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#### Abstract

Habitat loss and fragmentation represent two of the most significant threats to biodiversity. In some regions, like the Brazilian Cerrado, the deforestation rate can reach nearly 1 million hectares per year. Ecoacoustics and acoustic indices can be used to promote rapid assessments in threatened regions. We evaluated how two particular indices (the acoustic diversity index - ADI - and normalized difference soundscape index - NDSI) reflect bird species richness and composition in a protected area near Brasilia city. We hypothesized that ADI should reflect the characteristics of birds in the cerrado and in the gallery forest, i.e., with higher values in gallery forest than in the cerrado. Based on habitat structure, we also hypothesised that NDSI should be lower in less complex habitat, and lower in areas close to urbanized areas. We assessed 30 locations by installing automatic recorders to generate 15 min wave files (48 kHz, 16 bits, stereo). The manual hearing of the files revealed the presence of 107 bird species (74 in gallery forest and 47 in cerrado). Our results showed that ADI was significantly associated to species richness, being higher in gallery forest than in the cerrado. We found that NDSI values were lower in areas close to highways, an important source of impact for bird diversity. We argue that acoustics indices are a valid approach for rapid biodiversity assessment, however basic knowledge on species occurrences is essential to interpret the values provided by these indices.

## **1. Introduction**

Biodiversity loss caused by humans is a major and challenging problem globally (Pimm et al., 2006), and threats to species and ecosystems are set to continue in the future (Pereira et al., 2010). Deforestation and habitat fragmentation caused by expansion of croplands and pastures impose serious threats to species and native ecosystems, especially in South America, where the most globally significant changes in cropland expansion occurred between 1960-1990 (Ramankkutty et al., 2002). Most of cropland expansion has occurred in the Brazilian Cerrado (woodland savanna), a region, which has lost half of its original area (more than 1 million km<sup>2</sup>) in recent decades (Brasil, 2009; Françoso et al., 2015; Klink and Machado, 2005). How biodiversity responds to habitat loss and fragmentation is one of the key topics in ecology and conservation biology (Sala et al., 2000).

Considering the rapid deforestation rate with in the Brazilian Cerrado, (Brasil, 2009; Klink and Machado, 2005) it is important to develop and apply methods that can be effective for rapid biodiversity assessment. The field of Ecoacoustics, a emerging discipline that investigates spatial and temporal variation of the sounds associated with landscape structures (Sueur and Farina, 2015), may provide the answers to the ecological and conservation issues. Bioacoustics has been traditionally used in behavioural studies, and only recently has been applied in conservation biology (Ritts et al., 2016; Sueur et al., 2008b; Towsey et al., 2014a). The field of ecoacoustics, an emerging discipline that investigates spatial and temporal variation of the sounds associated with population, community or landscape structures (Sueur and Farina, 2015), may provide the answers to ecological and conservation issues.

Ecoacoustics may provide an excellent tool in conservation biology because it is noninvasive, it can synthesize a wide range of species and many locations can be surveyed simultaneously using automated recorders. In addition, the recordings can be sent to experts for thorough analysis, and data can be collected without human interference. Unlike the *in situ* observations made by researchers, recordings can represent a voucher material of biodiversity inventories, creating a permanent record of a field study. The availability of analytical tools for data processing has also increased in recent years, and semi-automatic or automatic signal extraction and even identification is now possible, although there are still obstacles in automatic identification of soundscapes entities. Acoustic indices for ecological studies and biodiversity monitoring are one of the automatic approaches for data analysis (Kasten et al., 2012; Pieretti et al., 2011; Pijanowski et al., 2011; Sueur et al., 2014), and acoustic indices can potentially be used as a surrogate for observational data when quick assessments are necessary. Another advantage of the use of acoustic indexes is to overcome the problem of analysing hours and hours of records obtained by passive recorder units.

