf litter invertebrates. Ten random sampling points of 1 m² were selected in each ecosystem and sampling period. At each collection point ('litter-only'), only the loose soil was gently scraped with a metal trowel, to include those HB into the samples that fell out of the leaf litter during this collection procedure. We refrained from simply separating leaf litter and soil, considering that when we remove the litter, HB might flee the litter and hide in the loose topsoil.

Assemblage analysis

Richness and abundance of HB were measured in each sampling site and differences among ecosystems were gauged using One-Way-Analysis-of-Variance (ANOVA). ANOVA was also applied to find possible differences in HB along the year. When the ANOVA indicated differences in the average among ecosystems and/or seasonality, a Tukey's post-hoc test was performed to find specific difference(s). The normality of the data was verified by the Shapiro-Wilk test.

The taxonomic composition of HB communities between ecosystems was compared using Permutational Multivariate Analysis of Variance (PERMANOVA). Non-metric Multidimensional Scaling (NMDS) plots were used to help interpret the results found with the PERMANOVAs (see Anderson, 2001 for similar procedure). In order to check for environmental variables with influence on the HB communities, associated with periods of the year, a correlation analysis (Pearson correlation) was applied between HB abundance and richness and meteorological data - temperature,

humidity and precipitation. All analyses were performed using PAST software version 3.14 (Hammer et al., 2001).

Results

Composition of Histeridae

A total of 1945 individuals Histerid beetles (HB) were captured, distributed by five genera and 14 species/ morphospecies (species). All species (14 species, 682 individuals) were monitored in the native vegetation (NV), 12 species (513 individuals) in the Mature Secondary succession (MS), 13 species (180 individuals) in Early Secondary succession (ES), 11 species (232 individuals) in Agriculture (Ag) and 13 species (338 individuals) in Pasture (Pa). The most abundant species were *Phelister* sp1 (302 individuals), *Phelister* sp4 (257 individuals), *Hister* sp1 (246 individuals) and *Phelister* sp2 (184 individuals). These four species represent more than 50 percent of all specimens collected. The genera *Phelister* and *Hister* were prevalent in all communities, with more 71 percent of all specimens captured (*Table 1*).

Table 1. Total numbers of Histerid beetles species in the environments different in the Brazilian Amazon. NV = Native Vegetation; MS = Mature Secondary succession (15 years of regeneration); ES = Early Secondary succession (5 years of regeneration); Ag = Agriculture (Cocoa); Pa = Pasture. R = Rainy Season; Intermediary Season; D = Dry Season.

Species	NV			MS			ES			Ag			Pa			Total
	R	I	D	R	I	D	R	I	D	R	I	D	R	I	D	
<i>Phelister haemorrhous</i>	14	4	6	-	-	-	-	-	-	-	-	-	6	4	3	37
<i>Phelister</i> sp1	60	68	-	16	80	11	17	6	3	-	6	8	7	20	-	302
<i>Phelister</i> sp2	26	25	-	26	20	-	9	-	2	3	2	34	7	15	15	184
<i>Phelister</i> sp3	-	25	-	10	72	22	1	2	1	-	6	10	10	5	6	170
<i>Phelister</i> sp4	28	20	6	35	30	-	2	10	2	-	-	26	19	20	59	257
<i>Hister punctifer</i>	24	8	7	14	10	3	6	6	7	6	6	5	3	2	1	108
<i>Hister</i> sp1	44	30	-	33	38	18	18	6	-	9	14	30	-	2	3	246
<i>Hister</i> sp2	6	40	-	2	2	1	-	-	-	8	1	8	2	-	18	88
<i>Omalodes marseuli</i>	11	8	7	13	2	6	8	2	1	7	3	3	10	4	4	89
<i>Omalodes</i> sp1	72	14	6	2	-	-	4	8	4	4	4	10	6	2	12	148
<i>Omalodes</i> sp2	30	24	16	10	10	0	1	-	8	-	-	-	3	-	24	126
<i>Euspilotus</i> sp1	16	8	8	5	2	1	6	6	5	6	6	2	5	4	-	80
<i>Euspilotus</i> sp2	6	6	4	10	5	4	12	11	7	6	3	1	14	7	7	103
<i>Operclipygus</i> sp	5	-	-	-	-	-	2	-	-	-	-	-	-	-	-	7

Histerid beetles (HB) richness and abundance presented significant differences between ecosystems ($F_{4,114} = 111.3$, $p < 0.001$; $F_{4,114} = 346$, $p < 0.001$) (Figs. 2a and 2b). Generally, less disturbed ecosystems had higher values of both indexes, although with exceptions: higher richness and abundance depicted in Pastures when comparing with Cocoa farms (Ag) and Early secondary succession (ES).

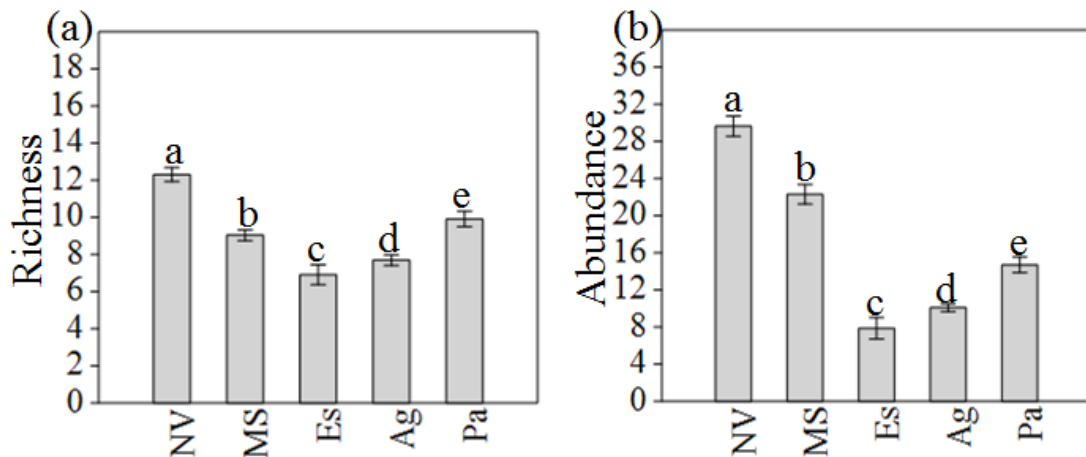


Figure 2. Differences in the projected values for (a) Richness (\pm SE), and (b) Abundance (\pm SE) of Histerid beetles (HB) community in the different ecosystems considered. The values followed by the same letters are not significantly different according to Tukey test. NV, Vegetation native; MS, Mature Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture; Pa, Pasture.

NV presented all species while *Phelister haemorrhous* was not captured in MS, ES and Ag; *Operclipygus* sp was not captured in MS, Ag and Pa; *Hister* sp2 and *Omalodes* sp2, were not found in ES and Ag, respectively.

The results of the NMDS showed that the HB assemblages of different ecosystems could be separated from each other by ordination of the species composition, suggesting that these assemblages change gradually from the most pristine to more disturbed ecosystems. The composition of the HB of NV and MS were more similar to each other than HB assemblages of other habitats: ES, Ag and Pa could be discriminated from NV and MS (Fig. 3). In fact, the Permutational Multivariate Analysis of Variance (PERMANOVA) showed that HB taxonomic composition of the ecosystems studied was significantly different ($F_{4,114} = 28.44$, $p < 0.0001$).

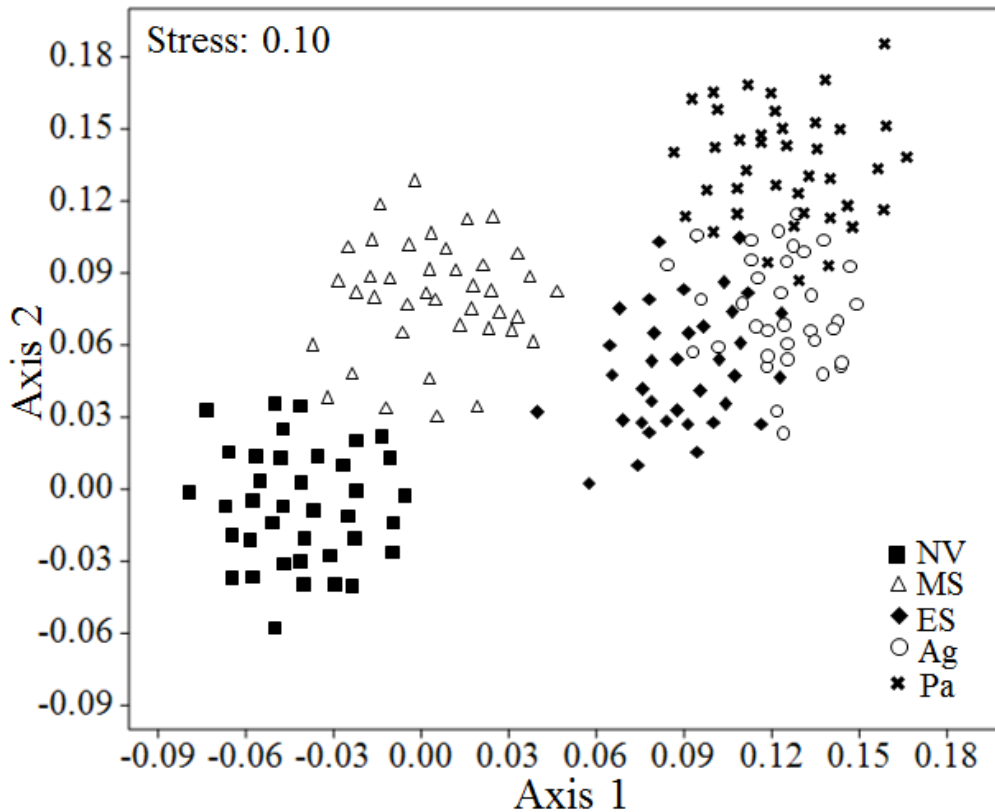


Figure 3. Non-metric multidimensional scaling (NMDS) showing *Histerid* beetles grouped in accordance with the ecosystems (using Bray-Curtis similarity). NV, Vegetation native; MS, Mature Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture (Cocoa); Pa, Pasture.

Seasonality patterns in Histerid beetles assemblages

With respect to the relation between the period of the year and HB assemblages, statistical differences in abundance and richness were identified ($F_{2,342} = 11.71$, $p < 0.001$; $F_{2,342} = 13.29$, $p < 0.001$): significant differences in abundance were found between the averages of the rainy and dry seasons (Tuckey test: $Q = 6.76$, $p < 0.001$), and the intermediary and dry seasons (Tuckey test: $Q = 5.64$, $p < 0.05$); significant differences in richness were found between averages of rainy and intermediary seasons (Tuckey test: $Q = 3.17$, $p < 0.01$), and intermediary and dry seasons (Tuckey test: $Q = 4.09$, $p < 0.05$) (Fig. 4a).

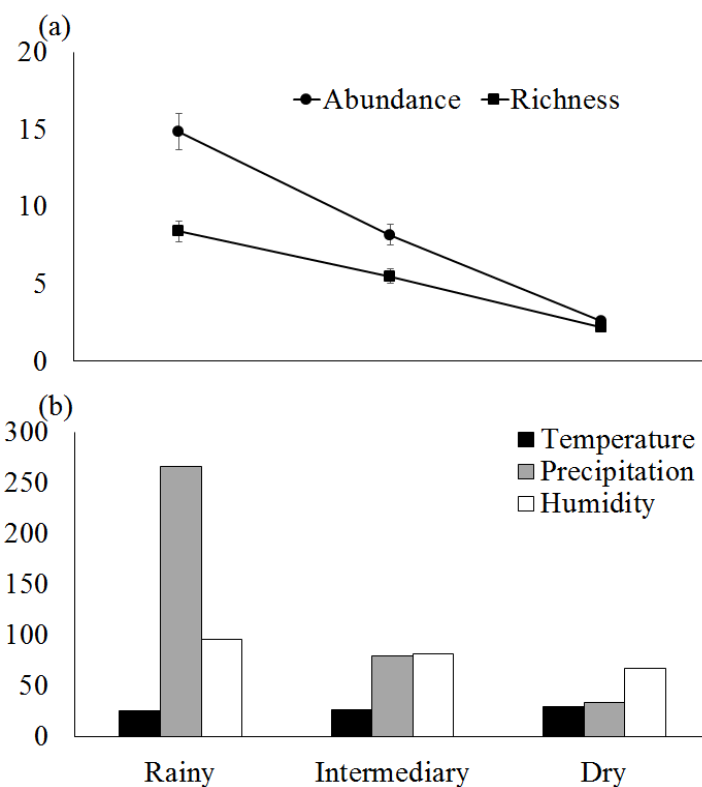


Figure 4. Abundance and richness of Histerid beetles (a) and distribution of environmental variables during the study period (b).

Among the climatic variables evaluated (Figure 4b), humidity and precipitation were positively correlated with the abundance and richness of Histeridae. In contrast, the air temperature negatively influenced these indexes (Table 2).

Table 2. Pearson correlation between environmental variables and ecological indices (abundance and richness) of Histeridae collected in Uruará, Pará, northing Brazil. Asterisks indicate statistical significance at level of * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$.

Environmental variables	Abundance	Richness
Humidity	0.57***	0.49***
Temperature	-0.29*	-0.29**
Precipitation	0.48***	0.44***

Discussion

Composition

Degallier et al. (2011) highlighted the shortage of information on Histerid beetles (HB) in Neotropical areas, where environmental and ecological conditions

should enhance their biodiversity. In fact, there is a lack of specific collections and specialized taxonomists for this group. Despite the lack of HB collections, the results obtained provide clues on the diversity of the group in the Amazon region (Leivas et al., 2013).

The genus *Phelister* (Marseul, 1853), which was the most abundant in the present study, represents a varied genus with 88 species described with mostly Neotropical distribution (Mazur, 2011). These species can be found on diverse substrates like faeces, carcasses, decomposing plants, plants, mammal burrows, bird nests, and in debris piles of leaf-cutting ants. Some species, associated with carcasses, have been considered relevant for forensic entomological studies (Almeida and Mise, 2009; Leivas et al., 2013). The enormous diversity, lack of taxonomic studies, and the possibility of description of many new taxa hinders the identification of species (Leivas et al., 2013). The genus *Hister* (Linnaeus, 1758), which was the second most abundant in the present study, is the most diverse genus of HB, with approximately 195 described species distributed across all zoogeographic regions (Mazur, 2011). These species occur in faeces, carcasses, decomposing plants and fungi, mammal burrows, and in debris piles of leaf-cutting ants. The small number of species recorded in Brazil suggests that new taxa and new records might be described in future (Leivas et al., 2013, 2015).

In the present study, both richness and abundance were the highest in the most pristine and/or conserved ecosystems (e.g. NV and MS). These results suggest the presence of differences among HB communities associated with specific ecosystems, probably related with the environmental conditions and bioecology of species (Moraes et al., 2013). Summerlin (1989) mentions a trend starting from low dominance and high richness of HB communities in pristine ecosystems to higher abundances and reduced richness of HB communities in open and degraded ones. Thus, diversity and abundance of HB communities are intimately related with differences in environmental characteristics of these ecosystems (Lopes et al., 2005).

Biotic and abiotic factors associated with most pristine/ heterogeneous environments, are, for example, mild climates, high canopy cover, and complex ground cover that provide environments for hibernation and more diverse prey, and

better conditions for larvae and adults. Cattle treading on pasture areas or the use of machinery in agriculture areas contribute to soil compaction, resulting in changes in the soil structure, reduction of soil litter and water, thereby decreasing and/or eliminating many species (Kruess and Tschardtke, 2002).

Seasonality

In the present study, the abundance and richness of HB collected at different times of the year (rainy season, intermediary season, and dry season) depicted a trend with higher diversity during the rainy season, corroborating the hypothesis that the abundance, richness, and diversity are associated with climatic conditions (Andresen, 2005). Nonetheless, microclimatic factors are not necessarily correlated with climatic conditions, but they play an important part in the activity of HB and associated taxa in different climates (Moraes et al., 2013).

The positive correlation between air moisture and the diversity of HB is expected, because higher air moisture may produce favourable microclimates for HB. Previous studies demonstrated that vegetation structure and its effects on microclimate (e.g. temperature and air moisture) might be one of the most important factors controlling and structuring the distribution of beetles (Magura et al., 2000). Apparently, the evolutionary life cycle strategy of beetles is optimized and synchronized with seasonal changes of microclimatic environmental conditions (Kotze et al., 2011; Wang et al., 2014).

Although temperature was negatively correlated with abundance and richness in the present study, annual temperature variation in this region is small, suggesting that rain and humidity are probably the main drivers affecting the dynamics of invertebrates (Andresen, 2008). Silva et al. (2010) presented two hypotheses to explain the lower abundance and richness of adult beetles in the dry season: (1) adults are sensitive to the effects of drought and remain underground during this period, or (2) adults die in the dry season and only the immature beetles survive in the nest, reaching the adult stage at the beginning of the wet season. Nevertheless, temperature is a major factor affecting activity, flight, foraging behaviour, and metabolism of beetles (Saska et al., 2010).

Additional studies are necessary to document the extent of sensitivity to microclimate and climate, and they could be particularly relevant in the light of the potential effects of climate change (Williams et al., 2007; Maveety et al., 2014). Understanding seasonal patterns of beetles in a given region is important for several reasons. Firstly, seasonal variations in abundance, richness, and species composition emphasize the role of phenology and the effect of survey timing on the results obtained for studying the association of HB with ecosystems. Further, seasonal information of HB might be a relevant ecological indicator, which might have importance in the management of ecosystems.

Conclusion

The Histerid beetles collected in this study demonstrate patterns of habitat preference and marked seasonal variations. In addition, the results indicate factors and conditions that might affect overall biodiversity distribution in the Brazilian Amazon. In fact, the present study should be complemented with studies linking Histerid beetles with other taxa for understanding and assessing the state of conservation of the diverse ecosystems of the studied region.

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Chapter 7

Are disturbance gradients in neotropical ecosystems detected using rove beetles? A case study in the Brazilian Amazon

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Abstract

In the Neotropics, Rove Beetles (Staphylinidae) are known to be especially diverse and abundant, but studies in this region are still incipient and the information is dispersed in the scientific literature. In this work the responses of the Rove Beetles to gradients of disturbance in representative ecosystems of the Brazilian Amazon were evaluated. Specifically, we assessed the possibility of using patterns in richness, abundance and taxonomic diversity of Rove Beetles as anthropogenic disturbance indicators. The obtained results show that the Rove Beetles richness was sensitive to the structural changes induced by increasing anthropogenic disturbance. Additionally an increasing trend in community complexity from more to less disturbed ecosystems was observed. Differences in responses can be explained by differences in the intensity and extent of the change from pristine systems. Overall, the composition of Rove Beetles communities is ecosystems' specific and sensitive to anthropogenic induced structural changes and management actions and, therefore, should be considered a valuable ecological indicator for assessing the extent ecosystems' disruption in the Neotropics.

Keywords: Biodiversity; disturbance indicators; landscape heterogeneity; Neotropical forests; insect; Brazilian Amazon.

Introduction

Neotropical forests are biologically diverse ecosystems, representing some of the richest areas of the planet (Lindenmayer et al., 2002). Species from these forests are threatened by deforestation, fragmentation, conversion to forest monocultures, climate change and other stressors like fire and even fire suppression (Carnus et al., 2006; Loskotová and Horák, 2016). There is an enormous concern and speculation about the effects of human disturbances in the biodiversity of neotropical ecosystems, namely the impacts on species composition and the modifications in the ecological services provided (e.g. Morris, 2010; Aerts and Honnay, 2011; Cajariba et al., 2017). Deforestation in the Brazilian Amazon (Amazonia), has been causing a sharp erosion of biodiversity and the disruption of complex global climate cycles (e.g. Morris, 2010; Cajariba et al., 2017). The most significant landscape changes identified for the Amazonia are forest logging, the establishment of extensive livestock operations and intensive farming, but also the expansion of road nets and urban areas (Mertens et al., 2002; Tabarelli et al., 2004), power generation, and mining (Soares-Filho et al., 2005). These anthropogenic activities are considered strong environmental stresses whose impacts were not fully anticipated (Cajariba et al., 2017). In fact, these landscape changes promote ecosystems' substitution and might isolate taxa and even cause extinctions due to interactions in which species are engaged (Aizen et al., 2012; Valiente-Banuet et al., 2015). Understanding the impacts of ecosystem transitions in the Amazonia will ultimately support the best conservation strategies for the region, considered fundamental for sustaining earth's functioning and ultimately humankind (Cajariba et al., 2017).

In a conservation perspective, the condition of an ecosystem might be evaluated using the “difference” from reference situations, i.e. the disparity of a specific ecosystem from pristine ecosystems (Costanza, 2012). Considering the unfeasibility of measuring all aspects in a specific ecosystem, several scientists advocate the use of surrogates, usually termed ecological indicators (Costanza, 2012; Heath, 2013). Species, populations and communities might prove useful as indicators if sensitive to ecosystem changes, anticipative and easily monitored (Gardner et al., 2008; Rapport and Hildén, 2013). Besides the universal need for developing ways to assess status and trends in environmental condition of ecosystems (Niemi and McDonald, 2004), selecting organisms as indicators of anthropogenic disturbance to help conservation decisions is still a challenge in most biodiverse countries, where taxonomic and natural history knowledge is deficient (Kim and Byrne, 2006). This task is considered particularly urgent in megadiverse neotropical countries, since their natural systems are being eroded at unprecedented speeds and scales (Uehara-Prado et al., 2009). Even though a steady stream of scientists and a large number of publications and projects have been investigating the ecosystem changes versus disturbance, they provide a very fragmented picture concerning the environmental changes and its ecological consequences. Additionally and considering its vastness, most of the Amazonia’ ecosystems were still marginally studied (Cajaiba et al., 2017).

Terrestrial invertebrates and especially insects play a crucial role in most ecological processes and are key components of ecosystems’ structure and functioning (Bicknell et al., 2014; Cajaiba and Silva, 2015; Cajaiba et al., 2015; Campos and Hernández, 2015). In this context, insects’ abundance, composition and richness are related with other taxa, climate and soil characteristics, thus representing potential

target indicators of environmental changes (e.g. Nichols et al., 2008; Cajaiba et al., 2017). Understanding the ecological relevance of insects' communities in the humid neotropics could even support decision-making and robust management/recovery of imperiled ecosystems in the scope of the need for rapid, standardized and cost-saving assessment methodologies (Cajaiba et al., 2017). Among insects, Rove beetles (RB) of the family Staphylinidae (Insecta: Coleoptera) have been proposed as disturbance indicators (Anderson and Ashe, 2000; Pohl et al., 2007) given their abundance and diversity (Vásquez-Vélez et al., 2010) and variety of ecosystems used. Additionally they are easy to collect because the majority of species are found among leaf litter and fallen trunks. RB are sensitive and respond rapidly to abiotic, biotic and anthropogenic disturbances (Magura et al., 2013) and are considered ecologically significant, particularly of the soil-related fauna (Vásquez-Vélez et al., 2010). Moreover most are predators, sensitive to prey densities - although, parasitoids, saprophytes, omnivores, and opportunists can be found (Caballero et al., 2009).

In the Neotropics, RB are considered to be diverse and abundant, although supported mostly by incipient works (Gutiérrez-Chacon et al., 2009), dispersed within scientific and grey literature (Newton et al., 2005). The purpose of this study was to evaluate the responses of RB communities to increasing levels of stress in the Amazonia, in the scope of its usefulness as ecological indicators of anthropogenic disturbance. For that several ecosystems with increasing levels of anthropogenic stress were monitored, such as: primary forest, our reference condition, secondary forests within different stages of recovery, farmland and pasture for extensive livestock. We were particularly interested in assessing the RB communities' sensitivity to anthropogenic induced structural changes, i.e. how does richness, abundance and

taxonomic composition change? Is the response of these indexes correlated with increasing trends in disturbance? In addressing these questions we attempt to build a framework of reference, gauging its effectiveness as ecological indicators of disturbance. New insights in RB diversity responses to ecosystem degradation and/ or recovery could also provide standards guiding the most efficient conservation management actions in the Neotropics (Kotze et al., 2003).

