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Differentiation and characterization of burrows of two species of armadillos in the Brazilian Cerrado

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Abstract

Background: Signs left by some mammals such as tracks, trails, burrows, scratches, feces, hair, and others can be an important tool for species identification. This study was conducted in the Itirapina Ecological Station, in the southern Brazilian Cerrado, whose main objectives were to (1) compare the burrow morphometric variables of *Cabassous unicinctus* and *Euphractus sexcinctus* and (2) check for differences in the occurrence of foraging burrows in two different environments: areas with vegetation (shrubby grassland) and areas without vegetation (roads). The collected measures of armadillo's burrows have been width, height, depth, and angle relative to the soil. To record foraging burrows (<70 cm), the linear transect methodology was used simultaneously in roads and shrubby grassland environments.

Results: In a shrubby grassland environment of the Itirapina Ecological Station, a density of 121 burrows/ha pertaining to *C. unicinctus* and of 277 to *E. sexcinctus* was observed. Although there is overlap between them, morphometric variables were used to measure the burrows; significant differences were observed in all: depth, $p < 0.001$; ratio, $p < 0.001$; perimeter of the ellipse (perimeter), $p < 0.001$; and angle of excavation relative to the soil, $p < 0.001$. There was an observed interaction between the variables $p < 0.001$ for Wilk test. The PCA analysis evidenced two highly distinct groups of burrows, which corresponded to the initial visual classification. The variance explained by axes 1 and 2 was 83.18 %. The variable that most influenced the distinction of the burrows of the species was the perimeter. There was a density of 114 foraging burrows/ha across the roads and 42 in shrubby grassland. The results of the ANOVA, $p < 0.05$, and Tukey test, $p < 0.05$, demonstrate significant difference between the mean densities of foraging burrows in the said environments. This indicates that armadillos use the area devoid of vegetation to feed.

Conclusions: Thus, this study provides information that can assist in the surveys of these species as well as to clarify aspects of its ecology in the Cerrado.

Keywords: Burrows, *Cabassous unicinctus*, Cingulata, Dasypodidae, *Euphractus sexcinctus*, Xenarthra

Background

Medium and large mammals found in the Cerrado (Brazilian Savanna) generally have a wide geographical distribution, and they are recorded commonly in other biomes. This fact is observed for most species of the order Cingulata, family Dasypodidae (Fonseca et al. 1996) that in the Cerrado are represented by five genera and eight species of the ten that are found in Brazil. These species are *Cabassous tatouay*, *Cabassous*

unicinctus, *Dasyus novemcinctus*, *Dasyus septemcinctus*, *Euphractus sexcinctus*, *Priodontes maximus*, *Tolypeutes matacus*, and *Tolypeutes tricinctus* (Redford 1994; Marinho-Filho et al. 2002; Medri et al. 2011). Apparently, species distribution and abundance within this biome are associated with phytophysiological formations and soil characteristics (Anacleto et al. 2006).

Armadillos in general have been studied in the Cerrado since the decade of 1980 (Carter and Encarnaç o 1983; Encarnaç o 1987). Nevertheless, there are gaps despite the biology and ecology of the species in the biome. According to Abba et al. (2007), the lack of attention given to the

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Cingulata order is due to the fact that these species are not charismatic. This occurs even regarding species classified globally as “vulnerable” (*P. maximus* and *T. tricinctus*) in the IUCN Red List of Threatened Species (Abba and Superina 2010). Thus, the field research on armadillos should be intensified to increase knowledge and conservation (Superina et al. 2014).

Armadillos, like most neotropical species, are rarely observed in nature. However, during their various activities, these species often leave characteristic signs in the environment: trails, burrows, and feces (Brito et al. 2001). Becker and Dalponte (1999) reported that when these signs are interpreted correctly, they can provide reliable identification of the species and also may reveal ecological features such as the following: habitat use, food habits, activities performed, and relative abundance. Animals of the genera *Cabassous*, *Euphractus*, *Priodontes*, and *Tolypeutes* are associated with drier and higher environments within the Cerrado (Redford 1985), whereas *Dasytus* sp. shows a preference for more humid sites (Schaller 1983), riparian forests, and footpaths. According to Arteaga and Venticique (2008), in the Brazilian Central Amazon, armadillos prefer to build their nests in areas of low elevation, such that topography is one piece of information that should be considered when evaluating habitat use and estimates of species density. Another important fact observed in this region is that armadillos use areas of primary vegetation more intensively (Arteaga and Venticique 2012).

