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Comportamento de cópula e evolução das estruturas de coleta de óleo nos machos de abelhas das tribos Tetrapediini e Tapinotaspidini (Hymenoptera, Apidae)

Brasília (DF) - Brasil
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Tese de Doutorado apresentada ao Programa de Pós-Graduação em Zoologia da Universidade de Brasília, como parte dos requisitos para a obtenção do título de Doutor em Zoologia.

ORIENTADOR: PROF. DR. ANTONIO J. C. AGUIAR

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Tese de Doutorado apresentada em 18 de fevereiro de 2021.

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“Na busca por descobertas científicas, cada problema é uma oportunidade.
Quanto mais difícil o problema, maior a importância de sua solução.”
(Edward O. Wilson)

À minha mãe, Maria Izabel,
companheira em todos os momentos.
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RESUMO GERAL

As tribos Tapinotaspidini e Tetrapediini são os únicos grupos de abelhas em que os machos coletam óleo floral. A coleta do óleo ocorre através da utilização de adaptações morfológicas especializadas localizadas nas pernas e metassoma. Contudo, a função do óleo para os machos é desconhecida, embora seja esperado que esteja relacionada ao acasalamento. No presente estudo, as adaptações morfológicas e o comportamento de coleta dos machos de abelhas Tetrapediini e Tapinotaspidini foram analisados. Dessa forma, machos de todos os gêneros de Tapinotaspidini e diferentes espécies de Tetrapediini foram estudados quanto à: morfologia, comportamento de coleta de óleo e capacidade de estabelecer agregações temporárias. A tese consiste em quatro capítulos que abordam: 1) a evolução das estruturas associadas à coleta, transferência e armazenamento de óleo dos machos de Tapinotaspidini; 2) o comportamento de coleta de óleo dos machos de Tapinotaspidini e Tetrapediini; 3) o comportamento de machos de abelhas que dormem em agregações e/ou dormitórios e 4) abelhas e vespas que nidificam em cavidades pré-existentes em uma área de Cerrado do Distrito Federal (DF). Os resultados dos capítulos destacam a elevada diversidade morfológica de estruturas coletoras de óleo presente nos machos de Tetrapediini e Tapinotaspidini. O óleo está possivelmente associado à cópula, podendo ser utilizado na forma de: presente nupcial, camuflagem química ou demarcação e de território. Os machos de Tetrapediini e Tapinotaspidini podem apresentar o comportamento de patrulhar flores ou áreas de nidificação em busca de parceiras reprodutivas. Por fim, a utilização do óleo pode ter uma função alternativa, sendo utilizado como uma estratégia de defesa contra predadores noturnos por machos que dormem em agregações. Os resultados são discutidos a partir de uma perspectiva morfológica, ecológica e evolutiva e contribuem para a compreensão da evolução das adaptações morfológicas para coleta de óleo dos machos de abelhas Tetrapediini e Tapinotaspidini.

Palavras-chave: Abelhas solitárias, estados ancestrais, evolução, morfologia, Neotropical.

GENERAL ABSTRACT

The tribes Tapinotaspini and Tetrapediini are the only groups of bees where males are floral oil collectors. The collection of the oil occurs through the use of specialized morphological adaptations located in the legs and metasoma. However the function of the oil for the males is unknown, although it is expected to be related to mating. The collection of the oil occurs through the use of specialized morphological adaptations located in the legs and metasoma. However the function of the oil for the males is unknown, although it is expected to be related to mating. In this study, the morphological adaptations and collection behavior of Tetrapediini and Tapinotaspidini male bees were performed. Thus, males of all genera of Tapinotaspidini and different species of Tetrapediini were studied in terms of: morphology, oil collection behavior and ability to establish aggregations. The thesis consists of four chapters that address: 1) the evolution of structures associated with the collection, transfer and storage of oil from Tapinotaspidini males; 2) the oil collection behavior of Tapinotaspini and Tetrapediini males; 3) the behavior of male bees that sleep in aggregations and/or dormitory and 4) trap-nesting bees and wasps in a Cerrado area of the Federal District (DF). The results of the chapters emphasize the elevated morphological diversity of oil collecting structures present in Tetrapediini and Tapinotaspidini males. The oil is possibly associated with mating and can be used in the form of: nuptial gift, chemical camouflage or defended territory. Tetrapediini and Tapinotaspidini males may present the behavior of patrolling flowers or nesting sites in search of reproductive partners. Finally, the use of oil can have an alternative function, being used as a defense strategy against nocturnal predators, by males sleeping in aggregations. The results are discussed from a morphological, ecological and evolutionary perspective and contribute to the understanding of the evolution of morphological adaptations for collecting oil from Tetrapediini and Tapinotaspidini male bees.