In order to be useful, however, an acoustic index should have some congruence with the patterns of biodiversity or species diversity of the taxonomic group under investigation. If the acoustic index can be associated with species diversity or species activity, it can provide a valid tool to rapidly assess biodiversity using a soundscape approach.

The soundscape, according to <u>Dumyahn and Pijanowski (2011)</u>, represents the entire acoustic environment of a particular landscape. In this paper, we used acoustic data to (a) characterize species richness and composition of birds associated with the Cerrado in Brazil and (b) to compare the behaviour of two indices, an acoustic diversity index (ADI) (<u>Villanueva-</u> <u>Rivera et al., 2011</u>) and the normalized difference soundscape index (NDSI) (<u>Kasten et al., 2012</u>) in two different Cerrado habitat types: the cerrado *stricto sensu* (in lower case to indicate the phytophysiognomy) and the gallery forest (evergreen forest-like vegetation that exists along rivers and streams).

Gallery forest is a narrow strip of forest along the rivers and streams with three vegetation layers and a closed canopy, reaching 20-25 meters in height. On the other hand, the cerrado *stricto sensu* is a woodland savanna-like vegetation with sparse trees reaching 6-10 meters high, with unconnected canopy coverage from 50-70%, allowing sunlight to directly reach the soil (Eiten, 1972; Ribeiro and Walter, 1998).

We tested predictions from three hypotheses concerning the potential associations between biodiversity data and the acoustic indices. First, we predicted that acoustic indices would reflect the characteristics of bird communities associated with the cerrado stricto sensu (s.s.) and gallery forest, the two habitats with highest species richness in the Cerrado biome. The basis for this prediction was that the values of the acoustic diversity index [ADI; is a measurement of the degree of acoustic complexity found at a site (see Villanueva-Rivera and <u>Pijanowski (2014)</u> and the Methods for details)] should be higher in gallery forest, which has greater structural complexity compared with the cerrado. Second, we predicted that the biophony/technophony ratio (i.e. the ratio of sounds produced by wildlife and by human activities respectively) should be lower in the cerrado than in gallery forest due to the higher attenuation of anthropophonic sound by the complex vegetation in gallery forest. We used the normalized difference soundscape index (NDSI) to indicate the amount of biophony relative to technophony in the two habitat types (see Gage and Axel (2014) and the Methods for more details). Finally, we predicted that the spatial arrangement of technophony noise sources such as roads, houses and buildings or airports, should influence the values of the NDSI, such that lower values would be observed near such noise sources. Thus, the NDSI would be a valid proxy for

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the impact of technophony on biodiversity regardless of the type of habitat (cerrado or gallery forest), and areas located close to technophony sources will have lower values of NDSI.

#### 2. Materials and methods

## 2.1 Study area

We conducted this study in a protected area named the Environmental Protection Area of Gama e Cabeça do Veado (hereafter referred to as EPA), which is equivalent to the category V Protected Area Category defined by the International Union for Conservation of Nature (Phillips, 2002; Thomas and Middleton, 2003). The EPA covers 25,000 ha and was created in 1986. It is located south of Brasilia city ( $15^{\circ} 51'16'' - 15^{\circ} 58' 17'', 47^{\circ} 59' 39'' - 47^{\circ} 40' 09'' Long W$ ) (Figure 1), and comprises four reserves: Água Lima Farm, Roncador Ecological Reserve, Botanic Garden Ecological Station and a training centre area managed by the Brazilian Navy. In total 286 bird species have been recorded in the area (Braz and Cavalcanti, 2001), representing 33.6% of the bird species registered in the Brazilian Cerrado (Silva and Santos, 2005). The natural vegetation of the area is a mosaic of phytophysiognomies ranging from grasslands to cerrado (*s. s.*) and cerradão (tall savanna) (Eiten, 1972; Ribeiro and Walter, 1998). Along the rivers and streams there is a strip of gallery forest (up to 200 m wide) with the canopy reaching 20-25 meters (Eiten, 1972).