Species belonging to the genera *Cabassous*, *Dasytus*, *Euphractus*, and *Priodontes* are efficient diggers. In contrast, *Tolypeutes* do not dig burrows, rather of this, use the burrows dug by other animals (Medri et al. 2011). The ability of armadillos to dig burrows is directly related with the construction of shelter (housing, raising offspring), protection (escape from predators), and feed (foraging burrows) (Redford 1985; McDonough and Loughry 2003; Bagagli et al. 2004). The species that dig their own burrows produce ones with peculiar shapes and sizes (Srbek-Araujo et al. 2009; Ceresoli and Fernandez-Duque 2012), a fact that is related to the anatomical differences (present in the osteology, muscle, and myology) (Vizcaino et al. 1999; Koneval 2003) and morphological characteristics (weight, size, carapace format and members) of these animals (Marinho-Filho et al. 1998; Medri et al. 2011).

Bonato et al. (2008) recorded in the Itirapina Ecological Station (*Estação Ecológica de Itirapina* (EEI)) four species of armadillos: *C. unicinctus*, *D. novemcinctus*, *D. septemcinctus*, and *E. sexcinctus*. They observed that the *Dasytus* occur at a low population density. The same was also seen for the density burrows of this genera in a pilot study (Trovati personal communication) because *Dasytus* dig burrows with a triangular entrance

(Anacleto and Diniz Filho 2008). The two most common species of armadillos in EEI *C. unicinctus* and *E. sexcinctus* may present total length (head, body, and tail similar - 64 cm) (Eisenberg and Redford 1999; Nowak 1999). However, on average, the first are smaller and lighter than the second (Redford and Wetzel 1985; Haysen 2014). The differences between species are kept in many other morphological and anatomical features that appear related to burrowing, as previously mentioned. Other differences are in the diet, *E. sexcinctus* is omnivorous, feeding on vegetal material (plants, roots and fruits, invertebrates, vertebrates, and carrion (Dalponte and Tavares-Filho 2004; Vaz et al. 2012)). While *C. unicinctus* is insectivorous, the diet of these consists of more than 90 % arthropods (chiefly ants and termites) (Bonato et al. 2008), but acarina and isoptera are also found (Anacleto 2007).

Based on the introductory context that was thought in the following hypothesis, the burrows of armadillos have different morphometrics and that the two species of armadillos here studied, as well as some carnivores, use the roads to get food. So this study has two objectives. The first was to compare the burrow morphometric variables of *C. unicinctus* and *E. sexcinctus*. The second was to determine whether foraging burrows occur more frequently in areas with vegetation (shrubby grassland) or in areas without vegetation (roads).

Methods

Study site

The Itirapina Ecological Station belongs to the geomorphological unit of the “Cuesta Basalt Province,” which is inserted in a region called the Plateau of São Carlos, wherein the altitude ranges from 705 to 750 m. This is located in São Paulo State, between the municipalities of Itirapina and Brotas at 22° 00' to 22° 15' S and 47° 45' to 48° 00' W (Motta-Junior et al. 2008; Trovati and Munerato 2013).

