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Ants Sleep, Plants do not: The Variation in Species' Activity Influences the Topology of Interaction Networks

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Abstract

The emergence of graph theory allowed using the complex network approach to aggregate detailed information about interactions between species. Although the use of the complex network approach has improved the understanding about community structuring, few aspects such as the temporal variation in the species' activity pattern in the networks' topology have been explored, throughout the 24 hours of the day, until now. The current study used the ecological network approach to investigate ants interacting in the extrafloral nectary (EFN) of plants in order to test the hypothesis that the temporal variation in the foraging behavior of these animals affects the networks' topology. In order to assess the temporal effect on the interaction networks, 24-hour collections divided in two 12-hour shifts (day and night) were performed in 20 plots, thus totaling 288 collection hours over 6 months. The ant-plant interaction networks presented similarity among the topological metrics assessed throughout the day. Different ant species presented distinct foraging times. Thus, two modules referring to the day and night shifts emerged from the network and presented specific species at each foraging shift. On the other hand, the plants kept on providing the resource (active EFNs) throughout the day and night, one module. The results found in the current study have shown that ecological networks keep their structures constant; however, the ecological processes ruling these networks can better respond to the effects caused, for example, by the temporal variation in species' activity. Therefore, it is worth always taking into consideration the importance of ecological processes at the time to analyze interactions in the nature.

Introduction

The emergence of graph theory (Euler & Euler, 1736) allowed using the complex network approach to aggregate detailed information about interactions between species in biological communities (Amaral & Ottino, 2004; Bascompte et al., 2010). The network approach allows inferring information about each interaction between pairs of species, as well as assessing patterns at community level. This approach provides quantifiable metrics to discuss cause and effect relationships at lower hierarchical levels, as well as to discuss their consequences to the community as a whole (Lewinsohn et al., 2006b; Bascompte et al., 2010).

Using the complex network theory metrics as a tool allows describing patterns such as nesting, modularity, connectance and specialization in interactions between communities. Such patterns help making inferences about the processes underlying the communities' structure (Guimarães et al., 2007a, b; Bascompte et al., 2010). The nesting occurs when the species involved in the process present three intrinsic features, namely: generalist species interacting with each other; generalist species interacting with specialist species, and lack or rare occurrences of interactions between specialist species (Guimarães et al., 2006; Bascompte, 2007; Bascompte et al., 2010). The high modularity occurs when the network presents groups of species interacting more often with each other than

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with other species in the community (Lewinsohn, Ina & Prado, 2006a). The connectance, in turn, is the proportion between the observed interactions and the possible number of network interactions (Jordano, 1987). Finally, the specialization is based on information theory and assesses the least amount of interactions between two different species at population or community level (Blüthgen, Menzel & Blüthgen, 2006). The use of such metrics allows assessing the impact of the interaction that a given species exerts along with and on the other species in a given community.

Although the use of the complex network approach has enabled advances in the understanding of community structuring, there are still unexplored points (Blüthgen, 2010). For example, many networks describe relations between animals and plants, but few take into account the variations in the temporal activity patterns of species (Dáttilo, Díaz-Castelazo & Rico-Gray, 2014). Networks based on the direct observation of interactions (Gove, Majer & Dunn, 2007; Araujo et al., 2008; Allesina & Levine, 2011; Fagundes, Del-Claro & Ribeiro, 2012) may be biased if the observed species present specific activity times and if the observations do not contemplate such variations. It could exclude some species from the networks (which would not be registered because they were out of their activity period) and, consequently, the network structuring ecological processes inferred from the pattern obtained in the observations may be biased.

Plant-ant interaction networks are a very common example of networks that may present topologies with temporal variation. Many plant species are associated with several ant species that potentially provide protection against herbivory (Bentley, 1977; Miller, 2007; Fagundes et al., 2012). In this type of interaction, the plant provides nectar as food resource through extrafloral nectaries, also known as EFNs – nectar secreting structures, which are not involved in pollination (Fahn, 1988; Schmid, 1988). On the other hand, when ants search the plant looking for nectar, they end up attacking herbivores, thus establishing a facultative mutualistic interaction (Boucher, James & Keeler, 1982).

It is common for many ant species to show temporal pattern variation in their activities (Oliveira et al., 1999; Falcão, Dáttilo & Izzo, 2013). Some species may particularly present exclusively diurnal or nocturnal activity (Aschoff, 1966; Oliveira et al., 1999). Different species may show variations in their activity period in response to variations in temperature, humidity or solar irradiation, even in a single period (Retana & Cerda, 2000). Plants may also present temporal variations in their activity, such as opening or closing the stomata or EFNs at different periods in order to avoid water stress, or changing the amount and/or quality of the nectar produced in the EFNs in order to attract more efficient ant species to protect them during high predation periods (Falcão et al., 2013; Dáttilo et al., 2015). Therefore, the current study has conducted collections during 24-h periods in order to assess whether the changes in species' activity patterns could affect the structure of interaction networks. In order to do so, an EFN plant-ant interaction network was used as study model. The herein presented hypothesis is that the temporal variation in species' behavior affects the network topology and/or presents temporally varying interaction patterns.

Materials and Methods

Studied area

The study was conducted in the municipal protection area of the *Sempre Viva – Mucugê* Project; the area is located in Chapada Diamantina, Bahia State, Brazil, (12°59'31.43" S, 41°20'32.15" W). The region is formed by a mountainous complex, which is characterized by hills with altitude higher than 900 m above the sea level. The local landscape is dominated by rupestrian fields (Giulietti, A.M., Pirani, J.R. & Harley, R.M. 1997; Vasconcelos, 2011). According to Köppen's classification, the climate in the region is Cwa. The mean air temperature in the three coolest months of the year ranges from -3 °C to 18 °C, whereas the mean air temperature in the hottest month is higher than or equal to 22 °C; it rains during the summer.