#### 2.2 Data collection

We established 30 sampling sites (15 in gallery forest and 15 in cerrado). The locations were separated by at least 1 km, a distance more than three times that normally used in point census studies (e.g. 300 m in <u>Anjos (2007)</u>, 200 m in <u>Cavarzere et al. (2013)</u>, or 150 m in <u>Wilson</u> et al. (2000)). We used the package *ape* (Paradis et al., 2004) to conduct an *a posteriori* analysis

to test if our sampling locations were spatially correlated or not. We calculated the Moran's I using an inverted matrix of distance between all locations and the number of species registered at each point as a dependent variable. The result indicated that there was no spatial autocorrelation in our locations (observed =-0.0233; expected = -0.0345; p=0.7759) and, therefore, we assumed that our locations represent independent sampling units.

We used 10 SongMeter SM2 digital recording devices (Wildlife Acoustics, Maynard, MA, USA) to obtain records of birds during three consecutive days in each month from January to May of 2014. We first put the recorders on locations 1 to 10, and then we moved the recordings to the subsequent locations, completing the 30 sampling locations on the third day. In order to avoid river sounds, which could dominate the recordings, we put the recorders in locations away from any rapids or cascades. The recorders were programmed to start operating at sunrise and to stop two and half hours later. Recordings were made as wav files (48 kHz sampling rate, 16 bits, stereo mode) divided into recordings of 15 min duration with intervals of 5 min between them. Therefore, for each location we obtained eight 15 min recordings per month, which correspond to 60 hours/month (30 locations x 2,5 hours per point) or 300 hours of recording in total.

#### 2.2 Data analysis

We analysed the recordings, manually and automatically. We randomly selected 30 files of 15 min duration (one per location) to identify the species present. We use Audacity v.2.06 software (<u>Ash et al., 2014</u>) for annotation process. This procedure consists of listening to the recorded sounds while simultaneously analysing spectrograms created with Audacity (fast Fourier transformation with a 1024 window size and Hanning window). All records were identified, when possible, to species level. For the cases where we could not identify a species, we assigned it as 'unidentified 1', 'unidentified 2' and so on.

## 2.3 Automated data analysis

We used the packages *tuneR* (Ligges, 2014) and *soundecology* (Villanueva-Rivera and Pijanowski, 2014), both used in R v3.0.2 (R Core Team, 2015). The latter is dependent on *seewave* package (Sueur et al., 2008a). Initially, we selected 30 wave files with 15 min duration (one file per point for January) and used the Kaleidoscope software (Wildlife Acoustics Inc., USA) to split each of the 15 min recordings into recordings with 1 min duration providing 450 files (225 for each habitat type).

We calculated six different indices (Acoustic Complexity Index - ACI, Acoustic Diversity Index - ADI, Acoustic Evenness - AEI, Bioacoustic Index - BI, Total Entropy - H, and Normalized Difference Soundscape Index – NDSI) for all files using *soundecology* (Villanueva-<u>Rivera and Pijanowski, 2014</u>) and *seewave* (Sueur et al., 2008a) packages. We used the standard parameters available for each index for all calculation.

First, we performed a descriptive statistics (mean, standard deviation, frequency distribution) to inspect the calculated indices (Supplementary Material – figure S1). We also used the Spearman rank correlation coefficient, to evaluate the association between all indices, since most of the relation showed a non-linear pattern (Supplementary Material – figure S2). A preliminary analysis revealed that Acoustic Complexity – ACI and Bioacoustics – BI indices were less correlated with other indices (Supplementary Material – table S2). Additionally, ACI and BI had a significant variation of the values of the sites, which was not consistent with other indices (Supplementary Material – figure S3). On the other hand, the Acoustic Diversity Index

was high correlated with indices H and AEI (or -0.999 and 0.703, respectively) and had lower association with BI (-0.517) (Supplementary Material – table S2). So, we choose Acoustic Diversity Index as a representative of the different acoustic aspects measured by the indices. Thus, considering our objectives, we restricted our analyses only to the Acoustic Diversity Index and to the Normalized Difference Soundscape Index.

