



## RESEARCH ARTICLE - BEES

## Does Seasonality Affect the Nest Productivity, Body Size, and Food Niche of *Tetrapedia curvitaris* Friese (Apidae, Tetrapediini)?

ES CAMPOS<sup>1</sup>, TN ARAÚJO<sup>1</sup>, LS RABELO<sup>1</sup>, EMA BASTOS<sup>2</sup>, SC AUGUSTO<sup>1</sup>

1 - Universidade Federal de Uberlândia, Uberlândia, Minas Gerais, Brazil

2 - Fundação Ezequiel Dias, Belo Horizonte, Minas Gerais, Brazil

### Article History

#### Edited by

Cândida Aguiar, UEFs, Brazil

Received 10 April 2018

Initial Acceptance 18 June 2018

Final acceptance 18 August 2018

Publication date 11 October 2018

#### Keywords

Pollen sources, bee, trap-nest, Brazilian savanna.

#### Corresponding author

Solange Cristina Augusto

Universidade Federal de Uberlândia

Instituto de Biologia

Rua Ceará s/nº, Bloco 2D, Umuarama

CEP 38400-902 - Uberlândia-MG, Brasil.

E-Mail: solange.augusto@ufu.br

### Abstract

*Tetrapedia curvitaris* Friese is a widely distributed species, frequently attracted by trap-nests. Previous studies have revealed a higher frequency of nesting in the wet season and dimorphism between the sexes, with females exhibiting larger body size than males. We evaluated the effects of seasonality on the production of nests, food niche, and body size of *T. curvitaris*. The study was conducted from April 2009 to March 2010 and from April 2012 to March 2013 at the Água Limpa Experimental Station, located in the Triângulo Mineiro, Minas Gerais State. The number of cells was positively correlated with length and diameter of trap-nests. However, the number of nests and the number of cells produced did not differ between the seasons. The females demonstrated a larger head width than males and both presented greater body size in the hot/wet season. A higher food niche breadth was observed in the hot/wet season and low similarity in the use of pollen sources between seasons (PS=39.05%). Thus, it is concluded that the season has no effect on the production of nests or cells, but rather on the body size of males and females and food niche breadth. There was no interaction between sex and season, *i.e.*, both factors influenced the individuals' size independently. The production of smaller individuals in the dry season could be related not only to the quantity but also the quality of food offered to immature bees.

### Introduction

*Tetrapedia curvitaris* Friese is a widely distributed species, frequently found in surveys in the states of Minas Gerais and São Paulo (Camillo, 2005; Alves-dos-Santos et al., 2007; Pinto et al., 2015). Like other species of the genus *Tetrapedia*, this species has the behavior of nesting in pre-existing cavities (Garófalo et al., 2004; Camillo, 2005; Martins et al., 2012), allowing more detailed study of its nesting behavior.

The species of *Tetrapedia* with known nests, *Tetrapedia diversipes* Klug, *T. curvitaris*, *Tetrapedia rugulosa* Friese, and *Tetrapedia garofaloi* Moure present more than one generation per year, higher activity of individuals in the wet season, and the occurrence of pre-pupal diapause (Camilo, 2005). Additionally, it was observed a biased female sexual ratio for *T. diversipes*, *T. rugulosa*, and *T. garofaloi*, but not

for *T. curvitaris*. All the species studied present females larger than males, except *T. diversipes* (Camilo, 2005).

Availability of food resources is one of the factors that can affect body size of bees. Studies have shown that the size of solitary bees is strongly influenced by larvae feeding (Bosch & Vicens, 2002; Radmacher & Strohm, 2010), with females generally receiving more food than males (Stark, 1992; Strohm, 2000; Peruquetti & Del Lama, 2003).

*Tetrapedia* species, in addition to pollen and nectar, also use floral oil for the nourishment of bees' immature. Malpighiaceae species are important oil sources for *Tetrapedia* (Camillo, 2005; Alves-dos-Santos et al., 2007). Studies on pollen analyses of larval food from nests of *T. diversipes* also revealed the presence of pollen grains of Malpighiaceae species (Menezes et al., 2012; Neves et al., 2014). However, species of the genus *Dalechampia* (Euphorbiaceae) and *Ludwigia* (Onagraceae) were identified as the main sources of pollen for this species.



In the Brazilian savanna, the most important sources of pollen and oil sources for *Tetrapedia* bloom mainly in the hot/wet season (Carpentieri-Pípolo et al., 2008; Coelho & Spiller, 2008; Mendes et al., 2011), such as most woody and herbaceous species (Batalha & Mantovani, 2000). Thus, considering the available information on nesting behavior of *T. curvitaris*, we verified the possible effect of seasonality on nest productivity, food niche breadth, and body size of individuals in a Brazilian savanna area.

