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RESEARCH ARTICLE - ANTS

Ant assemblages (Hymenoptera: Formicidae) in three different stages of forest regeneration in a fragment of Atlantic Forest in Sergipe, Brazil.

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Abstract

In this study we compared the epigeic ant assemblages in forest fragments with three different status of plant recovery (an area reforested in 2007, another reforested in 2005 and another one of secondary forest, with over 35 years of plant regeneration), located in the municipality of Laranjeiras, Sergipe, Brazil. The ants were sampled in February (dry season) and June (rainy season) of 2012. We tested the following hypotheses: (1) the species richness of ants increases with time after the process of forest restoration; and (2) there are significant changes in species composition of ants among the three stages of forest regeneration. Twenty-five pitfall traps were installed in each area. A total of 82 morphospecies of ants were sampled, distributed in 31 genera and seven subfamilies. The richness of ants was similar among the three sites ($F = 1.71$, $p = 0.19$). The composition of ant species, however, was different in the area of late regeneration (35 years) compared to other areas of early reforestation ($p < 0.05$). Thus, epigeic ants were partially sensitive to changes in the habitat studied in response to reforestation, presenting changes in species composition but no differences in ant species richness among areas. We conclude that seven years after reforestation are not enough to restore the same ant diversity in disturbed environments.

Introduction

Remaining forests are important for the maintenance of favorable environmental conditions for the establishment and persistence of native fauna (Gibson et al., 2011; Ulyshen, 2011). Several studies have shown that part of the worldwide decline in biodiversity, threatening the functioning of ecosystems, is related to anthropogenic modification of the landscape (Dirzo & Raven, 2003; Colombo & Joly, 2010; Tabarelli et al., 2010), including the Atlantic Forest, one of the main hotspots in the world (Myers et al., 2000).

Deforestation of Atlantic Forest is considered a constant threat to biological diversity (Melo et al., 2009; Oliveira et al., 2004), including ant assemblages (Leal et al., 2012) and the monitoring of areas in process of plant recovery can be an important tool in the diagnosis of these threats (Conceição et al., 2006; Delabie et al., 2006).

Due to its abundance in most terrestrial ecosystems ants are considered ecologically dominant and play complex ecological roles such as ecosystem engineers, predators, herbivores and seed dispersal agents (Hölldobler & Wilson, 1990; Folgarait, 1998). In tropical ecosystems the importance of ants is more evident because they can represent up to 60% of all arthropod biomass, and approximately 90% of their abundance (Hölldobler & Wilson, 1990; Floren & Linsenmair, 1997).

Species richness and structure of ant assemblages can be used as response variables in environmental monitoring, as these insects are sensitive to anthropogenic activities, including agricultural practices (Hernández-Ruiz & Castaño-Meneses, 2006) and reforestation (Pais & Varanda de 2010; Schmidt et al., 2013). Therefore, the study of these insects is useful to assess the success of forest restoration practices (Sobrinho et al., 2003; Silva & Silvestre, 2004; Holway & Suarez, 2006; Wetterer, 2012).

Although recovery of degraded areas is commonly used to reduce the negative environmental impacts on forest remnants (Metzger, 2009; Calmon et al., 2011) and, in spite of several studies on the role of the replanting of native species in accelerating the recovery of degraded environments, there are still many questions about the time required for the recovering of ant fauna along a gradient of forest regeneration

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of fragments dominated previously by an agricultural matrix (Neves et al., 2010; Teodoro et al., 2010; Leal et al., 2012).

Processes that influence the structure and species diversity of epigeic ants in agroecosystems are still poorly known (Neves et al., 2010; Teodoro et al., 2010), despite the increasing conversion of forest fragments in less diverse and structurally simple habitats (Primack & Corlett, 2005; Barona et al., 2010).

In this study, we investigated the response of epigaeic ant assemblages in forest fragments with three different status of plant recovery, aiming to test the following hypotheses: (1) Species richness of ants increases with time after the process of forest restoration (following an increase in habitat complexity) and (2) the composition of ant species undergoes changes along a gradient of regeneration of reforested area, with reduction of generalist species.