Material and methods

Study sites

The study area was located in the municipality of Uruará, state of Pará, northern Brazil (Fig. 1). Primary forest (69% of the area) is the dominant land use/ land cover while deforestation is concentrated mainly in the south-central part of territory and near the main roads. Extensive livestock production and the exploitation of timber (mostly illegal) are currently considered the most negative environmental pressures (Cajaiba et al., 2015). The studied areas contain the most representative ecosystems of the region, in terms of biophysical and ecological characteristics, for understanding the response of RB communities, such as Native Vegetation (NV), Early Secondary succession (ES - secondary vegetation with five years of regeneration), Maturing Secondary succession (MS - secondary vegetation with 15 years of regeneration), Agriculture (Ag - cocoa plantations, *Theobroma cacao* L.) and Pasture for extensive livestock (Pa) (Fig. 1, For generic characteristics of the ecosystems sampled, see Table S1). The climate of the study area is classified as Aw (Köppen), hot and humid with an average annual rainfall of 2000 mm.

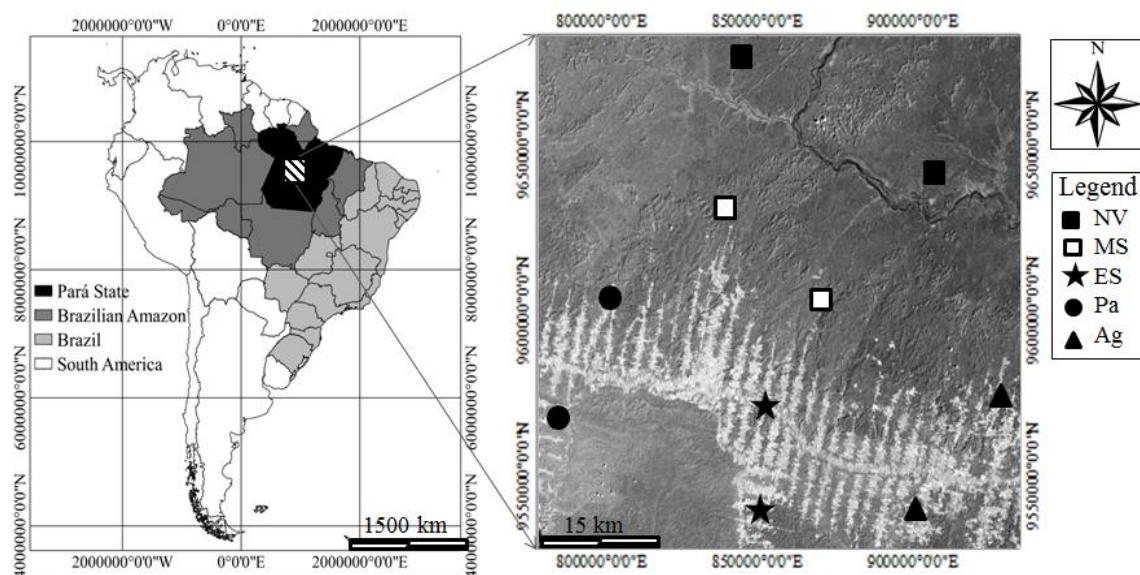


Fig. 1. Location of the study region in the municipality of Uruará, state of Pará, northern Brazil. Location of the ecosystems sampling: NV, Native vegetation; MS, Maturing secondary succession; ES, Early secondary succession; Pa, Pasture; Ag, Agriculture.

Staphylinidae sampling

Sampling was carried out during the year 2015, in the months of February/March (rainy season), June (final of rainy season and early dry season) and September/October (dry season), allowing the integration of annual seasonal differences in the activity of RB. The sample points were placed at a minimum distance of 100 meters from ecotones, to ensure that most beetles captured were associated to the ecosystem in study (Cajaiba et al., 2017). Pitfall traps with 75 mm diameter and 110 mm deep were filled with preservative liquid consisting of formalin, alcohol, water and a few drops of detergent to break the surface tension. A roof was attached to each trap to prevent rainwater from entering the trap, remaining installed for 48 h prior to collection. Sampling survey period and intensity, although apparently inadequate for definitive inventory, served the purpose of comparing RB general sensitivity to

ongoing changes in the scope of their application as ecological indicators of disturbance (Dale and Beyler, 2001; Santos and Cabral, 2004).

Each pitfall was associated to a different treatment in order to attract RB in accordance with to their feeding ecology: non-baited, human faeces, meat and banana. In each studied site four sample points were placed 100 m apart. Each sample point contained the four pitfall treatments, separated by 5 meters. The distance between pitfall traps by location was determined in order to select individuals according to their favoured food resources, as suggested by related studies (Almeida and Louzada, 2009; Silva et al., 2012; Campos and Hernández 2015; Qodri et al., 2016). This protocol was applied to all ecosystems and periods of collections, totalizing a sampling effort of 480 traps. The identification was based on the taxonomical keys proposed by Navarrete-Heredia et al. (2002) and by comparison of material deposited in the DZUP (Entomological Collection Pe Jesus Santiago Moure of the Department of Zoology of the Federal University of Paraná).

Environmental variables

Fourteen microclimate and ecosystem variables were measured by location: Temperature, Humidity, Precipitation, Circumference at Breast Height, Circumference at ankle height, Canopy cover, Number of plant species, Number of plants, Number of species of shrubs, Number of shrubs, Percentage of exposed soil, Percentage of green (vegetation) cover, Percentages of leaf litter cover, Height of leaf litter. The Temperature, Humidity and Precipitation of each point were measured during the traps installation and removal with a portable weather station (model Oregon Scientific WMR200A). To assess the environmental complexity of each sampling site, the

quadrat-section method was adopted (Campos and Hernández, 2015). Using a cross as a reference, four quadrants (northeast, northwest, southeast, southwest) were marked and, in each quadrant the following variables were measured: the distances to the centre of the cross, height, crown diameter and trunk diameter of all trees with Circumference at breast height greater than 15 cm and all shrubs with Circumference at ankle height less than 15 cm and height greater than 1 m. Trunk diameter was taken at breast height (1.3 m) for the trees and ankle height (Circumference at ankle height = 0.1 m) for the shrubs. In each quadrant, the Height of leaf litter in 1 m × 1 m marked square (using PVC pipe) was measured with a ruler, and the percentages of Percentages of leaf litter cover, Percentage of green (vegetation) cover and Percentage of exposed soil were measured by visual estimation using the following classes, 0–5%, 6–25%, 26–50%, 51–75%, 76–95% and 96–100% (Campos and Hernández, 2015). Using these same classes, the percentage of Canopy cover in the four quadrats was calculated with a convex spherical densiometer (D) Lemmon (Lemmon 1954). Information concerning the methodology associated with each variable monitoring is depicted in Table S2, Supplementary Material.

Assemblage analysis

Rarefaction curves were used to assess whether the sampling effort was enough to monitor all the species by ecosystem, and the average efficiency of sampling was calculated based on Chao 1. Although rarefaction interpolates data back, non-parametric species estimators such as Chao 1 (Chao et al., 2009) extrapolate from the data to find what the 'true' number of species may have been by using the number of rare species - singletons and doubletons - as a way of calculating how likely it is there

are more undiscovered species. Species richness, Abundance, Shannon-Weiner diversity index and dominance Berger Parker index of each ecosystem RB were measured, and differences among ecosystem were gauged using One-Way-Analysis-of-Variance (ANOVA) and subsequent Tukey post hoc tests to check for specific differences. Before applying ANOVA, the normality of the data was verified by the Shapiro-Wilk test. The taxonomic composition of RB communities among ecosystems was compared using Permutational Multivariate Analysis of Variance (PERMANOVA). Non-metric Multidimensional Scaling (NMDS) plots were used to help in interpreting the results found with the PERMANOVAs (see Anderson, 2001 for similar procedure).

A beta diversity partition analysis was applied to verify the dissimilarity between ecosystems. Using the partitioning framework proposed by Baselga (2010), the pairwise dissimilarity index ($\beta_{s\sigma r}$) was partitioned into two components: turnover (β_{sim}) and nestedness (β_{nes}): $\beta_{s\sigma r} = \beta_{sim} + \beta_{nes}$. This method was applied in order to evaluate whether dissimilarities in the composition of the RB communities occurred through the substitution of some species by others (β_{sim}) or by the formation of nested subsets of more diverse communities (β_{nes}). All analyses were performed using the functions `beta.pair` from `betapart` package (Baselga et al., 2017) within R 3.2.4 program (R Core Team 2016).

Association of Rove Beetle assemblages with environmental variables

Canonical Correspondence Analysis (CCA) was used to estimate the influence of the fourteen environmental variables on RB communities. Principal Components Analysis (PCA) was used to reduce variables to 4 principal components (PC). The

CCA was performed with the Bray-Curtis dissimilarity index on two axes. Species singletons and doubletons were omitted from ordination analyses but considered for calculations of species richness. Deleting rare species is a useful way to reduce the statistical noise in the data set without losing much information (McCune and Grace, 2002).

Species indicator value - IndVal

To determine possible indicator species, single value indicator (IndVal) developed by Dufrêne and Legendre (1997) was calculated, combining specificity (patterns of relative abundance) of a given species in a given ecosystem with its fidelity within that ecosystem (patterns of incidence). Species with a high specificity and high fidelity within a habitat are considered to achieve the highest indicator value. This analysis was performed in R 3.2.4 program (R Core Team 2016) using the *indicspecies* package 1.7.5 (De Cáceres and Jansen, 2015) with 9999 permutations, using data for RB abundance.

Results

Assemblages analysis

A total of 1,493 individuals were collected, grouped into 11 subfamilies and 71 species and morphospecies (from now on morphospecies). From these, 43 morphospecies (488 individuals) were identified in the Native Vegetation (NV), 32 morphospecies (192 individuals) in the Maturing Secondary succession (MS), 36 morphospecies (300 individuals) in Early Secondary succession (ES), 26 morphospecies (406 individuals) in Agriculture (Ag) and 13 morphospecies (103 individuals) in Pasture (Pa). The most abundant morphospecies were: *Hoplandria* sp

(350 individuals), *Aleochara repetita* Sharp, 1887 (294 individuals) and *Aleochara* sp1 (200 individuals), together represent approximately 57% of total abundance. The subfamilies that contributed most to richness and abundance were Aleocharinae (S=15; N=966), Staphylininae (S=20; N=140) and Paederinae (S=13; N=169) (Morphospecies taxonomic and guilds classification is depicted Table S3, Supplementary Material).

The curve of accumulation of species presented an asymptote only for Pa, the other ecosystems would require larger sample sizes to determine all diversity (Fig. 2). The Chao 1 estimator demonstrated that the number of species would be of 76 species for NV, 47 for MS, 70 for ES, 45 species for Ag and 14 species for Pa.

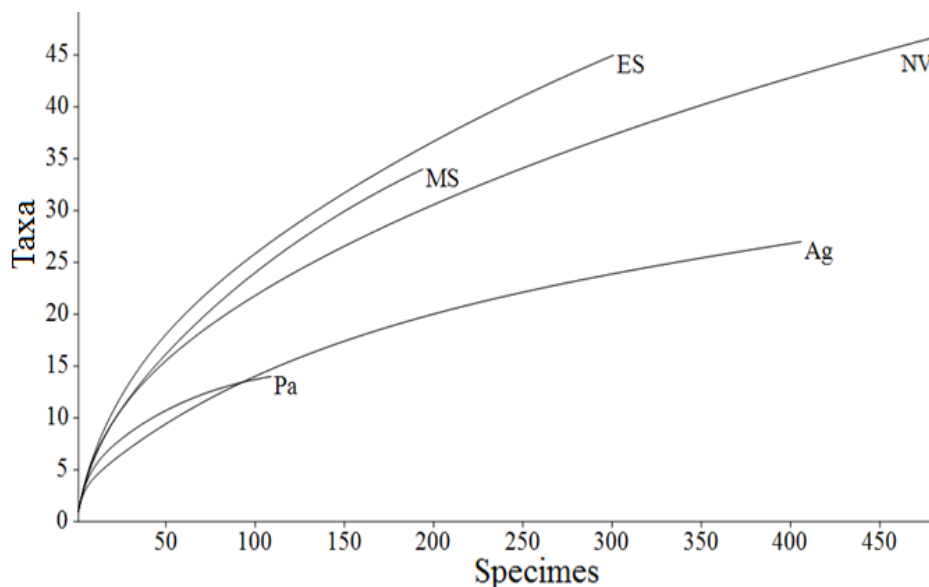


Fig. 2. Rove Beetles individual-based rarefaction curves for the studied sites. NV, Vegetation native; MS, Maturing Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with 5 years of regeneration); Ag, Agriculture; Pa, Pasture.

Rove Beetle (RB) richness was significantly different between ecosystems ($F_{4,205} = 31.25$, $P < 0.0001$): the average RB richness was significantly higher in NV

by comparison with the other ecosystems ($P < 0.01$). RB richness was also high in ES, although not differing from Ag ($P > 0.05$) but higher than the MS and Pa ($P < 0.05$) (Fig. 3a). RB abundance varied significantly between the ecosystems ($F_{4,205} = 65.72$, $P < 0.001$). Mean abundance was significantly higher in NV ($P < 0.01$), differing from all ecosystems but not from Ag ($P > 0.05$). Pa presented lower abundance than all the other ecosystems ($P < 0.01$) (Fig. 3b).

Shannon diversity was higher in NV ($H' = 2.69$), while Pa showed dominance of few morphospecies ($D = 0.88$). The Shannon diversity was significantly different among ecosystems ($F_{4,205} = 35.25$, $P < 0.0001$): significantly higher in NV ($P < 0.01$) and differing from the other ecosystem. On the other hand, MS, ES and Ag were similar to each other and different from NV and Pa ($P < 0.001$) (Fig. 3c). There was a significant difference in the Berger-Parker dominance index for the five ecosystems studied ($F_{4,205} = 17.03$, $P < 0.01$). Pa presented the highest index, being different from the other sites ($P < 0.01$) while NV was significantly lower than the others ($P < 0.001$) (Fig. 3d).

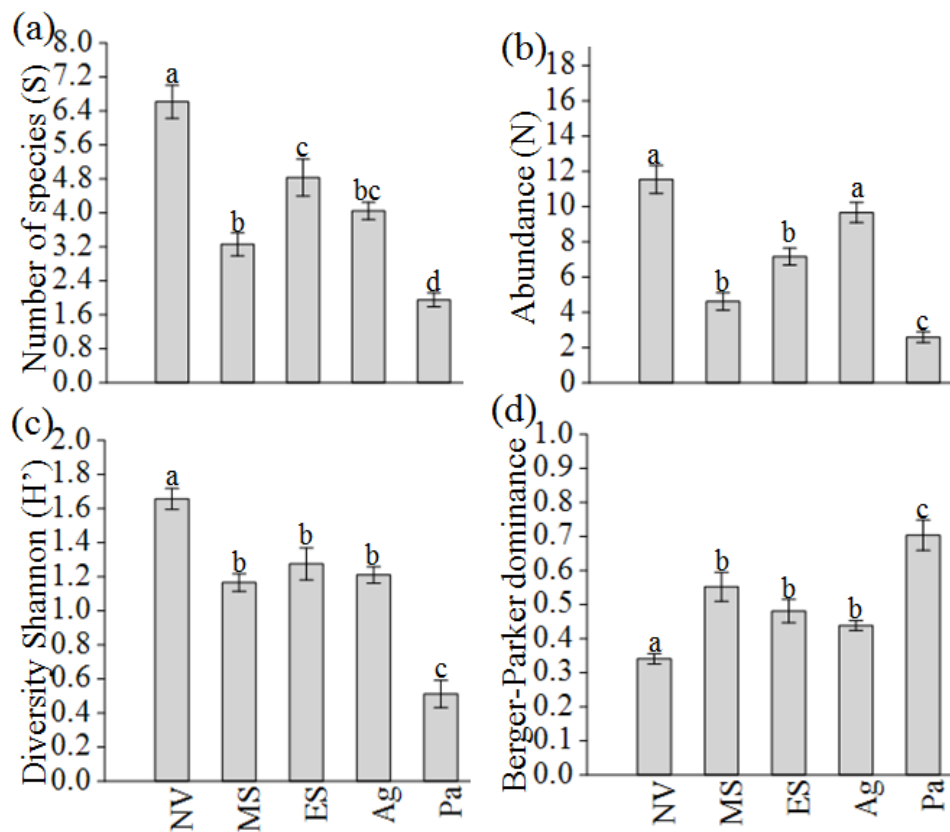


Fig. 3. Barplot expressing the differences in the projected values for (a) Number of morphospecies, (b) number of individuals, (c) Shannon diversity, and (d) Berger-Parker dominance of Staphylinidae community in the different ecosystems considered. Error bars indicate 95% confidence intervals. The values followed by the same letters are not significantly different according to Tukey test. NV, Vegetation native; MS, Maturing Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with 5 years of regeneration); Ag, Agriculture; Pa, Pasture.

Composition

Of the 71 morphospecies sampled, 12 morphospecies were collected exclusively on NV, followed by ES with 9 unique morphospecies and MS with 7 morphospecies. Only morphospecies *Hoplandria* sp and *Lithocharis* sp1 were sampled in all ecosystems (Table S3, Supplementary material). The results of the NMDS showed that the RB assemblages of different ecosystems could be separated from each

other by ordination of the species composition. The composition of the RB samples of NV and MS were more similar to each other than RB assemblages of Ag, Pa and ES. Ag and Pa also form two group isolated from NV and MS. The NMDS results depict an arch in the diagram, suggesting that the RB assemblages change gradually from the most pristine to more disturbed ecosystems (Fig. 4). The Permutational Multivariate Analysis of Variance (PERMANOVA) presented significant differences in the RB taxonomic composition between ecosystems ($F_{4,205} = 45.71, P < 0.001$).

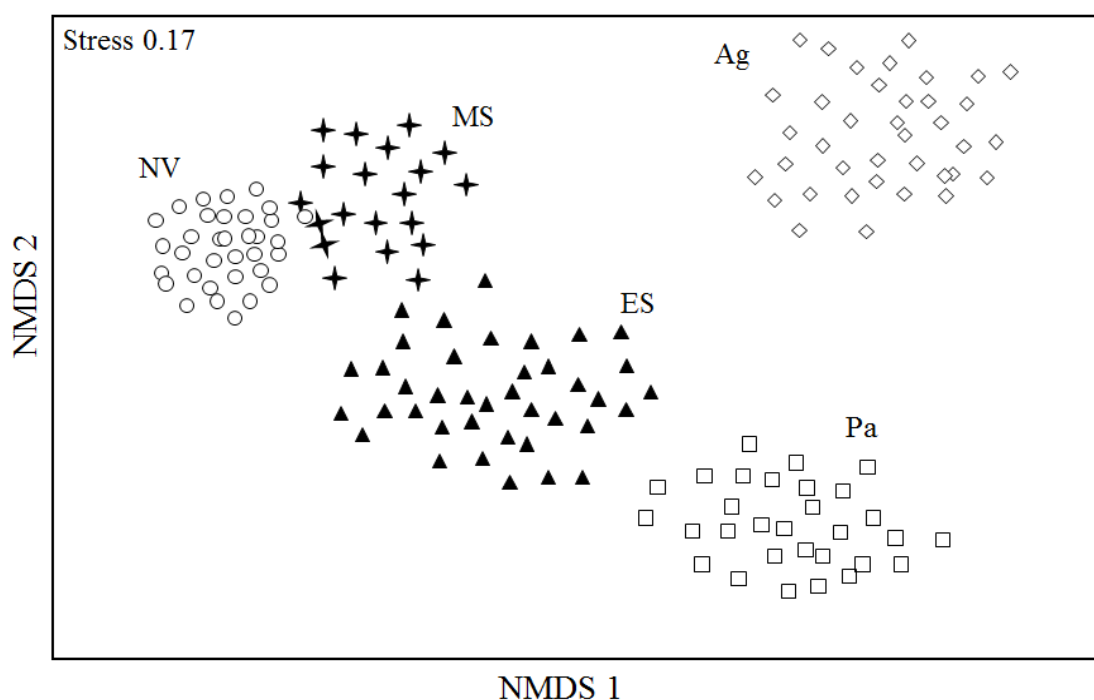


Fig. 4. Non-metric multidimensional scaling (NMDS) showing RB grouped in accordance with the ecosystems (using Bray-Curtis similarity). NV, Native vegetation; MS, Maturing Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture (Cocoa); Pa, Pasture.

For all the pairwise comparisons the turnover was the predominant component. The highest turnover values (β_{sim}) were found when comparing Pa with MS (β_{sim} : 0.80), followed by Ag with MS (β_{sim} : 0.68) and Pa with Ag (β_{sim} : 0.67). The lowest

β_{sim} was found comparing NV with MS ($\beta_{sim} = 0.38$) (Fig. 5 and Table S4, Supplementary material). Pa ecosystem presented the higher values of turnover. An increase in the turnover gradient was detected from NV to MS, then to ES and Ag and finally to Pa, although not clear when we comparing NV with ES and Ag (β_{sim} : 0.48 and 0.49, respectively) (Fig. 5 and Table S4, Supplementary material). If we consider all the studied ecosystems Pa seems to be most divergent in term of species composition. The highest nestedness values (β_{nes}) were found when comparing NV with MS (β_{nes} : 0.25), NV with Pa (β_{nes} : 0.20), NV with ES (β_{nes} : 0.16) and ES with Ag (β_{nes} : 0.16). The lowest nestedness was found when comparing MS with Ag ($\beta_{nes} = 0.07$) and MS with Pa (β_{nes} : 0.07) (Fig. 5 and Table S4, Supplementary material). Beta diversity (β_{sor}) presented higher values (up to 0.6) for the five ecosystems (Fig. 5). The highest dissimilarity values (β_{sor}) were found when comparing Pa with MS, NV, ES and Ag (β_{sor} : 0.87; 0.83; 0.83; 0.79, respectively), followed by Ag with MS (β_{sor} : 0.76) and ES with MS (β_{sor} : 0.71). Intermediate values were obtained for comparisons between NV and MS (β_{sor} : 0.64); NV and ES (β_{sor} : 0.64); NV and Ag (β_{sor} : 0.63) (Fig. 5 and Table S4, Supplementary material).

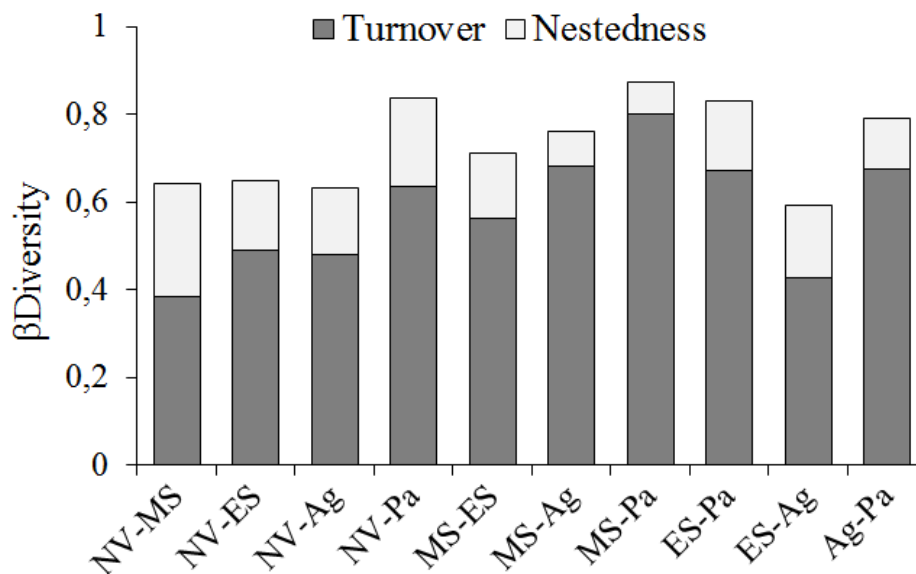


Fig. 5. Comparison of dissimilarity values for β_{sor} (overall dissimilarity), β_{sne} (dissimilarity resulting from nestedness), and β_{sim} (turnover) for the different ecosystems. NV, Native vegetation; MS, Maturing Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture (Cocoa); Pa, Pasture.