## Material and methods

### Study areas and trap-nests

This study was conducted at the Água Limpa Experimental Station (ALES) (19°05'48" S and 48°21'05" W) belonging to the Federal University of Uberlândia and located in the city of Uberlândia, Minas Gerais state. The area covers a mixed system with 104 ha of natural areas of the Brazilian savanna and 194.72 ha of pastures, crops, and orchards (Neto, 2008).

The area also contains 17 ha of predominantly fruit crops, including mango, *Mangifera indica* L., West Indian cherry, *Malpighia emarginata* DC., passion fruit, *Passiflora edulis* Sims, pineapple, *Ananas comosus* (L.) Merr., guava, *Psidium guajava* L., and tamarindo, *Tamarindus indica* L.

The area presents a seasonal tropical climate. It is characterized by cold/dry season, from April to September, and a hot/wet season from October to March (Klink & Machado, 2005).

In the study area, two bee shelters, 2.5 m high and 1.5 m long, were built from wood rafters and plastic canvas covers, and they were situated 200m from each other. These bee shelters were supplied with trap-nests, consisting of bamboo sticks, commercially used for fishing. These bamboo canes were closed at one end by the node, with a length ranging from 8.00 cm to 20.10 cm, and diameters between 0.45 cm and 1.00 cm. New trap-nests were frequently added to maintain the availability of these substrates. The nests were organized inside building bricks, placed on shelves in the shelters.

### Number of nests and cells

The nests of *T. curvitaris* were collected in two periods, from April 2009 to March 2010 (Period 1 = P<sub>1</sub>) and from April 2012 to March 2013 (Period 2 = P<sub>2</sub>). During P<sub>1</sub>, all operculated nests were collected at monthly visits. In P<sub>2</sub>, biweekly visits were made and only half of the total operculated nests were collected.

The collected nests were taken to the Laboratory of Ecology and Behavior of Bees, Federal University of Uberlândia (LECA-INBIO/UFU). Only the individuals from P<sub>2</sub> were used in the analysis regarding the number of cells, according to the season.

The length and diameter of each trap-nest collected in P<sub>2</sub> were measured with the aid of a pachymeter. After

emergence, all individuals were sacrificed, mounted with an entomological pin, and deposited in the collection of LECA-INBIO/UFU. Subsequently, the trap-nests were opened and their contents analyzed.

### Body size of males and females

All individuals of *T. curvitaris* that emerged from nests collected during P<sub>2</sub> were measured. The size of males and females was determined from the maximum head width (in millimeters) (Fig 1) using the ImageJ 1.44p program (Rasband, 1997-2011). For this, all individuals were photographed using a digital camera, positioned parallel to the measuring structure. The program was calibrated using the 10 mm scale in each photo.

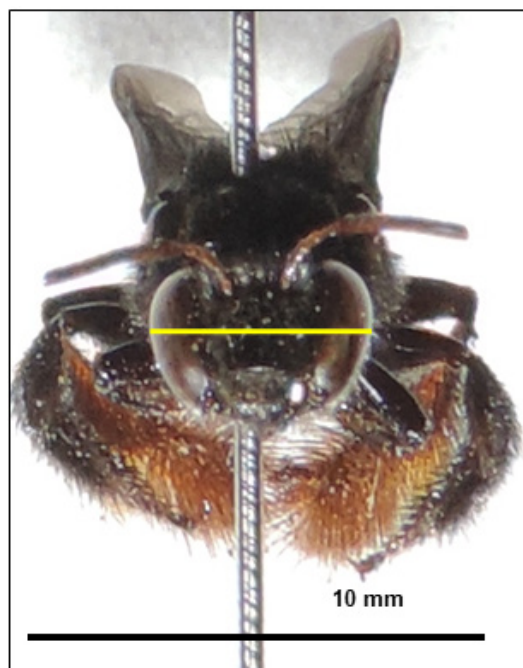


Fig 1. Measure of the head maximum width of *Tetrapedia curvitaris*.

### Food niche

During the P<sub>2</sub> samples of larval food were removed from 22 active nests of *T. curvitaris*, 11 from the cold/dry season and 11 from the hot/wet season. The samples were obtained by introducing pollen collectors made of wooden sticks and pins at the nest's entrance. The larval food samples were stored into 15 mL centrifuge tubes, with 2 ml 70% alcohol, and processed using the acetolysis method (Erdtman, 1960).