The EEI is a fragment of approximately 2.400 ha of the Cerrado, inserted in an array of areas of cultivated *Pinus* spp. (pine), *Eucalyptus* spp. (eucalyptus), *Saccharum officinarum* (sugarcane), *Citrus aurantium* (orange), and areas of exotic grassland (predominating *Brachiaria decumbens*) (Trovati and Munerato 2013). The principal physiognomies of this fragment are the following: 1.250 ha of shrubby grassland (*campo sujo*) and 370 ha of shrubby grassland with trees (*campo cerrado*). The landscape is also composed of flood plains and fragments of woody savanna (*cerrado sensu stricto*), gallery forests, swamp forest, and forest-like savanna (*cerradão*). Besides the native physiognomies mentioned, there are two areas in which *Pinus* sp. is cultivated and savannah vegetation native to the Cerrado is currently in the phase of regeneration. A third area of approximately 25 ha is

still used for *Pinus* spp. silviculture. Moreover, *Pinus* spp., *Eucalyptus* spp., *B. decumbens*, and *Melinis minutiflora* are invasive species in virtually all the EEI environments (Trovati 2009). The soil of the drier interfluves is primarily composed of sand (over 85 %); soils in wetter areas contain 60–70 % of clay or silt (Brasileiro et al. 2005).

Sample area

Sampling was conducted in an area that included shrubby grassland and the roads that cut through this environment. The soil in both road and shrubby grassland environments sampled showed similar compaction characteristics and the same composition (sand over 85 %). This area was chosen because it is the predominant physiognomy in the EEI, and according to Redford (1985), the species studied (*C. unicinctus* and *E. sexcinctus*) prefer the open, dry areas of the Cerrado. Shrubby grassland areas present a cover that is predominantly composed of grasses interspersed with sparse shrubs, typical country species, and a few individual small tree species. Among the shrubs, *Annona crassiflora* (marolo), *Annona coriacea* (araticum-liso), *Annona dioica* (Marolorasteiro), *Campomanesia pubescens* (guabi-roba), and *Caryocar brasiliense* (pequi) stand out, which according to Paulino-Neto (2014) constitute an important food resource to fauna, while among the small tree species, *Jacaranda caroba* and the palm trees *Attalea geraensis* (Indaiá-rasteiro) and *Syagrus petraea* (coco-de-vasoura) are worth highlighting.

Sampling and burrow morphometric variables

The sampling was performed using the linear transect methodology, with each one measuring 3 m width by 200 m length (600 m² per transect). The area surveyed considered only the roads that cut through the shrubby grassland physiognomy, with 11 sampling sites, in which only recently excavated burrows of *C. unicinctus* and *E. sexcinctus* were counted. An initial visual classification

of the burrows of *C. unicinctus* and *E. sexcinctus* was performed at each sampling point. This classification of armadillo burrows was based on the description proposed by Carter and Encarnaç o (1983), who described the burrows of *C. unicinctus* as circular and those of *E. sexcinctus* as an “inverted U” (Fig. 1).

However, visual classification is considered subjective, since its effectiveness is related to the observer’s experience (McDonough et al. 2000). Abba et al. (2007) indicated that the dimensions, shape of the burrows, and the observation of some of these being excavated could be a determinant in discriminating the excavator species. Thus, the current belief is that the morphometric characteristics of burrows can assist in making this visual classification less empirical.

Following the initial visual classification, the burrows were measured to determine the following morphological characteristics: depth, largest diameter (LD) or width, smallest diameter (SD) or height, and angle of excavation in relation to the soil. The width and height of the burrow opening were measured using a caliper constructed with 30-cm rulers. The ratio of these diameters (LD/SD) and the perimeter of the ellipse (perimeter) were calculated using the division of the largest diameter by the smallest diameter. To measure the angle of burrow excavation in relation to the soil, an inclinometer was used.

To assess the relationship between foraging burrows in the road and shrubby grassland environments, the linear transect methodology was used. The specific measure of each of linear transects was 600 m². However, this time, the area surveyed included both the road and shrubby grassland physiognomy; 21 points were haphazardly sampled in each environment totaling 1.26 ha. Again, only recently excavated burrows were counted in order to estimate the density in each environment. In addition, the depth of these burrows was measured. Burrows to a depth of less than 70 cm were considered foraging burrows because the armadillo species studied