The Acoustic Diversity Index (ADI), an index derived from the H index proposed by Sueur and colleagues (<u>Sueur et al., 2008b</u>), quantifies the proportion of sounds that are above a certain threshold (-50 dBFS in our case) and then applies the Shannon index to represent the recorded sounds. The index is calculated according to the following formula:

$$H' = \sum_{i=1}^{3} p_i \ln p_i$$

where  $p_i$  is the fraction calculated in each frequency band and S is the number of defined frequency bands (see <u>Villanueva-Rivera et al. (2011)</u> for details). It is important to emphasize that the ADI takes into account the sounds that exists in the files and it is not related to the bird species *per se*.

The Normalized Difference Soundscape Index (NDSI) is the ratio between biophony (the sounds produced by animals), and technophony (sounds produced by machinery, such as vehicles and airplanes). NDSI index is calculated according the following formula:

$$NDSI = \frac{\beta - \alpha}{\beta + \alpha}$$

where  $\beta$  represents the biophony (i.e. biophony power spectral density ( $\Sigma$  2-11 kHz)) and  $\alpha$  represents the technophony (i.e. technophony power spectral density ( $\Sigma$  1-2 kHz)). See <u>Gage and</u> <u>Axel (2014)</u> for details about the NDSI index.

For the calculation of both indices we used a Fast Fourier Transformation based in a 1024 bin of signal sampling and frequency intervals of 1 kHz (the standard parameter on the *soundecology* package). For the ADI calculations, we set 12 kHz as the maximum frequency to be analysed and for the NDSI calculations we considered the frequency intervals of 1-2 kHz for technophony calculations and 2-12 kHz for biophony sounds.

## 2.4 Statistical Analysis

Bird communities associated with the two environments in our study area were characterized according to two parameters: species richness and composition. Considering that species richness per location had a normal distribution (Shapiro test W = 0.9801, p=0.831), we applied a *t* test to verify if the number of species found in each location varied between the two habitat types (cerrado s.s. and gallery forest). The comparison of species composition of each area was done by creating an Euclidian matrix of distances between all locations based on the Jaccard index. Then we used the packages *vegan* (Oksanen et al., 2013) and *cluster* (Maechler et al., 2015) in R (R Core Team, 2015) to produce a representative dendrogram of bird communities (Unweighted Pair Group Method with Arithmetic Mean – UPGMA).

The two studied habitats were also characterized in terms of their vegetation structure. We used the values of the normalized difference vegetation index (NDVI) as a proxy for the vegetation structure, because it has been demonstrated that the alpha and beta diversity biodiversity parameters can respond to NDVI variation. This relation had been tested for plants (<u>He et al., 2009</u>) and for birds and mammals (<u>Toranza and Arim, 2010</u>). Areas covered by dense vegetation, such as gallery forest, will present higher values of NDVI whereas areas with less biomass, such as cerrado and grasslands, will present lower values. Therefore, we also characterized our set of sampling locations accordingly to the NDVI values extracted in each one of them. Since the distribution values of NDVI in our locations had a normal distribution (Shapiro test W = 0.9396, p-value = 0.0885) we used parametric statistics (*t* test) to test possible differences in biodiversity indices in cerrado and gallery forest in relation to NDVI. The creation of the NDVI image is described below.

We evaluate the association of acoustic diversity index and biodiversity by applying a linear model between the mean ADI value and the bird species richness (represented in log scale) registered in each location. To test the prediction from our first hypothesis (ADI index should be higher in the gallery forest) and our second hypothesis (NDSI should be lower in the cerrado) we compared the median values based on 450 recordings used in the analysis. ADI values were not normally distributed (W = 0.851, p=0.486) so we used a Wilcoxon test to test for potential differences in the ADI index between habitats and used a t test for the comparisons of NDSI values as the data were normally distributed.