Three slides were prepared from each larval food sample ( $n = 66$  slides). The slides were deposited in the pollen slide collection of the Laboratory of Plant Morphology, Microscopy and Image of the Federal University of Uberlândia (LAMOVI-INBIO/UFU). The characteristics of the pollen grains outlined by the acetolysis were used to compare the grain morphology of collected samples with the literature (Salgado-Labouriau, 1973; Roubik & Moreno, 1991), a database of pollen grain images (Bastos et al., 2008), and a reference slide collection from LAMOVI-INBIO/UFU.

Quantitative analysis of the larval food sample was performed by scanning the entire microscope slide using 800 $\times$  of magnification. For this, the cover slip was divided into four quadrants and approximately 400 pollen grains were counted, 100 grains in each quadrant and all grains present in quadrants that had less than 100 grains (Vilhena et al., 2012). Approximately 1,200 grains were counted per nest. Pollen types occurring in abundances lower than 3% in each sample were excluded from the analysis, as they were considered as contaminants.

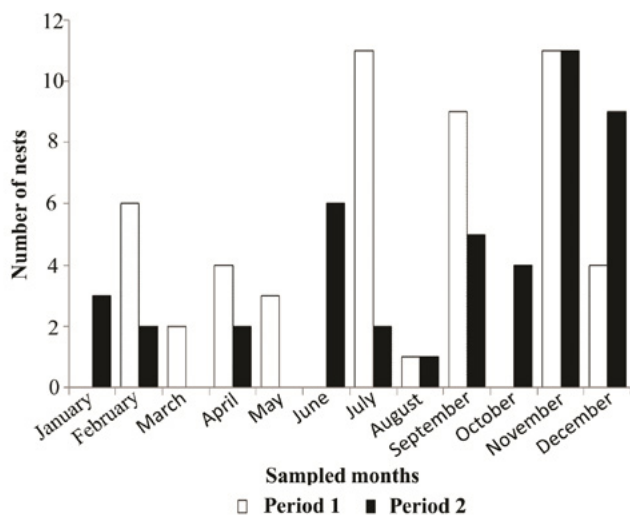
#### Data analysis

To verify possible differences in the number of nests between the hot/wet and cold/dry seasons, a t-test was performed for the data collected in P<sub>1</sub> and the Mann-Whitney test for the data collected in P<sub>2</sub>. A Pearson correlation test was performed to verify if there was an association between the length and diameter of trap-nests and the number of brood cells produced, considering all nests, independent of the season. A t-test was also performed to evaluate differences between the number of cells produced and the seasons of the year.

To evaluate if there was a difference in the size of the individuals between the sexes, according to the season, and whether there was an interaction between these factors, a Two-Factor ANOVA was performed using the maximum head width measurement.

All analyzes were performed in the R Studio program (version 3.4.0) (R Core Team, 2017) and the normality was tested using Kolmogorov–Smirnov (Lilliefors;  $P > 0.05$ ).

The food niche breadth in each season was calculated from the Shannon-Wiener Diversity Index (Camillo & Garófalo, 1989; Aguiar et al., 2013), and later compared by the Hutcheson t-test. In order to establish the degree of uniformity of pollen collection in the species of plants visited by bees, the Pielou Index ( $J'$ ) was used. These analyzes were performed in the



**Fig 2.** Abundance of *Tetrapedia curvitaris* nests obtained in Period 1 (April 2009 to March 2010) and in Period 2 (April 2012 to March 2013), at the Água Limpa Experimental Station, Uberlândia-MG.