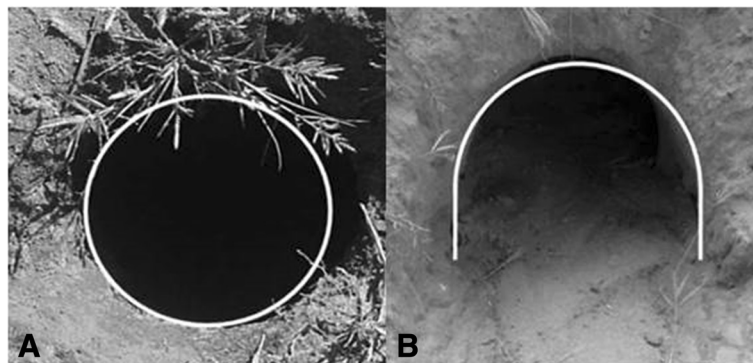


Fig. 1 Burrows of *C. unicinctus* (a) and *E. sexcinctus* (b) in the Estação Ecológica de Itirapina

here generally have the size (length of body and tail) near this measure (Redford and Wetzel 1985; Eisenberg and Redford 1999; Hayssen 2014).

Data analysis

Comparison between the variables measured, depth, ratio, perimeter, and angle of excavation in relation to the soil, for the burrows of *C. unicinctus* and *E. sexcinctus*, was performed by the multivariate analysis of variance (generalized MANOVA). Furthermore, to test the initial visual classification of the burrows, Principal Component Analysis (PCA) was performed using the Multivariate Statistical Package (MVSP) program (Kovach 1999).

Finally, to compare the occurrence of the burrows in the road and shrubby grassland environments, an analysis of variance (ANOVA) was carried out, followed by Tukey test. The generalized MANOVA, ANOVA, and Tukey test were performed using the MINITAB program, version 17.0 (Minitab 2015).

Ethics committee

Permit to this study were granted by Instituto Brasileiro do Meio Ambiente e dos Recursos Naturais Renováveis (IBAMA 036/2007- CGFAU) and the Instituto Florestal de São Paulo with the research license for the project entitled “Mammals burrowing (Dasypodidae and Echimyidae) Cerrado of Itirapina region and its role in terrestrial vertebrate communities”.

Results

A total of 180 burrows (80 previously classified as pertaining to *C. unicinctus* and 100 to *E. sexcinctus*) were recorded, with a mean density of 7.27 and 16.63 burrows per transect, corresponding to a density of 121 and 277 burrows/ha to respective species.

Although some degree of overlap occurred among the morphometric variables of burrows of *C. unicinctus* and *E. sexcinctus*, significant differences were observed for all the variables: depth, $p < 0.001$; ratio, $p < 0.001$; perimeter, $p < 0.001$; and angle of excavation relative to the soil, $p < 0.001$ (Table 1). There was an observed interaction between the variables, $p < 0.001$ for Wilk test.

The PCA analysis evidenced two highly distinct groups (Fig. 2), which corresponded to the initial visual classification, except for a single *C. unicinctus* burrow, which

was classified as *E. sexcinctus*. The identity of the only burrow misidentified in the initial classification was corrected for all the analysis performed. The variance explained by axes 1 and 2 of the PCA was 83.18 %. The variable that most influenced the distinction of the burrows of the species studied here was the perimeter, followed by the variables, ratio, angle of excavation in relation to the ground, and burrow depth (Table 2).

Regarding foraging burrows found in the different environments studied (road and shrubby grassland), there were recorded 144 burrows in the road and 53 burrows in the shrubby grassland. The mean density of foraging burrows for 21 points sampled on the roads and shrubby grassland were 6.86 (standard error (SE) ± 1.53) and 2.52 (SE ± 0.642), respectively. The results of the ANOVA, $p < 0.05$, and Tukey test, $p < 0.05$, demonstrate a significant difference between the mean densities of foraging burrows in the said environments. This shows that the shrubby grassland environment of the EEI has an estimated density of 114 foraging burrows/ha in areas with vegetation (shrubby grassland) and 42 in areas without vegetation (roads).