Data Collection

Ants associated with plants were collected, from April to October 2013, in 20 plots (10 x 10 m), thus totaling a 2-km² area in a 5-km transect (Fig 4). The collection has followed the protocol by Agosti et al. (2000), who recommended plots at least 50m distant from each other in order to avoid overlapping ant nests. Monthly inventories of plants with extrafloral nectaries (EFNs) were performed in each plot. Whenever ants were found feeding on EFN plants during the monitoring conducted in each plot, one to three individuals belonging to each species were collected. In addition, the time the ant visited the plant and the region in the visited plant (leaf, flower bud, leaf knot; stem, flower, fruit, leaf peduncle, fruit peduncle and bud peduncle) were recorded. All plant species visited by the ants were collected for identification purposes at the end of the study.

The 24-h collections were divided in two 12-h shifts (day and night) in order to assess the temporal effect on the interaction networks. Ten (10) plots were inspected in each incursion during the day and 10 at night in order to avoid the effects caused by human presence disturbance. The daytime collections were conducted between 7:00 a.m. and 7:00 p.m., with 2-h intervals between samplings. The nocturnal collections were conducted between 7:00 p.m. and 7:00 a.m. in the following morning, also with 2-h intervals between samplings. It totaled 288 collection hours during 6 months. Each plot was inspected for at least 10 minutes and for at most 24 minutes during collection. This procedure aimed at minimizing the possible sub- or super-sampling effects on a single plot. The biological material samples were properly identified (the ants were identified by PhD Professor Rodrigo Feitosa (UFPA), whereas the plants were identified by experts from the herbarium of Feira de Santana State University – UEFS) and deposited in the entomological collection Prof. Iohan Backer at the UEFS Zoology Museum and in the herbarium belonging to the same institution.

Data analysis

Data concerning the presence/absence of ants associated with EFN plants were used to generate ant-plant incidence matrices: 1) six complete matrices comprising all the visiting ants and visited plants, in each month; 2) six diurnal matrices comprising just ants found in the plants throughout the day, in each month; 3) six nocturnal matrices comprising just ants found in the plants throughout the night, in each month; 4) one general matrix comprising all the visiting ants and visited plants, and grouping the six incursions to the field; 5) one general diurnal matrix comprising all interactions recorded during the six incursions performed at daytime; and 6) one general nocturnal matrix comprising all interactions recorded during the six incursions performed at night; in a total of 21 matrices.

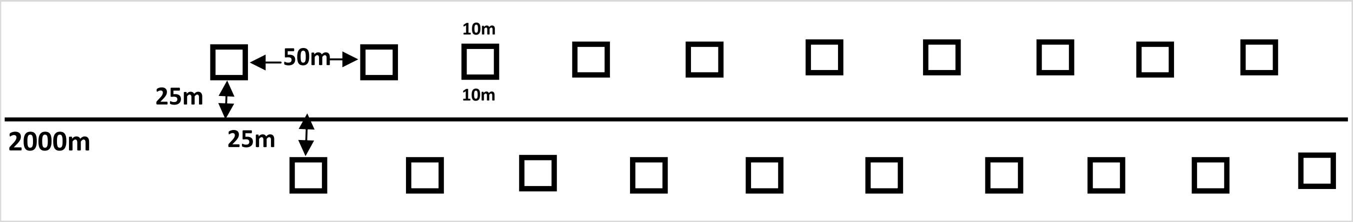

Fig 4. Scheme of sampling design used showing the transects subdivided into 20 plots (10 x 10m) each, distant 50m from each other.

Data referring to the intervals between 5:00 and 7:00 a.m. (dawn) and between 5:00 and 7:00 p.m. (twilight) were not taken into consideration in the construction of the diurnal and nocturnal matrices in order to avoid overlapping the species record between the nocturnal and diurnal periods. As the collections performed in these periods incorporated the light change between day and night, they overlapped species at the beginning and at the end of their activity (see the results for further details). Thus, two other matrices were generated: 1) matrix "ant x hour", comprising the 10 intervals and all ant species interacting with plants during the six incursions to the field; 2) matrix "plant x hour", comprising the 10 intervals and all plant species interacting with ants during the six incursions to the field. These two matrices incorporated the period during which the temporal variations of the species (ants and plants) were observed in the networks.

An abundance graph containing the 12 intervals/ collection hour, as well as the two light transition periods (between 5:00 and 7:00 a.m. and between 5:00 and 7:00 p.m.), was generated in order to assess whether the abundance of ant species varied temporally. A null model-based approach was used to assess whether the nesting pattern in each network (general, diurnal and nocturnal) could be obtained at random. In order to do so, the occurrence of interactions in each network was randomized 1000 times and the likelihood of each NODF value to be generated by chance was calculated. The NODF value may range from 0 (indicating a non-nested network) to 100 (indicating a perfectly nested network) (Almeida-Neto et al*.,* 2008). The Shannon index (H_2') was used to calculate the specialization value at community level (Blüthgen et al*.,* 2006). This index assesses the maximum network generality value ($H_2' = 0$) and the maximum specialization value ($H_2' = 1$). The connectance (C), which is the proportion between the observed interactions and the possible number of network interactions (Jordano, 1987), was also assessed in the current study.

The connectivity, connectance, specialization at community level (H_2'), modularity (M) and nesting (NODF) values concerning the 6 "ant x plant" diurnal networks and the 6 "ant x plant" nocturnal networks were calculated in order to assess whether the grouped, general, diurnal and nocturnal networks presented topology changes according to the collection period. These metrics were calculated for each collection in each period (night or day). Subsequently, a paired t-test was conducted in order to compare the metrics calculated for the diurnal and nocturnal networks. The metric values of the diurnal matrices were compared to the metric values of the nocturnal matrices belonging to the same collection month. All analyses were performed in the R software (R Core Team 2012) using Bipartite packages (Dormann et al., 2008) in order to generate the networks and calculate the nesting degree; SNA (Carter T. Butts 2013) and IGRAPH (Csardi G, Nepusz T 2006) were used to generate the network graphs.

