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Temporal variation in production and nutritional value of pollen used in the diet of *Apis mellifera* L. in a seasonal semideciduous forest

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Abstract

The flora of mountain formations in the Caatinga biome is composed predominantly by semi-deciduous species with representatives of both Atlantic and Amazon forest. Information on the potential for bee pollen production of these species is limited. In this study we evaluated the potential of production, the temporal variation, the botanical origin and the nutritional value of bee pollen produced in a seasonal semideciduous forest in northeastern Brazil. We identified a total of 252 flowering plant species throughout the year. The diet of *Apis mellifera* consisted of 74 pollen types distributed in 58 genera and 27 families. We identified two production peaks of bee pollen, the highest occurring in the rainy season. Nutritional value considering crude protein, carbohydrates, lipids and mineral matter changed over the study period, with influence of rainfall on the dry matter level. Some taxonomic groups of plants showed a strong relationship with nutrients, suggesting that although the diet of *A. mellifera* is broadly diversified, this species devoted most of its pollen foraging effort on the genus *Mimosa* and the palm tree species of *Attalea speciosa*. The results show that the seasonal semideciduous forest of the mountain range in the Northeast Brazil presents plant species: *Mimosa caesalpiniifolia, Baccharis trinervis*, *Mimosa tenuiflora Myracrodruom urundeuva*, *Cecropia pachystachya*, *Attalea speciosa*, with high nutritional level and potential for the pollen production.

Introduction

The quantity and quality of pollen collected by bees are closely related to the type of vegetation and availability of floral resources. Bee pollen is the agglutination of flower pollen that receives small amounts of nectar and other salivary substances from *Apis mellifera* (Villanueva et al., 2002). This process facilitates the attachment of pollen to the corbicula of foraging bees and transportation to the colony. Bee pollen contains more than 200 substances (Komosinska-Vassev et al., 2015), consisting mainly of proteins, amino acids, lipids, fibers, enzymes, minerals, sugars and vitamins (Arruda et al., 2013; Avni et al., 2014; Bogdanov, 2015; Sattle et al., 2015). This composition makes pollen essential for feeding brood as well as for maintenance of the colony of *A. mellifera* (Marchini et al., 2006).

Bee pollen is also an important source of income for beekeepers in different countries. Worldwide, pollen production is approximately 1,500 tons/year, with Spain and China as the two major producers (Estevinho et al., 2012; Yang et al., 2013). However, global production of bee pollen is concentrated on temperate countries and regions and little is known about the potential production in tropical and subtropical areas of the world (Estevinho et al., 2012). In these warmer regions, beekeeping has faced consistent growth in recent years, but focusing on honey production.

In Brazil, for instance, much of the beekeeping production is based on semi-arid parts of the country (Barreto et al., 2006), where the physiognomy varies from sparsely vegetated desert to areas with dry forests covered by dense tree layers (Araujo-Filho, 2013). Nevertheless, pollen production

in the region is limited especially by the lack of knowledge about the identity of polliniferous plants with potential to sustain beekeeping activity throughout the year (Milfont et al., 2011).

The present study investigates the potential of bee pollen production in a seasonal semideciduous forest of the semi-arid area of northeastern Brazil and evaluates the temporal variation in the production of pollen used in the diet of *A. mellifera* and its nutritional value.

Material and methods

Study area

Samplings were carried out from November 2012 to October 2013 in a seasonal semideciduous forest, located in an environmental protection area in the municipality of Meruoca (3°35'40.63" S and 40°24'11.91" W), state of Ceará, Brazil (Fig 1A). The climate of the region is Aw', characterized as hot, humid and rainy in the summer (Köppen, 1948). The average annual rainfall is 1,530.3 mm (Fig 1B). The rainy season is concentrated between January and April, extending to June and with annual average rainfall of 1,194.3 mm (Carvalho, 2013). In this type of geomorphological formation, a predominance occurs of deep soils, moderately drained with good fertility. The rainwater accumulated in the soil (eutrophic Red-Yellow Argisols) favors the establishment of arboreal species. The original prevailing vegetation is pluvial-nebular subperennifolia (humid forests, serranas), but nowadays, besides the natural areas, there are also subsistence crops and a poorly developed beekeeping (FUNCEME, 2015).

Floristic composition and bee plants

In order to survey the floristic composition in the study area we set up a radius with approximately 1,000 meters (Krebs, 1999), having the apiary as the central point. Then, we examined the study area on a monthly basis and identified the flowering plant species, considering the entire vertical stratification (except Poaceae species) (Silva et al., 2012). We took three samples of each flowering plant species for the preparation of vouchers and subsequent identification of the

species by experts. Vouchers are deposited in the Herbarium Professor Francisco José de Abreu Matos-HUVA, State University Vale do Acaraú (UVA), Ceará Brazil.