Material and Methods

The study was conducted in three sites: two sites were previously plantations of sugar-cane that were reforested, one with 32ha in 2005 (RF1) and another with 30.7ha and reforested with native species in 2007 (RF2). The third area is a secondary Atlantic Forest fragment with 55ha (FF) used as "Area of permanent preservation" (APP). All sites are located at Fazenda Boa Sorte, a large sugar-cane company, located in the municipality of Laranjeiras (10° 48' 44"S, 37° 10' 16" W), state of Sergipe, Brazil.

The studied region is dominated by agricultural land with altitude ranging from 30 to 68 m a.s.l. The mean annual temperature is 25.5 °C and annual average rainfall of 1,200 mm. The rainy season usually lasts from May to October. The original vegetation was dominated by Atlantic forest and all remnants are embedded in a 20 year-old, homogeneous matrix of sugar-cane fields (Cuenca & Mandarino, 2007). The soil type is Spodozol, mainly sandy clay, deep, with low fertility and high porosity (draining rainfall). The area reforested in 2005 (RF1) with 32 ha, is at an intermediate stage of development, with seven years of planting and composed of trees with canopy of approximately 4-6 meters (10°49'15,8"S; 37°09'41"W). The other area, reforested in 2007 (RF2), with 30.7 ha is in early stage of development, with five years of planting and composed by sparse patches of woody vegetation, shrubs, herbs and grasses with a single layer of treetops with up to 4 m tall (10°49'01,6"S; 37°09'40,7"W).

Fourteen species of trees native to the Atlantic Forest were used in reforestation: *Tapirira guianensis*, *Caesalpinia echinata*, *Genipa amerciana*, *Spondias lutea*, *Schinus terebinthifolius*, *Erythrina velutina*, *Enterolobium contorsiliquum*, *Cleome tapia*, *Caesalpinia leiostachya*, *Inga marginata*, *Cassia grandis*, *Lonchocarpus sericeus*, *Anadenanthera macrocarpa* and *Hymenaea courbaril*

The fragment (FF) of Atlantic Forest is an area of secondary forest, protected from logging for over 35 years and consists of trees with 7-20 meters in height that forms a closed canopy (10°49'17"S; 37°11'13"W).

Epigeic ants were sampled in 15 transects of 50 m, being five transects per area. We established a minimum distance of 150 m between each transect. Ant sampling was conducted using pitfall traps on the ground surface. In each transect, five pitfalls were installed at a distance of 10 m, totaling 25 pitfalls/site. Pitfalls consisted of 1,000 cm^3 plastic pots containing approximately 120 cm^3 water with detergent and were kept for 48 h in the field (Schmidt & Solar, 2010).

Sampling was conducted in two periods, one during the dry season (February 2012) and another during the rainy season (June 2012). All ants collected were sorted to species level when possible or morphospecies, using identification keys from Bolton (1994) and Fernandez (2003) and later the identification was confirmed through comparison with specimens from the collection of the Laboratório de Ecologia de Comunidades (Ant collection), of the Universidade Federal de Viçosa and Laboratório de Mirmecologia of the CEPEC/CEPLAC, Ilhéus, Bahia, Brazil. Voucher specimens of all species are deposited in Laboratório de Pragas Florestais of the Universidade Federal de Sergipe.

To verify the effect of habitat type (RF1, RF2, and FF) and sampling period (wet or dry season) (response variables) on the species richness of ants (explanatory variable) the linear mixed effect (LME) was used, followed by residuals analysis to verify the adequacy of the error distribution and the fit of the model. Fixed factors were sampling sites (RF1, RF2, and FF), while samples (nested within sites) were treated as random factors. A minimum adequate model (MAM) was obtained by extracting non-significant terms ($P < 0.05$) from the full model arranged by all variables and their interaction (Crawley, 2007), using the software R (R Development Core Team, 2009).

A non-metric multidimensional scaling (NMDS) analysis was carried out to verify differences in the composition of ant fauna among the forest regeneration types (Neves et al., 2010). The ordination was conducted using the Jaccard index. Additionally, similarity analysis (ANOSIM; Clarke, 1993) were conducted to compare the difference between two or more groups of sampling units among sites. Differences between R-values were used to determine similarity patterns among ant assemblages in the three sites. The analysis were conducted using the software PAST (Hammer et al., 2001).