Before we tested our third hypothesis (a spatial association of NDSI and sources of anthropogenic sounds), we evaluated whether NDSI values would be influenced by species richness. Thus, we used a simple linear model to verify if NDSI values for cerrado s.s. and gallery forest would be correlated to species richness. We then used model selection analysis, based in a generalized linear model, to verify how different spatial variables would influence the NDSI values. The explanatory variables used in this analysis were: distance from airport (air), distance from highways (disth), distance from offices (administrative builds and visitors center) (disto), normalized vegetation difference index (ndvi), distance from residential districts (neigh), distance from roads (local access unpaved roads) (roads) and bird species richness (spp). Details about the explanation of each variable and basic statistic (mean, s.d., and range) can be found in

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the Table S1 (Supplementary Material). We used the package MuMIn (<u>Barto'n, 2016</u>), which uses a second order Akaike Information Criterium (AICc), to perform a model selection.

We created the NDVI image by combining the red (b4) and near infrared (b5) bands of the Landsat 8 OLI (Operational Land Imager). We obtained the images (dated from Sep/2014) from the United States Geological Service (USGS) (<u>http://earthexplorer.usgs.gov</u>). We used the following formula to produce the NDVI image is:

$$NDVI = \frac{(b5 - b4)}{(b5 + b4)}$$

All maps created by us had the same spatial characteristics (cell size and extent) of the LandSat 8 images. We used the package *raster* (Hijmans et al., 2014) to generate all maps used as explanatory variables. We created a 250 m buffer around each location to calculate the mean value of the NDVI image. For all other explanatory variables we created a distance map with values starting from each feature (airport, highways, roads, offices, residential districts). Each feature (roads, highways, airport) was produced by manually digitizing over a Google Earth image. We considered differences to be significant for p<0.05 for all statistical tests. Results are presented as means  $\pm$  SD, unless otherwise noted.

#### 3. Results

Acoustic identification by listening to recordings and viewing spectrograms revealed the presence of 107 bird species in the 30 sampling locations, where 74 and 47 bird species were identified in the gallery forest the cerrado, respectively (Supplementary Material - table S3). From this total, 18 birds, mostly with a single call, were not identified at species level. We used two song parameters (duration and frequency) and the spectrogram shape to ensure that they were different from the identified species. The mean number of species registered per point per

habitat was significantly higher in gallery forest than in the cerrado (t=-2.8073, df=27.878, p=0.009) (Figure 2). A total of 46 species were heard exclusively in the gallery forest, whilst 29 species were heard exclusively in the cerrado. The number of species recorded per location ranged from 5-17 and 2-15 in the gallery forest and cerrado, respectively.

A cluster analysis based on species composition revealed low similarity between the cerrado and the gallery forest (Fig. 3). The most frequent species associated with the gallery forest were Burnished-buff Tanager (*Tangara cayana*), Golden-crowned Warbler (*Basileuterus culicivorus*) and Buff-breasted Wren (*Cantorchilus leucotis*). In the cerrado, the most frequent species identified were Turquoise-fronted Parrot (*Amazona aestiva*), Glittering-throated Emerald (*Amazilia fimbriata*) and Rufous-collared Sparrow (*Zonotrichia capensis*).

The two environments also differed in terms of vegetation structure. The mean NDVI was much lower in the cerrado compared with the gallery forest (NDVI<sub>cerrado</sub>=0.0400, NDVI<sub>gallery forest</sub>=0.300, t=-12.954, p<0.001) (Figure 4-left).

The ADI was greater in the gallery forest (ADI<sub>gallery forest</sub> = 1.276, N=450, s.d.=0.581) than in the cerrado (ADI<sub>cerrado</sub> = 1.109, N=450, s.d.=0.653, W=75764, p<0.01) (Figure 4-middle), confirming the prediction that the ADI index should be higher in the gallery forest because of its greater structural complexity. The mean NDSI values in the cerrado s.s. were lower than those in the gallery forest (t=-12.854, df=52.547, p<0.01) (Figure 4-right), confirming the prediction of our second hypothesis that higher values of NDSI would be found in more complex habitats. The mean value of NDSI (January to May) in the cerrado s.s. was -0.0542 (N=450, s.d.=0.382) whilst the mean value in the gallery forest was 0.1184 (N=450, s.d.=0.335).