Effects of environmental variables on Rove Beetle assemblages

PC1 was positively correlated with the following environmental variables: Circumference at breast height; Number of plant species and Canopy cover, PC2 was positively correlated with Precipitation (P) and Humidity (H), PC3 positively correlated with Percentages of leaf litter cover and negatively correlated with Circumference at breast height and PC4 negatively correlated with Percentages of leaf litter cover and Number of plant species (NPS) (Fig. 6 and Table S5, Supplementary Material).

The first two axes generated by CCA explained 88.27% (55.89% in axis 1 and 32.38% in axis 2) of the total variation in the RB composition. Among the PC's, environmental variables represented by PC1 ($R=-0.6283$) were considered the most

important factors affecting the first axis, and the Percentage of leaf litter cover (represented by PC3) ($R=0.6631$) was highly correlated with the second axis (Fig. 6).

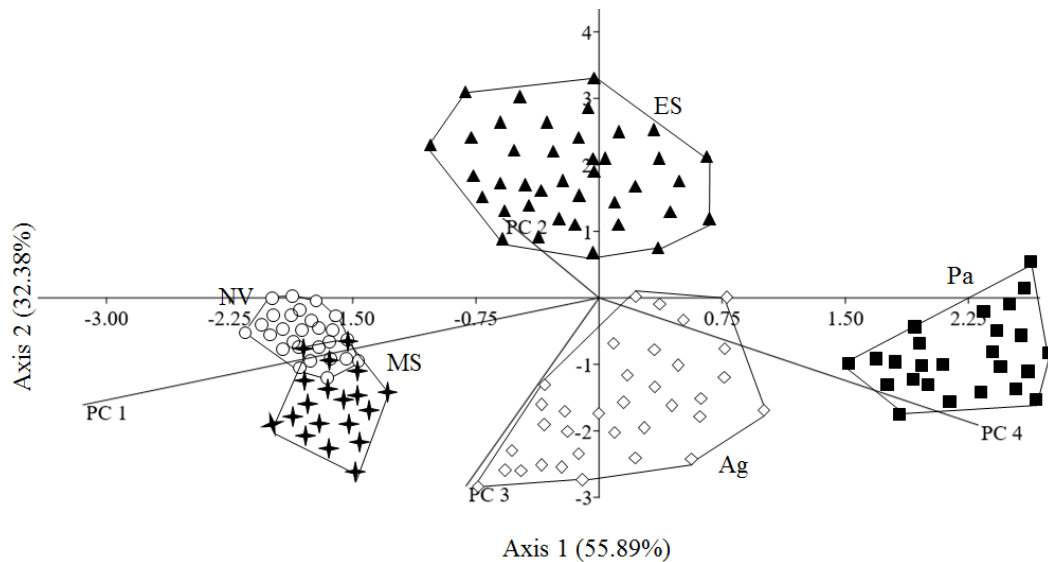


Fig. 6. Canonical Correspondence Analysis (CCA) used to estimate the influence of environmental variables on Rove beetle (Staphylininae) communities. Four axes were considered in the Principal Components Analysis (PCA), including 4 principal components (PC). NV, Native vegetation; MS, Maturing secondary succession (15 years of natural regeneration of vegetation); ES, Early secondary succession (5 years of natural regeneration of vegetation); Ag, Agriculture (Cacao plantations); Pa, Pasture.

The communities of RB in NV and MS were mainly determined by precipitation and humidity (0.591 and 0.522, respectively) (PC2) and Percentages of leaf litter cover and negatively correlated with Circumference at breast height (PC3); Ag, ES and Pa was positively correlated with bare soil (0.22) (PC4). The fourteen environmental variables separated the five ecosystems in four distinct groups. ES and the Ag were more similar to each other, although in opposite sides of axis 1. NV and MS were partially superimposed, while Pa is located far from the other ecosystems. In

fact, the distribution of some morphospecies of beetles is deeply linked to certain characteristics: *Oligotergus* sp2 and *Diachus* sp1 occurred mostly in areas with bare soil. Conversely, *Xantholinus* sp1 and *Xantholinus* sp2 occurred in areas with lower temperatures and *Rusilus* sp1, *Nordus* sp in wet areas; *Anotylus* sp1 and *Piestus aper* were mainly associated with high cover (Leaf litter cover and Height of leaf litter); *Hoplandria* sp occurred in more diverse areas (Number of shrubs), while *Diachus* sp2 and *Thinoharis* sp1 related to high precipitation (Fig. S1, Supplementary Material).

IndVal method

Of the 71 morphospecies found in the present study, 13 species were selected as ecosystem indicators. According to IndVal, 3 RB morphospecies were associated with NV, 3 RB morphospecies with ES, 3 RB morphospecies with Ag, 2 RB morphospecies with Pa and 2 with NV/MS. The subfamilies that presented the highest number of indicator morphospecies were Paederinae, Staphylininae and Oxytelinae, with 3 morphospecies each (Table 1).

Table 1 Analysis of RB' indicator species (IndVal) of NV, Vegetation native; MS, Maturing Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with 5 years of regeneration); Ag, Agriculture; Pa, Pasture. A, Mean Abundance; B, Relative frequency of occurrence (for details of A and B, see De Cáceres and Legendre, 2009).

Morphospecies	A	B	Indicator value	P	Site
<i>Aleocharinae</i> sp11	0.76	0.33	0.50	0.005	NV
<i>Piestus aper</i>	0.90	0.26	0.48	0.005	NV
<i>Neohypnus</i> sp	0.80	0.16	0.36	0.005	NV
<i>Diachus</i> sp1	0.86	0.29	0.47	0.005	ES
<i>Coproporus</i> sp4	0.90	0.24	0.47	0.005	ES
<i>Lithocharis</i> sp2	0.74	0.15	0.33	0.05	ES
<i>Aleochara</i> sp1	0.73	1	0.85	0.005	Ag
<i>Oligotergus</i> sp1	0.78	0.17	0.36	0.005	Ag
<i>Aneucamptus</i> sp	0.80	0.10	0.27	0.05	Ag
<i>Oxytelus</i> sp2	1	0.35	0.60	0.005	Pa
<i>Oxytelus</i> sp1	0.74	0.33	0.50	0.005	Pa
<i>Thinocharis</i> sp1	0.94	0.54	0.72	0.005	NV + MS
<i>Anotylus</i> sp1	0.98	0.42	0.64	0.005	NV + MS

Discussion

Assemblages analysis

Although considerable effort has been made to identify the most appropriate disturbance indicators for assessing the status of ecosystems, this exercise was many times unsatisfactory due to different views on what to measure and quantify. Additionally, the conventional measures regarding ecosystem studies require multiple specialization levels that are difficult to implement due to resource constraints (Cajaiba et al., 2015). Our study suggested using the composition of Rove beetles (RB) assemblages as stress – response indicators to key changes in such ecosystems. This proposal can only be completely addressed when the sampling effort associated with the heterogeneity of the ecosystems is considered in the scope of the need for rapid, standardized and cost-saving assessment methodologies (Scott and Anderson, 2003;

Cajaiba et al., 2017). Although we used several techniques for monitoring complementary phenological periods, the rarefaction curves of our work did not reach their asymptote values, indicating the high diversity of RB fauna in the monitored ecosystems (Colwell and Coddington, 1994). Anyway, the responses to anthropogenic disturbances found with non-specific sampling methods are applicable per se, and should be improved in later studies with specific methods for this group (Uehara-Prado et al., 2009). With hyperdiverse groups such as insects, it is difficult to obtain the complete inventory of the species that inhabit a given area (Jiménez-Valverde and Hortal, 2003). That is the case of Staphylinidae, which is the second most diverse family of the Order Coleoptera (Navarrete-Heredia et al., 2002). In the scope of disturbance indicator/surrogate works, the sampling protocol was designed in such a way that it could be led by one or two scientists in the field, minimizing operational costs associated to remote areas and increasing the chance of replication in future studies (see Paoletti, 1999; Gardner et al., 2008). In fact, indicator / surrogate studies depend, above all, on the common species that are introduced more rapidly into the inventory (Jiménez-Valverde and Hortal, 2003; Vásquez-Vélez et al., 2010).

The monitored richness (71 species) is comparatively high when paralleled to the values reported from other regions of the world (e.g. Sanabria et al., 2008; Yamamoto et al., 2014; Jahnová et al., 2016). The subfamilies Staphylininae, Aleocharinae and Paederinae, respectively, which had the highest richness, are also the ones that have the largest number of species worldwide (Navarrete-Heredia et al., 2002; Vásquez-Vélez et al., 2010). In general, the individuals of Pselaphinae, Aleocharinae, Paederinae and Staphylininae are generalist predators and species can be found in leaf litter, decomposing trunks, carrion, excrement, remains of fruits, caves,

banks of bodies of water, etc. There are also genera that are highly specialized in what they eat (e.g. some Pselaphinae) or where they live, such as those that are myrmecophilous (Aleocharinae, Paederinae and Staphylininae) (Navarrete-Heredia et al., 2002; Vásquez-Vélez et al., 2010) and the genus *Aleochara*: feeds on cyclorrhaphous fly puparia, acting as biological control for many dipteran pests (Yamamoto et al., 2014). However, and as stated before, the diversity of RB found in this work corresponds only a proportion of the probable diversity of this region (Asenjo et al., 2013).

Composition of Staphylinidae

Ecosystem degradation and loss are currently the most serious threats to richness and diversity (Fahrig, 2001) although this connection is not fully understood in the tropics (Rondinini and Chiozza, 2010). Establishing species lists for each ecosystem and for all potential “cold-spot” and “hot-spot” areas to evaluate their contribution to the conservation or depletion of the overall biodiversity would be too expensive (Lachat et al., 2012). We suggest, therefore, that studies for the purpose of relating disturbance with biodiversity should be carried out in specific areas and then extrapolated to other regions with similar characteristics.

In our study, pristine ecosystems were associated with higher species diversity and richness of RB, conforming to previous patterns reported for other groups of animals, and particularly beetles (González-Megías et al., 2011; Cajaiba et al., 2017). The applied indexes (Shannon diversity and richness) attained higher values for native vegetation, probably resulting from greater environmental complexity observed in relation to areas of increasing anthropic influence (e.g. gradient from native vegetation

to pastures). In fact, environmental complexity allows the coexistence of a larger set of species in pristine areas, due to the variety of resources offered and micro climatic and ecological conditions (Casas et al., 2016; Cajaiba et al., 2017). This also justifies the extraordinary abundance of a few species in pastures and agriculture (for details see Table S3, Supplementary material), adapted to homogeneous microclimatic and more homogenous structural factors (Quintero and Halffter, 2009).

Anthropogenic disturbance may affect species richness and diversity in several ways, and responses may vary within studies among taxonomic or functional groups or among studies within the same group (Basset et al., 2008). This variation may be attributed to several factors, such as the sensitivity of species to sampling effort, the spatial and temporal scale of the study, disturbance intensity, frequency and type (Basset et al., 2008).

We found similarities in the abundance of RB between ecosystems (e.g. NV and Ag), although different compositions (demonstrated by PERMANOVA and NMDS). Thus, species composition and or morphological traits (e.g. dispersal ability) should be used to complement species richness and/or abundance for characterizing the disturbance status and even the conservation value of an ecosystem in this region (Niemelä, 1997). Furthermore, the lower number of RB in secondary forests (ES and MS) indicates either reduced resource availability in the young stands or the colonization stage of the RB in young forest stands (Lange et al., 2014). We found a high degree of species turnover between ecosystems, which represented approximately 80% of the overall beta diversity. This indicates that each ecosystem presents a fairly distinct subset of species, although agroecosystems (agriculture and pastures) typically retain a non-random set of species that often replaces core forest habitat and dietary

specialists (Lyra-Jorge et al., 2010). The obtained results also seem to show agroecosystems act, in this region, as a selective filter on Rove beetles across the landscape (Chiarello, 2000), affecting matrix fluxes according to the dispersal capacity of each species (Lees and Peres, 2009). “Agroecosystems” resilient species are also more likely to survive in fragmented landscapes than forest specialists (Tabarelli et al., 2012; Magioli et al., 2016) representing the residual fauna in the aftermath of habitat fragmentation.

Effects of environmental variables on Rove Beetle assemblages

The PCA/CCA analysis revealed that RB assemblages of highly disturbed ecosystems (e.g. Pasture and Agriculture) are mainly associated with bare soil and high temperatures. In contrast, the communities of pristine ecosystems (native vegetation) are positively influenced by the complex structural vegetation, soil and humidity, which are also important determinants of RB communities’ ecology (Silva and Hernández, 2016; Cajaiba et al., 2017). Vegetation complexity is therefore the key environmental driver of RB community structure in this region. This finding is consistent with previous studies that demonstrated that plant community composition, productivity, and diversity are the primary factors affecting arthropod abundance, diversity, and community composition (Liu et al., 2016; Cajaiba et al., 2017).

In our study, richness, diversity and composition followed a “linear” trend in line with the disturbance gradient: a decrease as disturbance increased. RB assemblages are highly distinct between ecosystems and in that way easily separated from each other (Moraes et al., 2013). Litter type and litter depth are important ecological determinants, providing hunting and foraging niches, protection from

predators and desiccation (Cajaiba et al., 2017). Additionally litter provides attachment points for web building (Leclerc and Blandin, 1990). Coarse woody debris is considered fundamental as overwintering site, for ovipositioning and larval development for many RB species (Lövei et al., 2006; Cajaiba and Silva, 2017). Cattle trampling in grassland areas contributes to soil compaction, resulting in reduced pathways through the litter layer, hiding surfaces and hunting places. Moreover, reduced cover affects foraging or maintenance of water balances in RB (Lövei et al., 2006).

The RB richness observed in forest ecosystems and/ or maturing secondary succession (NV/MS) could be associated with the availability of niches and food resources. On the other hand, disturbed ecosystems support mostly generalist and tolerant RB species (Cajaiba et al., 2017). In fact, the distribution of RB along different environmental characteristics seems to demonstrate discrete associations with particular biotypes within the landscape (Silva and Hernández, 2016). Evidence suggests that species richness and abundance (and also biomass) are negatively impacted by disturbed ecosystems (Silva and Hernández, 2016; Cajaiba et al., 2017) especially those without tree canopy cover. Overall changes in vegetation characteristics affect RB diversity and community composition indirectly, through pathways or mechanisms, such as changing microclimatic conditions and soil properties (Schaffers et al., 2008; Liu et al., 2016).

Value indicator - IndVal

A species restricted to one or a few ecosystem types potentially represents a better indicator of environmental change than an ecosystem generalist, owing to the

greater susceptibility of the specialist to local or regional characteristics. Indicator species are used to monitor environmental changes and assess the impacts of disturbances (De Cáceres and Legendre, 2009). RB are considered to be mostly generalists, although it is increasingly found that many species are specialists (Buse and Good, 1993), with a significant indicator value for a particular ecosystems (Pohl et al., 2007; Gutiérrez-Chacon et al., 2009).

The Indicator Value (IndVal) has been used in several works (e.g. Bicknell et al., 2014; Cajaiba et al., 2017), due to its efficiency in identifying specific indicators. The applications of indicator species analysis are many, including conservation, land management, landscape mapping, or design of natural reserves (Cajaiba et al., 2017). In our study, RB indicators were considered useful tools to define the disturbance status of ecosystems, using the native vegetation as our reference. This means, for instance, that one can use indicator species to classify sites of pristine ecosystems (e.g. in vegetation mapping) or to monitor the succession/ degradation of a particular area or even to detect an environmental gradient from natural to disturbed areas in time and space (e.g. recover after pollution, disturbance by fire, climatic changes, etc) (De Cáceres et al., 2010).

Considering the little knowledge of the biodiversity and ecology of RB in the Neotropics, the indicator species detected in this study represents a contribution to the characterization of the Neotropical fauna, specifically of this important but unappreciated taxon. Additional information is urgently needed to define further the ecological roles of RB (Gutiérrez-Chacon et al., 2009).

Contributions for conservation/ challenges and final remarks

The characterization of general diversity patterns and responses to disturbance at the continental scale may be a very difficult task, due to functional and structural differences among biomes and ecosystems, and idiosyncrasies (e.g., history of disturbance) of different regional communities (Uehara-Prado et al., 2009). However, general patterns emerging from studies focused on specific biomes within regions (Kim and Byrne, 2006) could validate the use of ecological indicators within specific geographical limits (Uehara-Prado et al., 2009). The results obtained show that the RB richness was sensitive to the structural changes induced by increasing pressure from human actions. Additionally an increasing trend in community complexity from more to less disturbed ecosystems was observed. Highly disturbed systems had detrimental effects on the abundance of specialists and benefited generalist species. Differences in responses can be explained by differences in the intensity and extent of the change from pristine systems. Overall, the composition of Rove Beetles communities is ecosystems' specific and sensitive to anthropogenic induced structural changes and, therefore, should be considered a valuable ecological indicator in the Neotropics. Nevertheless our work should be complemented by ecological and behavioural studies to understand and assess the state of degradation and or conservation of the ecosystems considered (Brown, 1997), considering that ecological assessment and monitoring are important procedures to address effective management, in which the use of essential indicators is considered crucial to measure and evaluate the status and trends of target environmental systems (Cajaiba et al., 2017). Additionally, future studies concerning disturbance patterns and its connection with potential ecological indicators in the neotropics and specifically in Amazonia should focus on increasing the geographical

sampling coverage of this biome, in search of well-defined patterns of response to disturbance gradients.

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Supplementary material

Table S1. Generic characteristics of the ecosystems sampled in the municipality of Uruará, state Pará, northern Brazil.

Habitat	N. areas	Characteristics
Native Vegetation – NV	02	Composed of vegetation whose facies is an upper canopy formed mainly by green trees, with crowns that touch each other, creating a dense and enclosed canopy all year round. In this ecosystem no traces of anthropic action were verified (for example, trails, residues, fires, selective cutting of wood, agricultural activities, among others).
Maturing secondary succession - MS (vegetation with 15 years of regeneration)	02	A secondary forest (or second-growth forest) is a forest or woodland area which has re-grown after abandonment of agriculture. Biological diversity gradually increases as long as there are primary remnants to supply seeds. The average height of the trees is over 12 meters and the average diameter is over 14 centimeters.
Early Secondary succession – ES (vegetation with five years of regeneration)	02	Same as above. This phase typically lasts for up to six years and in some cases up to ten years, depending on soil quality and / or seed bank. The average height of trees is no more than four meters and the diameter of the main trees can reach eight centimeters.
Agriculture - Ag (cocoa plantations, <i>Theobroma cacao</i> L.)	02	Agroecosystem represented by areas planted with cacao crops (<i>Theobroma cacao</i> L.).
Pasture for extensive livestock – Pa	02	Agroecosystem dominated by herbaceous vegetation (mainly species of the genus <i>Brachiaria</i> spp) used for extensive livestock farming.

Table S2. Specification of the methodology used for monitoring the environmental variables.

Variables	Specification	Monitoring Methodology
Temperature	Celsius (°C)	Measured during the traps installation and removal with a portable weather station (model Oregon Scientific WMR200A).
Humidity	Humidity (%)	Measured during the traps installation and removal with a portable weather station (model Oregon Scientific WMR200A).
Precipitation	Precipitation (mm)	Measured during the traps installation and removal with a portable weather station (model Oregon Scientific WMR200A).
Circumference at Breast Height	Centimeters (cm)	Trunk diameter was taken at breast height (1.3 m) for the trees.
Circumference at Ankle Height	Centimeters (cm)	The diameter was measured at the ankle height (CAH = 0.1 m) for the shrubs.
Canopy Cover	Percentage (%)	Calculated with a convex spherical densiometer (D) Lemmon and assigned the following classes: 0–5%, 6–25%, 26–50%, 51–75%, 76–95% and 96–100%
Number of Plant Species	Number of Plant Species/ m ²	The number of tree species was counted in an area of 100 m ² (10 x 10 m) in the vicinity of each pitfall trap.
Number of Plants	Number of Plants/ m ²	The number of tree was counted in an area of 100 m ² (10 x 10 m) in the vicinity of each pitfall trap.
Number of Species of Shrubs	Number of Species of Shrubs/ m ²	The number of shrubs species was counted in an area of 100 m ² (10 x 10 m) in the vicinity of each pitfall trap.
Number of Shrubs	Number of Shrubs/ m ²	The number of shrubs was counted in an area of 100 m ² (10 x 10 m) in the vicinity of each pitfall trap.
Percentage of Exposed Soil	Percentage (%) / m ²	The percentage exposed soil in each quadrant was estimated in different percentage classes (0-5, 6-25, 26-50, 51-75, 76-95, 96-100%)
Percentage of Green Cover (vegetation)	Percentage (%) / m ²	The percentage green cover (vegetation up to 1 m height) in each quadrant was estimated in different percentage classes (0-5, 6-25, 26-50, 51-75, 76-95, 96-100%)
Percentages of Leaf Litter Cover	Percentage (%)	The percentage of litter in each quadrant was estimated in different percentage classes (0-5, 6-25, 26-50, 51-75, 76-95, 96-100%).
Height of Leaf Litter	Centimeters (cm)	Litter height was measured using a ruler at five points inside the square (near each corner and in the center)

Adapted Da Silva and Hernández (2016)

Table S3. Total numbers of Staphylinidae morphospecies/species in the different ecosystems studied in the Brazilian Amazon. NV = Native Vegetation; MS = Maturing secondary succession (15 years of regeneration); ES = Early secondary succession (5 years of regeneration); Ag = Agriculture (Cocoa); Pa = Pasture. RA (%) = Relative Abundance. FG = Feeding Guild (Pr, Predaceous; Ps, Parasitoids; S, Saprophagous; M, Mycophagous;

Subfamily Morphospecies/Species	NV	MS	ES	Ag	Pa	RA (%)	FG	Total
Aleocharinae (S=15; N=966; N%=64.70)								
<i>Aleochara repetita</i>	114	1	43	136	0	19.69	Pr, Ps	294
<i>Aleochara</i> sp1	1	0	21	145	33	13.40	Pr, Ps	200
<i>Aleochara</i> sp2	8	0	8	7	0	1.54	Pr, Ps	23
Aleocharinae sp1	20	19	15	0	2	3.75	Pr	56
Aleocharinae sp2	2	0	0	0	0	0.13	Pr	2
Aleocharinae sp3	2	2	5	0	0	0.60	Pr	9
Aleocharinae sp4	0	1	0	0	0	0.07	Pr	1
Aleocharinae sp5	0	0	0	0	5	0.33	Pr	5
Aleocharinae sp6	0	1	0	0	0	0.07	Pr	1
Aleocharinae sp7	0	0	1	0	0	0.07	Pr	1
Aleocharinae sp8	0	1	0	0	0	0.07	Pr	1
Aleocharinae sp9	0	0	1	0	0	0.07	Pr	1
Aleocharinae sp10	1	0	0	0	3	0.27	Pr	4
Aleocharinae sp11	14	3	0	1	0	1.21	Pr	18
<i>Hoplandria</i> sp	115	68	91	63	13	23.44	Pr	350
Paederinae (S=13; N=169; N%=11.32)								
<i>Thinocharis</i> sp1	73	22	0	4	0	6.63	Pr	99
<i>Rugilus</i> sp1	1	6	7	6	0	1.34	Pr	20
<i>Pinophilus</i> sp1	1	1	0	0	0	0.13	Pr	2
<i>Biocrypta</i> sp1	0	3	0	0	0	0.20	Pr	3
<i>Palaminus</i> sp1	0	0	1	0	0	0.07	Pr	1
<i>Ronetus</i> sp1	1	0	1	3	0	0.33	Pr	5
<i>Xenaster</i> sp1	0	0	0	2	0	0.13	Pr	2
<i>Echiaster</i> sp1	0	0	0	3	0	0.20	Pr	3
Paederini sp1	0	0	1	0	0	0.07	Pr	1
<i>Achenomorphus</i> sp1	0	1	2	0	0	0.20	Pr	3
<i>Lithocharis</i> sp1	6	2	4	4	2	1.21	Pr	18
<i>Lithocharis</i> sp2	2	1	6	1	0	0.67	Pr	10