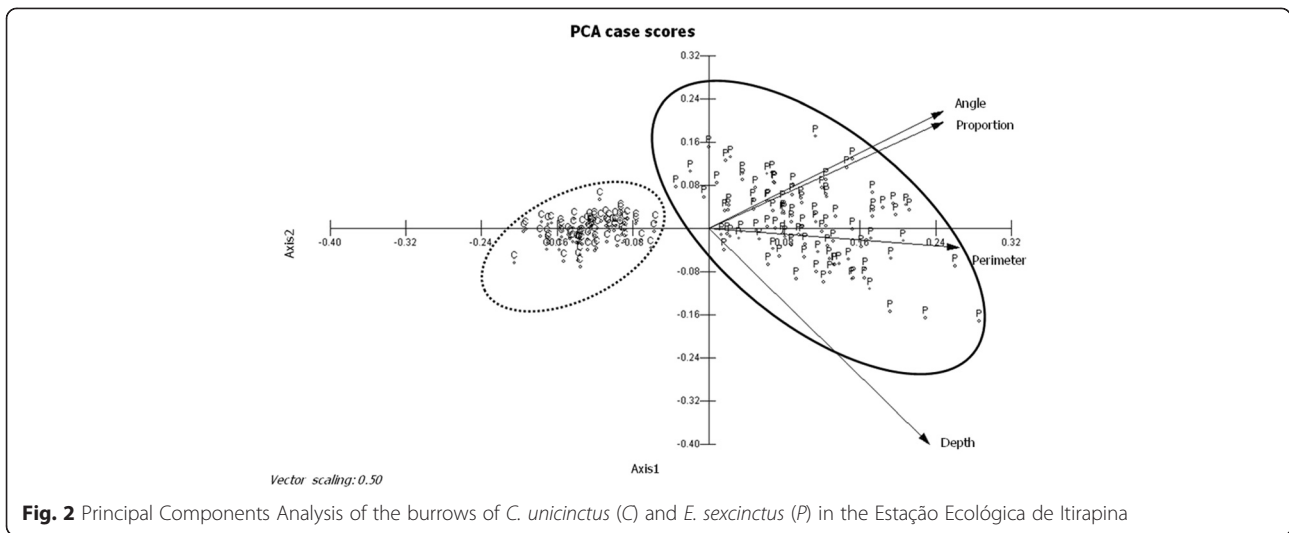
Discussion

The density of armadillo burrows in the shrubby grassland area probably was considered high for both species in the EEI. However, even though he knew the importance of comparison of densities with similar studies to have parameter settings, it did not find any information about it in the literature. The density of burrows of *E. sexcinctus* was twice higher than that registered to *C. unicinctus*. Lima Borges and Thomas (2004) report, that *E. sexcinctus* can make agglomerates burrows in open areas. (2004) report that *E. sexcinctus* can make agglomerate burrows in open areas. Additionally, Redford and Wetzel (1985) cite that armadillo species can reuse old burrows. Some species of the genus *Cabassous* change new burrows all night (Encarnaç o 1987; McDonough and Loughry 2003). However, there is no information about the theme to *C. unicinctus*. Based on the information that has to home range, which is up to 101.06 ha for *C. unicinctus* and up to 958.0 for *E. sexcinctus* (Encarnaç o 1987), it is believed that these species dig various burrows during their lifetime. This probably makes *E. sexcinctus* have to dig more burrows.

Table 1 Number and morphometry of burrows of *C. unicinctus* and *E. sexcinctus* (mean \pm standard error and minimum and maximum measurements) at Estaç o Ecol gica de Itirapina

Dasypodidae		Morphometric variables			
Species	N	Depth (cm)	Ratio (cm)	Perimeter (cm)	Angle (�)
<i>C. unicinctus</i> ^a	80	37.50 \pm 0.84 (18–60)	1.01 \pm 0.01 (0.9–1.0)	38.40 \pm 0.36 (32.9–39.8)	65.00 \pm 1.22 (45–90)
<i>E. sexcinctus</i> ^b	100	65.36 \pm 2.7 (18–150)	1.19 \pm 0.01 (0.9–1.4)	59.32 \pm 0.82 (38.6–75.3)	54.56 \pm 0.97 (30–90)

Letters a and b indicate the significant difference between the burrows of the species for all variables



The burrows excavated by armadillos, which are used for sleeping, nesting, escaping predators, thermoregulation, or creating an insect reservoir, are dug in well-drained soils or into ant and termite mounds (McDonough and Loughry 2003, 2008).