The Visweb command was used in the matrices "ant x hour" and "plants x hour" in order to visualize the distinct modules related to the occurrence of ants and plants interacting in the diurnal or nocturnal intervals. The modularity (M) represents the degree of subgroup formations within a given community. The Modular Software (Marquitti et al*.,* 2014) based on Simulated Annealing (S), with 0-1 interval (Guimerà, Sales-Pardo & Amaral, 2004), was used to calculate the modularity index (M). This index varies from 0 (absence of modules) to 1 (network split in modules) (Olesen et al*.,* 2007).

A null model approach was used to assess whether the M value found in the matrices “ants x hour” and “plants x hour” could be generated at random. The occurrence of the interactions of the matrices “ant x hour” and “plant x hour” in each network was randomized 1000 times. The dissimilarity between ant and plant species in the general diurnal and general nocturnal networks was also quantified using the Morisita index. Morisita Overlap Index is a measure of how similar or different two sets of data are. The index ranges from 0 (no similarity) to 1 (complete similarity) (Morisita, M. 1962).

Results

A total of 46 ant species associated with 56 plants was collected. Thirty-eight (38) ant species interacting with 44 plant species were collected in the nocturnal period, whereas 33 ant species interacting with 47 plant species were collected in the diurnal period. The ants were distributed in different parts of the plant, according to the EFN location, at the following proportions: leaf = 37.50%, bud = 22.41%; leaf node = 12%; stem = 10.41%; flower = 7.1%; fruit = 4.51%; flower peduncle = 4.41%; fruit peduncle = 0.1%; and bud peduncle = 0.1%.

The general network comprising all species found during the nocturnal and diurnal collections was based on the record of 2883 ant occurrences. This network has shown nesting greater than that expected at random (NODF = 54.39, p <0.001, Fig 1A). The general diurnal and general nocturnal networks have shown nesting values similar to those of the general network; the NODF values were 49.04 (p <0.001, Fig 1B) and 52.3 (p <0.001, Fig 1C), respectively. The general network has also presented mean specialization level ($H_2{'}$= 0.194) similar to that of the general diurnal ($H_2{'}$ = 0.212) and general nocturnal networks ($H_2{'}$ = 0.226). The connectance of ants and plants in the three networks was also similar (general = 0.20; general diurnal = 0.20; general nocturnal = 0.18). The three networks have also presented similar modularity values (general = 0.20; general diurnal = 0.24; general nocturnal = 0.23).

The ants found in the EFN plants have shown well-marked daily activity pattern. The diurnal-species community was particularly different from the nocturnal-species community (“ant x hour” network modularity = 0.21; p <0.001; Morisita = 83%) (Fig 2A). A group of ant species (17%) has presented visitation activity regardless of the time and formed a type of timeless group (Fig 2A). However, the plant community did not present significant differences (“plant x hour” network