Keywords: Ancestral states, evolution, morphology, Neotropical, solitary bees.

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INTRODUÇÃO GERAL

Estruturada no formato de “Revisão” a ser submetida ao periódico “Oecologia australis”, em português, intitulada “*Seleção sexual em abelhas solitárias: o enigma dos machos coletores de óleo*”.

Seleção sexual

A seleção sexual representa uma importante etapa da seleção natural que envolve estratégias e/ou evolução de características que permitam a obtenção de parceiros sexuais, acasalamentos e descendentes (Searcy 1982, Alcock 2011). O termo "seleção sexual" foi proposto por Darwin (1859, p. 83), ao propor que a seleção dependeria não da luta pela existência em relação a outros organismos ou condições externas, mas da luta entre os indivíduos do mesmo sexo, em geral machos, pela conquista do sexo oposto. Segundo Darwin:

> Natural selection will be able to modify one sex in its functional relations to the other sex, or in relation to wholly different habits of life in the two sexes, as is sometimes the case with insects. And this leads me to say a few words on what I call Sexual Selection. This depends, not on a struggle for existence, but on a struggle between the males for possession of the females; the result is not death to the unsuccessful competitor, but few or no offspring.

Quando falamos sobre seleção sexual dois importantes pontos precisam ser considerados: (1) a competição de machos pelo acesso às fêmeas; (2) o comportamento das fêmeas em escolher parceiros reprodutivos (Parker 1979, Thornhill & Alcock 1983). Os diferentes interesses reprodutivos de machos e fêmeas tendem a gerar comportamentos distintos que visam potencializar o sucesso reprodutivo do indivíduo (Parker 1979, Chapman 2003). Dessa forma, enquanto os machos investem em realizar o maior número de cópulas possíveis, as fêmeas são mais seletivas na escolha dos parceiros reprodutivos (Alcock 1980, Krebs & Davies 1996). O estudo de Bateman (1948), sobre o comportamento reprodutivo de *Drosophila melanogaster*, ilustra bem a diferença de estratégias entre machos e fêmeas. Tanto pela diferença na variação na taxa de sucesso reprodutivo como pelo comportamento dos machos que tentaram copular com o maior número de fêmeas possível, embora a maior parte delas tenha sido receptiva a apenas um ou dois – dos três a cinco – machos disponíveis. Assim, mesmo com os machos produzindo um número muito maior de gametas, as fêmeas geralmente são consideradas o sexo limitante pela capacidade de escolher os parceiros reprodutivos (Thornhill & Alcock 1983).

Seleção sexual em abelhas

Os insetos representam um interessante modelo biológico para o estudo da evolução por seleção sexual, seja pela sua elevada diversidade no comportamento de acasalamento e/ou por apresentarem uma considerável distribuição e abundância (Alcock 1980, Cueva Del Castillo 2007). Segundo Thornhill & Alcock (1983), a maior parte dos insetos tem uma vida adulta curta, não sendo necessário acompanhá-los por longos períodos para avaliação do sucesso reprodutivo desses organismos. Diferentes estudos abordam o comportamento de cópula (abelhas e vespas), a utilização de feromônios (borboletas e mariposas) e presentes nupciais (dípteros e ortópteros) por insetos no processo de acasalamento (Alcock 1978, Alcock 1980, Eickwort & Ginsberg 1980, Thornhill & Alcock 1983, Turgeon *et al.* 1983, Vahed 1998, Gwynne 2008). Entretanto, mesmo com a região Neotropical apresentando elevada diversidade de insetos, estudos sobre seleção sexual para muitos desses organismos ainda são escassos e/ou mal relatados (Cueva Del Castillo 2007, Contreras-Garduño & Lazcano 2013).