Nonetheless, in the same study area, Bonato et al. (2008) obtained inverse values regarding the density of individual armadillos, estimated using a mark-recapture method, showing 0.26 individuals/ha for *C. unicinctus* and 0.14 individuals/ha for *E. sexcinctus*. Probably, the higher density of *E. sexcinctus* burrows is related to the behavior of digging more burrows compared to *C. unicinctus*, and not with its abundance (density), which could be explained by the fact that the latter armadillo species is more fossorial (Wetzel 1980). When it is assumed that *C. unicinctus* is more fossorial than *E. sexcinctus*, it is believed that this can spend longer periods of time in the subsurface and therefore produces less burrows. This is possibly explained by its anatomic difference. The relative development of the olecranon process is interpreted as an improvement of the mechanical advantage of the triceps muscle, the forearm extensor. *C. unicinctus* has a better anatomical development than *E. sexcinctus* (Vizcaíno et al.

1999). But differences in morphological characteristics (Marinho-Filho et al. 1998; Medri et al. 2011) and feed between the two species should not be forgotten (Dalponte and Tavares-Filho 2004; Anacleto 2007; Bonato et al. 2008; Vaz et al. 2012).

The differences in depth, ratio, and perimeter also can be explained by anatomical and morphological characteristics of each species. *C. unicinctus* measures 34–44 cm, with a tail of 16.5–20 cm (Eisenberg and Redford 1999; Nowak 1999; Hayssen 2014), and weighs between 2.2 and 4.8 kg when adult (da Merrit 1985; Redford 1994; Hayssen 2014). The carapace has 10 to 13 movable bands (Emmons and Feer 1997). The tail is leather (devoid of corneal plates), but the main feature is the strong claws (Marinho-Filho et al. 1998), with the largest claw being sickle shaped (Medri et al. 2011). In contrast, *E. sexcinctus* is larger, measuring 46–54 cm in rostrum-anal length, with a tail of 22.2 cm on average, and weighs around 3 to 6 kg (Redford and Wetzel 1985; Eisenberg and Redford 1999; Anacleto et al. 2006). The head of this kind is conical and flat on top. The carapace is semicircular in shape, with 6 to 8 movable bands and long hair. The tail is long and protected by corneal rings (Silva 1994). However, these same characteristics are also overlapped, which can be explained by the juxtaposition of the morphological measurements of these species.

Minimum burrow depth may be controlled primarily by factors other than the short-term energy cost of burrowing. Shallow tunnels in sandy soil readily cave in as the soil dries out. Shallow tunnels dry out more quickly, are subject to greater temperature fluctuations, and are easily destroyed by any large animals stepping on the surface above them (Vleck 1981). Thinking about the ratio and perimeter of the burrows, the energy cost of burrowing is directly proportional to the mass of soil removed. Thus, cost of burrowing is roughly proportional to the square

Table 2 Comparison of values of the Principal Components Analysis for the variables (depth, angle, ratio, and perimeter) of the burrows of armadillos *C. unicinctus* and *E. sexcinctus* in the Estação Ecológica de Itirapina

Variables	Axis 1	Axis 2
Percentage	69.81	13.37
Depth ⁴	0.47	-0.80
Ratio ²	0.49	-0.39
Perimeter ¹	0.53	-0.07
Angle ³	0.49	0.43

Numbering from 1 to 4 indicates the order of influence of the variable in separating the burrows, 1> influence 4< influence

root of body mass (Vleck 1981). This reflects a lower energetic cost of *C. unicinctus* in relation to *E. sexcinctus* to remain in the subsoil.