### Preparation of reference pollen collection

We collected the anthers of all species of flowering plants and maintained in 70% ethanol for 24 hours (Silva et al., 2014). The material was then ground, subjected to the acetolysis procedure (Erdtman, 1960) and maintained in 50% glycerin. For each plant species, we mounted three slides using Kisser gelatin and sealed with paraffin (Silva et al., 2014). Slides were properly identified according to Silva et al. (2010) and deposited in the pollen collection of the Bee Laboratory, Department of Animal Sciences, Federal University of Ceará (UFC). Pollen grains were photographed with a digital camera attached to a trinocular microscope. Based on the images, we made measurements of pollen grains for morphological descriptions. Subsequently, we organized a Pollen Catalogue with 252 species that was used to identify, by comparison, the pollen collected by *A. mellifera* foragers.

Pollen collection from *Apis mellifera* colonies

Pollen samples were obtained installing pollen collectors in the entrances of ten Langstroth hives, standardized to the number of bees (28 thousand ± 2 thousand), number of combs (n=10) and queen age (one year). These pollen collectors require bees to enter the hive through small holes that scrape the pollen from the bee corbiculae. We sampled pollen every other day, always at 5:30 pm, totaling 15 monthly collections for each colony. Fresh pollen samples were cleaned by removing the debris attached to the pollen (dead bees, bee larvae, propolis) and weighed to the nearest 0.1 g and then cooled in a freezer at less than 20 °C.

Pollen analysis

Pollen of the 15 samples taken every month from each hive was pooled, forming a single monthly composite sample per hive. Each composite sample was divided into two parts, one for chemical analysis and another for botanical origin
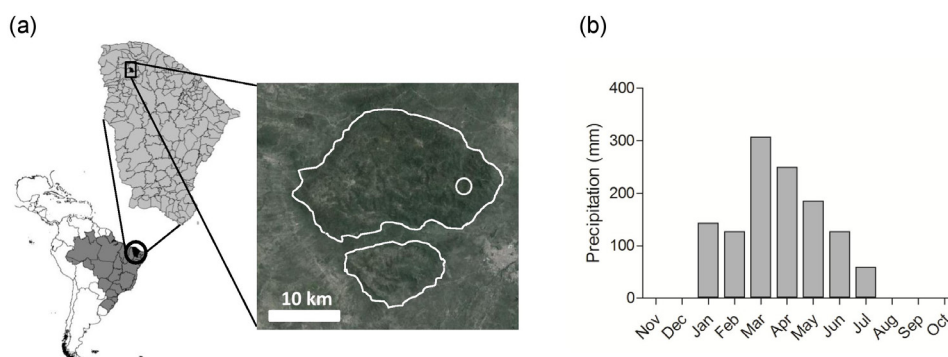
Fig 1. a) The study area (white circle - 3 ° 35'40.63 "S, 40 ° 24'11.91" W) is located within the environmental protection area of the Serra da Meruoca (bounded by the white line). (b) Rainfall in the municipality of Meruoca during the study period (FUNCEME - Cearense Foundation of Meteorology and Water Resources 2012-2013)

identification. Thus, for the same composite monthly sample, we obtained information about the visited plants and the nutritional value of the diet of *A. mellifera*.

Chemical analysis

Chemical analyses were carried out in the Laboratory of Animal Nutrition (LANA-DZO-UFC) in Department of Animal Sciences at the Federal University of Ceará. The bee pollen collected was kept in falcon tubes of 15 mL and frozen until the time of chemical analysis. Pollen samples were thawed, dried in a forced air oven at 55°C for 72 hours, and ground to pass through a 1 mm screen (Wiley Mill, Arthur H. Thomas, Philadelphia, PA, USA). All samples were analyzed for dry matter (DM; AOAC 1990; method number 930.15), mineral matter (MM; AOAC 1990; method number 924.05), crude protein (CP; AOAC 1990; method number 984.13), lipids (FAT; AOAC 1990; method number 920.39). The total carbohydrate content (TC) was calculated by the formula TC (%) = 100 - %CP - %FAT - % MM), according to Sniffen et al. (1992).

Identification of the botanical origin of pollen

Pollen samples, about two grams, were kept in 4 mL of 70% alcohol, in falcon tubes with capacity for 15 mL, for 24 hours, then they were centrifugated and the supernatant was discarded. After the alcohol was discarded, the material was kept for 24 hours in 4 mL glacial acetic acid (Silva et al., 2014) and acetolyzed, for each sample, 5mL of the acetolysis mixture was used, nine parts of acetic anhydride for one-part sulfuric acid (9:1) following the method described by Erdtman (1960). After acetolysis, the material was kept in 50% glycerin. We mounted three slides for each sample, using Kisser gelatin and transparent lacquer for reading fast.

In the qualitative analysis, the pollen types found on the slides were identified by comparison with the pollen types of the reference slides of the plants that flourished in the area during the study period. We also used specific literature for the identification of pollen collected by bees (Silva et al., 2010; Bauermann et al., 2013; Silva et al., 2014).

In the quantitative analysis, we identified and counted the first 400 pollen grains found on each slide (Montero & Tormo, 1990). Then, we calculated the relative frequency (percent) of each pollen type and classified the material in accordance to occurrence, as proposed by Barth (1970) and Louveaux et al. (1970,1978): predominant pollen (DP, > 45% of the total pollen grains present on the slide), accessory pollen (AP, from 15 to 45%), important isolated pollen (IIP, 3 to 15%) and occasional isolated pollen (OIP, < 3%). We also measured the total volume of pollen grains of each plant species in each sample used by *A. mellifera* based on the mean length of the longitudinal axis measured on 25 pollen grains. Based on these values, the mean volume pollen grain for each plant species was calculated according to the method proposed by Villanueva-G and Roubik (2004). Total pollen volume expresses the pollen dominance in the diet of *A. mellifera*.