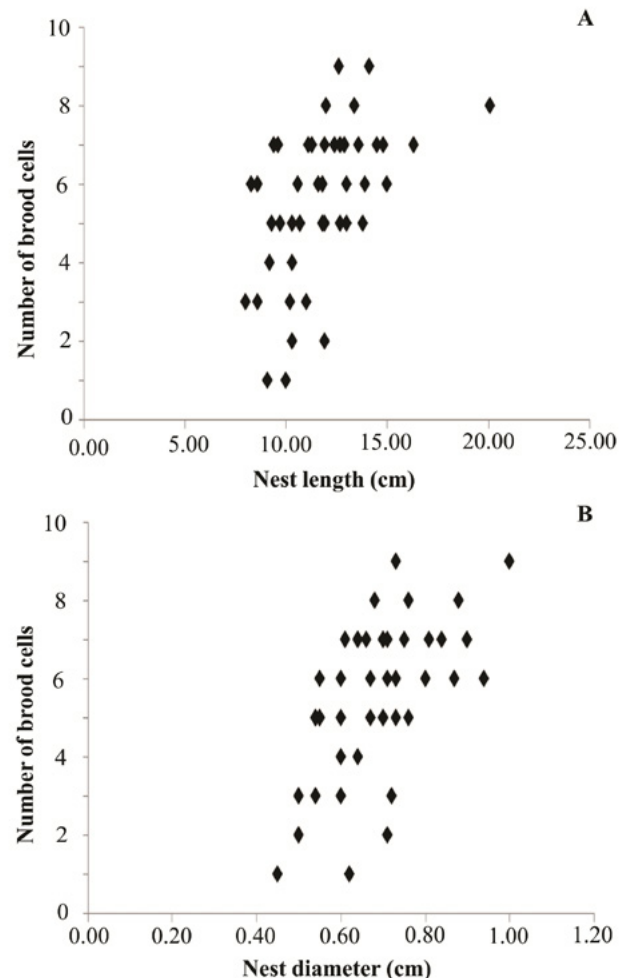
PAST program 2.13 (Hammer et al., 2001; Zar, 2013). To verify the similarity in the use of pollen sources between the seasons of the year, the percentage of similarity index  $PS = \Sigma$  of the lowest percentage of each pollen type was calculated (Brower et al., 1997).

## Results

#### Number of nests and cells

A total of 51 nests of *T. curvitaris* were collected during P<sub>1</sub> and 45 nests in P<sub>2</sub>. Nests were collected throughout the year, and no significant difference was observed in the number of nests between the hot/wet and cold/dry seasons (P<sub>1</sub>:  $t = 0.33$ ,  $df = 10$ ,  $p > 0.05$ ; P<sub>2</sub>:  $U = 12.50$ ,  $n_1 = 6$ ,  $n_2 = 6$ ,  $p > 0.05$ ) (Fig 2).

The number of cells per nest ranged from 1 to 9. Nests containing 5 to 7 cells were the most frequent, representing 66.67% of nests collected. The number of cells was positively correlated with bamboo length ( $r = 0.52$ ,  $df = 43$ ,  $p < 0.05$ ) (Fig 3A) and diameter ( $r = 0.58$ ,  $df = 43$ ,  $p < 0.05$ ) of trap-nests (Fig 3B). However, as the number of nests produced, the number of cells did not differ significantly between seasons ( $t = 0.49$ ;  $df = 43$ ;  $p > 0.05$ ).



**Fig 3.** Number of brood cells produced in *Tetrapedia curvitaris* nests, according to length (A) and diameter (B) of trap-nests.

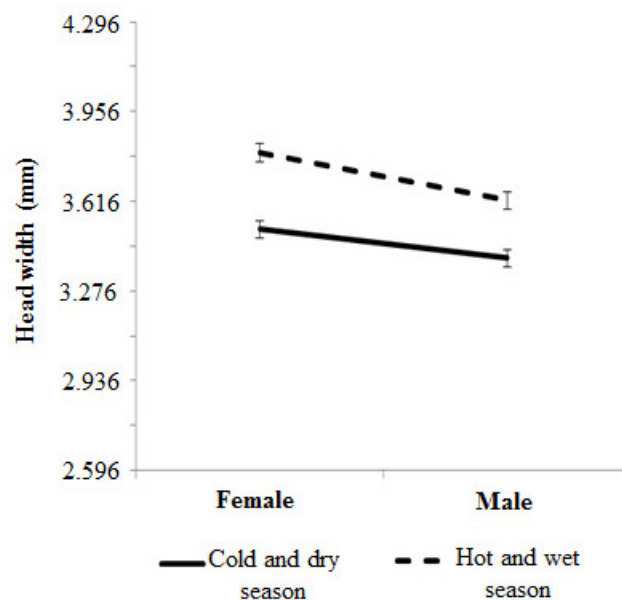
*Body size of males and females*

A total of 112 individuals were measured, 44 males and 68 females, 46 individuals from the cold/dry season and 66 from the hot/wet season. The females presented larger head widths than the males ( $F_{1,108} = 9.36$ ;  $p < 0.05$ ) (Fig 4). Both males and females presented a higher maximum head width in the hot/wet season ( $F_{1,108} = 28.46$ ;  $p < 0.05$ ) (Fig 4). However, there was no interaction between sex and season ( $F_{1,108} = 0.56$ ;  $p > 0.05$ ), i.e., both factors influenced the individuals' size independently.