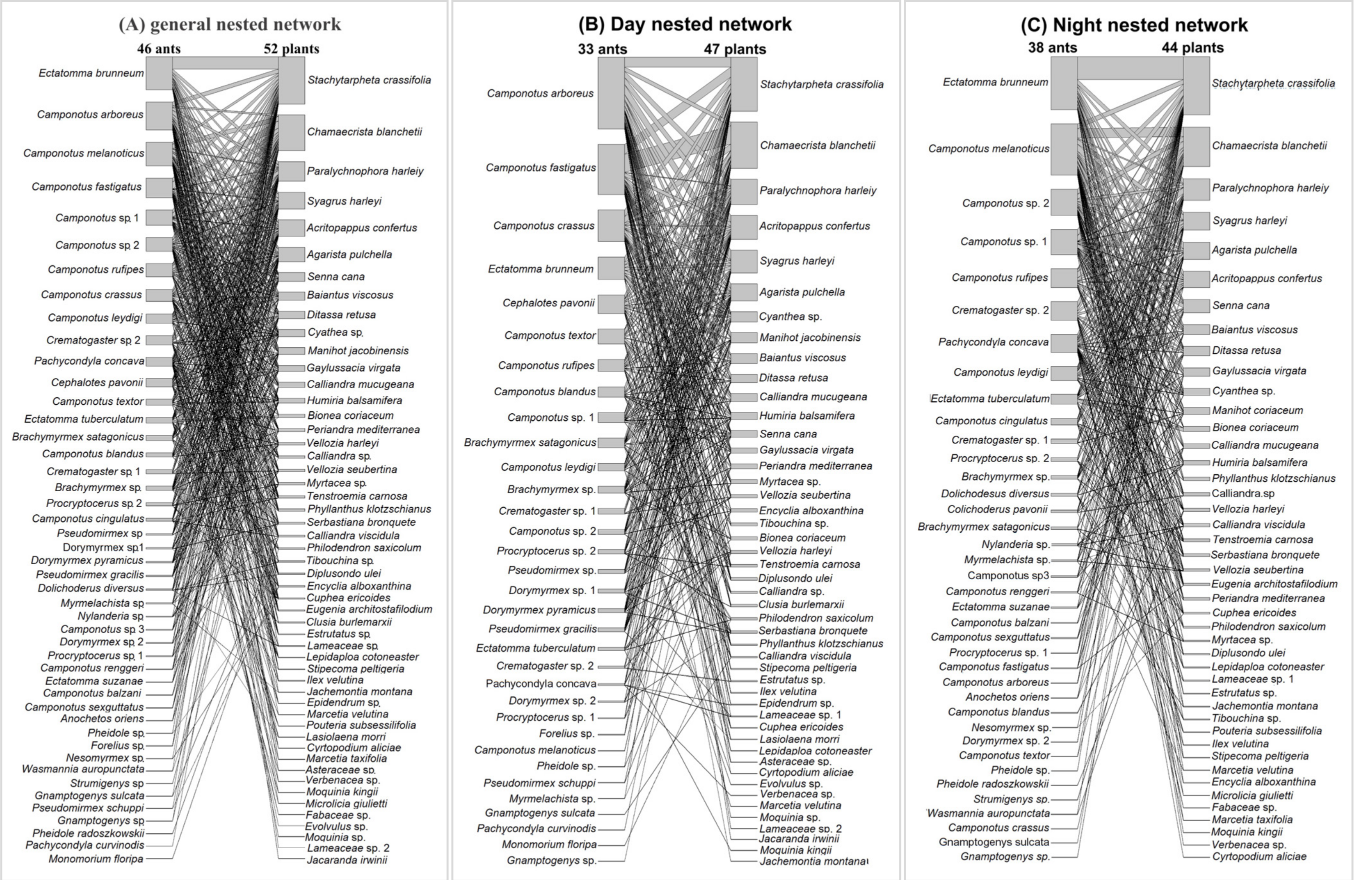

Fig 1. Mutualistic Interaction Networks, weighted by the presence and abundance between ants and plants with EFNs in Chapada Diamantina, Brazil. (A) general nested network with all interactions between 46 ant species and 52 plant species without taking into consideration the temporal variation in species' activity. (B) nested network with species active just during the day, from 07:00 a.m. to 07:00 p.m.; the network comprises 33 ant species interacting with 47 plant species. (C) nested network with species active from 07:00 p.m. to 07:00 a.m.; the network comprises 38 ant species interacting with 44 plant species.

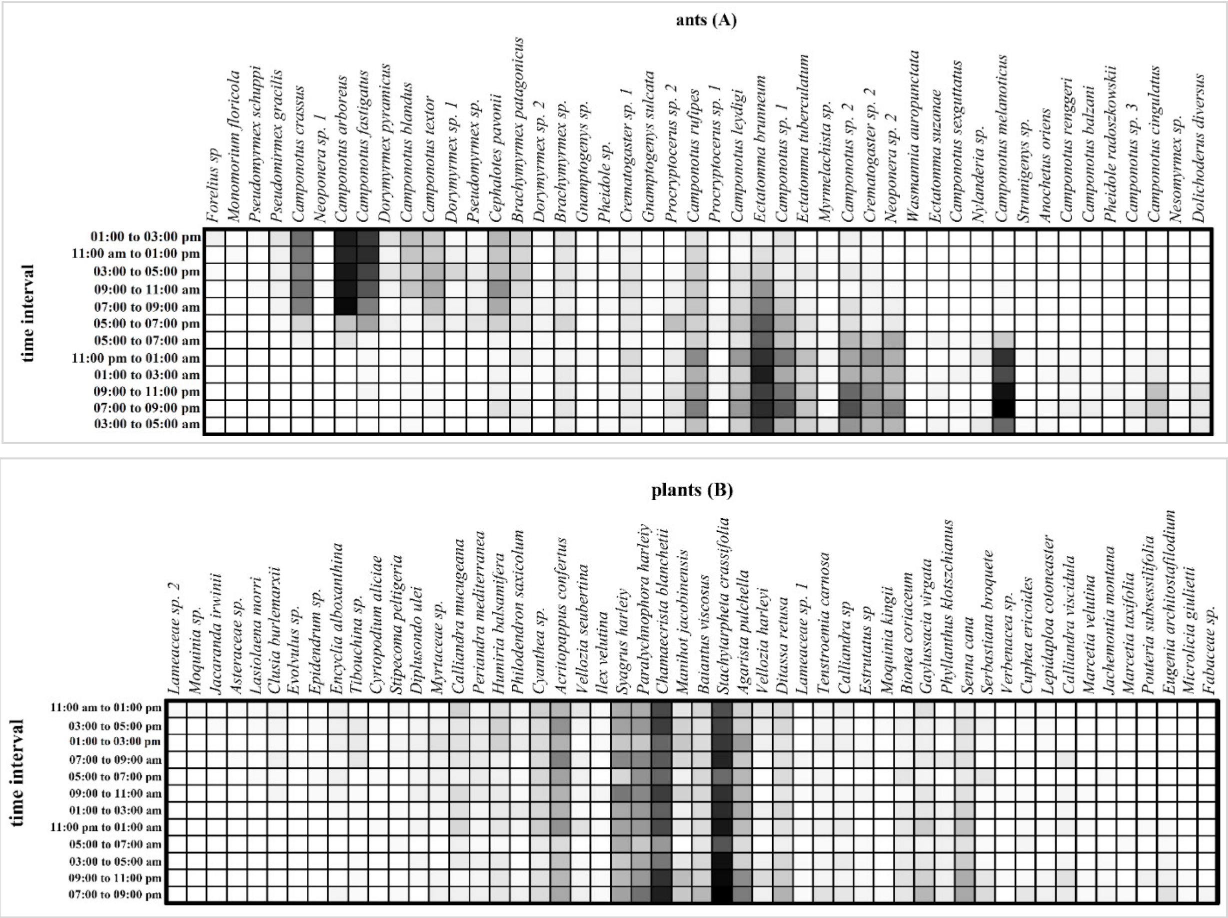

Fig 2. Interaction matrices comprising ant and plant species found in the 12-hour sampling period in Chapada Diamantina, Bahia State, Brazil. (A) matrix with 46 ant species interacting with EFN plants. (B) matrix with 52 plant species interacting with ants along 12 sampling intervals. The sequence of times is established by the abundance and frequency of ants in the plants. The gray scale was established by the R program according to the presence and abundance of ants in the plants according to each interval of couple of hours. The greater the intensity of gray, the greater the incidence of the species.

modularity = 0.01; p = 0.998; Morisita = 1.1%; Fig 2B). The connectance, specialization (H_2’), modularity and nesting values of the diurnal and nocturnal matrices in each month were similar (Table 1); however, there was significant variation in the ant species’ composition (Fig 2A).