Data analysis

Generalized linear models (GLM) were used to evaluate how the independent variables 'number of pollen types' and 'precipitation (mm)' influence the following response variables: (a) pollen production (g/month), (b) crude protein, (c) total carbohydrates, (d) lipids, (e) mineral matter and (f) dry matter. A model for each response variable was elaborated as follows: response variable = 'number of pollen types' + 'precipitation (mm)'. As the response variables are continuous and normal, we used a GLM with Gaussian distribution and identity binding function. We performed *a posteriori* diagnostic tests to verify if the models were adequate (such as the analysis of the normality of residuals and verification of the influence of atypical values). A Spearman correlation (rs) was performed to check if the dominance of some botanical groups (families or genera) is related to the chemical variables (crude protein, total carbohydrates, lipids, mineral matter and dry matter). Dominance was estimated using the total number and volume of pollen grains of each plant species used by *A. mellifera*. A cluster analysis was run to classify pollen samples in accordance with colonies of *A. mellifera*, by using the Bray-Curtis similarity index and the paired group algorithm. The consistency of grouping pattern was tested by means of cophenetic correlation, in which values close to unity indicate good representativeness. The analyses were run using R 2.13.1 (R Development Core Team, 2014). For the grouping and similarity analyses, the 'vegan' package was used (Oksanen, 2013). For the other analyses the native functions of the R software were used.

Results

During the study period the diet of *A. mellifera* consisted of 74 pollen types distributed in 58 genera and 27 families. Families with the highest number of species were Leguminosae (n = 16), Asteraceae (11) and Rubiaceae (6) (Table 1). The genus with the highest number of species (n = 7) in bloom during the study period was *Mimosa* (Leguminosae).

Regarding the proportion of pollen types in the samples during the rainy season (Table 1), the best represented species in January were *Mimosa tenuiflora* (34.88% = AP) and *Psidium cattleianum* (22.23% = AP); in February, March and April, *Mimosa caesalpiniifolia* was dominant (47.38%, 74.75% and 72.88% = DP); *Wedelia calycina* was relevant only in March (16.25% = AP); in May, pollen of *Leucaena leucocephala* and *Mimosa niomarlei* was accessory; in June, pollen of *Baccharis trinervis* was accessory, but very close to dominant (44.93 = AP).

In July, which is the transition period between the rainy and dry seasons, foraging bees collected pollen mostly on *Mimosa tenuiflora* (78.90 = DP). In the dry season, the number of pollen types was lower compared to the rainy season (Table 1). In August, *Attalea speciosa* (46.38% = DP), *Borreira spinosa* (23.88% = AP) and *M. tenuiflora* (17.60 = AP) had the highest proportions in samples.

Table 1. Temporal variation in the diet of *Apis mellifera* in a seasonal semideciduous forest in the State of Ceará, in the period from November 2012 to October 2013. Months highlighted in gray correspond to the rainy season and months highlighted in white represent the dry season. The values in each month correspond to the percentage of each plant species of the whole pollen collected during each month.

Family	Species/Pollinic types	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct
Acanthaceae	*Ruellia asperula* Lindau.									0.05	0.10		
Amaranthaceae	*Alternanthera brasiliana* (L.) O. Kuntze							0.08					
	Alternanthera tenella Colla						0.15	0.20	1.45	0.05	0.73		
Anacardiaceae	*Anacardium occidentale* L.	9.13	2.70							0.08	0.05	0.18	0.95
	Myracrodrum urundeuva Allemão								0.20	1.35	**95.33**	13.00	
Arecaceae	*Attalea speciosa* (Mart.) Barb. Rodr.	**53.80**	**64.85**	7.78	4.18	0.85	0.45	0.08	6.23	2.83	**46.38**	2.88	20.03
Asteraceae	*Acanthospermum hispidum* DC.								0.03				
	Baccharis trinervis Pers.				2.88	5.05	0.38	6.90	44.93	0.70	0.18	0.40	
	Bidens subalternans DC.							0.08					
	Emilia sonchifolia (L.) DC. ex Wight						0.23	0.20		0.03			
	Melanthera latifolia (Gardner) Cabrera						0.65		4.65	0.15			
	Pithecoseris pacourinoides Mart.								5.30	0.55	0.10		
	Stilpnopappus tomentosus Mart. ex DC.							0.03	0.03				
	Trichogonia salviifolia Gardner					0.10			0.05				
	Tridax procumbens L.					0.10		2.00	4.60	0.13	2.60		
	Vernonanthura brasiliana (L.) H. Rob.	0.08							0.08		1.03	0.28	0.05
	Wedelia calycina Rich.		4.15	9.35	8.60	16.25	11.40	3.60	4.68		0.05		
Boraginaceae	*Cordia trichotoma* (Vell.) Arrab		0.70	0.93	0.15				0.68	1.28	1.15		
Commelinaceae	*Aneilema brasiliense* C.B. Clarke							0.05	0.53	0.05			
	Commelina benghalensis L.					0.15	3.50	0.80	3.55	0.65			
	Commelina diffusa Burmf.				0.15	0.05	0.33	0.03					
Convolvulaceae	*Ipomoea piurensis* O´Donell								0.03				
	Merremia macrocalyx O'Donell									0.03	0.45		
Convolvulaceae	sp1								0.03		0.03		
	Turbina cordata Choisy						0.03						
Euphorbiaceae	*Croton floribundus* Spreng				0.25	0.03							
	Croton jacobinensis Baill			0.30		0.08							
	Croton microcalyx Mull. Arg.					0.10	0.08						
Lamiaceae	*Hyptis pectinata* (L.) Poit.				0.03				1.78				
	Hyptis suaveolens (L.) Poit							0.30	6.70	4.05			
	Ocimum gratissimum L.			0.03	0.10	0.18	0.40	0.38	0.48	0.10			
Leguminosae	*Anadenanthera columbrina* (Vell) Brenan	0.15	2.83	0.50	0.33								0.25
	Bauhinia cheilantha (Bong) Steud.						0.03			0.05	0.35	0.05	
	Bauhinia ungulata L.								0.05				
	Delonix regia (Bojer ex hook.) Raf.				0.53		0.55	0.30	0.10			0.25	
	Dioclea grandiflora Mart ex Benth								0.08				