Since the values of NDSI were not significantly associated with species richness  $(F_{1,28}=0.823, R^2=0.028, p=0.372)$  we assumed that the technophony was the main driver of the

observed differences between sampling locations. The correlation of ADI with bird species richness also showed a significant relationship, although bird species richness explained only 20.7% of the ADI variation ( $F_{1,28}$ =7.328,  $R^2$  = 0.207, p=0.0114) (Figure 5).

The model selection that tested the association of NDSI values and anthropogenic sources generated 45 possible models by combining the explanatory variables, but only two of them showed the  $\Delta$ AICc lower than 2 (Table 1). The most important variable explaining the spatial variation of NDSI is the distance from highways, followed by distance from offices and distance from local unpaved roads. The weights of those variables to compose the models were 0.96, 0.91 and 0.77, respectively (Table 1). Distance from highways explained 44.3% of the variance observed in NDSI values and was significantly associated to the response variable (i.e., locations away from highways tends to present higher values than those close to highways (F<sub>1.28</sub>=22.34, R<sup>2</sup>=0.445, p<0.01 – Fig. 6)). Hence the prediction from our third hypothesis, that lower values of NDSI would be expected near places with more human activities was confirmed in both habitat types.

#### 4. Discussion

As far as we know, this is the first study comparing the composition of bird communities by using acoustics indices in South America and in the Brazilian Cerrado. An earlier study in Brazil aimed to define sampling schemes for general passive acoustic monitoring (<u>Pieretti et al.</u>, <u>2015</u>) did not include biodiversity data for comparisons. We believe that the knowledge of biodiversity characteristics allows a better understanding of the behaviour of acoustic indices, which is essential for their interpretation and for drawing meaningful extrapolations. Our results indicate that acoustic indices could be a valid surrogate for traditional studies of birds in the Cerrado biome when rapid assessments are required because acoustic indices are significantly correlated. Considering the characteristics of the bird communities associated to the cerrado and gallery forest, i.e., different diversity and bird species composition in both habitats, the behaviour of the acoustic diversity index and the normalized difference soundscape index reflected the patterns that were predicted.

Despite the fact that cerrado habitat occupies an area four times larger than the gallery forest in the EPA, the number of bird species is higher in gallery forest habitat (Braz and Cavalcanti, 2001; Machado, 2000). This pattern is not a particular characteristic of birds of the Cerrado biome. The same pattern has been found in mammals (Redford and Fonseca, 1986), in contrast to groups such as lizards (Nogueira et al., 2009) and plants (IBGE, 2004) where the number of species in the cerrado tends to be higher than in gallery forest.

It is important to note that the species composition is totally different between the two habitats. Our results showed that there is a low similarity between the habitats, reflecting differences in the ecological requirements for birds or differences on the origin of species associated with forest environments in the Cerrado biome (Silva, 1995; Silva and Bates, 2002). The higher richness of bird species in the gallery forest in relation to the cerrado can be explained by the differences in the vegetation structure, which is a traditional interpretation in traditional methods of interpretation of bird species richness (MacArthur, 1964; Wiens et al., 1989).

The NDVI (normalized difference vegetation index) obtained for our area reflects such differences in the vegetation structure. Thus, the results for direct evaluation (hearing the sounds) and automatic (ADI) are consistent with the vegetation structure. We observed the congruence in the relation of ADI with vegetation structure, reported in other studies (e.g. <u>Pekin et al. (2012)</u>,

and this observation confirms that acoustic indices are strongly associated with avian species richness (<u>Towsey et al., 2014b</u>) or with the structure of the environment (<u>Farina and Pieretti, 2014</u>; <u>Pekin et al., 2012</u>).