<i>Scopobium</i> sp	0	2	0	0	0	0.13	Pr	2
Oxytelinae (S=4; N=97; N%=6.50)								
<i>Carpelimus</i> sp1	3	1	0	0	0	0.27	S	4
<i>Anotylus</i> sp1	28	14	0	0	1	2.88	S	43
<i>Oxytelus</i> sp1	7	0	0	1	23	2.08	S	31
<i>Oxytelus</i> sp2	0	0	0	0	19	1.27	S	19
Tachyporinae (S=6; N=85; N%=5.69%)								
<i>Coproporus</i> sp1	33	19	14	1	0	4.49	Pr, M, S	67
<i>Coproporus</i> sp2	0	0	3	0	0	0.20	Pr, M, S	3
<i>Coproporus</i> sp3	0	1	1	0	0	0.13	Pr, M, S	2
<i>Coproporus</i> sp4	0	1	10	0	0	0.74	Pr, M, S	11
<i>Mycetoporus</i> sp1	0	0	0	1	0	0.07	Pr, M, S	1
<i>Sepedophilus</i> sp1	1	0	0	0	0	0.07	Pr, M, S	1
Piestinae (S=1; N=12; N%=0.8)								
<i>Piestus aper</i>	11	1	0	0	0	0.80	S	12
Staphylininae (S=20; N=140; N%=9.38)								
<i>Lissohypnus</i> sp	1	6	17	4	0	1.88	Pr	28
<i>Neohypnus</i> sp	8	0	1	1	0	0.67	Pr	10
<i>Xantholinus</i> sp1	2	1	8	0	0	0.74	Pr	11
<i>Xantholinus</i> sp2	2	0	1	0	2	0.33	Pr	5
<i>Heterothops</i> sp	0	0	2	0	0	0.13	Pr	2
<i>Nordus</i> sp	0	0	2	0	0	0.13	Pr	2
<i>Bolitogyrus</i> sp	0	0	2	0	0	0.13	Pr	2
<i>Platydracus</i> sp	1	0	0	0	0	0.07	Pr	1
<i>Oligotergus</i> sp1	0	0	2	7	0	0.60	Pr	9
<i>Oligotergus</i> sp2	0	0	2	2	0	0.27	Pr	4
<i>Acylophorus</i> sp	1	2	0	1	0	0.27	Pr	4
<i>Philonthus</i> sp1	0	0	1	1	0	0.13	Pr	2
<i>Philonthus</i> sp2	1	0	1	0	2	0.27	Pr	4
<i>Philonthus</i> sp3	0	2	0	0	0	0.13	Pr	2
<i>Diochus</i> sp1	5	2	19	3	0	1.94	Pr	29
<i>Diochus</i> sp2	2	0	2	0	1	0.33	Pr	5
<i>Belonuchus</i> sp1	2	0	1	0	0	0.20	Pr	3
<i>Belonuchus</i> sp2	0	3	2	0	0	0.33	Pr	5
<i>Belonuchus</i> sp3	0	1	1	3	0	0.33	Pr	5

<i>Belonuchus</i> sp4	3	2	1	1	0	0.47	Pr	7
Pselaphinae (S=3; N=6; N%=0.4)								
<i>Pselaphinae</i> sp1	2	0	0	0	0	0.13	Pr	2
<i>Pselaphinae</i> sp2	1	0	0	0	0	0.07	Pr	1
<i>Pselaphinae</i> sp3	3	0	0	0	0	0.20	Pr	3
Scydmeninae (S=1; N= 2; N%=0.14)								
<i>Scydmeninae</i> sp1	1	1	0	0	0	0.13	Pr	2
Euasthetinae (S=1; N=1; N%=0.06)								
<i>Octavius</i> sp	1	0	0	0	0	0.07	Pr	1
Megalopsidiinae (S=1; N=1; N%=0.07)								
<i>Megalopinus</i> sp1	1	0	0	0	0	0.07	Pr	1
Osoriinae (S=6; N=14; N%=0.94)								
<i>Holotruchus</i> sp	1	0	0	0	0	0.07	S	1
<i>Aneucamptus</i> sp	1	0	0	4	0	0.33	S	5
<i>Tannea</i> sp	3	0	0	0	0	0.20	S	3
<i>Thoracophorus</i> sp	1	1	0	1	0	0.20	S	3
<i>Osorius</i> sp	0	0	0	0	1	0.07	S	1
<i>Clavilispinus</i> sp	1	0	0	0	0	0.07	S	1
Abundance	488	192	300	406	107	100		1.493
Richness	43	32	36	26	13	-		71

Table S4. The turnover (β_{sim}) and nestedness (β_{sor}) components for each ecosystem pairwise comparisons. The (β_{sim}) results are depicted in the dashed part of the table and (β_{sor}) results are depicted without highlight. NV, Native vegetation; MS, Maturing Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture (Cocoa); Pa, Pasture.

	NV	MS	ES	Ag	Pa
NV	---	0.39	0.49	0.48	0.64
MS	0.26	---	0.56	0.68	0.80
ES	0.16	0.15	---	0.43	0.67
Ag	0.15	0.08	0.16	--	0.67
Pa	0.20	0.07	0.16	0.12	---

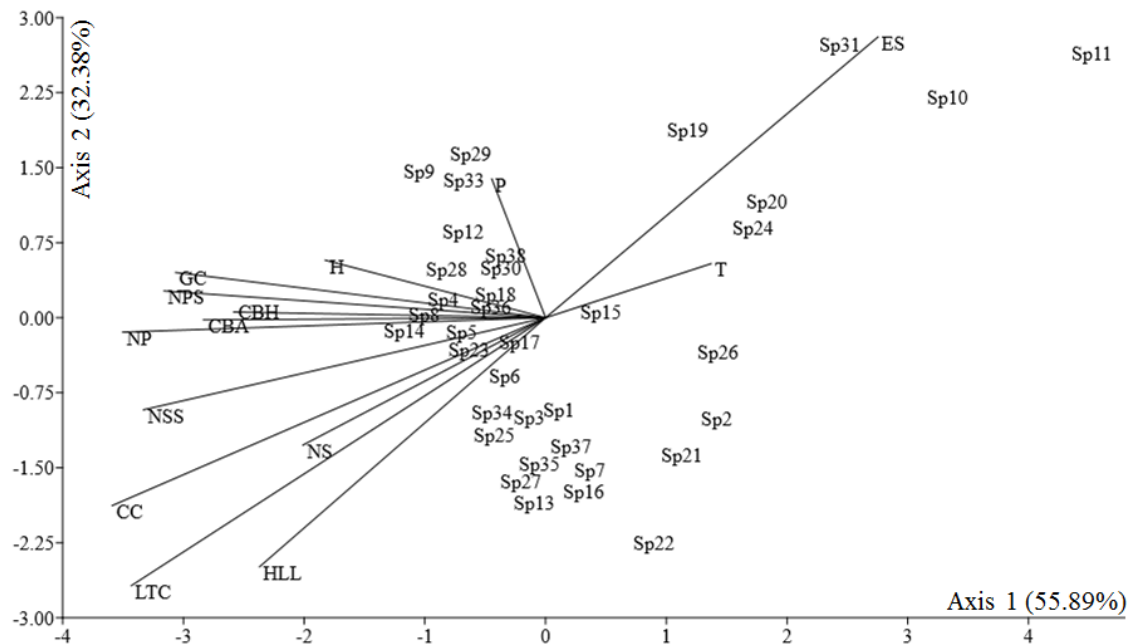
Table S5. Eigenvalues of the Principal Components Analysis (PCA) for the first four axes. T. Temperature; H. Humidity; P. Precipitation; CBH. Circumference at Breast Height; CAH. Circumference at Ankle Height; CC. Canopy Cover-CC; NPS. Number of Plant Species; NP. Number of Plants; NSS. Number of Species of Shrubs; NS. Number of Shrubs; PES. Percentage of Exposed Soil; PGC. Percentage of Green (vegetation) Cover; PLC. Percentages of Leaf Litter Cover; HLL. Height of Leaf Litter.

Variables	Components			
	PC 1	PC 2	PC 3	PC 4
T	-0.0063	-0.0126	-0.0174	-0.0056
H	0.0486	0.1046	0.0561	-0.0435
P	0.0168	0.9938	-0.0099	0.0043
CBH	0.7763	-0.0193	-0.4947	0.3663
CAH	0.0147	-0.0005	-0.0029	0.0008
CC	0.1730	-0.0030	0.4348	0.3564
NPS	0.5401	-0.0148	0.3173	-0.6257
HLL	0.0066	-0.0014	0.0198	0.0402
PLC	0.1178	0.0014	0.5961	-0.5072
PGC	0.1256	0.0223	0.0464	-0.1284
PES	-0.0162	0.0036	-0.0331	-0.0835
NP	0.2061	-0.0061	0.2776	-0.2493
NS	0.0025	0.0010	0.1593	-0.0235
NSS	0.0139	-0.0002	0.0204	0.0073

Figure S1. CCA for Staphylinidae species using 14 environmental variables (Species singletons and doubletons were omitted from ordination).

Variables: Temperature-T, Humidity-H, Precipitation-P, Circumference at Breast Height-CBH, Circumference at Ankle Height-CAH, Canopy Cover-CC, Number of Plant Species-NPS, Number of Plants-NP, Number of Species of Shrubs-NSS, Number of Shrubs-NS, Percentage of Exposed Soil-PES, Percentage of Green (vegetation) Cover-PGC, Percentages of Leaf Litter Cover-PLC, Height of Leaf Litter-HLL.

Species: Sp1-*Aleochara repetita*; Sp2-*Aleochara* sp1; Sp3-*Aleochara* sp2; Sp4-*Thinocharis* sp1; Sp5- *Thinocharis* sp2; Sp6-*Rusilus* sp1; Sp7-*Ronetus* sp1; Sp8-*Carpelimus* sp1; Sp9-*Anotylus* sp1; Sp10-*Oxytelus* sp1; Sp11-*Oxytelus* sp2; Sp12-*Coproporus* sp1; Sp13-*Coproporus* sp4; Sp14-*Piestus aper*; Sp15-*Lithocharis* sp1; Sp16-*Lithocharis* sp2; Sp17-*Lissohypnus* sp; Sp18- *Neohypnus* sp; Sp19-*Xantholinus* sp2; Sp20-*Quedius* sp2; Sp21-*Oligotergus* sp1; Sp22-*Oligotergus* sp2; Sp23-*Acylophorus* sp; Sp24-*Philonthus* sp2; Sp25-*Diochus* sp1; Sp26-*Diochus* sp2; Sp27-*Diochus* sp3; Sp28-*Aleocharinae* sp1; Sp29-*Aleocharinae* sp2; Sp30-*Aleocharinae* sp3; Sp31-*Aleocharinae* sp5; Sp32-*Aleocharinae* sp10; Sp33-*Aleocharinae* sp11; Sp34-*Belonuchus* sp2; Sp35-*Belonuchus* sp3; Sp36-*Belonuchus* sp4; Sp37-*Aneucamptus* sp; Sp38-*Hoplandria* sp.



Chapter 8

Diversity of Scolytinae (Coleoptera: Curculionidae) in different landscapes in northern Brazil

Diversidade de Scolytinae (Coleoptera: Curculionidae) em diferentes paisagens no norte do Brasil

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Abstract

A survey of beetle assemblages of the subfamily Scolytinae was conducted in habitat gradients ranging from primary forest to secondary forests, cocoa plantations, and pastures in the Amazonian biome. Sampling was carried out in 2015, during February/March (rainy season), June (intermediary season), and September/October (dry season). A total of 4,534 Scolytinae was captured, comprising four genera and nine species. The most abundant species were *Xyleborus affinis* and *Xyleborus volvulus*, representing approximately 34% and 31% of the total abundance, respectively. Species of Scolytinae in general showed seasonality and greater abundance and richness in regions with greater coverage of plant litter.

Keywords: Amazon, Biodiversity studies, Habitat preference, Insects.

Resumo

Assembleias de besouros da subfamília Scolytinae foram estudadas em gradientes de habitats que variam de floresta primária a florestas secundárias, plantações de cacau e pastagens no bioma Amazônia. A amostragem foi realizada durante o ano de 2015, nos meses de fevereiro/março (estação chuvosa), junho (estação intermediária) e setembro/outubro (estação seca). Um total de 4.534 Scolytinae foi capturado, compreendendo quatro gêneros e nove espécies. As espécies mais abundantes foram *Xyleborus affinis* e *Xyleborus volvulus*, representando aproximadamente 34% e 31% da abundância total, respectivamente. As espécies de Scolytinae em geral mostraram sazonalidade e maior abundância e riqueza em regiões com maior cobertura de serapilheira.

Palavras-chave: Amazônia, Estudos de biodiversidade, Preferência de habitat, Insetos.

Introduction

The Amazon biome is a unique area; it holds the world's largest rainforest and is considered a biodiversity hotspot. It is also important for carbon storage and regional/global regulation of the hydrological cycles and climate (Fernandes, 2009). However, human action has caused, directly or indirectly, the reduction and loss of biodiversity (Cajaiba *et al.*, 2015) due to changes in the natural ecological processes and the emergence of new combinations of species, mainly by habitat transformation, fragmentation, and damage (Marques *et al.*, 2002). Several factors have been identified including the suppression of natural habitats, ecosystem fragmentation, overexploitation of natural resources, climate change, and introduction of alien species. This biodiversity loss threatens the sustainability of the region, with a consequent reduction of environmental services such as pest control, nutrient cycling, and maintenance of soil structure (Cajaiba and Silva, 2015; Valiente-Banuet *et al.*, 2015).

Predicting the ecological consequences of land-use/land-cover changes is, therefore, a subject of great scientific and political interest in order to support strategic options for sustainable development, land-use planning, and natural resource management (Turner *et al.*, 2007). In this context, ecological assessment and monitoring are important tools to support effective management of ecosystems and natural resources, and the use of pertinent indicators is crucial to measure and evaluate the status and trends of target environmental systems (Cajaiba *et al.*, 2015).

Beetles have characteristics that qualify them as appropriate for ecological studies (Carlton and Robinson, 1998; Didham *et al.*, 1998; Cajaiba and Silva, 2015) and for monitoring different compartments of the ecosystem (Marinoni, 2001). Several studies suggest that beetles are strongly associated with habitat structure and factors associated with microclimates (Schwarzkopf and Rylands, 1989; Bishop *et al.*, 2009) that can be altered by anthropic activities. For example, beetles respond to environmental disorder, soil depth, local humidity, temperature, soil pH, and pollution (Pearce *et al.*, 2003; Kaiser *et al.*, 2009; Garcia *et al.*, 2010; Negro *et al.*, 2010; Vasquez-Velez *et al.*, 2010). Beetles have also been shown to indicate successful management and restoration of habitats (Jacobs *et al.*, 2010; Paoletti *et al.*, 2010; Gomez, 2010).

Beetles of the subfamily Scolytinae (Coleoptera: Curculionidae), with more than 6,000 described species, constitute one of the largest groups of Coleoptera (Eroğlu *et al.*, 2005; Sarikaya and Avci, 2011). These beetles, commonly known as bark and ambrosia beetles, colonize living and dead trees (Gandhi *et al.*, 2010), with some species causing widespread mortality of coniferous and deciduous trees in forests (Oliver and Mannion, 2001; Gandhi *et al.*, 2010). In general, bark beetle larvae feed on the phloem of their host trees, whereas ambrosia beetle larvae feed on the symbiotic fungus inoculated by the mother beetles when they bore into the xylem of their host trees (Rabaglia *et al.*, 2006). These insects are well-known forest pests and also play an important role in wood degradation (Gray, 1972). In the Amazon biome, these beetles were also found to attack several forest and fruit trees (Abreu, 1992; Barbosa, 1994; Dall'Oglio and Peres-Filho, 1997; Abreu *et al.*, 2012).

The factors that influence the biodiversity of beetles are not clearly known, but the literature shows that structural complexity of vegetation and different environmental disturbances play major roles. Several studies have shown the biology, ecology, and feeding behavior of Scolytinae (Franceschi *et al.*, 2005; Rabaglia *et al.*, 2006; Fettig *et al.*, 2007). However, there is a gap in the knowledge on the Brazilian fauna of Scolytinae, as well as that in the Amazon biome. Thus, we conducted a survey of Scolytinae assemblages, over a wide range of habitats from primary forest to secondary forests, cocoa plantations, and pastures in the Amazon biome, with the aim of supporting future research and adding information about these economically important insects. Specifically, we addressed the following questions: 1) Does the composition of Scolytinae assemblages vary among the different habitats? 2) Does the diversity of Scolytinae in the Amazon biome differ between seasons?

Material and Methods

This study was performed near the city of Uruará, state of Pará, northern Brazil (Figure 1). The territorial extension of the municipality is 10796 km² and its population encompasses circa 44789 inhabitants. The dominant land use/ land cover (LU/LC) is natural forest (69% of the area) and deforestation is concentrated mainly in the south-central part of territory and near the main roads. Extensive livestock production and

the exploitation of timber at a large scale (mostly illegal) are currently considered the most serious environmental threats (Cajaiba, 2014). The studied habitats contain pertinent gradients, in terms of biophysical and ecological characteristics, for testing the response of Scolytinae communities facing the main anthropogenic drivers. These gradients encompass: Native Vegetation - NV, Early Secondary succession – ES (vegetation with five years of regeneration), Mature Secondary succession - MS (vegetation with 15 years of regeneration), Agriculture - Ag (cocoa plantations, *Theobroma cacao* L.) and Pasture for extensive livestock - Pa. The climate is classified as Aw (Köppen), hot and humid and the average annual rainfall is 2000 mm (Cajaiba *et al.*, 2015).

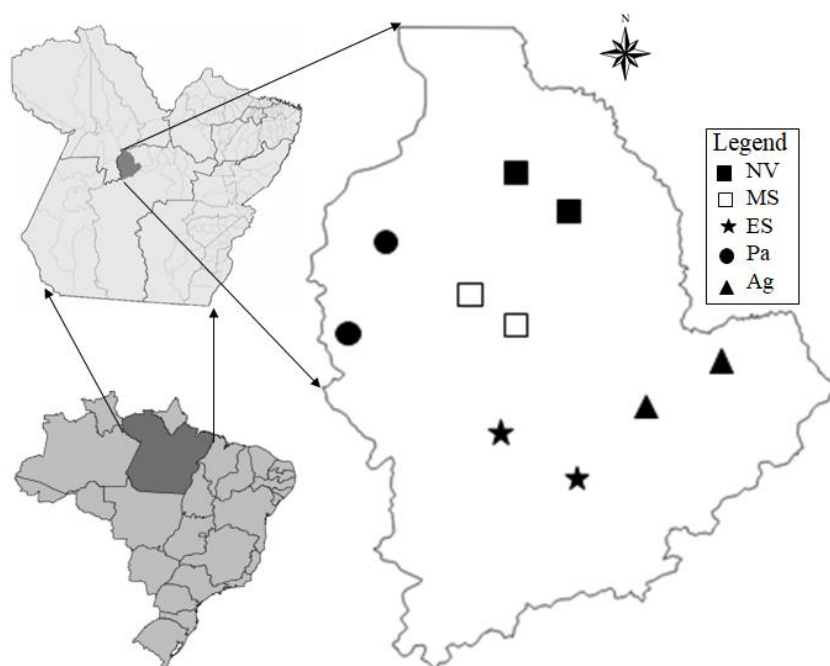


Figure 1. Location of the study area in the municipality of Uruará, state of Pará, northern Brazil with sampled habitats: Ag, Agriculture; ES, Early secondary succession; MS, Mature secondary succession; NV, Native vegetation; Pa, Pasture.

Sampling was carried out during the year 2015, in the months of February/March (rainy season), June (final of rainy season and early dry season) and September/October (dry season). This allowed integrating eventual seasonal differences in the activity of Scolytinae. The sample points were placed at a minimum distance of 100 meters from ecotones, to ensure that most beetles captured were associated to the

habitats in study. Pitfall traps with 75 mm diameter and 110 mm deep were filled with preservative liquid consisting of formalin, alcohol, water and a few drops of detergent to break the surface tension. A roof was attached to each trap to prevent rainwater from entering the trap, remaining installed for 48 h prior to collection.

In each study site (NV, MS, ES, Ag and Pa) seven sample points were placed 100 m apart. Each sample point contained four pitfall traps separated by 5 meters. This protocol was applied to all areas and periods of collections, totalizing a sampling effort of 840 traps.

The Temperature-T, Humidity-H and Precipitation-P of each point were measured during the traps installation and removal with a portable weather station (model Oregon Scientific WMR200A). Data about Percentage of Exposed Soil-PES, Percentages of Leaf Litter Cover-PLC, Height of Leaf Litter-HLL were also collected. To assess the environmental complexity of each sampling site, the quadrat-section method was adopted (Campos and Hernández, 2015). Using a cross as a reference, four quadrants (northeast, northwest, southeast, southwest) were marked and, in each quadrant the following variables were measured: the HLL was measured with a ruler in 1 m × 1 m marked squares (using a PVC pipe), and the percentages of PLC and PES were measured by visual estimation using the following classes, 0–5%, 6–25%, 26–50%, 51–75%, 76–95% and 96–100% (Campos and Hernández, 2015).

Richness (S) and abundance (N) of each sampling site were measured and differences among sites were gauged using One-Way-Analysis-of-Variance (ANOVA). ANOVA was also applied to find differences in the seasonality of Scolytinae, using a Tukey's post-hoc test. The normality of the data was verified by the Shapiro-Wilk test.

The Bray-Curtis cluster analysis was applied to verify the similarity between different habitats, and UPGMA algorithm was used to draw the distance based on Bray-Curtis index. This index ranges between 0 (indicating no similarity in community composition between sites) and 1 (indicating complete overlap), and it is considered one of the most robust measures of community similarity (Magurran, 2004). The cophenetic correlation coefficient was used to verify the result significance of the cluster analysis. Multiple multivariate regressions were applied to analyze the

relationship between the dependent (abundance and richness of Scolytinae) and predictive (T, H, P, PLC, HLL and PES) variables.

Results and Discussion

A total of 4,534 Scolytinae, distributed in four genera and nine species, was captured. Of these, we identified 854 individuals of all species in natural vegetation (NV), eight species (1116 individuals) in mature secondary succession (MS), seven species (244 individuals) in early secondary succession (ES), four species (2,028 individuals) in agriculture (Ag), and two species (292 individuals) in pasture (Pa). The genus *Xyleborus*, with 4,514 individuals, was prevalent in all ambrosia beetle communities, comprising 99.5% of all specimens captured. The most abundant species were *Xyleborus affinis* EICHHOFF 1868, with 1,526 individuals (approximately 34% of the total abundance), and *Xyleborus volvulus* FABRICIUS 1775, with 1,386 individuals (approximately 31% of the total abundance). *Xyleborus volvulus* was more abundant in Ag (506 individuals) and NV (300 individuals), whereas *X. affinis* was more abundant in MS (382 individuals), followed by Pa (210 individuals) and ES (58 individuals) (Table 1).

Table 1. Species of Scolytinae collected in different habitats and their abundance in different seasons in the municipality of Uruará, state of Pará, northern Brazil. Ag, Agriculture; ES, Early secondary succession; MS, Mature secondary succession; NV, Native vegetation; Pa, Pasture. The letters R, I and D following the vegetation types correspond to the rainy, intermediary and dry seasons, respectively.

Species	NV			MS			ES			Ag			Pa		
	R	I	D	R	I	D	R	I	D	R	I	D	R	I	D
<i>Xyleborus volvulus</i>	104	196	32	144	116	24	34	20	18	260	246	110	34	38	10
<i>Xyleborus affinis</i>	74	130	38	210	156	16	38	20	18	250	238	128	110	90	10
<i>Xyleborus ferrugineus</i>	32	64	16	58	64	18	24	0	2	156	136	48	0	0	0
<i>Xyleborus</i> sp1	14	38	30	72	56	8	18	8	0	134	132	38	0	0	0
<i>Xyleborus</i> sp2	18	12	24	58	42	8	12	0	0	70	82	0	0	0	0
<i>Xyleborus</i> sp3	8	6	8	24	30	6	18	10	0	0	0	0	0	0	0
<i>Xylosandrus</i> sp	0	4	0	0	2	0	4	0	0	0	0	0	0	0	0
<i>Coccotrypes</i> sp	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0
<i>Hypothenemus</i> sp	2	2	0	2	2	0	0	0	0	0	0	0	0	0	0

The fact that the genus *Xyleborus* presented the greatest abundance and richness is consistent with the results of several studies (Abreu *et al.*, 2012; Kendra *et al.*, 2011; Meurer *et al.*, 2013). Although beetles of this genus may be considered beneficial, because they favor natural disbranching and contribute to degradation of wood waste in reforested areas, some species may cause high negative economic impacts, damaging large amounts of fresh-cut or stored wood (Wood, 1982; Meurer *et al.*, 2013).