*Food niche*

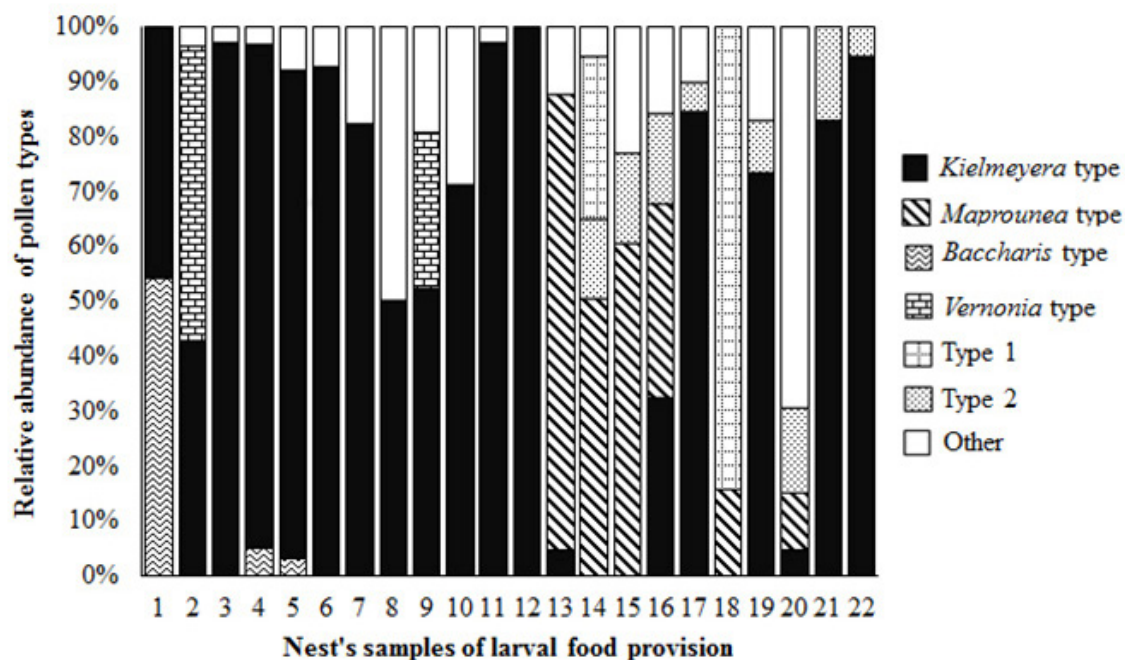
*T. curvitaris* used 22 pollen types as sources of pollen for larval food, belonging to eight botanical families: Asteraceae, Clusiaceae, Euphorbiaceae, Fabaceae, Lamiaceae, Malpighiaceae, Myrtaceae, and Sapindaceae. According to abundance and frequency data, *Kielmeyera* type was abundantly collected and presented in all nests in the cold/dry season and 72.72% of the nests in the hot/wet season. In the hot/wet season, 15 pollen types were identified. The most abundant pollen types were *Kielmeyera* (37.24%), *Maprounea* (27.55%), type 1 (12.93%), and type 2 (9.35%) (Fig 5). Twelve pollen types were identified in nests collected in the cold/dry season. The most abundant pollen types in this season were *Kielmeyera* (73.89%), *Vernonia* (9.04%), and *Baccharis* (6.19%) (Fig 5).

*T. curvitaris* presented a higher food niche breadth in the hot/wet season ( $H' = 1.83$ ) compared to the cold/dry



**Fig 4** - Maximum head width in millimeters (averages  $\pm$  standard error) of *Tetrapedia curvitaris* collected between the seasons (cold and dry season and hot and wet season) in the period from April 2012 to March 2013, at the Água Limpa Experimental Station, Uberlândia-MG.

season ( $H' = 1.04$ ) ( $t = -43,947$ ,  $df = 17.516$ ,  $p < 0.05$ ). Higher uniformity in the collection of pollen was also observed in the hot/wet season ( $J' = 0.66$ ) than cold/dry season ( $J' = 0.42$ ). In addition, similarity in the use of pollen sources between the seasons was low ( $PS = 39.05\%$ ).



**Fig 5.** Relative abundance of the pollen types sampled in the larval food of *Tetrapedia curvitaris* in cold/dry (samples 1 to 11) and hot/wet seasons (samples 12 to 22) in the period from April 2012 to March 2013, at the Água Limpa Experimental Station, Uberlândia-MG. \*The category "Other" represents the other pollen types which presented relative abundance less than 5%.

## Discussion

In the present study, we did not observe an effect of seasonality in the nest productivity. Camillo (2005) suggests that during the cold/dry season, mainly between June and September, immature of *T. curvitaris* probably present a pre-pupal diapause, thus reducing the activity of the bees during this period. In our study, although the number of nests and cells found in the cold/dry season was numerically smaller, there was no significant difference between the seasons, as registered in other studies (Camillo, 2005; Mesquita & Augusto, 2011).