The abundance of ants visiting the plants reduced from 05:00 to 07:00 a.m. and from 05:00 to 07:00 p.m. Soon after, there was a peak in the subsequent intervals: from 07:00 a.m. to 5:00 p.m. and from 7:00 p.m. to 5:00 a.m. (Fig 3). It was a clear indication that there was change of shifts between ant species. It is worth highlighting that some generalist ant species in the network such as *Ectatomma brunneum* (the most generalist species in the nocturnal network (Fig 1C) and the fourth most generalist species in the diurnal network (Fig 1B)) did not visit some plant species such as *Chamaecrista blanchetii* (the second most generalist species in the three networks) and *Agarista pulchella* (the sixth most generalist species in the network). Similarly, *Camponotus fastigatus* (the second most generalist species in the diurnal network and not found in the nocturnal network) did not visit *Paralychnophora harleiy* (the second most generalist plant species in the diurnal network) (Fig 1B). Finally, *Camponotus arboreus* (the first most generalist species in the diurnal network and not found in the nocturnal network) did not visit *Senna cana* or *Baiantus viscosus*, which are also generalist plant species in the general network.

Discussion

The current study has shown that the topological metrics of the network of interactions between ants and EFN plants did not change between day and night shifts. However, there was great temporal variation in the composition of ant species found in the networks. It indicated that the structure of the interactions remained constant, but the characters (ant species) performing ecological services varied. These species eventually played similar roles in the network because the network topology remained constant even when the foraging varied temporally. However, the change of shift observed in different ant species has indicated a possible specificity (aggressiveness, for example) in the plant protection service. Another relevant factor lies on the fact that this

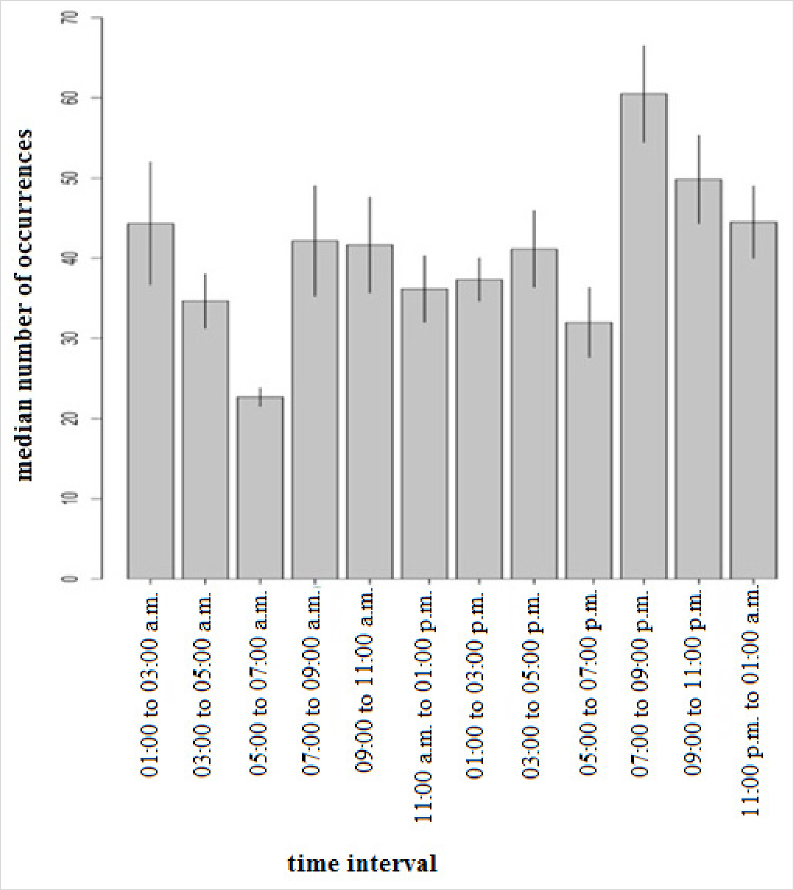

Fig 3. Average number of occurrences of ants found in EFN plants in Chapada Diamantina, Bahia State, Brazil, divided in 12 sampling intervals. Bars indicate Standard Error.

temporal variation did not take place in the plant community. It indicated that the plants remained active and provided the nutritional resource for 24 hours. Eventually, the need of protection against potential predators, which was provided by different ant species, justified the constant EFN activity.

An intriguing point about the herein obtained patterns concerns what could lead the ant community to shift between the day and night periods. Ants may become more or less aggressive when they catch herbivores depending on the sugar concentration in the consumed food (Grover et al*.*, 2007; Kaplan et al*.*, 2015). Likewise, the sugar concentration in the nectar may vary within a single plant (Herbert G . Baker, 1978). Eventually, if the chemical composition of the nectar varies at different times of the day, perhaps it could explain the temporal variation in the activity of ants, since this variation is influenced by the variation in the EFN quality. Different studies have shown that the variation in the extrafloral nectar composition may affect the behavior of certain ant species and make them more or less active/aggressive, as well as make them abandon or become dependent on the resource (Grover et al*.*, 2007; Wilder & Eubanks, 2010; Heil et al*.*, 2014). Thus, it is possible assuming the reason why generalist ants do not visit generalist plants in the network, since the nectar of these plants does not hold some attractive or repellent compound to these ant species.