The species *X. affinis* and *X. volvulus* were the most abundant in all studied habitats, corroborating the results of Meurer *et al.* (2013). *X. affinis*, considered one of the most aggressive ambrosia beetle species in Brazil and other tropical regions (Beaver, 1976; Meurer *et al.*, 2013), is abundant above the Tropic of Capricorn (Meurer *et al.*, 2013). Beaver (1976), in a study conducted in a native forest of Mato Grosso, stressed the importance of *X. affinis* in the area, noting that the wetter the region, the better was the adaptation. According to this author, *X. affinis* apparently has no preference for hosts and may attack a wide range of plants, including herbs and lianas. The strong dominance of *X. affinis* can be explained by interspecific competition, considering that fast-growing species are more destructive and reduce the resources available for other plant species (Abreu *et al.*, 1997).

The Scolytinae species richness varied significantly among the study sites ($F_{4,415} = 223.1$, $p < 0.0001$). The average species richness was significantly higher in NV and Ag ($p < 0.01$) than in the other habitats. The second largest species richness was observed in MS ($p < 0.05$). The lowest species richness was found in Pa ($p < 0.05$) (Fig. 2a). The Scolytinae abundance also varied significantly among the study sites ($F_{4,415} = 123.8$, $p < 0.0001$). The mean abundance was significantly higher in Ag ($p < 0.01$) than in the other sites. The second largest mean abundance rate was observed in MS ($p < 0.05$) (Fig. 2b).

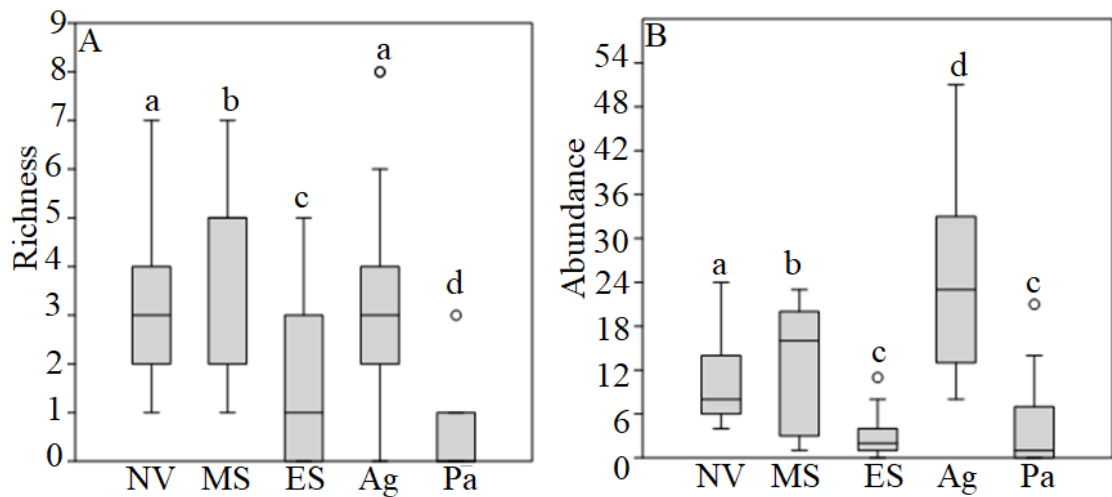


Figure 2. Box and Whisker plots expressing the differences in the projected values for (a) species richness (\pm SE) and (b) number of individuals (\pm SE) of Scolytinae in different habitats in the municipality of Uruará, state of Pará, northern Brazil. The values followed by the same letters are not significantly different according to Tukey test. Ag, Agriculture; ES, Early secondary succession; MS, Mature secondary succession; NV, Native vegetation; Pa, Pasture. Circles in the figures indicate the outliers.

According to Bray-Curtis similarity index and cluster analysis, a greater similarity was observed between NV and MS (0.77) (cophenetic correlation coefficient = 0.90) (Fig. 3).

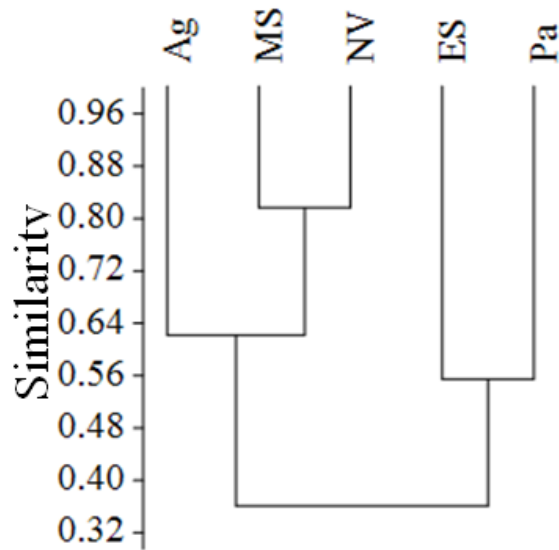


Figure 3. Bray-Curtis similarity index for the different habitats sampled (cophenetic correlation coefficient = 0.90) in the municipality of Uruará, state of Pará, northern Brazil. Ag, Agriculture; ES, Early secondary succession; MS, Mature secondary succession; NV, Native vegetation; Pa, Pasture.

Predictive variables were significant for Scolytinae abundance ($F_{6,413} = 84.5$, $R^2 = 0.55$, $p < 0.001$) and richness ($F_{6,413} = 158.9$, $R^2 = 0.70$, $p < 0.001$). The variable HLL best explained the abundance and richness of Scolytinae (Table 2).

Table 2. Multivariate regressions between environmental variables and ecological indices (abundance and richness) of Scolytinae collected in the municipality of Uruará, state of Pará, northern Brazil. H, Humidity; HLL, Height of Leaf Litter; P, Precipitation; PES, Percentage of Exposed Soil; PLC, Percentages of Leaf Litter Cover; T, Temperature.

Variables	Abundance				Richness			
	Coeff. (\pm SE)	<i>T</i>	<i>P</i>	R^2	Coeff. (\pm SE)	<i>T</i>	<i>p</i>	R^2
T	-0.718 (\pm 0.37)	-1.92	0.53	0.12	0.135 (\pm 0.04)	2.72	0.001	0.24
H	-0.050 (\pm 0.05)	-0.88	0.05	0.12	0.074 (\pm 0.00)	9.83	0.001	0.28
P	0.032 (\pm 0.00)	5.89	0.37	0.08	-0.002 (\pm 0.01)	-3.79	0.0001	0.04
HLL	6.750 (\pm 0.43)	15.35	0.001	0.35	0.776 (\pm 0.05)	13.28	0.000	0.44
PLC	-0.130 (\pm 0.02)	-5.39	0.001	0.17	0.020 (\pm 0.01)	6.40	0.001	0.41
PES	0.964 (\pm 0.22)	4.23	0.01	0.18	0.333 (\pm 0.03)	11.02	0.001	0.25

The fact that Ag had the highest abundance and species richness can be explained by the high HLL in these areas. On the other hand, ES and Pa are typically characterized by a relatively more open canopy with reduced occurrence of sub-forest warm and dry environment, as well as lower amounts of litter on the ground, which may explain the impoverishment observed in the Scolytinae communities. The influence of environments with greater litterfall rates on the distribution of the Scolytinae community can be confirmed by the high density of these individuals in Ag, MS, and NV. These habitats have greater amounts of material that can favor the development of vegetable debris, trunks, and broken branches, providing ideal conditions for the rapid population growth of Scolytinae species. The diversity of arthropod communities is related to the structural complexity of the habitat. More complex environments have a greater number of beetle species because of the greater supply of ecological niches for these organisms (Schaffers *et al.*, 2008).

With respect to the seasonality of Scolytinae, of the nine species sampled, six (approximately 67%) were available in the three periods surveyed. *Hypothenemus* sp. was collected only in the rain season, whereas *Xylosandrus* sp. and *Coccotrypes* sp. were exclusive to the intermediary season (Table 1). A statistically significant difference in abundance was noted among the sampling seasons ($F_{2,41} = 81.76$, $p < 0.001$), but no differences were noted between the averages of the rainy and intermediary seasons ($p > 0.05$). The dry season showed lower abundance, being statistically shorter than the other seasons ($p < 0.001$) (Fig. 4a). Species richness also showed differences between the sampling seasons ($F_{2,41} = 21.53$, $p < 0.001$). No differences were noted between the averages of the rainy and intermediary seasons ($p > 0.05$), but the dry season showed lower species richness, being statistically shorter than the other seasons ($p < 0.001$) (Fig. 4b).

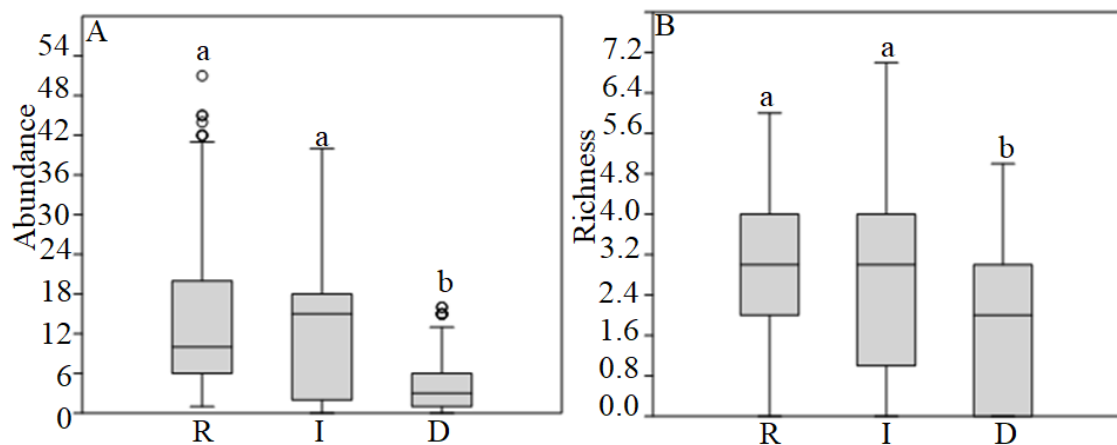


Figure 4. Scolytinae community in different periods of sampling in the municipality of Uruará, state of Pará, northern Brazil: (a) number of individuals (\pm SE) and (b) number of species (\pm SE). The values followed by the same letters are not significantly different according to Tukey test. D: Dry; I: Intermediary; R: Rain. Circles in the figures indicate outliers.

The abundance and richness of Scolytinae tended to show a seasonal distribution in a year, with less number of individuals and species occurring during the dry season than during the heavy rainfall season. This significant difference in the population of Scolytinae throughout the year was also observed by Flechtmann *et al.* (2001). According to Wood (1982), temperature and humidity are the two most important factors in the micro-climate of the galleries of Scolytinae species. Those belonging to the genus *Xyleborus*, in particular, reduced the growth of fungi they feed on. However, additional studies are needed to understand the extent of these changes, especially in light of the potential effects of climate change (Maveety *et al.*, 2014). Seasonal variations in diversity and composition emphasize the influence of phenology on survey timing in studying Scolytinae/habitat associations. Moreover, seasonal information on Scolytinae beetle is essential to understand the relevant ecological processes and, thus, the related management aspects. Asynchronous seasonal cycles may contribute to the reduction of interspecific competition among Scolytinae.

Our study should be complemented by ecological and behavioral studies to understand and assess the preservation status of the habitats considered (Brown, 1997). Ecological assessment and monitoring are important procedures to ensure effective

management of ecosystems and natural resources. The use of indicators of ecosystem integrity is considered crucial to measure and assess the status and trends of target environmental systems (Kandziora *et al.*, 2013).

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Chapter 9

How informative is the response of Ground Beetles' (Coleoptera: Carabidae) assemblages to anthropogenic land use changes? Insights for ecological status assessments from a case study in the Neotropics

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Abstract

Ground Beetles (Carabidae) have been suggested to be used as ecological indicators of biodiversity ecological status under the effects of anthropogenic land use and land cover changes in highly biodiverse regions such as the Neotropics. In this study Ground Beetles' assemblages from a region in the Brazilian Amazon were investigated for evaluating their applicability as disturbance indicators. Differences in assemblages between ecosystems, discriminated by complimentary methodologies, demonstrate a sensitive reaction from the most pristine forests to increasingly disturbed systems. Besides capturing the influence of different prevailing conditions between ecosystems, being easy to communicate and to link with the other components of the system, the Ground Beetles are easily and routinely measurable using standard methodologies. This study represents a step forward in using Ground Beetles for the purposes of planning, management and public reporting on the ecological status of Neotropical ecosystems. Additionally, the methods depicted could support projections for trends of relevant ecosystem attributes under realistic social-ecological change scenarios, which can be used to guide effective conservation planning.

Keywords: Biodiversity conservation; Land use changes; Brazilian Amazon; Landscape heterogeneity.

Introduction

Neotropical rainforests are considered biodiversity hotspots (Gardner, 2010), vital for carbon storage and regional/global regulation of the hydrological cycles and climate (Berenguer et al., 2015). However, anthropogenic activities have been causing massive loss of biodiversity in these systems (Kim et al., 2015), associated with new ecosystem processes and combinations of species/morphospecies that are ultimately linked with land use/land cover changes (LUCC) (Titeux et al., 2016). Biodiversity loss threatens the sustainability of regional ecosystems, with major implications in the socio-ecological services delivered (Cajaiba and Silva, 2015, 2017). Predicting the ecological consequences of LUCC is therefore subject to scientific and political studies aimed at supporting strategic options for landscape planning and natural resources management (Turner et al., 2007). In this context the selection of the most pertinent indicators for ecological assessments and monitoring is crucial, namely to detect and evaluate possible trends with relevance to guide conservation decision-making in the scope of disturbed ecosystems management (Cajaiba et al., 2017a).

Terrestrial invertebrates and especially insects play a critical role in most ecological processes and are key components of ecosystems' structure and functioning (Bicknell et al., 2014; Viegas et al., 2014; Campos and Hernández, 2015). Insects' diversity is intimately related with other taxa diversity and abiotic characteristics, thus representing potential ecological indicators of the ecosystem as a whole (e.g. Nichols et al., 2008). Therefore, understanding the ecological relevance of insects in the Neotropics could even support decision-making and robust management/recovery of imperilled ecosystems in the scope of the need for rapid, standardized and cost-saving

assessment methodologies (Godfray et al., 1999; Cajariba et al., 2017b). Ground beetles (GB) of the Carabidae family have been already suggested as promising ecological indicators in biodiversity studies and ecological integrity/status assessments, considering their morphological, behavioural, taxonomic composition, ecological traits, abundance and sensitivity to environmental changes (Koivula et al., 2002; Rainio and Niemelä, 2003; Koivula, 2011). GB represent an ecologically and taxonomically well-known group, which is straightforward to collect by using pitfall traps (e.g. Rainio and Niemelä, 2003; Niemelä et al., 2007; Koivula, 2011; Kotze et al., 2011). Furthermore, in forest ecosystems GB exhibit important trophic roles in the food webs, either as predators, decomposers or phytophagous (Qodri et al., 2016). Several GB are generalist predators (Nitzu et al., 2008), feeding on a variety of arthropod preys, such as Collembola, Acari, and larvae of Diptera (Ribera et al., 1999), providing valuable ecosystem services by controlling forest and even agriculture pests (Holland, 2002) and weeds (Bohan et al., 2011). The GB trends also reflect the consequences of ecosystem changes given their dependence on strict microhabitat conditions and specific diets (Koivula, 2002).

Diverse studies have used GB for indicating successional stages, of the degree of ecosystems fragmentation, urbanization pressures, metal contamination in soils, disturbance gradients, responses to environmental characteristics, regeneration in natural areas and ecosystem “health” (Fujita et al., 2008; Silva et al., 2008; Uehara-Prado et al., 2009; Moraes et al., 2013). Despite the recognized importance as ecological indicators, their application in the Neotropics, such as the Brazilian Amazon (Amazonia), has been extremely limited. As a consequence, the effects of human actions (e.g. deforestation, fire, animal grazing, logging and mining) on the GB

communities and their relationships with the structure and functioning of ecosystems are still poorly understood (Samu et al., 2010). Another problem is the replicability of studies' results since many areas in the region have low accessibility and several logistical and practical difficulties, which restrict the application of standardized methods of sampling and replication (Cajaiba et al., 2015a).

Our study examined the concealed patterns of GB assemblages through a gradient of ecosystem disturbance, including areas of primary forest, secondary forest, in different stages of recovery, cocoa plantations and pastures in a region of the Amazonia. The main objective was to analyse the dissimilarity of GB assemblages by testing the following hypotheses: (1) the GB community composition, using morphospecies, is sensitive to disturbance of reference ecosystems such as primary forest; (2) increasing disturbance can be assessed by an increasing dissimilarity of the selected ecological indicators from pristine forests, namely morphospecies richness, abundance, taxonomic diversity and ecological composition. These hypotheses were tested through representative gradients in order to provide new insights for the GB diversity standard assessments as indicators of disturbance in Neotropical ecosystems.

Material and Methods

Study sites

The study area was located in the municipality of Uruará, state of Pará, northern Brazil (-03°43'27" S - 53°44'8" W, Figure 1). Forest (69% of the area) is the dominant land use/ land cover (LULC) while deforestation is concentrated mainly in the south-central part of territory and near the main roads. Extensive livestock production and the exploitation of timber (mostly illegal) are currently considered the most serious environmental pressures (Cajaiba et al., 2015b). The studied areas contain the most

representative habitats of the region, in terms of biophysical and ecological characteristics for understanding the response of Ground Beetles (GB) assemblages to landscape disturbance, such as Native Vegetation (NV), Early Secondary succession (ES - secondary vegetation with five years of regeneration), Mature Secondary succession (MS - secondary vegetation with 15 years of regeneration), Agriculture (Ag - cocoa plantations, *Theobroma cacao* L.) and Pasture for extensive livestock (Pa). The climate is characterized as hot-humid (Köppen's classification), with annual average temperature and precipitation of 26 °C and 2000 mm respectively (Peel et al., 2007).

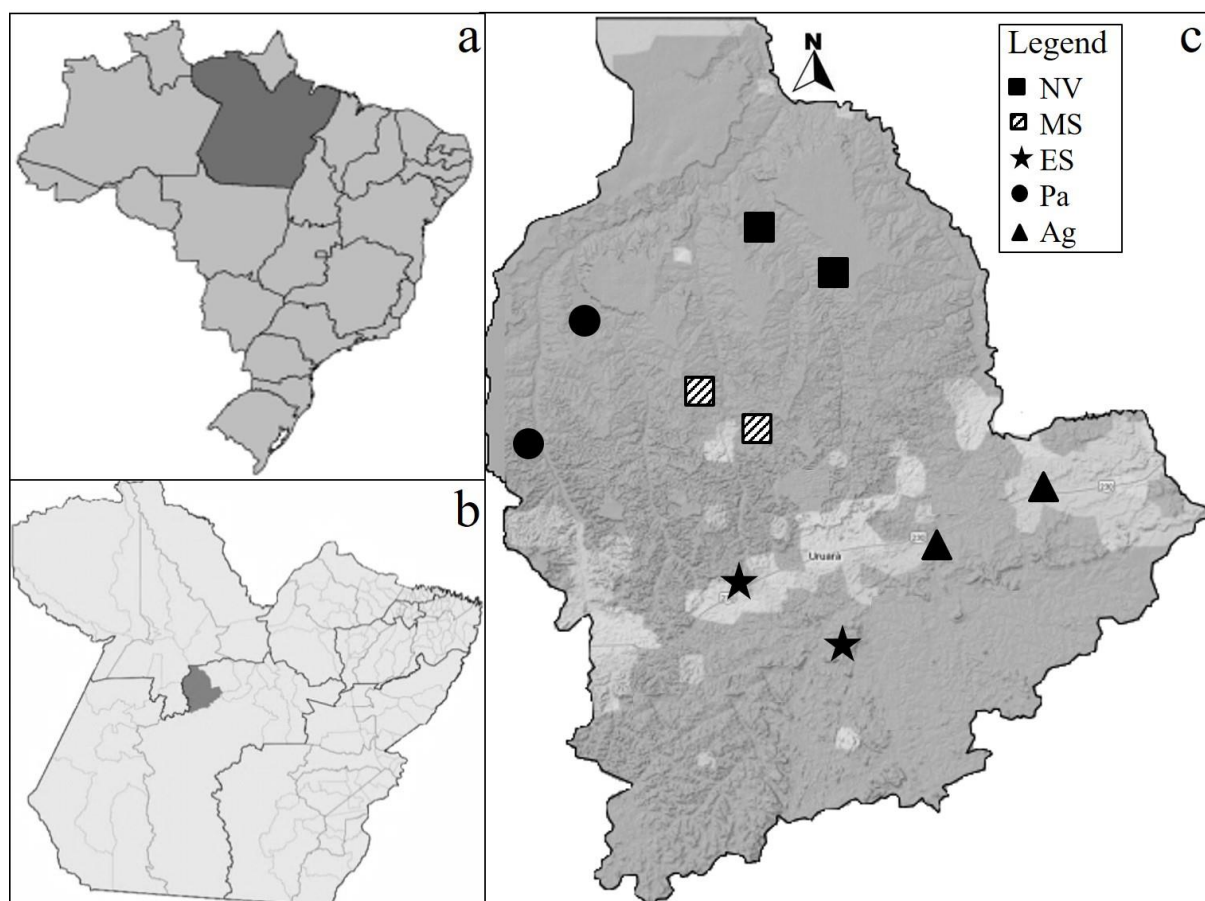


Fig. 1. Location of the study region, (a) Brazil, (b) Pará state, and (c) city of Uruará. Location of the sampling areas: NV, Native vegetation; MS, Mature secondary succession; ES, Early secondary succession; Pa, Pasture; Ag, Agriculture.

Carabidae sampling

Fieldworks were carried out during the year 2015 in the rainy season (February/ March), intermediate season (June) and dry season (September/ October), for inspecting eventual periodic differences in the activity and composition of GB. The selected locations were positioned at minimum distance of 100 meters from ecotones, guaranteeing that most GB captured were associated to the monitored ecosystem. GB were trapped using pitfalls with 75 mm diameter and 110 mm deep, filled with preservative liquid consisting of formalin, alcohol, water and a few drops of detergent to break the surface tension. A roof was attached to each pitfall trap in order to prevent the destructive effect of direct rainwater. In each ecosystem per season, fifty-six traps were placed (100 m from each other), remaining installed for 48 h prior to collection. This protocol was applied to all areas and periods of collection, totalizing a sampling effort of 840 traps (see Table S1, Supplementary material). Sampling survey period and intensity, although inadequate for definitive inventory, served the purpose of comparing GB general sensitivity to ongoing changes in the scope of their application as ecological indicators of disturbance (Dale and Beyler, 2001).

Environmental variables

Fourteen environmental variables, considered with potential influence on GB communities, were measured in the sampled ecosystems (Felton et al., 2006): temperature (T), humidity (H), precipitation (P), circumference at breast height (CBH), circumference at ankle height (CAH), canopy cover (CC), richness of plants (RP), abundance of plants (AP), richness of shrubs (RS), abundance of shrubs (AS), percentage of exposed soil (PES), percentage of green (vegetation) cover (GC), percentages of leaf litter cover (LLC), height of leaf litter (HLL). The air temperature,

relative humidity and rainfall of each point were measured during the traps' installation and removal by a portable weather station (model Oregon Scientific WMR200A). To assess the environmental complexity of each ecosystem, the quadrat-section method was adopted (Campos and Hernández, 2015). Using a cross as a reference, four quadrants (northeast, northwest, southeast, southwest) were marked and in each quadrant the following variables were measured: trees with circumference at breast height (CBH) greater than 15 cm, shrubs with CBH less than 15 cm and with height greater than 1 m were selected and the distances to the centre of the cross, height, crown diameter and trunk diameter. Trunk diameter was taken at breast height (1.3 m) for the trees and ankle height (CAH = 0.1 m) for the shrubs. In each quadrant, the height of leaf litter in 1 m × 1 m marked square (using PVC pipe) was measured with a ruler, and the percentages of leaf litter layer, green and exposed soil area (no vegetation or leaf litter) were measured by visual estimation using the following classes, 0–5%, 6–25%, 26–50%, 51–75%, 76–95% and 96–100% (Campos and Hernández, 2015). Using the same classes, the percentage of canopy cover in the four quadrats was calculated with a convex spherical Lemmon densiometer (D). Information concerning the methodology associated with each variable is depicted in Table S2, Supplementary Material.