Table 1. Temporal variation in the diet of *Apis mellifera* in a seasonal semideciduous forest in the State of Ceará, in the period from November 2012 to October 2013. Months highlighted in gray correspond to the rainy season and months highlighted in white represent the dry season. The values in each month correspond to the percentage of each plant species of the whole pollen collected during each month. (Continuation)

Family	Species/Pollinic types	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct
Leguminosae	*Indigofera suffruticosa* Mill.							0.30					
	Leucaena leucocephala Wit.							37.78	2.78		0.05		
	Mimosa caesalpiniifolia Benth		1.55		**47.38**	**74.75**	**72.88**	3.05	0.08				
	Mimosa candolei R. Grether									0.33	0.45		
	Mimosa invisa Mart					0.18	2.35		2.68				
	Mimosa niomarlei Afr. Fer.						0.40	28.00	1.85				
	Mimosa sensitiva L.							0.08	0.55		0.95		
	Mimosa setosa Benth							0.23	1.50	0.28	0.58		
	Mimosa tenuiflora (Willd) Poir.	4.18	5.30	34.88		0.28	0.80	4.65	2.20	**78.90**	17.60		6.50
	Piptadenia stipulacea (Benth) Ducke					0.13							
	Senna splendida (Vogel) H.S.I						0.38						
Loranthaceae	*Struthanthus syringifolius* (Mart.) Mart.		5.33										
Malvaceae	*Guazuma ulmifolia* Lam			6.80	0.50	0.05	0.03						
	Sida spinosa L.								0.03	0.08	0.03		
	Triumfetta rhomboidea Jacq									0.03	0.45		
Melastomataceae	sp1							0.10					
Meliaceae	*Azadirachta indica* A. Juss.			5.30	0.05								
	Cedrela odorata L.			4.13									
Myrtaceae	*Eucalyptus citriodora* Hook		0.08			0.18	1.08	0.33					0.18
	Eugenia uniflora L.												0.13
	Psidium cattleianum Sabine			22.23	28.08	0.78							0.10
	Psidium guajava var. pomifera L.			7.00	6.35		0.03		0.33		0.25		1.28
Nyctaginaceae	*Boerhavia difusa* L.								0.08	0.25	0.25		
Onagraceae	*Ludwigia octovalvis* (Jacq.) P.M		0.08										
Passifloraceae	*Passiflora cincinnata* Mast.		0.15		0.03	0.10	0.10	0.03	0.03		0.28		
Poaceae	*Zeamays* L.		0.03			0.65	1.33	0.03					
Rubiaceae	*Borreria latifolia* (Aubl.) K. Schum						2.10						
	Borreira spinosa (L.) Cham							3.93	3.08	13.08	23.88		
	Diodella apiculata Delpret.						0.03	0.05	0.05				
	Manettia cordifolia Mart.									0.05	0.20	0.05	0.03
	Spermacoce sp.										0.05		
	Spermacoce verticillata L								1.10				
Rutaceae	*Citrus limonia* Osbeck		1.28	0.68	0.13								
Sapindaceae	*Cardiospermum corindum* L.						0.08	0.03	0.10	0.10	0.18		
Solanaceae	*Brugmansia suaveolens* (Humb)										0.03		
Turneraceae	*Turnera* sp.							0.05	0.28	0.03	0.03	0.03	
Urticaceae	*Cecropia pachystachya* Trécul	32.68	11.00	0.13								0.83	**57.53**
Verbenaceae	*Lantana camara* L.				0.33								
Taxa (S)		**6**	**14**	**14**	**18**	**20**	**27**	**30**	**38**	**26**	**31**	**9**	**12**
Shannon index (H')		**1.065**	**1.343**	**1.909**	**1.480**	**0.885**	**1.169**	**1.866**	**2.280**	**0.838**	**1.591**	**0.246**	**1.230**
Equitability (J')		**0.595**	**0.509**	**0.724**	**0.512**	**0.295**	**0.355**	**0.549**	**0.627**	**0.257**	**0.463**	**0.112**	**0.495**
Berger-Parker (D)		**0.538**	**0.649**	**0.349**	**0.474**	**0.748**	**0.729**	**0.378**	**0.449**	**0.789**	**0.464**	**0.953**	**0.575**