The optimal defense theory predicts that, in order to obtain greater protection during the highest predation risk period, plants may vary the extrafloral nectar supply and quality during a certain period of the day or in a certain part of the plant (Falcão et al*.*, 2013; Dáttilo et al*.*, 2014c; Millán-Cañongo, Orona-Tamayo & Heil, 2014) to attract ants able to provide more efficient protection. Therefore, such variation in nectar composition may be beneficial to plants if the defense behavior of the ants found during the day and at night is complementary (high-quality EFN/most aggressive ants; low-quality EFN/less aggressive ants), since it is likely that the herbivores eating plants throughout the day are different from and less abundant than the nocturnal ones (Wilder & Eubanks, 2010; Dáttilo et al., 2014c, 2015).

Competition is also an important component that may be associated with the temporal variation in ant behavior (Retana & Cerda, 2000). In this case, the nutrient supplied by the plants would have little relevance to the role played by the change of shifts between ant species. Thus, ants temporally vary their activity to reduce the impact of competition and, consequently, to reduce niche overlapping. The temporal variation in ant activity could also be a simple behavioral response of each ant species. The problem in considering it to be a behavioral response lies on the fact that the variation takes place in many species and in a very synchronized way.

Therefore, it is more plausible thinking that the competition is also a factor responsible for such patterns (Dáttilo et al., 2014b). The Morisita indices data provide robustness for this discussion when, for the ants x plants networks, they indicate no similarity between the ant species over the 24 h, which does not occur in the NEF supply networks of the plants. Another relevant factor, which is possibly associated with competition, is that four ant species – *Ectatomma brunneum, Camponotus rufipes, Camponotus leydigi and Camponotus sp. 1* - remained active for 24 hours, although they were more abundant at night. These four species belong to two subfamilies – Ectatomminae and Formicinae – described in the literature as presenting highly aggressive behavior in comparison to other nectariferous species (Dáttilo et al., 2014).

The 24-hour collections performed in the current study aimed at investigating to what extent the temporal variation in species' activity affects the topology of ecological networks, as well as at clarifying whether some studies, which did not take into consideration the species' activity period, could be biased in the interpretation of data. It was possible seeing that the network topological metrics remained very similar, fact that corroborated the results of several studies about nested networks (Burgos et al., 2007; Dattilo, 2014; Pires et al., 2011). However, there was great difference in the composition of one of the parts (ants) in this bipartite network. It remains to be seen whether such change may affect the community functioning. Thus, the temporal pattern of the species may have important repercussions on how the species interact in the community, even in a similar topology. Many network studies have made projections about the impact that the extinction of a given species may have on the other species in the network (Jordano, Bascompte & Olesen, 2003; Bascompte, Jordano & Olesen, 2006; Bascompte & Jordano, 2007). These results were presented in a general ecological network without taking into consideration whether the studied species temporally varied their interactions. However, if there are temporal modules, some of the connections described when all species are grouped would cease to exist. Thus, it is essential assessing the systems by taking the variation in the species' activity pattern into consideration in order to avoid interpretation bias in extinction projections, for example. In addition, other networks such as seed dispersal and pollination ones would be as subjected to this type of temporal variation as networks of interactions between EFN plants and ants.

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References

Agosti, D., Majer, J.D., Alonso, L.E. & Schultz, T.R. (2000). Ants: standard methods for measuring and monitoring biodiversity. Smithsonian Institution Press. Washington. 280 p.

Allesina, S. & Levine, J.M. (2011). A competitive network theory of species diversity. Proceedings of the National Academy of Sciences of the United States of America, 108: 5638-5642. https://doi.org/10.1073/pnas.1014428108

Almeida-Neto, M., Guimarães, P., Guimarães, J.P.R., Loyola, R.D., Ulrich, W., Guimarães, P.R.J., Loyota, R.D. & Ulrich, W. (2008). A consistent metric for nestedness analysis in ecological systems: reconciling concept and measurement. Oikos, 117: 1227-1239.
https://doi.org/10.1111/j.0030-1299.2008.16644.x

Amaral, L.A.N. & Ottino, J.M. (2004). Complex networks: Augmenting the framework for the study of complex systems. European Physical Journal B, 38: 147-162.
https://doi.org/10.1140/epjb/e2004-00110-5

Araujo, M.S., Guimarães, P.R., Svanback, R., Pinheiro, A., Guimaraes, P., dos Reis, S.F. & Bolnick, D.I. (2008). Network Analysis reveals contrasting effects of intraspecific competition on individuals vs. population diets. Ecology, 89: 1193-1981. https://doi.org/10.1890/07-0630.1

Aschoff, J. (1966). Circadian Activity Pattern with Two Peaks. Ecological Society of America, 47: 657-662.
https://doi.org/10.2307/1933949

Bascompte, J. (2007). Networks in ecology. Basic and Applied Ecology, 8: 485-490. https://doi.org/10.1016/j.baae.2007.06.003

Bascompte, J. & Jordano, P. (2007). Plant-Animal Mutualistic Networks: The Architecture of Biodiversity. Annual Review of Ecology, Evolution, and Systematics, 38: 567-593.
https://doi.org/10.1146/annurev.ecolsys.38.091206.095818

Bascompte, J., Jordano, P. & Olesen, J.M. (2006). Asymmetric Coevolutionary Networks Facilitate Biodiversity Maintenance. Science, 312: 431-433.
https://doi.org/10.1126/science.1123412

Bascompte, J. (2010). Structure and dynamics of ecological networks: Understanding the architecture of species relationships may help predict how ecosystems respond to change. Science, 329: 765-766.
https://doi.org/10.1126/science.1194255

Bentley, B. (1977). The protective function of ants visiting the extrafloral nectaries of *Bixa orellana* (Bixaceae). The Journal of Ecology, 65: 27-38. https://doi.org/10.2307/2259060

Blüthgen, N. (2010). Why network analysis is often disconnected from community ecology: A critique and an ecologist's guide. Basic and Applied Ecology, 11: 185-195. https://doi.org/10.1016/j.baae.2010.01.001