Results

We collected 82 ant morphospecies, distributed in 31 genera (Table 1). The subfamilies Myrmicinae and Formicinae presented 66% of all ant species sampled, with 42 and 12 morphospecies, respectively. The genera *Pheidole* and *Camponotus* presented the higher richness with 11 (13.5%) and 10 (12%) morphospecies, respectively. Twelve species were restricted to RF1 site, six species were found exclusively in RF2 site while 34 species were restricted to FF site (Table 1).

Table 1. Relative frequency of epigeic ant species collected in pitfalls, during the wet and dry season of 2012 in three sites of different forest regeneration stages: a fragment of secondary forest (FF) one area of reforestation with five years (RF2) and other with seven years of reforestation (RF1).

Ant Subfamilies	Occurrence in each season					
	FF		RF1		RF2	
	Dry	Wet	Dry	Wet	Dry	Wet
DOLICHODERINAE						
Dolichoderus lutosus	-	0.2	-	-	-	-
Dolichoderus diversus	-	0.2	-	-	-	-
Dolichoderus attelaboides	-	0.2	-	-	-	-
Dorymyrmex biconis	-	-	-	-	0.2	-
Azteca sp. 1	0.8	0.4	-	-	-	-
Azteca sp. 2	0.2	-	-	-	-	-
Azteca sp. 3	-	0.2	-	-	-	-
ECITONINAE						
Labidus praedator	-	0.4	-	-	-	-
Labidus coecus	-	-	0.4	-	0.4	-
Nomamyrmex esenbeckii	-	-	-	0.2	-	-
Neivamyrmex diana	-	-	-	0.2	-	-
FORMICINAE						
Brachymyrmex pr. *patagonicus*	-	-	-	0.2	-	-
Camponotus trapezoideus	0.2	-	-	-	-	-
Camponotus renggeri	1	0.4	-	-	-	-
Camponotus bispinosus	0.2	-	-	-	-	-
Camponotus novogranadensis	0.6	1	-	-	-	-
Camponotus fastigatus	1	-	-	-	-	-
Camponotus arboreus	0.2	-	0.2	-	-	-
Camponotus cingulatus	0.8	-	-	-	-	-
Camponotus vittatus	-	0.4	0.8	1	0.6	0.8
Camponotus (*Myrmaphaenus*) sp.9	-	-	1	0.8	0.8	-
Camponotus rufipes	-	-	0.4	-	0.2	1
Nylanderia pr. *fulva*	-	-	0.4	-	0.2	-
MYRMICINAE						
Piramica pr. *perpava*	0.4	0.4	-	-	-	-
Piramica sp. 2	-	0.2	-	-	-	-
Cephalotes atratus	0.6	-	-	-	-	-
Cephalotes umbraculatus	0.2	-	-	-	-	-
Cephalotes minutus	-	-	-	0.2	-	-
Cephalotes maculatus	0.2	-	-	-	-	-
Cephalotes pusillus	-	-	-	0.2	-	-
Cephalotes depressus	-	-	-	-	-	0.4
Acromyrmex balsani	-	-	0.6	-	0.4	0.2
Acromyrmex rugosos rugosos	-	0.6	0.4	0.2	0.2	-
Atta sexdens rubropilosa	-	-	0.2	-	0.2	-
Cyphomyrmex minutus	-	0.2	-	-	-	-
Cyphomyrmex transversus	-	-	0.6	0.2	0.8	0.4
Mycetosoritis sp. 1 *	-	-	-	-	-	0.2
Mycocepurus obsoletus	-	0.4	-	-	-	-
Sericomyrmex sp. 1	0.2	0.2	-	-	-	-
Sericomyrmex sp. 2	-	0.4	-	-	-	-
Trachymyrmex sp. 1	0.2	-	-	-	-	-
Monomorium floricula	-	-	0.2	-	0.8	-
Solenopsis tridens	-	-	0.2	0.2	-	-
Solenopsis sp. 2	0.2	0.6	-	-	-	-
Solenopsis sp. 3	0.6	-	-	-	-	-
Solenopsis saevissima	0.2	-	0.6	0.6	0.8	0.4
Solenopsis globularia	-	-	0.2	1	-	0.8
Hylomyrma balzani	0.2	0.2	-	-	-	-
Pheidole radoszkowskii	0.4	0.4	1	1	0.6	1
Pheidole fimbriata	-	-	0.2	0.4	-	-
*Pheidole (*gr. *Diligens)* sp. 3	0.4	0.8	-	-	-	-
Pheidole sp. 4	0.6	1	-	-	-	-
*Pheidole (*gr. *Flavens)* sp. 5	0.2	0.4	-	0.4	-	-
*Pheidole (*gr. *Tristis)* sp. 6	0.8	0.4	0.2	-	-	-
*Pheidole (*gr. *Fallax)* sp. 7	-	-	1	0.4	0.8	1
*Pheidole (*gr. *Diligens)* sp. 8	-	-	-	0.2	1	0.2
Pheidole sp. 9	-	-	-	-	-	0.4
*Pheidole (*gr. *Fallax)* sp. 10	-	-	0.4	0.6	0.2	-
*Pheidole (*gr. *Fallax)* sp. 11	0.6	0.8	0.8	0.4	0.8	0.4
Crematogaster abstinens	-	-	1	0.4	0.4	1
Crematogaster sp. 2	-	-	0.2	-	-	-
Crematogaster pr. *Distans*	-	-	-	-	0.6	-
Crematogaster sp. 4	0.4	-	-	-	-	-
Crematogaster sp. 5	0.2	-	-	-	-	-
Cardiocondyla emeryi	-	-	0.2	-	0.2	-
PONERINAE						
Odontomachus haematodos	1	1	1	0.6	0.8	1
Leptogenys unistimulosa	1	0.6	0.6	0.8	1	0.4
Hypoponera sp. 1	0.2	-	-	-	-	-
Pachycondyla venerae	0.4	0.4	-	-	-	-
Pachycondyla harpax	0.4	-	-	-	1	-
ECTATOMMINAE						
Gnamptogenys acuminata	0.2	-	-	-	-	-
Gnamptogenys sulcata	-	-	0.4	-	0.4	-
Ectatoma bruneunn	-	-	0.4	-	0.6	0.6
Ectatoma tuberculatum	0.2	-	0.6	0.2	-	-
Ectatoma edentatum	0.4	0.8	0.2	0.6	-	-
PSEUDOMYRMECINAE						
Pseudomyrmex tenuis	0.8	0.6	-	-	-	-
Pseudomyrmex termitarius	-	-	0.2	-	0.2	-
*Pseudomyrmex sp. (*gr. *Pallidus)*	-	-	-	-	0.2	-
Pseudomyrmex sp. 4	-	-	-	0.4	-	-
Pseudomyrmex gracilis	-	-	0.2	-	-	-
Pseudomyrmex sp. 6	-	-	0.2	-	-	-
Pseudomyrmex sp. 7	-	-	0.2	-	-	-