In September, the diet was monofloral, composed almost exclusively by pollen of *Myracrodruom urundeuva* (95% = DP). In October and November, *Cecropia pachystachya* (57.53% = DP; 32.68 = AP, respectively) and *Attalea speciosa* (20.03% = AP; 53.80 = DP) stood out over the other species used by *A. mellifera*. In December, *Attalea speciosa* predominated in the diet of honeybees (64.85%).

The cluster analysis reliably showed (cophenetic correlation = 0.8545) that colonies have high similarity as to bee pollen, with a minimum similarity of approximately 60% (Fig 2).

Pollen production was greater in the rainy season (344.76 ± 43.33 g/col./month) than in the dry season (149.20 ± 20.45 g/col./month) (Fig 3a). In March and April, we found the highest productions of bee pollen (517.61 g/col. and 454.09 g/col., respectively). Pollen production was positively related to rainfall (β = 1.23, t = 8.95, p <0.001, Fig 4a), but the richness of pollen types was not associated with production (β = -2.40; t = -1.61, p = 0.142).

The nutritional value of pollen sampled also varied throughout the study period (Fig 3b-3f). The protein content showed higher levels in the dry-rainy-dry seasons transitions
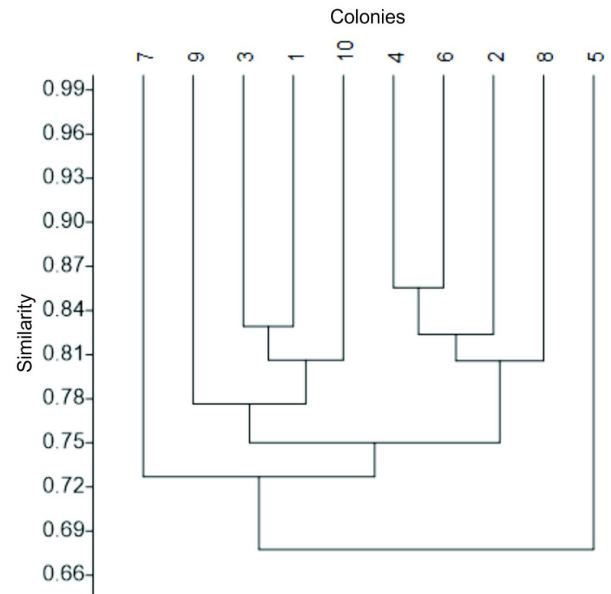
Fig 2. Similarity dendrogram (Bray-Curtis index and paired group algorithm) in the composition of pollen sampled in the colonies of *Apis mellifera* in a seasonal semideciduous forest in the State of Ceará, in the period from November 2012 to October 2013.
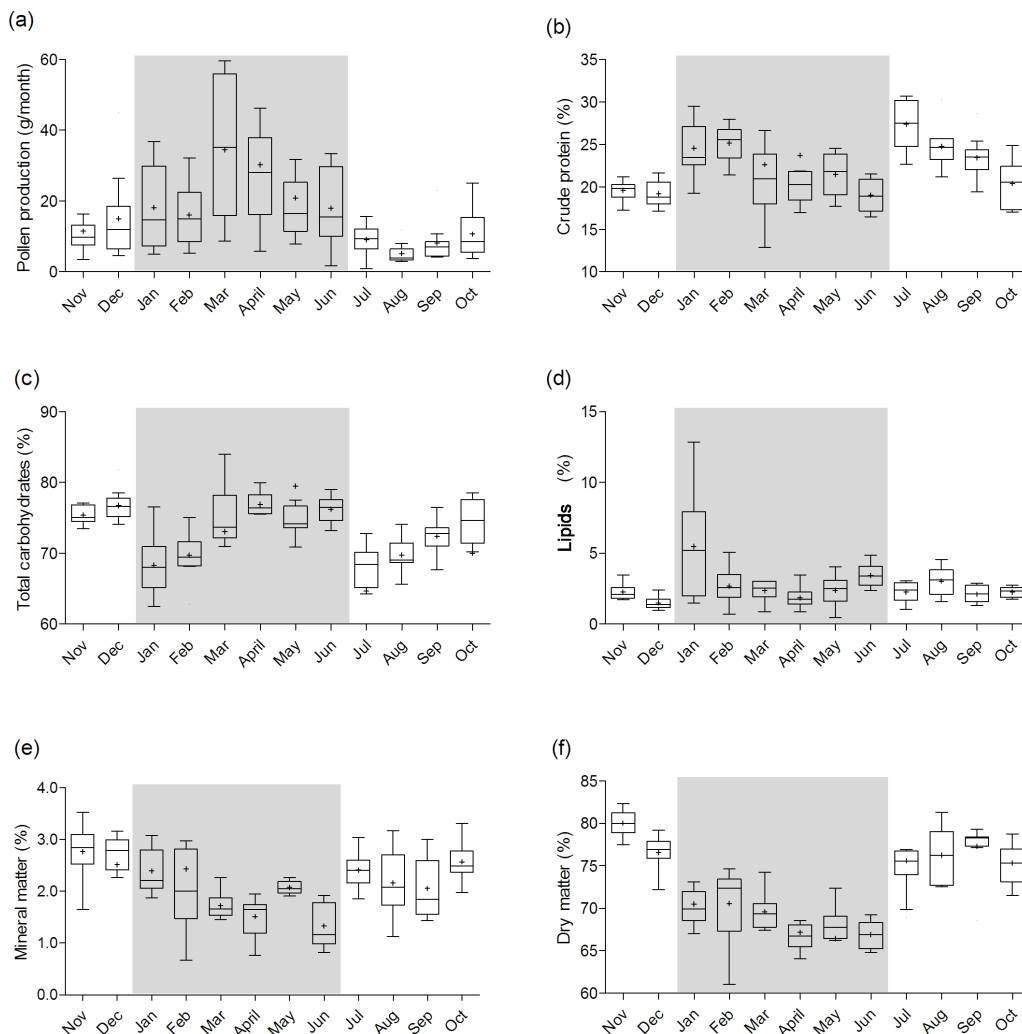
Fig 3. Temporal variation in production of bee pollen used in the diet of *Apis mellifera* and its nutritional value. (**a**) Annual production of bee pollen. (**b**) Total protein. (**c**)Total carbohydrate. (**d**) Lipids. (**e**) Mineral matter. (**f**) Dry matter. The symbol + represents the mean of each month. Months highlighted in gray correspond to the rainy season and months highlighted in white represent the dry season.