O comportamento de acasalamento das abelhas é bastante diverso, sendo esperado que as interações entre machos e fêmeas tenham evoluído de forma a maximizar o acesso dos machos às fêmeas receptivas, bem como das fêmeas aos machos disponíveis (Michener 2007, Danforth *et al.* 2019). Da mesma forma que ocorre com a maior parte dos himenópteros, os machos de abelhas possuem atividades menos complexas que as fêmeas (Alcock *et al.* 1978). Enquanto as fêmeas passam a maior do tempo investindo na prole (construção dos ninhos, provisão das células e postura de ovos), os machos investem energia na busca pelo maior número possível de parceiras receptivas (Eickwort & Ginsberg 1980, Paxton 2005). Como a maioria das fêmeas de abelhas são monândricas, e acasalam logo após emergirem, os machos que encontram parceiras receptivas primeiro apresentam maior possibilidade de sucesso reprodutivo (Alcock 1980, Eickwort & Ginsberg 1980), o que impulsiona uma busca intensa por fêmeas receptivas e, consequentemente, uma elevada diversidade de estratégias reprodutivas (Danforth *et al.* 2019).

Os machos investem tempo e energia em busca de fêmeas para acasalar (Thornhill & Alcock 1983). Essa busca não é aleatória, com os machos de diferentes espécies podendo adotar estratégias específicas para acessar fêmeas receptivas, sendo reconhecidas na literatura pelo menos quatro padrões: (1) patrulhar e/ou esperar flores visitadas por fêmeas; (2) disputar fêmeas em sítios de nidificação; (3) defender recursos coletado pelas fêmeas; (4) atrair fêmeas para sítios não relacionados aos recursos florais ou de nidificação (Alcock 1978, Alcock 1980, Eickwort & Ginsberg 1980, Paxton 2005) (Tabela 1), onde as táticas adotadas pelos machos geralmente estarão relacionadas a: disponibilidade de fêmeas, níveis de receptividade e a abundância de machos (Danforth *et al.* 2019).

Tabela 1. Estratégias dos machos para acessar parceiras reprodutivas.

Estratégia	Espécie	Referências
Disputa por fêmeas em sítios de nidificação	*Centris* (*Paracentris*) *burgdorfi* (Apidae, Centridini) *Centris* (*Paracentris*) *pallida* (Apidae, Centridini) *Epicharis* (*Epicharis*) *nigrita* (Apidae, Centridini) *Monoeca xanthopyga* (Apidae, Tapinotaspidini)	Sabino *et al.et al.* (2017) Alcock *et al.et al.* (1976) Martins *et al.* (2019) Cunha & Blochtein (2003)
Busca ou patrulha em flores visitadas por fêmeas	*Ancyloscelis apiformis* (Apidae, Emphorini) *Ancyloscelis gigas* (Apidae, Emphorini) *Arhysosage cactorum* (Andrenidae, Calliopsini) *Tetrapedia* diversipes (Apidae, Tetrapediini) *Tetrapedia* peckoltii (Apidae, Tetrapediini)	Alves-dos-Santos (1999) Alves-dos-Santos (1999) Cerceau *et al.* (2019) Cappellari *et al.* (2012) Cappellari *et al.* (2012)
Defesa dos recursos coletado pelas fêmeas	*Protomeliturga turnerae* (Andrenidae, Protomeliturgini) *Ptilothrix fructifera* (Apidae, Emphorini) *Tetraglossula anthracina* (Colletidae, Paracolletini) *Tetraglossula bigamica* (Colletidae, Paracolletini)	Medeiros & Schlindwein (2003) Oliveira *et al.* (2016) Gimenes (2002) Gimenes (1997)
Atração de fêmeas para locais não relacionados a recursos florais ou sítios de nidificação	*Eulaema meriana* (Apidae, Euglossini) *Euglossa imperialis* (Apidae, Euglossini) *Xylocopa hirsutissima* (Apidae, Xylocopini)	Kimsey (1980) Kimsey (1980) Velthuis & Camargo (1975a, 1975b)