FF34 ve/ RF1.12 vd/ RF2.6 p/ Comuns30; * New genus sp.1, R. Feitosa (personal communication, 19 September 2012)

There was no significant difference in species richness of ants among the three sites of forest regeneration ($F_{2,22}$ = 2.26, p = 0.12). However, the species richness of ants was lower in the wet season in the RF1 and RF2 sites, compared to FF area ($F_{1,22}$ = 11.19, p = 0.002) (Fig.1).

The NMDS analysis indicates the formation of two distinct groups (stress = 0.15) with one group represented by FF and another group formed by RF1 And RF2 (Fig.2). Besides, the analysis of similarity (ANOSIM) indicated significant difference in the structure of ant assemblages between FF *versus* RF1 (p = 0.003) as well as FF *versus* RF2 (p = 0.001) (Table 2).

The SIMPER analysis indicated that the morphospecies that contributed most to the differentiation among sites were *Pheidole* (group *Fallax*) sp.7, *Camponotus* (*Myrmaphaenus*) sp 9, *Crematogaster abstinens*, *Camponotus vittatus*, *Solenopsis saevissima*, *Pheidole* sp. 4, *Cyphomyrmex transversus, Solenopsis globularia*, *Ectatoma edentatum*, *Camponotus renggeri* and *Pseudomyrmex tenuis*. These morphospecies together contributed to 31.5% of cumulative dissimilarity among stages of plant recovery (Table 3).

Table 2. Analysis of similarity (ANOSIM) among three sites of different forest regeneration stages: a fragment of secondary forest (FF) one area of reforestation with five years (RF2) and other with seven years of reforestation (RF1).