(Fig 3b). Total carbohydrates were inversely proportional to the protein content (Fig 3b and 3c). Lipids concentration showed little variation, except for the January sample in which discrepant values when compared to the other months (5.46%) occurred (Fig 3d). The average annual mineral matter of bee pollen was 2.15%, with highest level in November (2.76%) and lowest in July (1.33%) (Fig 3e). Regarding dry matter, in the rainy season we obtained lower levels (68%) when compared to the months that there was no precipitation.

Analyzing the relationship between nutrients (CP, TC, FAT, MM, DM), number of pollen types and rainfall, we observed that the lipids was positively related to the number of pollen types (Table 2, Fig 4b). The mineral matter was negatively correlated with the number of pollen types (Fig 4c), while the dry matter was negatively correlated with rainfall (Table 2; Fig 3f and 4d). No relationship was detected between protein (or carbohydrate) with rainfall and the number of pollen types (Table 2). We also found a positive relationship between precipitation and pollen production (Fig 4a).

In addition, we found significant correlations between dominance of some taxonomic groups with some nutrients.

Table 2. Results of generalized linear models (GLM) of factors that influence chemical variables of bee pollen produced by *Apis mellifera* in a seasonal semideciduous forest in the State of Ceará, in the period from November 2012 to October 2013.

Dependent variables	Independent variables	Estimate (β)	Standard error	t	p
Crude protein	Number of pollen types	0.023	0.097	0.236	0.819
	Precipitation	0.003	0.009	0.349	0.735
Total carbohydrates	Number of pollen types	0.028	0.153	0.182	0.859
	Precipitation	0.009	0.014	0.671	0.519
Lipids	Number of pollen types	**0.037**	**0.016**	**2.321**	**0.049**
	Precipitation	-0.001	0.001	-0.843	0.424
Mineral matter	Number of pollen types	**-0.023**	**0.010**	**-2.385**	**0.041**
	Precipitation	-0.002	0.001	-2.003	0.076
Dry matter	Number of pollen types	-0.124	0.063	-1.969	0.085
	Precipitation	**-0.041**	**0.007**	**-5.684**	**0.000**
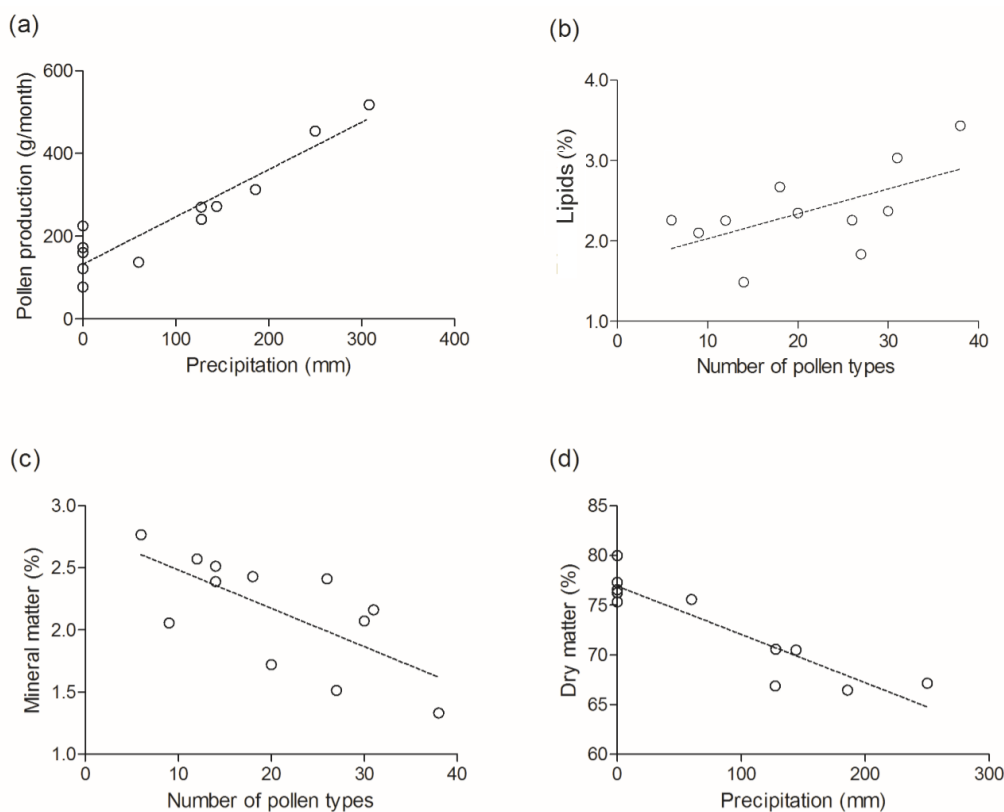
Fig 4. Spearman correlation between dependent variables. (**a**) Annual production of bee pollen and rainfall. (**b**) Lipids and number of pollen types (**c**) Mineral matter and pollen types (**d**) Dry matter and rainfall.