Although the species are not identified when ADI is calculated, the mean value of the index captured real differences between the habitats, being smaller for the cerrado and higher for the gallery forest, confirming our first hypothesis. Furthermore, we observed a significant positive correlation between ADI and bird diversity. However, it is important to highlight that ADI values change with species diversity, but also with species activity. Other studies have shown that acoustic indices change seasonally (Gage and Axel, 2014; Pijanowski et al., 2011; Ritts et al., 2016) with a peak in activity related to frequency band diversity. Previous bird studies in the EPA found a strong association of bird activity and climate seasonality (Machado, 2000) with four different patterns described. Therefore, the ADI value can change annually and complementary analyses are necessary to elucidate such relationships.

Our second result, related to the NDSI, indicated that cerrado and gallery forest could be distinguished based on this index. As we predicted, the cerrado had a lower NDSI values, an aspect that can be explained by a higher level of noise in relation to the gallery forest. Vegetation structure can provide a barrier for sound dispersion (Aylor, 1971; Lyon, 1973) and the spatial arrangement of different ecosystems in a landscape can affect acoustic indices (Farina and Pieretti, 2014; Villanueva-Rivera et al., 2011).

The observed difference for the NDSI between the two habitats could be caused exclusively by the biophony component if the two habitat types experience the same level of technophony. However, our third analysis, which investigated the spatial relation of NDSI with low frequency sound sources, indicated that regardless the type of habitat, lower values of NDSI were found near highways and local roads. Thus, the technophony values were effectively different over space and the difference in NDSI values cannot be explained by biophony alone. Anthropogenic noise may reduce species richness in urban areas (Proppe et al., 2013) and can impose new challenges for adaptation in vocal animals (Joo et al., 2011). Hence, analysis based on the relationship of biophony and technophony can be used to predict potential impacts of habitat loss or habitat fragmentation (Tucker et al., 2014).

Our study area is a set of reserves that were established in the 70s, when Brasilia city was at the beginning of its urbanization process. The city was created in early 1960s and was projected to hold 500,000 inhabitants, but since then a rapid expansion has occurred. Brasilia is now inhabited by 2.8 million people where urban areas have encroached upon the edges of the EPA. Our acoustic analyses indicate that some bird species might have responded to the impacts of urbanisation by avoiding the areas close to the reserve border. Alternatively, the anthropogenic sounds could be influencing the vocal behaviour of birds, making then sing less than they normally would. Some studies have shown noise-tolerant species tend to change their song structure (Hanna et al., 2011) or to compensate for increased technophony areas by increasing their song frequency (Slabbekoorn and Ripmeester, 2008) in order to allow a communication with other individuals. However, few species can tolerate higher levels of technophony and there is a tendency of species reduction in a gradient from native to urban areas (Patón et al., 2012; Proppe et al., 2013).

Nevertheless, the aspects highlighted above show the potential application of acoustic indices for the management of protected areas, especially due to its relatively rapid assessment of avian communities. Acoustic indices could potentially inform the zoning process or ecosystem integrity, and support decision makers, as also highlighted by other authors, including <u>Gage and</u>

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<u>Axel (2014)</u>. Overall, our results suggest that acoustic indices significantly reflect differences in species richness and composition in two habitats. Furthermore, our analyses indicate that acoustic indices show spatial arrangements that change accordingly to the proximity of anthropogenic sound sources.

## **5.** Conclusions

We demonstrate relationships between the characteristics of bird communities associated with different habitats in the Brazilian Cerrado analysed by using acoustic indices. As we initially expected, the acoustic diversity index (ADI) and the normalized difference soundscape index (NDSI) matched differences in species richness and composition that exist between gallery forest and cerrado, as well differences in the spatial arrangement of the sampling locations in relation to landscape characteristics.