Statistical analysis

Assemblage analysis

Estimated morphospecies/species (hereinafter designated by recognizable taxonomic units of ground beetles: RTU) richness was calculated for all sites together and for each individual site using the mean through the estimators Chao 1, Chao 2, Jackknife 1, Jackknife 2, and Bootstrap (and their confidence intervals at 95%) (Krell,

2004). The software EstimateS 9.0 was used for analyses (Colwell, 2013). The mean of these estimators was used in order to minimize any bias from any particular estimator, the performance of which often varies according to differences in richness, sampling effort, and community evenness (O'Hara, 2005).

RTU richness and abundance were determined and differences among ecosystems gauged using the Kruskal-Wallis test (a non-parametric equivalent to the multifactorial ANOVA), using the subsequent Dunn post hoc tests to check for specific differences (Sokal and Rohlf, 1995). The taxonomic composition of RTU communities between ecosystems was compared using Permutational Multivariate Analysis of Variance (PERMANOVA). Non-metric Multidimensional Scaling (NMDS) plots were used to help in the interpretation of the results found with the PERMANOVA (e.g. Anderson, 2001). These analyses were performed using the software Past 3.15 (Hammer et al., 2001).

Complementary, a beta diversity partition analysis was applied to verify the dissimilarity between ecosystems. Using the partitioning framework proposed by Baselga (2010), the pairwise dissimilarity index (β_{sor}) was partitioned into two components: turnover (β_{sim}) and nestedness (β_{nes}): $\beta_{sor} = \beta_{sim} + \beta_{nes}$. This method was applied in order to evaluate whether dissimilarities in the composition of the RTU communities occurred through the substitution of some species by others (β_{sim}) or by the formation of nested subsets of more diverse communities (β_{nes}). The β_{sor} index ranges from 0 (identical species assemblages) to 1 (different species assemblages). Using this approach with our dataset allowed testing (i) differences in the values of total dissimilarity (β_{sor}) between different sites studied, but also (ii) the relative contribution of species turnover (β_{sim}) and nestedness-resultant dissimilarity (β_{nes}) in

each site. Beta diversity analysis was performed using the functions *beta.pair* from *betapart* package (Baselga et al., 2017) within R 3.2.4 program (R Core Team, 2016).

Association of Ground Beetle assemblages with environmental variables

Canonical Correspondence Analysis (CCA) was used to estimate the influence of the fourteen environmental variables on RTU assemblages. Principal Components Analysis (PCA) was used to reduce variables to 4 principal components (PC). The CCA was performed with the Bray-Curtis dissimilarity index on two axes. These analyses were performed using the software Past 3.15 (Hammer et al., 2001).

Species indicator value – IndVal

To determine possible RTU indicators, single value indicator (IndVal) developed by Dufrière and Legendre (1997) was calculated, combining specificity (patterns of relative abundance) of a given RTU in a given ecosystem with its fidelity within that ecosystem (patterns of incidence). RTU with a high specificity and high fidelity within an ecosystem are considered to achieve the highest indicator value. Only taxa with $\text{IndVal} > 25\%$ were saved in the final lists (Dufrene and Legendre, 1997). This analysis was performed using R 3.2.4 program (R Core Team, 2016) using the *indicspecies* package 1.7.5 (De Cáceres and Jansen, 2015) with 9999 permutations, using RTU abundance.

Results

Assemblages analysis and composition of Ground Beetles

A total of 2378 ground beetles (RTU) were captured, distributed by 33 RTU. Overall, 29 RTU were identified within 859 specimens captured in Native Vegetation

(NV), 25 RTU were identified from the 414 specimens captured in the Mature Secondary (MS), 18 RTU were identified within 201 individuals captured in Early Secondary (ES), 26 RTU and 590 individuals were captured in Agriculture (Ag) and 13 RTU were identified within 314 individuals captured in Pasture (Pa). The most abundant GB RTU were: *Odontocheila* sp1 (185 individuals), *Galerita* sp1 (179 individuals), *Athrostictus* sp1 (155 individuals), *Pterostichini* sp1 (151 individuals) and *Pterostichus* sp1 (142 individuals), together representing approximately 35% of total abundance (Table S3, Supplementary material).

Considering all sampled sites together, the richness estimate Jackknife 1 and Bootstrap indicated a maximum of 35.40 and 34.58 species, respectively (Table S4a, Supplementary material). The average of these estimates and observed richness indicates that sampling efficiency was roughly 94.31% (Table S4b, Supplementary material).

Ground Beetles diversity and ecosystems

The average RTU richness revealed significant differences between ecosystems (Kruskal-Wallis nonparametric test, $KW = 251.8$, $df = 4$, $p < 0.001$). RTU richness was significantly higher in NV, followed by Ag, MS and finally by ES and Pa (Fig. 3a). RTU abundance had significant differences between the study ecosystems (Kruskal-Wallis nonparametric test, $KW = 220.2$, $df = 4$, $p < 0.001$). RTU abundance was significantly higher in NV, followed by MS and Ag (with equivalent medians), ES and lastly Pa (Fig. 3b).

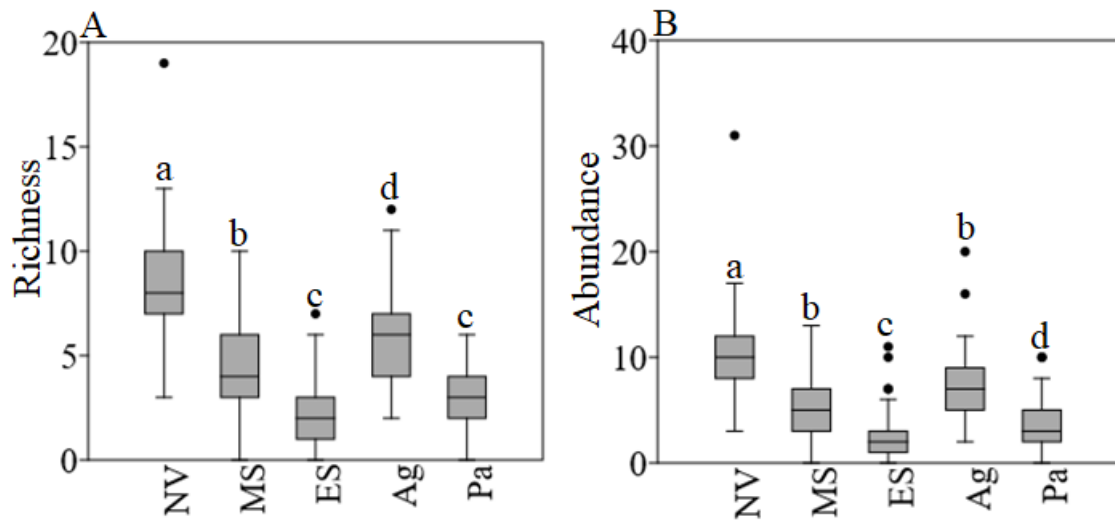


Fig. 3. Box and Whisker plots expressing the differences in Ground Beetles (A) recognizable taxonomic unit (RTU) richness and (B) RTU abundance for the different ecosystems. The values followed by different letters are significantly different according to the Dunn test. NV: Native vegetation; MS: Mature Secondary succession (vegetation with 15 years of regeneration); ES: Early Secondary succession (vegetation with five years of regeneration); Ag: Agriculture; Pa: Pasture.

The results of the NMDS showed that the GB assemblages of different ecosystems could be separated from each other by ordination of the RTU composition. The average composition of the GB samples of NV and MS was similar to each while RTU assemblages of Ag seem isolated from the other ecosystems. ES and Pa form a partially superimposed but divergent group from NV, MS and Ag. The NMDS results depict an arch in the diagram, suggesting that the RTU assemblages change gradually from the most pristine to more disturbed ecosystems (Fig. 4). The Permutational Multivariate Analysis of Variance (PERMANOVA) confirmed the previous results by

showing that RTU taxonomic composition among the ecosystems studied was significantly different from each other ($F_{4,415} = 17.43$, $p < 0.0001$).

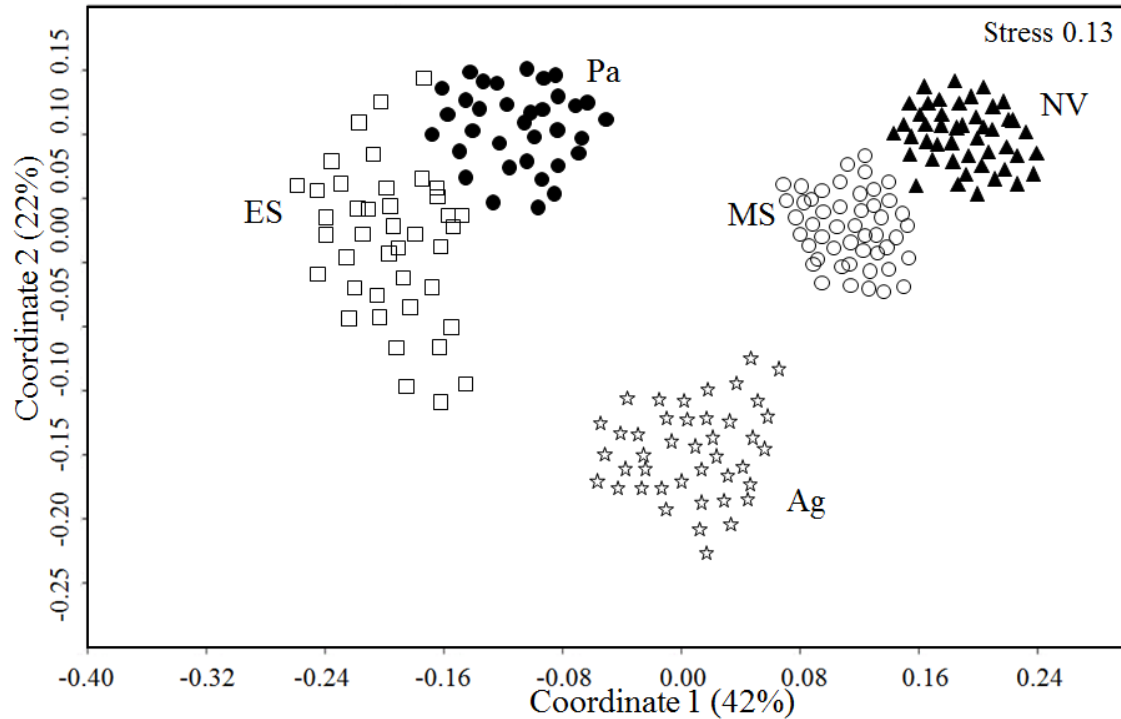


Fig. 4. Non-metric multidimensional scaling (NMDS) showing Ground beetles (GB) recognizable taxonomic units (RTU) grouped in accordance with the ecosystems (using Bray-Curtis similarity). NV, Native vegetation; MS, Mature Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture (Cocoa); Pa, Pasture.

The beta diversity (β_{sor}) presented relatively low values (β_{sor} maximum = 0.52) (Fig. 5, Table S5a, Supplementary material). The highest dissimilarity values (β_{sor}) were found when comparing Pa with NV ($\beta_{sor} = 0.52$), Pa with ES ($\beta_{sor} = 0.44$), Pa with Ag ($\beta_{sor} = 0.40$). The lowest β_{sor} was found when comparing NV with MS ($\beta_{sor} = 0.11$) and NV with Ag ($\beta_{sor} = 0.16$) (Fig. 5 and Table S5a, Supplementary material). The highest turnover values (β_{sim}) were found when comparing Es with Pa ($\beta_{sim} =$

0.36), and the lowest β_{sim} was found comparing NV with ES ($\beta_{sim} = 0$) (Fig. 5 and Table S5b, Supplementary material). The highest nestedness values (β_{nes}) were found when comparing NV with Pa ($\beta_{nes} = 0.39$) and the lowest nestedness was found when comparing MS with Ag ($\beta_{nes} = 0.02$) and MS with Pa ($\beta_{nes} = 0.07$) (Fig. 5 and Table S5b, Supplementary material).

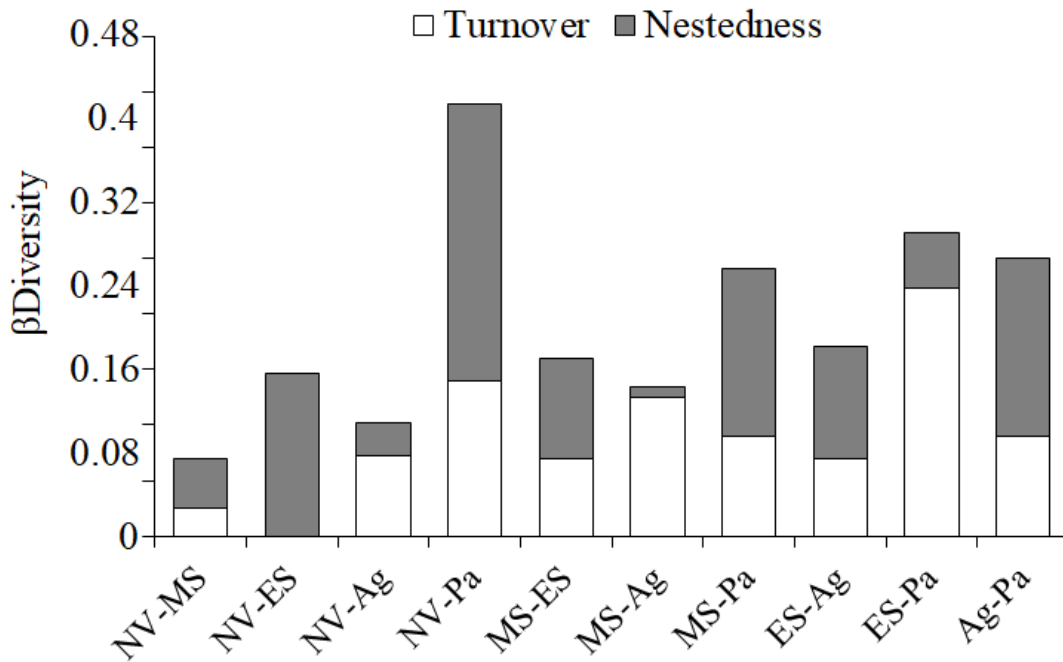


Fig. 5. Comparison of dissimilarity values for β_{sor} (overall dissimilarity), β_{sne} (dissimilarity resulting from nestedness), and β_{sim} (turnover) for the different ecosystems. NV, Native vegetation; MS, Maturing Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture (Cocoa); Pa, Pasture.

Effects of environmental variables on Ground beetle assemblages

The first two axes generated by CCA explained 83.71% (62.42% in axis 1 and 21.29% in axis 2) of the total variation in the RTU composition. Among the PC's,

environmental variables represented by PC1 ($R = -0.624$) were the most important factors affecting the first axis, and the Abundance of shrubs (AS) (represented by PC3) ($R = -0.809$) had a great correlation with the second axis (Fig. 6 and Table S6, Supplementary material). The RTU communities in Mature Secondary succession (MS) were mostly associated with the Abundance of shrubs (AS) (PC3); Pasture (Pa) was correlated with Humidity (H) (PC2) and Precipitation (P) (PC4); Agriculture (Ag) was correlated with H (PC2) and P (PC4), as well as environmental variables represented by PC1; Native Vegetation (NV) and Early Secondary succession (ES) were correlated with environmental variables represented by PC1. The distribution of RTU abundance according to the environmental variables seems to indicate that specific RTU respond to certain characteristics of the ecosystems. *Athrostictus* sp1, *Odontocheila* sp2 and *Selenophorus* sp1 occurred mostly in areas with bare soil. Conversely, *Odontocheila* sp1 occurred in areas with lower temperatures and *Carabus* sp1, *B. creptans* negatively in dry areas; *Carabus* sp3 and *Amara* sp1 were mainly associated with Leaf litter cover / Height of leaf litter; *Carabus* sp2 occurred in areas with higher Abundance of shrubs (Fig. 6 and Fig. 7).

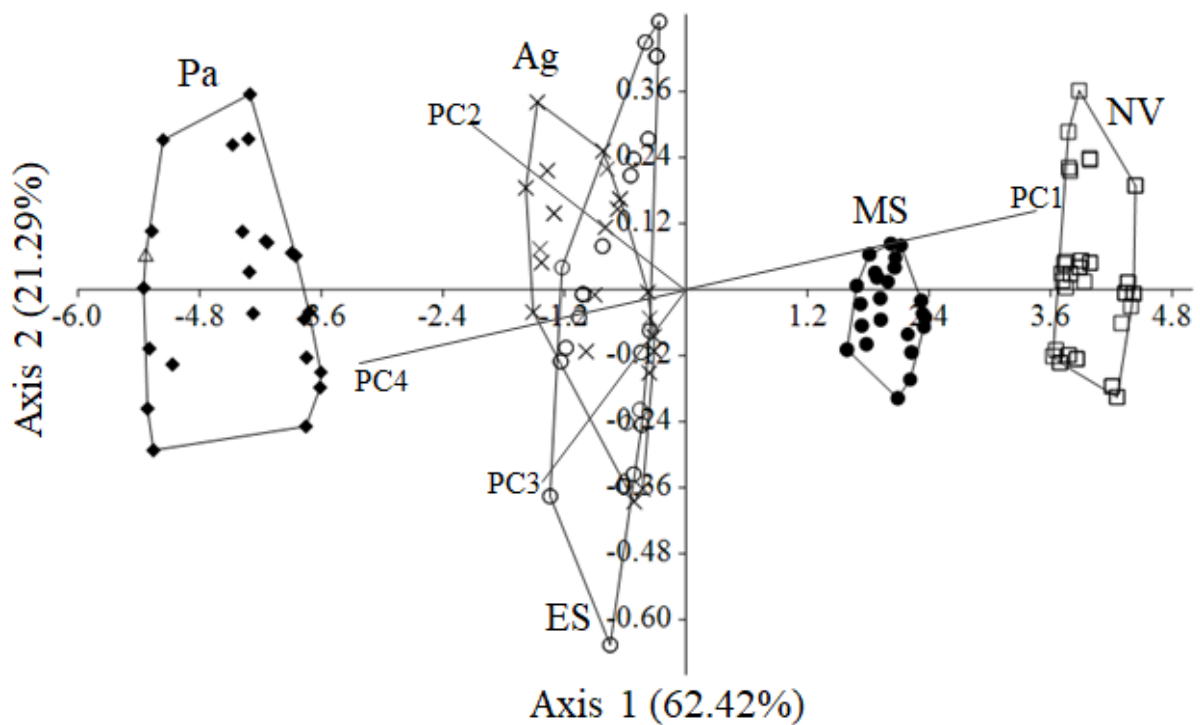


Fig. 6. Canonical Correspondence Analysis (CCA) used to estimate the influence of environmental variables on Ground Beetles communities. Four axes were considered in the Principal Components Analysis (PCA), including 4 principal components (PC). NV, Native vegetation; MS, Mature secondary succession (15 years of natural regeneration of vegetation); ES, Early secondary succession (five years of natural regeneration of vegetation); Ag, Agriculture (Cacao plantations); Pa, Pasture. PC1 was positive correlated with the following environmental variables: Circumference at Breast Height (CBH); Circumference at breast ankle (CBA); Canopy Cover (CC); richness of plants (RP); Height of Leaf Litter (HLL); Percentages of Leaf Litter Cover (PLC); Percentage of Green (vegetation) Cover (PGC); Abundance of Plants (AP); Richness of Shrubs (RS), and negative correlated with Temperature (T) and Percentage of Exposed Soil (PES). PC2 was positive correlated with Humidity (H), PC3 positive correlated with Abundance of Shrubs (AS) and PC4 with Precipitation (P), also positively.

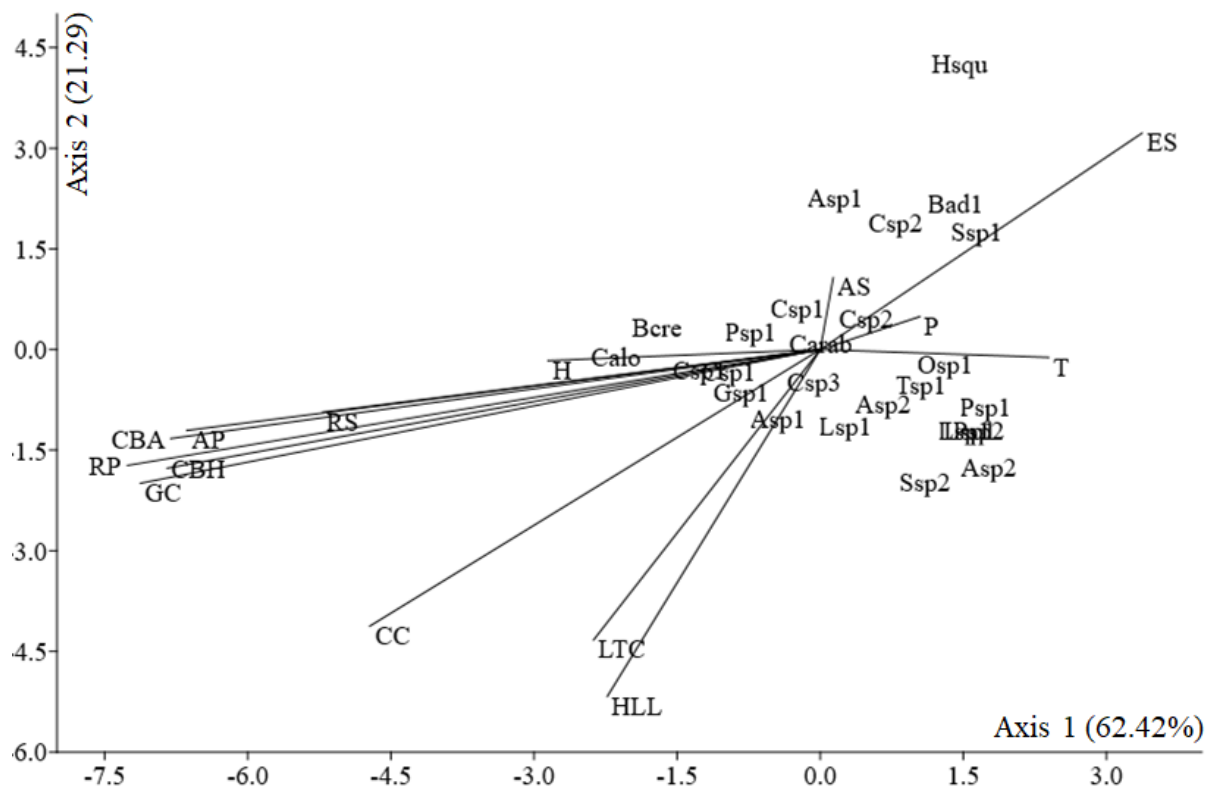


Fig. 7. CCA for Carabidae RTU using 14 environmental variables. For a description of the abbreviations of environmental parameters, see Table S2, and for abbreviations of GB RTU see Table S3, Supplementary material.

IndVal Method

Of the 33 RTU found in the present study, 12 RTU ($\approx 37\%$) were considered ecosystem indicators. According to IndVal, two RTU were significantly associated with NV, one with MS, six with Ag, and two with Pa. *Calosoma* sp1 which was an indicator of NV, was also associated with MS. No RTU was indicative for ES (Table 1).

Table 1 Recognizable taxonomic unit (RTU) selected by the IndVal method. NV, Native vegetation; MS, Mature Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture; Pa, Pasture (only taxa with IndVal>25% were considered).