There was a positive correlation between the protein content and dominance of pollen of the genus *Mimosa* (Leguminosae) and of the family Malvaceae (Table 3). Total carbohydrates were negatively correlated with the amount of Malvaceae pollen and positively with the amount of Poaceae pollen. The lipids was positively correlated with the presence of the family Malvaceae

species, while the mineral matter was negatively correlated with Asteraceae, Commelinaceae, Lamiaceae and positively with Arecaceae and Urticaceae (Table 3). These results were found in the correlation analysis based on the number of pollen grains of all plant species as well as in the analysis using the total pollen volume of each plant species in the bee diet.

Table 3. Correlation between taxonomic groups and chemical variables of bee pollen produced by *Apis mellifera* in a seasonal semideciduous forest in the State of Ceará, in the period from November 2012 to October 2013. The rs values are presented for dominance in the diet (% of pollen grains) and pollen volume of each plant group.

	Taxonomic groups	Crude protein		Total carbohydrates		Lipids		Mineral matter	
		rs	p	rs	p	rs	p	rs	P
Dominance (%)	*Mimosa*	**0.594**	**0.042**	-0.147	0.649	0.210	0.513	-0.329	0.297
	Asteraceae	-0.105	0.746	0.406	0.191	0.413	0.183	**-0.727**	**0.007**
	Commelinaceae	-0.011	0.972	0.377	0.227	0.168	0.602	**-0.664**	**0.018**
	Lamiaceae	0.015	0.964	0.370	0.237	0.348	0.268	**-0.653**	**0.021**
	Malvaceae	**0.723**	**0.008**	**-0.668**	**0.018**	**0.679**	**0.015**	-0.163	0.612
	Arecaceae	-0.329	0.297	-0.196	0.542	-0.035	0.914	**0.636**	**0.026**
	Poaceae	-0.184	0.568	**0.684**	**0.014**	-0.421	0.173	-0.388	0.213
	Urticaceae	-0.460	0.132	-0.008	0.981	-0.460	0.132	**0.663**	**0.019**
Volume pollen (cm³)	*Mimosa*	**0,678**	**0,015**	-0,413	0,183	0,476	0,118	-0,252	0,430
	Asteraceae	0,028	0,931	0,217	0,499	0,490	0,106	**-0,748**	**0,005**
	Commelinaceae	0,116	0,720	0,310	0,327	0,146	0,652	-0,575	0,051
	Lamiaceae	0,015	0,964	0,370	0,237	0,348	0,268	**-0,653**	**0,021**
	Malvaceae	**0,631**	**0,028**	**-0,638**	**0,026**	**0,653**	**0,021**	-0,254	0,426
	Arecaceae	-0,329	0,297	-0,196	0,542	-0,035	0,914	**0,636**	**0,026**
	Poaceae	-0,184	0,568	**0,684**	**0,014**	-0,421	0,173	-0,388	0,213
	Urticaceae	-0,460	0,132	-0,008	0,981	-0,460	0,132	**0,663**	**0,019**

Discussion

Bee flora of the seasonal semideciduous forest studied proved to be well diversified, and the foraging workers of *A. mellifera* interacted with many plant species, as observed in other biomes (Pacheco Filho et al., 2015). However, the workers of the colonies tended to constantly search for floral resources in plants of specific taxonomic groups, possibly because such plants provide further floral resources (nectar or pollen) at the time of collection or because of the quality of such resources (Hill et al., 1997).