We propose that acoustic indices, such as NDSI and ADI, provide potential not only for ecological studies, but also for evaluation of environmental impacts and protected area management. However, we emphasize that basic data on species occurrence and the description of ecological communities will be always necessary to enhance the interpretation of acoustic indices.

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## FIGURES

**Figure 1**. Location of the study area in Central Brazil and the sampling locations inside the Environmental Protected Area of Gama Cabeça-de-Veado (EPA), delimited with a white line. Dark grey areas are gallery forest and medium grey areas are the cerrado *stricto sensu*. The letters in the right top map represents the Brazilian states (DF=Federal District, GO=Goiás, BA=Bahia, MG=Minas Gerais and SP=São Paulo).

**Figure 2**. A boxplot comparing the ADI values between cerrado *stricto sensu* and gallery forest. The grey dots represent the mean value and the lines represent the median.

**Figure 3**. A dendrogram showing the similarity (Jaccard index) of the sampling locations accordingly to the species composition for the cerrado *stricto sensu* and the gallery forest.

**Figure 4**. Boxplots comparing the values of the Normalized Difference Vegetation Index (NDVI) (left), Acoustic Diversity Index – ADI (middle) and Normalized Difference Soundscape Index – NDSI (right) between cerrado *stricto sensu* and gallery forest. The grey dots represent the mean value and the lines represent the median.

**Figure 5**. A linear model of Acoustic Diversity Index with bird species richness in the Área de Proteção Ambiental Gama Cabeça de Veado. Squared and rounded symbols represent the gallery forest and cerrado locations, respectively.

**Figure 6**. A linear regression model between the Normalized Difference Soundscape Index (NDSI) and the distances from highways surrounding the Environmental Protected Area of Gama Cabeça-de-Veado (EPA). Squared points represent sampling locations set in the gallery forest and rounded points represent those located in the cerrado *stricto sensu*.

**Table 1**. Association between the Normalized Difference Soundscape Index (NDSI) and the

 explanatory variables. Values are resulting from a selection of models based on generalized

 linear models approach. The variables are ranked according to their relative importance.

#### Supplementary Material

**Figure S1**. Frequency distribution of values for the acoustic indices in the Environmental Protection Area of Gama e Cabeça de Veado (EPA), located in Brasília, Brazil. ADI = Acoustic Diversity Index, ACI = Acoustic Complexity Index, AEI = Acoustic Evenness Index, BI = Bioacoustic Index, H = Enthropy Index, NDSI = Normalized Difference Soundscape Index.

**Figure S2**. Correlation patterns between all pair of acoustic indices for the Environmental Protection Area of Gama e Cabeça de Veado (EPA), located in Brasília, Brazil. ADI = Acoustic Diversity Index, ACI = Acoustic Complexity Index, AEI = Acoustic Evenness Index, BI = Bioacoustic Index, H = Enthropy Index, NDSI = Normalized Difference Soundscape Index.

**Figure S3**. Median values for all indices according to the sampled sites (cerrado sites represented in white colour, gallery forest represented in grey colour) in the Environmental Protection Area of Gama e Cabeça de Veado (EPA), located in Brasília, Brazil Dashed line indicates the mean value the cerrado and continuous line for the gallery forest. ADI = Acoustic

Diversity Index, ACI = Acoustic Complexity Index, AEI = Acoustic Evenness Index, BI = Bioacoustic Index, H = Enthropy Index, NDSI = Normalized Difference Soundscape Index.

**Table S1**. Names and description of the explanatory variables used to evaluate the spatial

 variation of the Normalized Difference Soundscape Index (NDSI).

**Table S2**. Correlation values (Spearman coefficient) between calculated indices in theEnvironmental Protection Area of Gama e Cabeça de Veado (EPA), located in Brasília, Brazil.

**Table S3**. List of identified species in the recordings from the Environmental Protection Area of Gama e Cabeça de Veado (EPA), located in Brasília, Brazil.