The highest number of cells was verified in nests with longer bamboo length and larger diameter, indicating that the females can optimize the use of the substrates for nesting. The number of brood cells produced by solitary bee species on each nesting substrate tends to increase with the greater space availability (Pereira et al., 1999; Aguiar & Garófalo, 2004; Ramos et al., 2010). It is probable that this mechanism is related to the high energy cost of searching for new places for the construction of nests. The number of cells produced in a nest is also dependent on availability of food (Ramos et al., 2010).

In addition, the choice of cavities by foundress females can affect the body size of the offspring. Roulston and Cane (2002) verified that species of cavity-nesting bees presented significantly greater variation in body size compared to ground-nesting species. Thus, these authors suggest that the choice of cavity utilized may be a more important predictor of offspring body size than parental body size.

Females of *T. curvitaris* were larger than males, pattern observed for other species of solitary bees (Pereira et al., 1999; Camillo, 2005; Bosch & Vicens, 2006). The amount of food deposited in the brood cells strongly affects the size of the individuals produced (Roulston & Cane, 2002; Bosch & Vicens, 2002; Radmacher & Strohm, 2010). Female bees and wasps are usually larger than males, due to the greater amount of food supplied by the nesting female for their development (Stark, 1992; Strohm, 2000; Peruquetti & Del Lama, 2003).

Especially for females, the body size is a selective force that influences surviving and reproduction. A study realized with a solitary bee species, *Osmia cornuta*, recorded that small females have larger chances of dying during their development, disperse more frequently from their natal nesting site, present lower provision rates and produce more males (Bosch & Vicens, 2006).

In the present study, we also observed a seasonal effect in the size of both sexes males and females being significantly larger in the hot/wet season. Greater amount of food stored in the brood cell could explain the occurrence of larger individuals, as observed in other studies (Roulston & Cane, 2002; Bosch & Vicens, 2002; Radmacher & Strohm, 2010).

However, another possible explanation could be differences in composition and abundance of the pollen types used in the two seasons, as verified in the present study. While in the hot/wet season *Kielmeyera* type and *Maprounea*

type together represented, approximately, 65% of the pollen grains sampled, in the cold/dry season *Kielmeyera* type alone constituted 74% of the sample. In addition, in general, the similarity in pollen type diversity between seasons was low.

The flexible pollen foraging performed by polylectic bees such as *T. curvitaris* during the two different seasons can result in larval food consisting of pollen types that differ in protein content. It is known that the pollen quality of different plant families can vary, affecting bee growth and mortality (Roulston & Cane, 2002). Experimental study conducted with a solitary bee species *Lasioglossum zephyrum* (Halictidae) recorded increase of body sizes of the individuals submitted to diet contained rich-protein pollens (Roulston & Cane, 2002). Then, we suggest that changes in the pollen quality of larval food between seasons could also affect the body size of individuals of *T. curvitaris*.

In conclusion, although we did not observe an effect of season on the number of nests or cells produced we recorded an effect on the body size of both sexes and on food niche breadth. The results suggest the occurrence of a possible compensatory effect, in which the productivity of the nests is maintained, but there is a change in the quantity and/or quality of available food and, consequently, the production of smaller individuals. The influence of quantity and quality of floral resource, especially pollen, in the determination of body size is an issue that we intend to investigate in future studies.

## Acknowledgments

This study was supported by grants from Fundação de Amparo à Pesquisa do Estado de Minas Gerais (FAPEMIG) and Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq). This study was financed in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior - Brasil (CAPES) - Finance Code 001. ES Campos received a fellowship from CAPES and S.C. Augusto received research fellowship from CNPq (306298/2015-5).

## Authors' Contribution

ES Campos did field work and laboratory; LS Rabelo, EMA Bastos helped with pollen analyses; ES Campos, TN Araújo, LS Rabelo, SC Augusto worked in the statistical analysis. All authors contributed to the writing of the manuscript.