The family Leguminosae was the most frequent in the study area (Santos et al., 2014) and species such as *Mimosa caesapiniifolia* and *M. tenuiflora* were constantly present in the diet of *A. mellifera*. These two plant species are responsible for the highest availability of nectar and pollen in the Caatinga (Maia-Silva et al., 2012). The importance of the genus *Mimosa* in maintaining the diet of stingless bee species was reported for *Trigona spinipes*, *Partamona rustica*, in the State of Ceará (Blochtein et al., 2010), *Melipona subnitida* and *Melipona scutellaris* State of Rio Grande do Norte (Maia-Silva et al., 2015).

Pollen of *Myracrodrum urundeuva* was collected in high proportions during the dry season and in August it was monofloral. Its flowering in periods of low water availability highlights the importance of this species for the maintenance of *A. mellifera* colonies, as well as of native bees (Maia-Silva et al., 2012; Araujo-Filho, 2013).

Attalea speciosa was present in the diet of bees throughout the year, becoming dominant in November, December and August. Studies on pollen production in other regions of the northeastern Brazil reported that species of Arecaceae (e.g., *Cocos nucifera*) are always frequent throughout the year in the diet of honeybees, emphasizing the importance of this family for *A. mellifera* (Almeida-Muradian et al., 2005; Arruda et al., 2013; Alves & Santos, 2014).

Pollen production varied throughout the year with two peaks, one in the dry season with lower productivity and in the rainy season, with higher values attributed mainly to pollen provided by *Mimosa caesapiniifolia*. An important plant for the commercial production of bee pollen, as was certified in samples from Rio Grande do Norte, Bahia, Sergipe and São Paulo states (Melo et al. 2018). Fluctuations in pollen production are common, due to the influence of many productivity factors (Negrão et al., 2014), including the flowering of one or more species of abundant plants in the site, weather conditions, colony size, of brood area size and queen age (Dimou & Thrasyvoulou, 2007; Rebolledo et al., 2011; Avni et al., 2014).

Carbohydrates were the most abundant group of nutrients, followed by proteins. Both, the protein and carbohydrate contents varied throughout the year, probably due to the variation in the botanical source, environmental conditions, and factors related to the handling and storage (Villanueva et al., 2002; Yang et al., 2013; Sattle et al., 2015).

Protein increment in the bee diet is attributed mainly to the presence of *Mimosa* because the volume of pollen

grains of this genus was ten times greater than the volume of any other plant species found in the diet. Therefore, despite the positive correlation between total protein and Malvaceae pollen, the total pollen grain volume of this group is small suggesting that Malvaceae are less important protein sources when compared to *Mimosa*. Similarly, carbohydrate levels showed a negative correlation with Malvaceae and positive correlation with Poaceae, but their variability can be related mainly to the presence of Poaceae pollen due to its greater volume, as observed by Yang et al. (2013).

In turn, lipids remained constant throughout the year, and the lipids content was attributed to polliniferous sources. Bees select pollen with high levels of unsaturated fatty acids, which are better suited to bee metabolism (Estevinho et al., 2012; Avni et al., 2014). A fatty compound that contributes to levels of fatty acid esters is found in *pollenkitt* covering the whole grain surface, which is evident in many plant species (Pacini & Casadoro 1981; Dobson, 1988). In addition, the positive correlation of pollen types richness with lipids suggests that the larger the number of pollen types harvested by bees, the larger the chances of increasing the average lipids content in the pollen taken to the colony.

Bees cannot synthesize minerals, and other nutrients, and it is through pollen that they get the mineral quantities required for the structural maintenance of the individuals and the colony (Brodschneider & Crailsheim, 2010). The content of mineral matter in this study proved to be quite stable, with no discrepancy in values. High levels of mineral matter may be due to incorrect handling of the product by beekeepers, or inefficient cleaning process (Sattle et al., 2015). Nevertheless, a negative correlation between the mineral matter and the number of pollen types implies that the plant species that were used as resources in the bee diet during the dry season (when there are less pollen types and lower plant species richness) contributed with higher levels of minerals.

The level of dry matter is one of the main items to be monitored in pollen production. Herein, the water content was slightly increased in the months with higher rainfall and higher relative humidity. High levels of humidity can compromise the quality and potentially promote microbial growth because the bee pollen is a highly hygroscopic product (Barreto et al., 2005; Melo et al., 2015).

Floristic diversity, nutritional quality, reduced exposure to pesticides coupled with adequate management are factors that can contribute to colony productivity and performance (Colwell et al., 2017). Even so, the percentages based on the dry matter of CP, FAT, CT and MM are within the values found in the national and international literature (Yang et al., 2013; Negrão et al., 2014; Sattle et al., 2015).

Conclusion

It is known that *A. mellifera* has an extremely plastic behavior, and that its foraging on floral sources is mostly related to the abundance of resources as well to the floral density than to the species specific features (Stang, 2007). Here, we find that these honeybees invest most of its pollen foraging efforts in the genus *Mimosa* (Leguminosae) *Myracrodrum* (Anacardiaceae), *Cecropia* (Urticaceae) and the species *Attalea speciosa*. In this way, it is very important to keep these plants in the vicinity of the apiaries.

Although the study area is under semiarid climate regime and subjected to long periods of water deficit, the studied seasonal semideciduous forest allows production of bee pollen throughout the year without the use of artificial feeding. In addition, the bee pollen produced has a nutritional value similar to that observed in other countries, indicating that this product is within national and international standards.

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