	FMN	RF1	RF2
FMN	-	0.0003**	0.0001**
RF1	0.0003**	-	0.0732
RF2	0.0001**	0.0732	-

** significant difference, p< 0,01.

Discussion

In our study the species richness did not differ with time of restoration and, on the one hand, it suggests that five years are enough for the recovery of ant species richness. This time can be considered short compared to that from other studies conducted by Vasconcelos (1999) and Roth et al. (1994) (10 and 25 years, respectively). On the other hand, however, the differences found in species composition among FF and RF1 or RF2 indicate that other parameters, rather than species richness, are important to make decisions about the use of ants as bioindicators. An increase in species richness in FF might be correlated with a more complex environment, leading to an increase in availability of resources (Matos et al., 1994, Oliveira et al., 1995).

The restoration of RF1 and RF2 sites with native species of trees might have created favorable conditions for colonization of ants and have led to comparable values of species richness in FF area. The colonization of ants, however, seems to have been carried out by ant species from adjacent agroecosystems and not from nearby forested areas, since the composition of species between FF and the other two areas differ greatly. Difference in ant species composition between more complex areas and regenerating ones have been found by other authors (Vasconcelos, 1999; Schmidt et al., 2013). In general, generalist species have higher colonization rate of disturbed fragments than do specialist ants (Schoereder et al., 2004).

Although there were no differences in species richness among sites, there were differences between season of sampling, with higher values in the dry season of RF1 and RF2 (seasonality was not tested as a hypothesis in this study, and thus we used this variable only as a source of variation in the statistical model).

Table 3. SIMPER Analysis among three sites of different forest regeneration stages: a fragment of secondary forest (FF) one area of reforestation with five years (RF2) and other with seven years of reforestation (RF1).

Species	Cumulative percent of dissimilarity (%)
Pheidole (group *Fallax*) sp.7	3.407
Camponotus (Myrmaphaenus) sp.9	6.709
Crematogaster abstinens	9.905
Camponotus vittatus	12.89
Solenopsis saevissima	15.77
Pheidole sp. 4	18.58
Cyphomyrmex transversus	21.25
Solenopsis globularia	23.92
Ectatoma edentatum	26.59
Camponotus renggeri	29.05
Pseudomyrmex tenuis	31.51

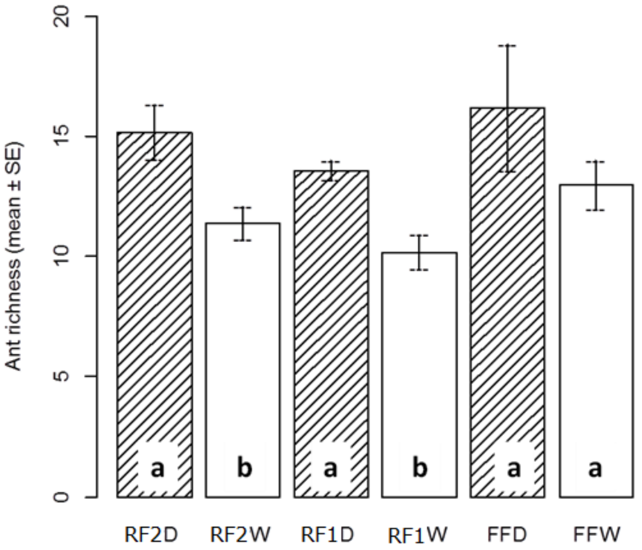

Figure 1. Species richness of ants in the dry season (white bars) and rainy season (hatched bars) (mean ± SE) sampled in three areas with different stages of forest recovery. RF1 = Fragment with 7 years of reforestation; RF2 = Fragment of 5 years of reforestation and FF = forest fragment with 35 years of plant recovery (D and W indicate sampling in dry and wet seasons, respectively). Different letters on bars indicate significant difference within the same site (p <0.05).