The difference observed in the angle of excavation of the burrows shows that, in general, *E. sexcinctus* prefers to dig burrows at a lesser angle of inclination than *C. unicinctus*, whose burrows are more perpendicular to the soil. This can once again be explained by the differences in anatomical and morphological structure of forelimbs and claws of the species. *C. unicinctus* has short forelimbs and particularly well-developed, strong, curved claws (Marinho-Filho et al. 1998), with the largest of these claws shaped like a sickle (Medri et al. 2011), while *E. sexcinctus* has members and less robust muscles and is prone to digging (Vizcaino et al. 1999). The forelimbs as hind limbs have five fingers, with claws, the second finger being the most developed (Pocock 1924).

Seabloom et al. (2000), using *Thomomys bottae* as a model, found that the cost of tunnel construction is independent of hill slope angle and that the costs of shearing soil and pushing soil horizontally through the tunnels were three orders of magnitude greater than the costs of lifting the soil against the force of gravity. This fact strengthens the increased ease of *C. unicinctus* to remain in the subsoil due to its lower mass corporeal. This provides excavation at an almost straight angle, thus decreasing energy expenditure. Thus, the optimum angle for constructing a burrow that involves the lowest energy cost is perpendicular to the surface (Arteaga 2004), especially in sandy soil (Vleck 1981). However, for *C. unicinctus* that dig their burrows in shallow angle seems to be more associated with their anatomy than the soil type. But this needs to be better studied.

The PCA showed a clear distinction between the burrows of these two species of armadillo. Generally, all the variables had some influence on differentiating the burrows, but the perimeter was a determining factor for distinguishing them. According to Carter and Encarnaç o (1983), Redford and Wetzel (1985), and Parera (2002), *E. sexcinctus* typically constructs burrows with an “inverted U”-shaped opening. The burrows of *C. unicinctus*, on the other hand, present a circular shape, since they dig while twisting their body in helical movements (Carter and Encarnaç o 1983). The difference in the burrows between *C. unicinctus* and *E. sexcinctus* is evident and easily identified in the field by the format of the burrow opening (Fig. 1). However, the observer does require some degree of experience. Thus, when the observer is inexperienced, calculating the perimeter can assist in identifying whether the burrow belongs to *C. unicinctus* or *E. sexcinctus*. According to Srbek-Araujo et al. (2009) and Ceresoli and Fernandez-Duque (2012), the most species that dig their own burrows, produce of specific size and shape.

The high density of burrows found in the road area in this study could be explained by the fact that armadillos also use this environment to feed. Redford (1985) mentioned that the shallower burrows of armadillos are commonly for foraging. It seems unlikely that shallow burrows, those with a depth of less than 70 cm, would provide shelter for these animals because they would make it easier to be caught by predators (Tozetti and Granzinolli 2000). Even though the road and adjacent areas are frequently disturbed and are often hostile environments for many species of wildlife, they can provide attractive features, including shelter, food, or nesting sites and could even facilitate the displacement of some species (Seiler 2001).

Additionally, according to Taraborelli et al. (2009), predation risk would be related to vegetation structure. For behavioral patterns of digging burrows in an area, beneath the cover provided by trees, shrubs, and herbaceous plants, shrubs and trees would afford vertical protection from raptors, but herbaceous plants would obstruct visual detection of terrestrial mammalian predators and of the shadow of raptors on the ground (Taraborelli et al. 2008). EEI has potential predators of *C. unicinctus* and *E. sexcinctus*, and these are the following terrestrial carnivores: *Leopardus pardalis*, *Puma concolor*, *Procyon cancrivorus*, *Cerdocyon thous*, and *Chrysocyon brachyurus* (Trovati 2009), with *Harpyhaliaetus coronatus* being the only possible raptor (Trovati 2009; Motta-Junior et al. 2008). This strengthens the idea that the preference of armadillos feed on the road in the study area is related to lower predation risk.

Conclusions

It can be concluded that this study provides information on relatively common species of armadillos in the Cerrado biome and which are usually forgotten. Thus, further studies are needed with fossorial mammals in the neotropical region, as these are possibly keystone species, given that their holes not only are influenced by biotic and abiotic factors but also act on these factors.

Competing interests

The author declares that he has no competing interests.

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