## References

- Aguiar, C.M.L. & Garófalo, C.A. (2004). Nesting biology of *Centris (Hemisiella) tarsata* (Hymenoptera, Apidae, Centridini). *Revista Brasileira de Zoologia*, 21: 477-486
- Aguiar, C.M.L., Santos, G.M.M., Martins, C.F. & Presley, S.J. (2013). Trophic niche breadth and niche overlap in a guild of flower-visiting bees in a Brazilian dry forest. *Apidologie*, 44: 153-162. doi: 10.1007/s13592-012-0167-4

- Alves-dos-Santos, I., Machado, I.C. & Gaglianone, M.C. (2007). História natural das abelhas coletoras de óleo. *Oecologia Brasiliensis*, 11: 544-557.
- Bastos, E.M.A.F., Thiago, P.S.S., Santana, R.M. & Travassos, A. (2008). Banco de imagens de grãos de pólen: mais de 130 espécies de plantas apícolas. (CD-ROM).
- Batalha, M.A. & Mantovani, W. (2000). Reproductive phenological patterns of Cerrado plant species at the Pé-De-Gigante Reserve (Santa Rita Do Passa Quatro, Sp, Brazil ): a comparison between the herbaceous and woody floras. *Revista Brasileira de Biologia*, 60: 129-145. doi: 10.1590/S0034-71082000000100016
- Bosch, J. & Vicens, N. (2002). Body size as an estimator of production costs in a solitary bee. *Ecological Entomology*, 27: 129-137. doi: 10.1046/j.1365-2311.2002.00406
- Bosch, J. & Vicens, N. (2006). Relationship between body size, provisioning rate, longevity and reproductive success in females of the solitary bee *Osmia cornuta*. *Behavioral Ecology and Sociobiology*, 60: 26-33
- Brower, J.E., Zar, J.H. & Von Ende, C.N. (1997). *Field & laboratory methods for general ecology*. EUA: Wm. C. Brown Publishers, 256 p
- Camillo, E. (2005). Nesting biology of four *Tetrapedia* species in trap-nests (Hymenoptera: Apidae: Tetrapediini). *Revista de Biología Tropical*, 53: 175-186. doi: 10.15517/rbt.v53i1-2.14412
- Camillo, E. & Garófalo, C.A. (1989). Analysis of the niche of two sympatric species of *Bombus* (Hymenoptera, Apidae) in southeastern Brazil. *Journal of Tropical Ecology*, 5: 81-92. doi: 10.1017/S0266467400003242
- Carpentieri-Pípolo, V., Neves, C.S.V.J., Bruel, D.C., Souza, S.G.H. de & Garbúglio, D.D. (2008). Frutificação e desenvolvimento de frutos de aceroleira no Norte do Paraná. *Ciência Rural*, 38: 1871-1876. doi: 10.1590/S0103-84782008000700011
- Coelho, M.F.B. & Spiller, C. (2008). Fenologia de *Heteropterys aphrodisiaca* O. Mach. – Malpighiaceae, em Mato Grosso. *Revista Brasileira de Plantas Mediciniais*, 10: 1-7. [http://www.sbpmed.org.br/download/issn\\_08\\_1/artigo1\\_v10\\_n1\\_pla7.pdf](http://www.sbpmed.org.br/download/issn_08_1/artigo1_v10_n1_pla7.pdf)
- Erdtman, G. (1960). The Acetolysis Method - A Revised Description. *Svensk Botanisk Tidskrift*, 54: 561-564.
- Garófalo, C.A., Martins, C. F. & Alves-dos-Santos, I. (2004). The Brazilian solitary bee species caught in trap nests. In B.M. Freitas & J.O.P Pereira (Eds), *Solitary Bees: conservation, rearing and management for pollination* (pp. 77-84). Fortaleza: Imprensa Universitária.
- Hammer, Ø, Haper, D.A.T. & Ryan, P.D. (2001). PAST: Paleontological Statistics software package for education and data analysis. *Palaeontologia Electronica*, 4: 1-9.
- Klink, C.A. & Machado, R.B. (2005). Conservation of the Brazilian Cerrado. *Conservation Biology*, 19: 707-713. doi: 10.1111/j.1523-1739.2005.00702
- Martins, C.F., Ferreira, R.P. & Carneiro, L.T. (2012). Influence of the orientation of nest entrance, shading, and substrate on sampling trap-nesting bees and wasps. *Neotropical Entomology*, 41: 105-111. doi: 10.1007/s13744-012-0020-5
- Mendes, F.N., Rêgo, M.M.C. & Albuquerque, P.M.C. (2011). Fenologia e biologia reprodutiva de duas espécies de *Byrsonima* RicH. (Malpighiaceae) em área de Cerrado no Nordeste do Brasil. *Biota Neotropica*, 11: 103-115. doi: 10.1590/S1676-06032011000400011
- Menezes, G.B., Gonçalves-Esteves, V., Bastos, E.M.A.F., Augusto, S.C. & Gaglianone, M.C. (2012). Nesting and use of pollen resources by *Tetrapedia diversipes* Klug (Apidae) in Atlantic Forest areas (Rio de Janeiro, Brazil) in different stages of regeneration. *Revista Brasileira de Entomologia*, 56: 86-94. doi: 10.1590/S0085-56262012000100014
- Mesquita, T.M.S. & Augusto, S.C. (2011). Diversity of trap-nesting bees and their natural enemies in the Brazilian savanna. *Tropical Zoology*, 24: 127-144. <http://www.fupress.net/index.php/tropicalzoology/article/view/10456>
- Neto, P.L. (2008). Levantamento planimétrico nº 36.243. Relatório Técnico. Uberlândia: Prefeitura Municipal de Uberlândia, Minas Gerais.
- Neves, C.M. de L., Carvalho, C.A.L. de, Machado, C.S., Aguiar, C.M.L. & Sousa, F.S. das M. (2014). Pollen consumed by the solitary bee *Tetrapedia diversipes* (Apidae: Tetrapediini) in a tropical agroecosystem. *Grana*, 53: 302-308. doi: 10.1080/00173134.2014.931455
- Pereira, M., Garófalo, C. A., Camillo, E., & Serrano, J. C. (1999). Nesting biology of *Centris (Hemisiella) vittata* Lapeletier in southeastern Brazil (Hymenoptera, Apidae, Centridini). *Apidologie*, 30: 327-338
- Peruquetti, R.C. & Del Lama, M.A. (2003). Alocação sexual e seleção sexo-dependente para tamanho de corpo em *Trypoxylon rogenhoferi* Kohl (Hymenoptera: Sphecidae). *Revista Brasileira de Entomologia*, 47: 581-588. doi: 10.1590/S0085-56262003000400008
- Pinto, C.E., Silva, A. da, Cordeiro, G.D. & Alves-dos-Santos, I. (2015). The body size of the oil-collecting bee *Tetrapedia diversipes* (Apidae). *Journal of Hymenoptera Research*, 47: 103-113. doi: 10.3897/JHR.47.4837
- Radmacher, S. & Strohm, E. (2010). Factors affecting offspring body size in the solitary bee *Osmia bicornis* (Hymenoptera, Megachilidae). *Apidologie*, 41: 169-177. doi: 10.1051/apido/2009064
- Ramos, M., Albuquerque, P. de & Rêgo, M. (2010). Nesting behavior of *Centris (Hemisiella) vittata* Lapeletier