GB RTU	Indicator value	P value	Habitat
<i>Calossoma</i> sp1	0.88	0.005	NV
<i>Brachynus creptans</i>	0.87	0.005	NV
<i>Pelecium</i> sp2	0.73	0.001	MS
<i>Calosoma</i> sp1	0.68	0.005	NV+MS
<i>Loxandrus</i> sp1	0.79	0.005	Ag
<i>Tetracha</i> sp1	0.68	0.005	Ag
<i>Athrostictus</i> sp2	0.67	0.01	Ag
<i>Pterostichini</i> sp2	0.63	0.02	Ag
<i>Selenophorus</i> sp2	0.59	0.01	Ag
<i>Amara</i> sp2	0.53	0.005	Ag
<i>Cymindis</i> sp2	0.76	0.005	Pa
<i>Odontocheila</i> sp2	0.63	0.005	Pa

Discussion

Neotropical Ground Beetles, ecosystems and environmental conditions

Assemblages analysis and composition of Ground Beetles

Our results demonstrate that GB diversity, represented by RTU richness and RTU abundance, was not indifferent to the ongoing structural changes in the study area, reaching higher values in the pristine ecosystems (NV - Native vegetation),

corroborating the results of Fujita et al. (2008). Although the RTU richness of cacao plantations (Ag) was considered high (even higher than MS - Mature secondary), RTU composition was quite dissimilar, as demonstrated by Permanova and NMDS. Additionally, the index $\beta_{s\text{or}}$ (global beta) depicted that Pa (Pasture) was the most “poor” and divergent within all ecosystems (Table S5a, Supplementary Material). Despite this, β_{sim} index (turnover) was only 22% of the NV RTU (our reference environment), i.e., circa 1/5 of species were replaced in Pa, showing that many of the RTU that inhabit NV can also be present in the other ecosystems. However, the β_{nes} (nestedness) value between NV and Pa was relatively high ($\beta_{\text{nes}} = 0.52$). Disturbed systems such as Pa and Ag may favour the presence of generalist species that tolerate the modification of their ecosystem and, thus, may allow the colonization of non-forest species from other adjacent ecosystems (Escobar, 2004). The conversion of the pristine ecosystems into Pastures (Pa) or monocultures (Ag) may lead to changes in abiotic factors, with probable detrimental effects on the structure and composition of the GB communities (Medri and Lopes, 2001). These changes seem also to decrease evenness in the assemblages, leading to the dominance of some RTU since the forest RTU are unable to adapt to the conditions of disturbed ecosystems (Davis et al., 2001; Liberal et al., 2011). More disturbed environments have reduced resources and microhabitats, supporting lower richness and are dominated by generalist and / or opportunistic RTU (e.g. Moraes et al., 2013).

Nonetheless, it should be noted that high diversity itself is insufficient in indicating "healthy" ecosystems, which should be complemented by RTU composition (Zou et al., 2015). In fact, many forest RTU are not able to occur in Cocoa agroforests, although the high diversity associated to these habitats (Schroth and Harvey, 2007).

Therefore, it is clear that relying on Cocoa agroforests for the conservation of biodiversity, in vast areas of Amazonia, would be ineffective and risky for the more vulnerable and specialized groups.

Effects of environmental variables on Ground beetle assemblages

Our results demonstrate that both RTU richness and abundance followed the trend expected for the disturbance gradient studied, i.e.: a decrease in the respective values as the disturbance level increase. Thus, different aspects of RTU diversity suggest that GB assemblages are distinct among ecosystem different land uses and soil conditions (Moraes et al., 2013), namely because: a) litter type and litter depth are important determinants, providing GB hunting and foraging niches, protection from predators, desiccation (Pearce et al., 2003) and providing attachment points for web building (Leclerc and Blandin, 1990); b) coarse woody debris is fundamental as overwintering site and for oviposition and larval development for many GB species (Lövei et al., 2006). For example, cattle trampling in grassland areas contributes to soil compaction, resulting in reduced pathways through the litter layer, hiding surfaces and hunting places. Additionally, reduced vegetation cover affects foraging or maintenance of water balances for GB species (Lövei et al., 2006).

It has also been suggested that the GB fauna may be affected not only by the shrub and herb cover (e.g. impeding its movement but protecting it from predation) (Brose, 2003; Taboada et al., 2008; Koivula et al., 2003) but also by plant diversity (i.e. increasing the availability of food resources) (Koricheva et al., 2000). Both the arboreal characteristics and the structural heterogeneity of the vegetation strongly influences the GB richness (Brose, 2003; Taboada et al., 2008), indicating the

importance of scattered trees within agricultural areas and late successional vegetation (Koivula et al., 2002; Magura et al., 2005). The GB richness observed in forest areas and/ or mature succession (NV/MS) could also be associated with the availability of niches and food resources. On the other hand, disturbed areas support mostly generalist and tolerant RTU (Kašák et al., 2017). In fact the distribution of GB along different environmental characteristics seems to demonstrate discrete associations with particular biotypes within the landscape (Silva and Hernández, 2016; Cajaiba et al., 2017a). Corroborating other studies, our results suggest that species richness and abundance (and also RTU richness, abundance and biomass) are negatively impacted by disturbed ecosystems (Taboada et al., 2008) especially those without tree canopy cover.

Neotropical Ground Beetles as ecological indicators of anthropogenic disturbances

The increasing human population in tropical areas implies that threats to biodiversity will be more vulnerable to ecosystem changes, which implies that effective sustainable management will be necessary to protect the tropical biodiversity against anthropogenic disturbances (Hulme, 2006). For this reason, effective sustainable management requires a detailed understanding of the complex relationships between disturbance levels and biodiversity response trends (Hulme, 2006). Thus, the use of GB metrics as ecological indicators to evaluate the effects of anthropogenic and natural disturbances in forests has been suggested as an important and reliable tool for defining sustainable forest management (Lindenmayer et al. 2000; Work et al., 2008). In fact, policy objectives have emphasized the use of species that are vulnerable or in other ways sensitive to changing forest conditions as ecological indicators (Work et al.,

2008) and the use of GB as promising ecological indicators has been supported by several studies (Niemelä et al., 2000; Larsen et al., 2003; Rainio and Niemelä, 2003; Silva et al., 2008; Work et al. 2008; Riley and Browne, 2011).

From our results, the IndVal signals indicator RTU for pristine and/or altered ecosystems was sensitive to the associated changes and might help the management decision-making in the scope of Neotropical systems affected by anthropogenic disturbances. In this study *Selenophorus* sp2 stands as good surrogate of disturbed environments, namely in Agriculture (Ag), being positively related to the number of shrub, while *Cymindis* sp2 and *Odontocheila* sp2 were good indicators for pasture areas (Pa), being positively related to the variables temperature and bare soil, respectively. On the other hand, *B. creptans* are indicators of native vegetation (NV) and *Calossoma* sp1 was also associated with NV + MS (Mature secondary) correlated negatively with dry areas, as identified By PCA / CCA. This relatively high number of indicator species (see Table 1) supports the applicability of GB metrics as ecological indicators for the use and management of forests for the region under study, indicating that several forest-dependent species disappear if the forests were completely transformed into an agricultural landscape (e.g. Pa or Ag).

Perspectives and challenges on using Neotropical Ground Beetles metrics for environmental management of forest landscapes

The conservation of biological diversity has become one of the important goals of managing forests in an ecologically sustainable way (Cajaiba et al, 2017b). Ecologists and forest resource managers need measures to judge the success or failure of management regimes designed to mitigate the biological diversity losses (Lindenmayer et al., 2000). Therefore, the use of ecological indicators for the

monitoring of environmental change/recovery becomes crucial. The use of GB represent several other environmental prevailing characteristics that have justified their relevance as ecological indicators, namely because they are influenced by temperature, moisture and shade (Thiele, 1977), food quality and abundance (Bilde et al., 2000; Bohan et al., 2001), habitat structure as reflected by the vegetation cover (Brose, 2003; Taboada et al., 2008), and substrate salts, sugars and acidity (Merivee et al., 2006; Milius et al., 2006) (see complete review in Koivula, 2011). Such studies demonstrated that, despite the different GB species pool of particular regions, the general patterns of their functional responses to anthropogenic disturbances were very similar (Niemelä et al., 2000). This suggested the possibility of using GB universally to monitor changes in terrestrial habitats (Eyre et al., 1990).

Three factors not mutually exclusive, associated with the alteration of the pristine ecosystems of the Amazon rainforest, may be responsible for different trends in diversity of RTU observed in the studied ecosystems (Cajaiba et al., 2017a): 1) the ability of some RTU in colonizing disturbed areas; 2) distinct ecological and ecophysiological tolerances of the RTU in relation to environmental conditions; and 3) dependence of some RTU on specific conditions and resources for reproduction. Thus, an important and urgent task is to find the GB diversity hotspots in in the Amazon forest, to ensure their conservation and to manage the surroundings in the most favourable way in order to allow their populations to survive and to spread (Warnaffe and Lebrun, 2004). Despite the proved ecological relevance of GB, problems in species identification and the lack of systematic natural history studies might originate constraints for their practical use as surrogates in routine ecological studies (Freitas et al., 2006). However, in our study most of the RTU were only identified until the level

of morphospecies (Maveety et al., 2011). We realize that this is a simplified, preliminary and demonstrative approach although we believe that RTU classification represents a reasonable trade-off between absolute taxonomic accuracy (which may take many years to achieve) and the ecological functional representativeness, necessary to ensure the applicability of GB diversity estimates for comparative analysis between different ecosystems (Maveety et al., 2014). In fact taxonomic surrogacy approaches have been developed to meet the short-term needs of providing scientific advice for resource managers and policy makers, reducing the time, costs and dependence on specialist taxonomists (Krell, 2004). Our work should be complemented by ecological and behavioural studies to understand and assess the ecological status and conservation priorities of the ecosystems considered (Brown, 1997). Anyway, the responses to anthropogenic disturbances found with non-specific sampling methods are applicable per se, and should be improved in further studies with standardized methods for this group (Uehara-Prado et al., 2009). Since for hyperdiverse groups, such as Carabidae, it is difficult to obtain the complete inventory of the species that inhabit in a given area, ecological assessment and monitoring based on indicators at the community level are important procedures to address effective management of ecosystems and natural resources. Therefore, the use of holistic ecosystem indicators is considered crucial to measure and evaluate the ecological status and trends of target components and environmental conditions (Kandziora et al., 2013; Cajaiba et al., 2017a).

Conclusions

Our approach represents a useful contribution to understand the relevance of key GB responses through a representative gradient of the structural and functional LUCC

dynamics under very complex and variable regional conditions. In fact, the present results indicate that pristine ecosystems are fundamental for conservation purposes, holding higher diversity and exclusive GB whose response was proved to be a reliable surrogate of other taxa as shown by Desender et al. (1999). Therefore, GB used as ecological indicators could gauge conservation actions, considering its ability to represent the overall community (Koivula et al., 2002; Niemelä et al., 2007). Nevertheless, since ecological integrity of the studied ecosystems can be only partly assessed by GB indicators, our results should be complemented with the information from other indicators, interactions and interferences (such as the specific agricultural practices disturbance) with precise applicability conditions. Despite the limitations inherent to a preliminary demonstration, the methodology proposed is applicable to other type of ecosystems affected by gradients of changes. In this perspective, this study seems to represent a useful contribution for the holistic understanding of the GB community role as surrogate of other taxa responses in this region. Moreover, this approach also provides a useful starting point, allowing the precise development of more instructive protocols for environmental managers and decision-makers, based on the potential added-value of our combined metrics approach, namely in order (1) to anticipate the impacts induced by anthropogenic pressures that will characterize most of this region in the future, and (2) to provide guidance for pertinent forest restoration strategies until the effects of diverse disturbances and regime shifts in the Amazonia are reasonably understood (Cobb et al., 2007; Cajas et al., 2017b).

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Supplementary material

Table S1. Ecosystems surveyed, number of sampled areas, number of traps per sampled area, sampling repetitions and the total number of traps by ecosystem. NV = Native Vegetation; MS = Mature secondary succession (15 years of regeneration); ES = Early secondary succession (5 years of regeneration); Ag = Agriculture (Cocoa); Pa = Pasture.

Ecosystem sampled	N° sampled areas	Number of pitfalls by sampled area	Repetitions (periods of the year)	Total traps / ecosystem
NV	2	28	3	168 (2 x 28 x 3)
MS	2	28	3	168 (2 x 28 x 3)
ES	2	28	3	168 (2 x 28 x 3)
Ag	2	28	3	168 (2 x 28 x 3)
Pa	2	28	3	168 (2 x 28 x 3)
Total number of pitfalls				840 (168 x 5)

Table S2. Specification of the methodology used for monitoring the environmental variables.

Code	Variables	Specification	Monitoring Methodology
T	Temperature	Celsius (°C)	Measured during the traps installation and removal with a portable weather station (model Oregon Scientific WMR200A).
H	Humidity	Humidity (%)	Measured during the traps installation and removal with a portable weather station (model Oregon Scientific WMR200A).
P	Precipitation	Precipitation (mm)	Measured during the traps installation and removal with a portable weather station (model Oregon Scientific WMR200A).
CBH	Circumference at Breast Height	Centimeters (cm)	Trunk diameter was taken at breast height (1.3 m) for the trees.
CAH	Circumference at Ankle Height	Centimeters (cm)	The diameter was measured at the ankle height (CAH = 0.1 m) for the shrubs.
CC	Canopy Cover	Percentage (%)	Calculated with a convex spherical densiometer (D) Lemmon and assigned the following classes: 0–5%, 6–25%, 26–50%, 51–75%, 76–95% and 96–100%
RP	Richness of Plant Species	Number of Plant Species/ m ²	The number of tree species was counted in an area of 100 m ² (10 x 10 m) in the vicinity of each pitfall trap.
AP	Abundance of Plants	Number of Plants/ m ²	The number of tree was counted in an area of 100 m ² (10 x 10 m) in the vicinity of each pitfall trap.
RS	Richness of Shrubs	Number of Species of Shrubs/ m ²	The number of shrubs species was counted in an area of 100 m ² (10 x 10 m) in the vicinity of each pitfall trap.
AS	Abundance of Shrubs	Number of Shrubs/ m ²	The number of shrubs was counted in an area of 100 m ² (10 x 10 m) in the vicinity of each pitfall trap.
PES	Percentage of Exposed Soil	Percentage (%) / m ²	The percentage exposed soil in each quadrant was estimated in different percentage classes (0-5, 6-25, 26-50, 51-75, 76-95, 96-100%)
PGC	Percentage of Green Cover (vegetation)	Percentage (%) / m ²	The percentage green cover (vegetation up to 1 m height) in each quadrant was estimated in different percentage classes (0-5, 6-25, 26-50, 51-75, 76-95, 96-100%)
PLC	Percentages of Leaf Litter Cover	Percentage (%)	The percentage of litter in each quadrant was estimated in different percentage classes (0-5, 6-25, 26-50, 51-75, 76-95, 96-100%).
HLL	Height of Leaf Litter	Centimeters (cm)	Litter height was measured using a ruler at five points inside the square (near each corner and in the center)

Table S3. Abundance of the different recognizable taxonomic unit (RTU) of Carabidae in the ecosystems monitored in the Brazilian Amazon. NV = Native Vegetation; MS = Mature secondary succession (15 years of regeneration); ES = Early secondary succession (5 years of regeneration); Ag = Agriculture (Cocoa); Pa = Pasture.

Code	RTU	NV	MS	ES	Ag	Pa	Total
Psp1	<i>Peleciium</i> sp1	65	14	0	24	32	130
Psp2	<i>Peleciium</i> sp2	5	37	0	0	0	42
Asp1	<i>Athrostictus</i> sp1	34	7	24	0	90	155
Asp2	<i>Athrostictus</i> sp2	7	0	0	30	0	37
Csp1	<i>Cymindis</i> sp1	38	4	14	4	12	72
Csp1	<i>Cymindis</i> sp2	0	0	0	4	38	42
Psp1	<i>Pterostichus</i> sp1	64	30	0	24	24	142
Psp2	<i>Pterostichus</i> sp2	28	11	3	12	19	73
Bcre	<i>Brachynus creptans</i>	42	5	0	0	0	47
Asp1	<i>Amara</i> sp1	39	19	2	21	0	81
Asp2	<i>Amara</i> sp2	4	0	6	23	0	33
Lsp1	<i>Laemostenus</i> sp1	4	14	0	13	0	31
Lsp2	<i>Laemostenus</i> sp2	16	20	3	5	0	44
Ssp1	<i>Selenophorus</i> sp1	42	22	12	9	0	85
Ssp2	<i>Selenophorus</i> sp2	4	0	0	31	0	35
Ssp3	<i>Selenophorus</i> sp3	32	20	3	15	0	70
Hsqu	<i>Helluomorphoides squiresi</i>	10	0	12	3	0	25
Lsp1	<i>Lebiini</i> sp1	17	7	0	21	0	45
Lsp2	<i>Lebiini</i> sp2	13	5	1	3	0	22
Pts1	<i>Pterostichini</i> sp1	35	1	35	63	17	151
Pts2	<i>Pterostichini</i> sp2	0	13	0	39	0	52
Pts3	<i>Pterostichini</i> sp3	24	22	4	30	0	80
Los1	<i>Loxandrus</i> sp1	0	0	0	39	0	39
Gsp1	<i>Galerita</i> sp1	76	42	13	47	1	179
Osp1	<i>Odontocheila</i> sp1	31	28	35	70	21	185
Osp2	<i>Odontocheila</i> sp2	0	0	0	0	42	42
Tsp1	<i>Tetracha</i> sp1	14	5	0	35	8	62
Tsp2	<i>Tetracha</i> sp2	9	8	3	3	2	25
Cas1	<i>Calosoma</i> sp1	56	16	0	0	0	72
Cas2	<i>Calossoma</i> sp2	48	36	10	2	0	54
Crs1	<i>Carabus</i> sp1	51	27	10	8	0	96
Crs2	<i>Carabus</i> sp2	0	21	13	20	8	28
Crs3	<i>Carabus</i> sp3	54	0	0	0	0	102
Abundance (N)		859	414	201	590	314	-
Richness (S)		29	25	18	26	13	-

Table S4a. Average estimates of richness of Ground beetles (GB) recognizable taxonomic units (RTU).

Estimators	Number of species	Standard deviation
Observed richness	33	±0.71
Bootstrap	34.58	±0.00
Chao 1	33	±0.00
Chao 2	33.51	±0.95
Jackknife 1	35.40	±0.98
Jackknife 2	34.05	±0.00
ACE	33	±0.00
ICE	34.2	±0.00

Table S4b. Observed and estimated (with standard deviation) richness of RTU for the sites sampled. NV, Native vegetation; MS, Mature Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with 5 years of regeneration); Ag, Agriculture; Pa, Pasture.

Estimators	Sites				
	NV	MS	ES	Ag	Pa
Observed richness	29	25	18	26	13
Bootstrap	30.25±0.00	26.81±0.00	21.14±00	27.85±0.00	15.00±0.00
Chao 1	29.00±0.24	25.66±1.07	18.00±0.24	26.00±0.53	14.00±0.16
Chao 2	30.33±2.03	26.01±0.11	23.00±4.42	26.60±0.98	15.50±2.95
Jackknife 1	31.66±0.66	28.33±0.66	24.66±3.71	28.33±2.40	18.00±1.15
Jackknife 2	31.85±0.00	28.50±0.00	26.16±00	27.31±00	18.05±0.00
ACE	29.00±0.00	25.00±0.00	18.42±0.00	26.00±0.00	14.75±0.00
ICE	31.00±0.00	28.01±0.00	24.15±0.00	28.15±0.00	17.57±0.00

Table S5a. β -diversity (total beta, β_{sor}) comparing the composition of RTU between the studied habitats. NV, Native vegetation; MS, Maturing Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture (Cocoa); Pa, Pasture.

	NV	MS	ES	Ag
MS	0.11	-	-	-
ES	0.21	0.26	-	-
Ag	0.16	0.22	0.27	-
Pa	0.52	0.38	0.44	0.40

Table S5b. The turnover (β_{sim}) and nestedness (β_{nes}) components for each ecosystem pairwise comparisons. The (β_{sim}) results are depicted in the dashed part of the table and (β_{nes}) results are depicted without highlight. NV, Native vegetation; MS, Maturing Secondary succession (vegetation with 15 years of regeneration); ES, Early Secondary succession (vegetation with five years of regeneration); Ag, Agriculture (Cocoa); Pa, Pasture.

	NV	MS	ES	Ag	Pa
NV	---	0.04	0.00	0.12	0.22
MS	0.07	---	0.11	0.20	0.14
ES	0.21	0.14	---	0.11	0.36
Ag	0.05	0.02	0.16	--	0.14
Pa	0.39	0.24	0.08	0.26	---

Table S6. Eigenvalues of the Principal Components Analysis (PCA) for the first four axes. T. Temperature; H. Humidity; P. Precipitation; CBH. Circumference at Breast Height; CAH. Circumference at Ankle Height; CC. Canopy Cover-CC; RP. Richness of Plants; AP. Abundance of Plants; RS. Richness of Shrubs; AS. Abundance of Shrubs; PES. Percentage of Exposed Soil; PGC. Percentage of Green (vegetation) Cover; PLC. Percentages of Leaf Litter Cover; HLL. Height of Leaf Litter.

Variables	Components			
	PC 1	PC 2	PC 3	PC 4
T	-0.0063	-0.0126	-0.0174	-0.0056
H	0.0486	0.1046	0.0561	-0.0435
P	0.0168	0.9938	-0.0099	0.0043
CBH	0.7763	-0.0193	-0.4947	0.3663
CAH	0.0147	-0.0005	-0.0029	0.0008
CC	0.1730	-0.0030	0.4348	0.3564
RP	0.5401	-0.0148	0.3173	-0.6257
HLL	0.0066	-0.0014	0.0198	0.0402
PLC	0.1178	0.0014	0.5961	-0.5072
PGC	0.1256	0.0223	0.0464	-0.1284
PES	-0.0162	0.0036	-0.0331	-0.0835
NP	0.2061	-0.0061	0.2776	-0.2493
AS	0.0025	0.0010	0.1593	-0.0235
RS	0.0139	-0.0002	0.0204	0.0073

Chapter 10

Synthesis and recommendations

The main objective of this thesis was to demonstrate the applicability of Coleoptera, namely the use of Scarabaeidae, Carabidae, Staphylinidae, Histeridae and Scolytinae (Curculionidae) families in ecological monitoring studies to evaluate how anthropogenic modification on neotropical forest ecosystems influences the composition, abundance, richness and diversity of these beetles, supporting their use as possible environmental indicators. In general, the chapters demonstrate that Coleoptera can be used to successfully predict the environmental condition of the dominant habitats/ecosystems in the studied region. Therefore, they are considered a step-forward in supporting decision-making and possibly robust ecosystem management/recovery in the context of the need for quick, standardized, and cost-effective assessment methodologies (LANGE et al., 2014; NAGY et al., 2016; BEIROZ et al., 2017; FRANÇA et al., 2018).

This last chapter provides a interplay of the main results obtained and groundbreaking associated, not addressing each chapter separately. It also includes recommendations for future studies and gaps to be filled regarding how diversity in communities of beetles can respond to changes in anthropogenic disturbed ecosystems.

General discussion and key findings

Simplification of neotropical forest habitats directly or indirectly results in biodiversity loss and reduction (BRAGA et al., 2015; SÁNCHEZ-DE-JESÚS et al., 2016) due to the change in natural ecological processes and the emergence of new combinations of species, particularly through the transformation, fragmentation and destruction of habitats (ALVARADO et al., 2018). The results of this thesis corroborate the findings of several other studies that demonstrated that the composition and structure of communities of beetles are highly modified in areas changed by human action, including habitat fragmentation and isolation, burning, deforestation, selective logging, pesticides, among others (see associated references in each chapter).