Blüthgen, N., Menzel, F. & Blüthgen, N. (2006). Measuring specialization in species interaction networks. BMC Ecology, 6: 9. https://doi.org/10.1186/1472-6785-6-9

Boucher, D.H., James, S. & Keeler, K.H. (1982). The Ecology of Mutualism. Annual Reviews of Ecology and Systematics, 13: 315-347. https://doi.org/10.1146/annurev.es.13.110182.001531

Burgos, E., Ceva, H., Perazzo, R.P.J., Devoto, M., Medan, D., Zimmermann, M. & María Delbue, A. (2007). Why nestedness in mutualistic networks? Journal of Theoretical Biology, 249: 307-313. https://doi.org/10.1016/j.jtbi.2007.07.030

Dáttilo, W., Aguirre, A., Flores-Flores, R. V., Fagundes, R., Lange, D., García-Chávez, J., Del-Claro, K. & Rico-Gray, V. (2015). Secretory activity of extrafloral nectaries shaping multitrophic ant-plant-herbivore interactions in an arid environment. Journal of Arid Environments, 114: 104-109. https://doi.org/10.1016/j.jaridenv.2014.12.001

Dáttilo, W., Díaz-Castelazo, C. & Rico-Gray, V. (2014a). Ant dominance hierarchy determines the nested pattern in ant-plant networks. Biological Journal of the Linnean Society, 113: 405-414. https://doi.org/10.1111/bij.12350

Dáttilo, W., Fagundes, R., Gurka, C.A.Q., Silva, M.S.A., Vieira, M.C.L., Izzo, T.J., Díaz-Castelazo, C., Del-Claro, K. & Rico-Gray, V. (2014b). Individual-based ant-plant networks: Diurnal-nocturnal structure and species-area relationship. PLoS ONE, 9: e99838. https://doi.org/10.1371/journal.pone.0099838

Dáttilo, W., Sánchez-Galván, I., Lange, D., Del-Claro, K. & Rico-Gray, V. (2014c). Importance of interaction frequency in analysis of ant-plant networks in tropical environments. Journal of Tropical Ecology, 30: 165-168. https://doi.org/10.1017/S0266467413000813

Dattilo, W. (2015). Mutualistic networks by Jordi Bascompte and Pedro Jordano. Journal of Complex Networks, 3: 158. https://doi.org/10.1093/comnet/cnu021

Dáttilo, W., Díaz-Castelazo, C. & Rico-Gray, V. (2014). Ant dominance hierarchy determines the nested pattern in ant-plant networks. Biological Journal of the Linnean Society, 113: 405-414. https://doi.org/10.1111/bij.12350

Del-Claro, K., Johnson, M. & Torezan-Silingardi, H.M. (2012). Plant-arthropod interactions: A behavioral approach. Psyche, 2012: 203208. https://doi.org/10.1155/2012/203208

Euler, L. & Euler, L. (1736). Solutio problematis ad geometrian situs pertinentis. Comentarii Academiae Scientarum Petropolitanae, 8: ID 128-140.

Fagundes, R., Del-Claro, K. & Ribeiro, S.P. (2012). Effects of the trophobiont herbivore *Calloconophora pugionata* (Hemiptera) on ant fauna associated with *Myrcia obovata* (Myrtaceae) in a montane tropical forest. Psyche, 2012: ID 783945. https://doi.org/10.1155/2012/783945

Fahn, A. (1988). Secretory tissues in vascular plants. New Phytologist, 108: 229-257. https://doi.org/10.1111/j.1469-8137.1988.tb04159.x

Falcão, J.C.F., Dáttilo, W. & Izzo, T.J. (2013). Temporal variation in extrafloral nectar secretion in different ontogenic stages of the fruits of *Alibertia verrucosa* S. Moore (Rubiaceae) in a Neotropical savanna. Journal of Plant Interactions, 9: 1-6. https://doi.org/10.1080/17429145.2013.782513

Giulietti, A.M., Pirani, J.R. & Harley, R.M. (1997). Espinhaço Range region, eastern Brazil. In S.D. Davis, V.H. Heywood, O. Herrera-MacBryde, J. Villa-Lobos & A.C. Hamilton, eds. Centres of plant diversity: a guide and strategy for their conservation. Information Press, Oxford, v. 3, p. 397-404.

Gove, A.D., Majer, J.D. & Dunn, R.R. (2007). A keystone ant species promotes seed dispersal in a " di V use " mutualism. Oecologia, 153: 687-697. https://doi.org/10.1007/s00442-007-0756-5

Grover, C.D., Kay, A.D., Monson, J.A., Marsh, T.C. & Holway, D.A. (2007). Linking nutrition and behavioural dominance: carbohydrate scarcity limits aggression and activity in Argentine ants. Proceedings of the Royal Society B Biological Sciences, 274: 2951-2957. https://doi.org/10.1098/rspb.2007.1065

Guimarães, P.R., Machado, G., de Aguiar, M.A.M., Jordano, P., Bascompte, J., Pinheiro, A. & dos Reis, S.F. (2007a). Build-up mechanisms determining the topology of mutualistic networks. Journal of Theoretical Biology, 249: 181-189. https://doi.org/10.1016/j.jtbi.2007.08.004

Guimarães, P.R., Rico-Gray, V., dos Reis, S.F. & Thompson, J.N. (2006). Asymmetries in specialization in ant-plant mutualistic networks. Proceedings of the Royal Society B Biological Sciences, 273: 2041-2047. https://doi.org/10.1098/rspb.2006.3548

Guimarães, P.R., Sazima, C., dos Reis, S.F. & Sazima, I. (2007b). The nested structure of marine cleaning symbiosis: is it like flowers and bees? Biology Letters, 3: 51-54. https://doi.org/10.1098/rsbl.2006.0562