(Hymenoptera: Apidae) in an area of the cerrado in the northeast of the State of Maranhão, Brazil. *Neotropical Entomology*, 39: 379-383. doi: 10.1590/S1519-566X2010000300011

Rasband, W.S. (1997-2011). ImageJ, U.S. National Institutes of Health, Bethesda, Maryland, USA. [http:// imagej.nih.gov/ij/](http://imagej.nih.gov/ij/). (accessed date: 22 February, 2018).

Roubik, D.W. & Moreno, J.E. (1991). The pollen and spores of Barro Colorado Island. St Louis (Missouri): Missouri Botanical Garden, 268 p

Roulston, T.H. & Cane, J.H. (2002). The effect of pollen protein concentration on body size in the sweat bee *Lasioglossum zephyrum* (Hymenoptera: Apiformes). *Evolutionary Ecology*, 16: 49-65. doi: 10.1023/A:1016048526475

R Core Team (2017). R: A Language and Environment for Statistical Computing. Reference Index. <http://softlibre.unizar.es/manuales/aplicaciones/r/fullrefman.pdf>. (accessed date: 28 February, 2018).

Salgado-Labouriau, M.L. (1973). Contribuição à palinologia dos Cerrados. Rio de Janeiro: Academia Brasileira de Ciências, 289 p

Stark, R.E. (1992). Sex ratio and maternal investment in the multivoltine large carpenter bee *Xylocopa sulcatipes* (Apoidea: Anthophoridae). *Ecological Entomology*, 17: 160-166. doi: 10.1111/j.1365-2311.1992.tb01174

Strohm, E. (2000). Factors affecting body size and fat content in a digger wasp. *Oecologia*, 123: 184–191. doi: 10.1007/s004420051004

Vilhena, A.M.G.F., Rabelo, L.S., Bastos, E.M.A.F. & Augusto, S.C. (2012). Acerola pollinators in the savanna of Central Brazil: temporal variations in oil-collecting bee richness and a mutualistic network. *Apidologie*, 43: 51-62. doi: 10.1007/s13592-011-0081-1

Zar, J.H. (2013). *Biostatistical Analysis*: Pearson New International Edition. Pearson Higher Ed.