The main outcomes by Coleoptera group are highlighted in the following paragraphs:

i. Scarabaeidae beetles (chapters 3, 4 and 5) - Our results showed that: (i) habitats differ in terms of species richness, taxonomic and ecological composition, with each habitat supporting exclusive indicator species; (ii) disturbed habitats (i.e., replacement of native forests by pasture or agriculture) are able to retain a small proportion of species associated with native forests or secondary forests; (iii) specialist forest species withdraw after depletion of pristine habitat (Chapter 3); (iv) conserved systems such as primary forest and late secondary vegetation, maintain a greater proportion of tunnelers, rollers and dwellers, the latter being absent in a pasture environments; (v) coprophages and necrophages were more abundant in more pristine environments (Chapter 4); (vi) the rainy season showed greater abundance and richness. However, several species were found exclusively in each of the three sampling periods (rainy season, intermediate and dry season) were encountered (chapter 5).

ii. Histeridae beetles (Chapter 6): The following results were obtained with the use of *Histeridae* beetles as the model of a biological study: (i) less disturbed habitats showed higher values of abundance and richness; (ii) the composition of communities was significantly different between pristine (e.g. primary forest and secondary vegetation) and anthropogenic habitats (agriculture and pasture); (iii) the rainy season showed greater abundance and richness, but each season exclusive species were monitored.

iii. Staphylinidae beetles (Chapter 7): (i) Primary forests were significantly richer and diverse when compared with the other habitats. However, there was no difference between primary forest and agriculture in terms of abundance. On the other hand, pasture presented the highest species dominance; (ii) the composition of the species changed among the different habitats, with higher dissimilarity between more pristine and more anthropogenic environments.

iv. Scolytinae (Curculionidae) beetles (chapter 8): (i) the greatest abundance of Scolytinae was found in environments with greater production of litter (cocoa plantations), and there was no difference in richness between primary forest and cocoa; (ii) there was greater abundance and richness of species in wetter seasons.

v. *Carabidae* beetles (Chapter 9): Like depicted in the previous groups, (i) primary forest attain the highest values of diversity and richness; (ii) beta diversity partitioning methods showed greater dissimilarity between primary forest and pasture.

As demonstrated in the previous topics, our results suggest a decrease in species richness and abundance of beetles' communities throughout gradients of increased anthropogenic disturbance of the native tropical forests in Amazonia. These anthropogenic disturbances may alter the composition of communities, affecting ecosystem services provided such as the decrease in the decomposition rates of organic materials through burial and removal, desequilibrium in the food webs by the removal of significant species associated with predation, herbivory and seed dispersal (ARELLANO, 2016; QODRI et al., 2016).

Collection methodologies described here provide an approach to improve the detection and description of the responses of beetles to disturbance and anthropogenic pressures, and facilitate a greater integration of ecological data collection efforts in tropical regions. Although spatially and temporally restricted, the results of this study highlight the attractiveness of several baits to different groups/ species of coleoptera (chapter 4). Although the human faeces baits presented greater abundance and richness (chapter 4) we suggest the use of several baits in future works. Traps with bait tend to be selective: if the aim of a specific work is an assessment of the overall community and estimate relative abundances, different baits should be used (RAFAEL, 2002); if the goal of the research is collecting specific species and/or groups (e.g. trophic/functional guilds), specific baits should be used (MARCHIORI, 2016).

An important outcome of this thesis was understanding the seasonality of species and communities associated (chapters 5, 6 and 9). Understanding seasonal/temporal patterns of ground-occurring beetles is of interest for several reasons. First, seasonal information can be used to determine optimal sampling periods when studying biodiversity and habitat associations. Second, seasonal information can be used to evaluate potential impacts of non-native fauna (WERNER; RAFFA, 2003; SHAKIR; AHMED, 2015; HILL et al., 2016), which can displace native congeners. Such displacement may be less likely where temporal separation exists (WERNER;

RAFFA, 2003; WANG et al., 2014). Finally, knowing the seasonal / temporal patterns of beetles is fundamental for support decision-making and robust management/recovery of imperilled ecosystems. Seasonality is an important component of ecosystems that should be taken into consideration in most biodiversity assessments. If the goal of a biodiversity study is to perform a complete inventory of a specific group, decisions concerning where (e.g. altitudinal gradient, location) and how (e.g. collection methods) to optimize the number of species sampled, but also when (e.g. time of year) collections should take place are quite relevant (MAVEETY et al., 2014).

Challenges and limitations

The realization of faunistic studies in diverse ecosystems such as the Amazonia is extremely challenging. Additionally the logistics associated with remote and vast areas monitoring is also complex and costly. Nevertheless and specifically for this region, several problems should also be considered:

i. Taxonomic challenges: Despite the proved ecological relevance of beetles, problems in species identification and the lack of systematic studies / natural history might originate unsuitability for its use as surrogate /indicator in routine ecological studies (FREITAS et al., 2006; and see also chapter two of this thesis). The lack of taxonomists and even taxonomic keys to identify the specimens collected is a common problem in this region (and in most neotropics/tropics) and, for several families even to get to the genera level (e.g. Tenebrionidae, Leiodidae, Chelovidae, Nitidulidae, ...). In our study most of the specimens were identified until the level of morphospecies. We realize this is a simplified, preliminary approach although we believe that morphospecies classification represents a reasonable trade-off between absolute taxonomic accuracy (which may take many years to achieve) and comparative estimates of beetles diversity for different ecosystems (MAVEETY et al., 2014). In fact taxonomic surrogacy approaches have been developed to meet the short-term needs of providing scientific advice for resource managers and policy makers, reducing the time, costs and dependence on specialist taxonomists (KRELL, 2004; COTES et al., 2009; HACKMAN et al., 2017).

ii. Challenges of sampling protocols for data comparison: Several studies with invertebrates have methodological limitations that may interfere with the interpretation of results (GARDNER et al., 2007a) and such problems are increasingly recognized in biodiversity studies (HAMER; HILL, 2000; HILL; HAMER, 2004; BARLOW et al., 2007a, b, c; GARDNER et al., 2007b; KOH, 2007; GARDNER et al., 2008). Few experiments carried out in the Amazon region sampled soil beetles using different types of traps at different times of the year. The methodological differences and the absence of scientifically tested and feasible general protocols may simplify complex realities, leading to erroneous results and eventually to erroneous decisions (SILVEIRA et al., 2010). We stress the importance of adequate inventories, suggesting broader discussion of general protocols and standardization of methods, taking into account the particularities of each group. However, more consistent studies are needed to understand the relationship between the beetles and the environment, in order to take more advantage of their use as ecological indicators, applied to environmental monitoring and assessment.

Priorities for future work and recommendations

In addition to providing greater knowledge of the responses of the beetles to environmental changes, this thesis also sheds light on several areas to be addressed in future studies contributing to use coleoptera as ecological indicators of disturbance. If this link is clarified, predictions and efforts to mitigate the impact of anthropogenic changes in natural systems will be more assertive. Among others, we consider some priority studies to be undertaken:

i. Expansion of sampling areas: Sampling should also be performed in other parts of the Amazonia (and Amazon). This may support more general conclusions, applicable to larger spatial scales and other neotropical ecosystems/habitat and landscapes.

ii. Elaboration of an identification key for the species that occur in the Amazon biome: Although we are aware of the difficulty of this task because of the vast territorial extension of the area and the megadiversity of the region, partnerships within researchers expertise in specific taxa and protocols of cooperation between partner

institutions (universities, research institutes and development agencies) should a priority in future projects and studies.

iii. Expansion of collection methodologies: Future studies should include additional methods of collection (e.g., flight intercept traps, sprinkling, soil sampling at different depths, manual collection on leaf litter and fallen logs), and the permanence of the use of several types of baits (as in our study). Is true, the use of different methodologies will depend on the purpose of the study and the interest group.

iv. Functional diversity analysis: Finally, but not least, we suggest that in future studies, functional diversity and functional traits should be addressed. Functional traits are behavioural, physiological, phenological or morphological characteristic measurable at the individual level (VIOLLE et al., 2007). They form the fundamental building blocks of functional diversity (FD), which is described as the value, range and relative abundance of functional traits in a given ecosystem (DÍAZ; CABIDO, 2001). A functional approach can be justified because there is growing evidence that FD is a better predictor of ecosystem processes than taxonomic diversity (DE BELLO et al., 2010; CLARK et al., 2012; GAGIC et al., 2015). This is likely because not all species contribute equally to all functions i.e. the relationship between taxonomic and functional diversity is not predictable and linear. A functional approach could increase the accuracy with which we link organisms to the ecological processes they govern and thus the accuracy with which we predict the consequences of species losses. In addition, the classification of species according to food guilds (e.g. predators, phytophagous, saprophagous, buffaloes, decomposers, mycophagous, among others) and Nesting guild - for dung beetles (rollers, tunnelers and dwellers) are of fundamental importance for the monitoring of anthropogenic action on tropical biodiversity.

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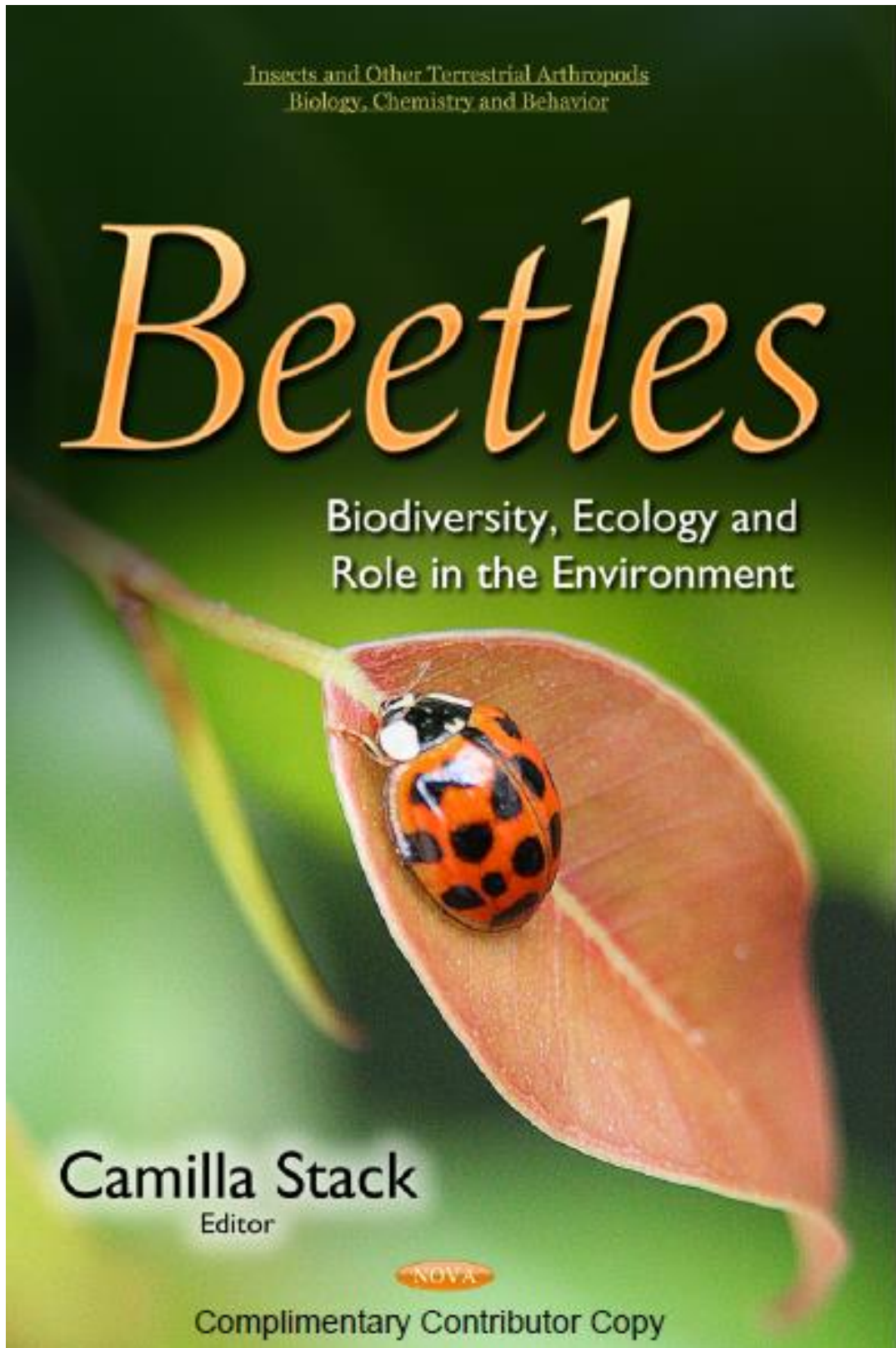
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APPENDICES – OTHER OUTCOMES

List of articles published, in press and / or submitted.

Publication referring to chapter two



Chapter 3

ASSESSING THE POTENTIAL ROLE OF GROUND BEETLES (COLEOPTERA) AS ECOLOGICAL INDICATORS IN TROPICAL ECOSYSTEMS: A REVIEW

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ABSTRACT

Emerging concerns with environmental issues has led researchers to look for indicators capable of providing information on the condition of ecosystems. Several animal species have been used for this role. Among these organisms, ground-beetles were found to be appropriate indicators, since they are adapted to all terrestrial habitats, being associated with ecosystem function and sensitive to ongoing changes. Several families of ground beetles, e.g., Carabidae, Cicindelidae, Scarabaeidae, Cerambycidae, Histeridae, Staphylinidae and Curculionidae were used in diverse ecological assessment studies due to its main characteristics, such as habitat fidelity and taxonomic diversity, and because they provide cheap and easy measurements from functionally diverse trophic chains.

Although the growing number of studies on ecological indicators little is known about the ecological patterns in the communities of ground beetles of tropical ecosystems. The present study provides a review on the use of ground beetles as ecological indicators in tropical ecosystems, with focus on the Brazilian Amazon region. We discuss its use for monitoring the effects of anthropogenic induced changes, namely the conversion of forests to agriculture and farming practices, among others on the functioning of tropical systems. We also highlight the major advances and applications employed, as well as proposals for future studies with this group.

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Does the composition of Scarabaeidae (Coleoptera) communities reflect the extent of land use changes in the Brazilian Amazon?



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ABSTRACT

Scarab beetles (Coleoptera: Scarabaeidae) have been used to investigate the effects of environmental disturbances on forest structure and diversity. This group is recognized as sensitive to habitat perturbations and ecosystem changes. Here we examine the effects of anthropogenic impacts on Scarabaeidae composition, testing the following hypotheses: (1) Scarab beetle communities react to land use disturbances with predictable trends, (2) disturbed habitats are able to retain only a part of the Scarab beetle community of native forests or late secondary forests; (3) habitats largely differ in terms of species richness, taxonomic diversity and ecological composition, supporting exclusive and indicator species. We selected areas of native forest, agriculture, pasture for extensive livestock and secondary forests in different stages of regeneration. Our results show that the Scarabaeidae species were not indifferent to the gradient of structural changes represented by the studied areas. In fact, their patterns of habitat preference reveals communities more abundant and diverse in pristine habitats. In contrast, disturbed habitats, dominated by agricultural activities and pasture, indicated clear detrimental effects on the abundance of all forest Scarab beetle specialists. On the other hand, the generalist species, mainly associated with open environments, seemed to be favoured by the prevailing conditions induced by agricultural activities. Overall, the composition of the Scarab beetle communities is variable and sensitive to those structural gradients and, therefore, capable of responding as useful ecological indicators for assessing the extent of land use change or degradation.

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Attraction of Scarabaeinae (Coleoptera: Scarabaeidae) to different baits in the Brazilian Amazon region

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Abstract: Dung beetles (Coleoptera: Scarabaeidae: Scarabaeinae) are considered essential for enhancing the physicochemical characteristics of soils, principally by changing organic materials (e.g. dead animals, faeces, fruits e leaf litter). This study compared the species richness and abundance of dung beetles captured using various types of baits, to demonstrate attractiveness differences in variable habitats of the Brazilian Amazon. Samplings were carried out with pitfall traps baited with human faeces, rotten banana, rotten meat and a non-baited trap, in February, March, June, September and October 2015. Habitats included native forests, agriculture areas, pastures and disturbed forests in different regeneration stages. A total of 13 736 Scarabaeinae beetles were captured, distributed over 98 species. Most individuals were captured using traps baited with faeces (76.7 % of individuals), followed by rotten meat baited traps (17.8 % of individuals), fermented banana baited traps (3.9 % of individuals) and finally by non-baited traps (1.6 % of individuals). A significant difference in attractiveness of the different baits used and habitats types was observed. Most of the captured assemblages were composed by coprophagous (42 %), generalist species (32 %), necrophagous (15 %) and none was classified as saprophagous. Approximately 54 % of the specimens were tunnelers, 25 % were rollers and 12 % were dwellers. The species of Scarabaeinae sampled in the region revealed qualitative and quantitative differences among their assemblages and the phytophysiognomies. The forest environments housed the greatest species richness observed, and a fraction of these is exclusive of these areas. We concluded that some species of Scarabaeinae have an important potential as disturbance indicators in the Amazonian ecosystem. *Rev. Biol. Trop.* 65 (3): 917-924. Epub 2017 September 01.

Key words: dung beetles, dung attractiveness, food preference, trophic guild, tropical forest.

The subfamily Scarabaeinae (Coleoptera: Scarabaeidae) includes dung beetles (DB), a globally distributed group of detritus-feeding insects, determinant of ecological functions such as nutrient recycling, secondary seed dispersal, bioturbation, and natural control of cattle parasites (Nichols et al., 2008; Simmons & Ridsdill-Smith, 2011). Their contribution to the improvement of the physico-chemical properties of the soil and plants is associated

with increased edaphic aeration and hydration (Andresen, 2002; Nichols et al., 2008).

Mammalian dung is one of the most important food resources for dung beetle communities, being also the dominant substrate for oviposition (Filgueiras, Liberal, Aguiar, Hernández, & Iannuzzi, 2009). Anyhow, in the Neotropics, fungi, plants and carcasses are also used by several species as food resource (Halffter & Matthews, 1966). This seems related





Can dung beetles (Scarabaeinae) indicate the status of Amazonia's ecosystems? Insights integrating anthropogenic disturbance with seasonal patterns

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Abstract

Temporal and spatial variation in dung beetles abundances is a pattern observed in many tropical forests. The present study evaluated the seasonal patterns of dung beetles in a range of increasingly disturbed ecosystems of the state of Pará, northern Brazil, to identify valuable disturbance indicators. The areas included native forest, agriculture, pasture for extensive livestock grazing and secondary forests. Fieldwork was carried out encompassing the complete range of environmental conditions encountered during the year. In total, 13,649 individuals were captured within 23 genera and 99 species but with pronounced differences among ecosystems and seasons. The obtained results seem to demonstrate that dung beetles can be used to help identify ecosystems under very complex and variable environmental conditions. The ecological drift observed also demonstrates the possibility of using dung beetles as ecological indicators of disturbance in Amazonia.

Keywords

Amazon rainforest; beetles; biodiversity; scarab; seasonality; tropical forest

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Are disturbance gradients in neotropical ecosystems detected using rove beetles? A case study in the Brazilian Amazon



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ABSTRACT

In the Neotropics, Rove Beetles (Staphylinidae) are known to be especially diverse and abundant, but studies in this region are still incipient and the information is dispersed in the scientific literature. In this work the responses of the Rove Beetles to gradients of disturbance in representative ecosystems of the Brazilian Amazon were evaluated. Specifically, we assessed the possibility of using patterns in richness, abundance and taxonomic diversity of Rove Beetles as anthropogenic disturbance indicators. The obtained results show that the Rove Beetle richness was sensitive to the structural changes induced by increasing anthropogenic disturbance. Additionally an increasing trend in community complexity from more to less disturbed ecosystems was observed. Differences in responses can be explained by differences in the intensity and extent of the change from pristine systems. Overall, the composition of Rove Beetle communities is ecosystem specific and sensitive to anthropogenic induced structural changes and management actions and, therefore, should be considered a valuable ecological indicator for assessing the extent ecosystems' disruption in the Neotropics.

1. Introduction

Neotropical forests are biologically diverse ecosystems, representing some of the richest areas of the planet (Lindemayer et al., 2002). Species from these forests are threatened by deforestation, fragmentation, conversion to forest monocultures, climate change and other stressors like fire and even fire suppression (Camus et al., 2006; Ledkovská and Horák, 2016). There is an enormous concern and speculation about the effects of human disturbances in the biodiversity of neotropical ecosystems, namely the impacts on species composition and the modifications in the ecological services provided (e.g. Morán, 2010; Aerts and Honeay, 2011; Cajalba et al., 2017). Deforestation in the Brazilian Amazon (Amazonia), has been causing a sharp erosion of biodiversity and the disruption of complex global climate cycles (e.g. Morán, 2010; Cajalba et al., 2017). The most significant landscape changes identified for the Amazonia are forest logging, the establishment of extensive livestock operations and intensive farming, but also the expansion of road nets and urban areas (Mertens et al., 2002; Tabarelli et al., 2004), power generation, and mining (Souza-Hilho

et al., 2005). These anthropogenic activities are considered strong environmental stressors whose impacts were not fully anticipated (Cajalba et al., 2017). In fact, these landscape changes promote ecosystems' substitution and might isolate taxa and even cause extinctions due to interactions in which species are engaged (Aron et al., 2012; Valiente-Banuet et al., 2015). Understanding the impacts of ecosystem transitions in the Amazonia will ultimately support the best conservation strategies for the region, considered fundamental for sustaining earth's functioning and ultimately humankind (Cajalba et al., 2017).

In a conservation perspective, the condition of an ecosystem might be evaluated using the "distance" from reference situations, i.e. the disparity of a specific ecosystem from pristine ecosystems (Costanza, 2012). Considering the unfeasibility of measuring all aspects in a specific ecosystem, several scientists advocate the use of surrogates, usually termed ecological indicators (Costanza, 2012; Heath, 2013). Species, populations and communities might prove useful as indicators if sensitive to ecosystem changes, anticipative and easily monitored (Gardner et al., 2008; Rapport and Hildén, 2013). Besides the universal need for developing ways to assess status and trends in environmental

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Publication referring to chapter seven

Cajaiba et al.: Diversity and seasonality of Histeridae in northern Brazil
- 1227 -

SEASONAL PATTERNS IN THE DIVERSITY OF HISTERID BEETLES (HISTERIDAE) ARE ECOSYSTEM SPECIFIC? A CASE IN PARA STATE, NORTHERN BRAZIL

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Abstract. The objective of this work was to characterize the seasonal variation in the community of Histerid beetles (Histeridae) of different ecosystems in the Brazilian Amazon, ranging from primary or secondary forests with different stages of regeneration to cocoa farms and pastures. Pitfall traps were distributed within the monitored ecosystems during different periods of the year. A total of 1945 Histerid beetles, belonging to five genera and 14 species were captured. Higher diversities were observed during the rainy season, although with differences among ecosystems. The genera *Phelister* and *Hister* were ubiquitous in all ecosystems, constituting 71% of all the specimens captured. Histerid beetle communities, discriminated by ordination methods, change gradually from the most conserved ecosystems to more disturbed ones. Moreover, the results showed that disturbed ecosystems, namely cocoa farms and pastures, have detrimental effects on the occurrence of Histerid beetles, which are considered forest specialists, while enhancing generalist species.

Keywords: Amazon rainforest, biodiversity, soil insect, fluctuation, tropical forest

Introduction

Currently, there is enormous concern and speculation about the effects of anthropogenic disturbance on the biodiversity of tropical ecosystems, including the effects on species composition and the modifications in the ecological services provided (e.g. Morris, 2010; Aerts and Honnay, 2011; Cajaiba et al., 2017a). Deforestation in the Neotropics, particularly in the Brazilian Amazon (Amazonia), has been causing a sharp erosion of biodiversity and disruption of the complex global climate phenomena (e.g. Hassan et al., 2005; Morris, 2010). Therefore, the conservation of the Amazonian ecosystems represents a growing core challenge for sustaining earth's functioning and ultimately that of mankind (e.g. Viana and Pinheiro, 1988; Viegas et al., 2014).

Predicting the ecological consequences of land use/land cover (LU/LC) changes is therefore a subject of scientific and political interest in order to support strategic options for sustainable development, land use planning and natural resources management

Publication referring to chapter eight

The screenshot shows the 'Active Submissions' page of the journal 'Neotropical Biology and Conservation'. The page features a navigation menu, a user login section, and a table of active submissions. The journal's ISSN is 2236-3777. The submission table lists one item with ID 13470, submitted on 12-02, by authors Cajaiba, Silva, and Périco. The title is 'DIVERSITY OF SCOUTINAE (COLEOPTERA: CURCULIONIDAE) IN...'. The status is 'IN EDITING'. The page also includes a search bar and a 'Start a New Submission' link.

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