Guimerà, R., Sales-Pardo, M. & Amaral, L.A.N. (2004). Modularity from fluctuations in random graphs and complex networks. Physical Review E - Statistical, Nonlinear, and Soft Matter Physics, 70: 1-4. https://doi.org/10.1103/PhysRevE.70.025101

Heil, M., Barajas-Barron, A., Orona-Tamayo, D., Wielsch, N. & Svatos, A. (2014). Partner manipulation stabilises a horizontally transmitted mutualism. Ecology Letters, 17: 185-192. https://doi.org/10.1111/ele.12215

Herbert G. Baker, P.A. O. and I.B. (1978). A comparison of the amino acid complements of floral and extrafloral nectars. Botanical Gazette, 139: 322-332. https://doi.org/10.1086/337008

Jordano, P. (1987). Patterns of Mutualistic Interactions in Pollination and Seed Dispersal: Connectance, Dependence Asymmetries, and Coevolution. American Naturalist, 129: 657-677. https://doi.org/10.1086/284665

Jordano, P., Bascompte, J. & Olesen, M.J. (2003). Invariant Properties in Coevolutionary Networks of Plant-Animal Interactions. Ecology Letters, 6: 69-81. https://doi.org/10.1046/j.1461-0248.2003.00403.x

Morisita, M. (1962). Iσ-index a measure of dispersion of individuals? Population Ecology, 4: 1-7. https://doi.org/10.1007/BF02533903

Kaplan, I., Eubanks, M.D., Kaplan, I.A.N. & Eubanks, M.D. (2015). Aphids Alter the Community-Wide Impact of Fire Ants, 86: 1640-1649. https://doi.org/10.1890/04-0016

Lewinsohn, T.M., Ina, P. & Prado, P.I. (2006a). Structure in plant-animal interaction assemblages. Oikos, 113: 1-11. https://doi.org/10.1111/j.0030-1299.2006.14583.x

Lewinsohn, T.M., Inácio Prado, P., Jordano, P., Bascompte, J. & Olesen, J.M. (2006b). Structure in plant-animal interaction assemblages. Oikos, 113: 174-184. https://doi.org/10.1111/j.0030-1299.2006.14583.x

Marquitti, F.M.D., Guimarães, P.R., Pires, M.M. & Bittencourt, L.F. (2014). MODULAR: Software for the autonomous computation of modularity in large network sets. Ecography, 37: 221-224. https://doi.org/10.1111/j.1600-0587.2013.00506.x

Millán-Cañongo, C., Orona-Tamayo, D. & Heil, M. (2014). Phloem Sugar Flux and Jasmonic Acid-Responsive Cell Wall Invertase Control Extrafloral Nectar Secretion in *Ricinus communis*. Journal of Chemical Ecology, 40: 760-769. https://doi.org/10.1007/s10886-014-0476-3

Miller, T.E.X. (2007). Does having multiple partners weaken the benefits of facultative mutualism? A test with cacti and cactus-tending ants. Oikos, 116: 500-512. https://doi.org/10.1111/j.2007.0030-1299.15317.x

Olesen, J.M., Bascompte, J., Dupont, Y.L. & Jordano, P. (2007). The modularity of pollination networks. Proceedings of the National Academy of Sciences of the United States of America, 104: 19891-19896. https://doi.org/10.1073/pnas.0706375104

Oliveira, P.S., Rico-Gray, V., Díaz-Castelazo, C. & Castillo-Guevara, C. (1999). Interaction between ants, extrafloral nectaries and insect herbivores in Neotropical coastal sand dunes: Herbivore deterrence by visiting ants increases fruit set in *Opuntia stricta* (Cactaceae). Functional Ecology, 13: 623-631. https://doi.org/10.1046/j.1365-2435.1999.00360.x

Pires, M.M., Guimarães, P.R., Araújo, M.S., Giaretta, A.A., Costa, J.C.L. & dos Reis, S.F. (2011). The nested assembly of individual-resource networks. Journal of Animal Ecology, 80: 896-903. https://doi.org/10.1111/j.1365-2656.2011.01818.x

Retana, J. & Cerda, X. (2000). Patterns of diversity and composition of Mediterranean ground ant communities tracking spatial and temporal variability in the thermal environment. Oecologia, 123: 436-444. https://doi.org/10.1007/s004420051031

Schmid, R. (1988). Reproductive versus extra-reproductive nectaries-historical perspective and terminological recommendations. The Botanical Review, 54: 179-227. https://doi.org/10.1007/BF02858528

Vasconcelos, M.F. de. (2011). O que são campos rupestres e campos de altitude nos topos de montanha do leste do Brasil? Brazilian Journal of Botany, 34: 241-246. https://doi.org/10.1590/S0100-84042011000200012

Wallace, A. R. 1889. Darwinism: an exposition of the theory of natural selection with some of its applications. London & New York: Macmillan & Co. https://doi.org/10.5962/bhl.title.2472

Wilder, S.M. & Eubanks, M.D. (2010). Extrafloral nectar content alters foraging preferences of a predatory ant. Biology Letters, 6: 177-179. https://doi.org/10.1098/rsbl.2009.0736

Supplementary Material

Table 1 - Index values of the interaction networks between plants with extrafloral nectaries and ants in Chapada Diamantina, Bahia State, Brazil. The general, diurnal and nocturnal values correspond to the values of each metric calculated from the general matrices that grouped data from six collections. On the other hand, the DIF values indicate the mean of the differences between the network metric measured during the day and that measured during the night. Values in brackets indicate the standard error.

Indices	General	Day	Night	DIF	*t*	DF	*P*
Connectance	21%	20%	18%	0.01 (0.03)	0.77	5	0.47
H_2' (specialization)	0.19	0.21	0.23	-0.02 (0.04)	1.76	5	0.14
M (modularity)	0.20	0.24	0.23	0.02 (0.03)	1.44	5	0.21
NODF (nestedness)	54.39	49.04	52.3	1.04 (6.51)	0.41	5	0.70