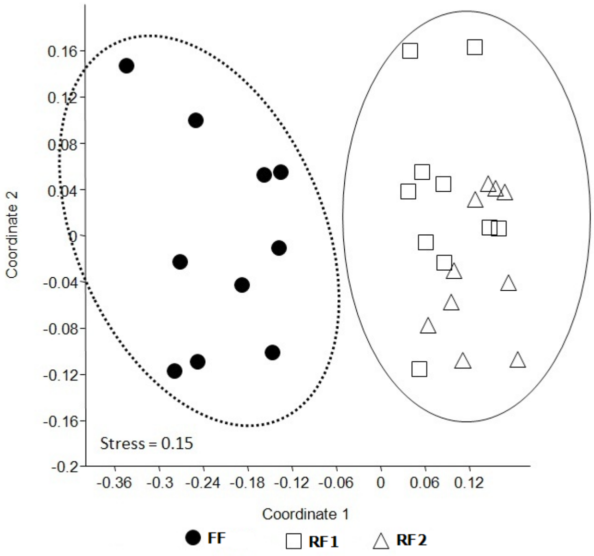

Figure 2. Analysis of non-metric multidimensional scaling ordination (NMDS) from the assemblage composition of ants in three sites of different forest regeneration stages: a fragment of secondary forest (FF) one area of reforestation with five years (RF2) and other with seven years of reforestation (RF1).

Changes in the frequency of foraging ants have been observed with environmental seasonality (Wolda, 1988; Kaspari, 2000; Castro et al., 2011; Cook et al., 2011). The availability of resources is reduced during the dry months of the year, and the increased mobility of ants in this period might explain the rise in species richness in the dry season (Andow, 1991; Dantas et al., 2011). These results, however, should be viewed with caution since the data were collected considering just a year and a longer period of collection is needed to establish more secure inferences about effects of seasonality on ant species.

Considering that thirty-four species were restricted to FF site (42% of all species found) our results make it clear that environmental differences among the areas were crucial for determining the composition of species.

The most common species in the FF were *Camponotus renggeri*, *Pheidole* sp.4 and *Pseudomyrmex tenuis*, indicating a preference of these species for less-disturbed environments. Notably, several studies also demonstrate the occurrence of *C. renggeri* in seasonal forest formations such as Cerrado (Del-Claro & Oliveira, 1999; Christianini et al., 2007; Neves et al., 2012) and forests in the semi-arid region of Brazil (Hites et al., 2005). Conceição et al. (2006) also have reported the presence of *P. tenuis* in environments with low disturbance. Besides, predatory species of the genus *Pyramica* (Masuko, 2009) were also found only in the FF, suggesting that this fragment has a more suitable resource availability than the other areas.

The genera *Pheidole* (group *Fallax*) sp.7, *Camponotus* (*Myrmaphaenus*) sp.9, *Crematogaster abstinens*, *Solenopsis globularia* and *Cyphomyrmex transversus*, were found in RF1 and RF2, indicating that these species could have preference for colonization in degraded areas. Other studies in the Atlantic Forest recorded *S. globularia* in a disturbed mangrove area (Delabie et al., 2006) and *C. transversus* in grasslands (Braga et al., 2010). In fact, the areas RF1 and RF2 have low density of tree species, allowing the establishment of herbaceous species.

Individuals of *Solenopsis saevissima* exhibit aggressive behavior and are also usually associated with disturbed environments (Silvestre et al., 2003). The presence of this species is favored in sites colonized by pioneer plant species, typically found in early sucessional areas (Vasconcelos, 2008; Schmidt et al., 2013). Although *Camponotus vittattus* was sampled in all three areas, this species had similar occurrence to *S. saevissima* being more frequent in samples from RF1 and RF2, thus suggesting its preference for opened sites. In contrast, we also reported the presence of some ant species in sites with late regeneration time (RF2 and FF), such as *Ectatoma edentatum*. Previous studies have associated the occurrence of this species with advanced stages of plant recovery (Ramos et al. 2003; Vasconcelos 2008).

The RF1 site had a similar species richness of epigeic ants compared with RF2 or FF sites. However, the species composition differs considerably among environments with similar historical disturbances *versus* a forest fragment with late regeneration. Our study shows that ant assemblages can vary greatly along a gradient of plant recovery and the conservation of forest fragments with different stages of reforestation is important to sustain more diverse ant fauna. Therefore, reforestation programs that prioritize the conservation and the adoption of native plant species in these areas are a good alternative (Gillespie et al. 2000), enabling the development of scientific studies and especially the maintenance of local biodiversity.

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