Busca por fêmeas em áreas de nidificação

A busca por parceiras reprodutivas em áreas de nidificação é comum em espécies de abelhas em que ocorre alta densidade de ninhos e as fêmeas são monogâmicas (cópula única) e emergem de forma aproximadamente sincronizada (Alcock 1980, Paxton 2005). Nesse tipo de situação, os machos geralmente emergem antes do que as fêmeas (protandria) e buscam por parceiras virgens recém-emergidas em áreas de nidificação (Alcock *et al.* 1978, Thornhill & Alcock 1983, Paxton 2005). A estratégia de buscar fêmeas receptivas em áreas de nidificação já foi descrita para os machos de diferentes espécies de abelhas solitárias, sendo comum o registro de centenas ou milhares de machos patrulhando áreas de nidificação (Danforth *et al.* 2019).

No Arizona (EUA), Alcock *et al.* (1976) estudaram o comportamento dos machos de *Centris* (*Paracentris*) *pallida* Fox, 1899. A espécie *C. pallida* é protândrica e os machos escavam o solo em áreas de nidificação em busca de fêmeas prestes a emergir. Quando uma fêmea de *C. pallida* era descoberta, uma intensa competição de machos era observada, com muitos deles lutando para obter acesso a potencial parceira reprodutiva. Em seguida, um único macho montado sobre a fêmea voava com a parceira para uma árvore próxima para dar continuidade ao processo de cópula. Na espécie *Amegilla dawsoni* (Rayment, 1951), amplamente distribuída na região central da Austrália Ocidental, os machos patrulham áreas abertas, onde muitas fêmeas construíram ninhos no inverno anterior (Alcock 1996a, 1996b). Os machos inspecionam o solo, às vezes, utilizando as antenas para inspecionar buracos no solo previamente escavados (Alcock 2013). Quando uma fêmea virgem surge é comum uma disputa intensa entre muitos machos disputando o acesso à fêmea, que será ignorada após a inseminação (Alcock 1996a, 2013).

Os machos de uma mesma população podem apresentar diferentes estratégias comportamentais na busca por parceiras reprodutivas (Thornhill & Alcock 1983). Do ponto de vista evolutivo, um bom exemplo, é o das espécies em que os machos apresentam morfos diferentes (Danforth *et al.* 2019). Na disputa por fêmeas, machos maiores de algumas espécies de *Centris* (denominados beta ou metândricos) disputam o acesso a fêmeas virgens nas áreas de nidificação (Alcock *et al.* 1977, Toro *et al.* 1991, Vinson *et al.* 1996). Por sua vez, resta aos machos menores (alfa), incapazes de disputar com os machos maiores, o patrulhamento de flores na tentativa de encontrar parceiras receptivas (Vinson *et al.* 1996, Simmons *et al.* 2000). Em *Amegilla dawsoni* (Rayment, 1951), machos menores também podem patrulhar áreas de nidificação, porém evitam os mesmos horários em que machos maiores são observados buscando as fêmeas (Alcock

1997). Todavia, o sucesso reprodutivo dos machos menores de *A. dawsoni* em áreas de nidificação não é garantido, uma vez fêmeas que já tenham acasalado não são receptivas (Simmons *et al.* 2000).

No Brasil, também são conhecidas espécies em que os machos buscam por fêmeas em áreas de nidificação como, por exemplo, *Monoeca xanthopyga* Harter-Marques, Cunha & Moure, *Epicharis* (*Epicharis*) *bicolor* Smith 1854 e *Epicharis* (*Epicharis*) *nigrita* Friese, 1900 (Cunha & Blochtein 2003, Rocha-Filho *et al.* 2008, Martins *et al.* 2019).

Busca por fêmeas em flores

Quando a densidade de ninhos agregados não é alta à busca por fêmeas na entrada dos ninhos não se mostra vantajosa devido a maior dificuldade em defender ou patrulhar grandes áreas (Alcock 1980). Desse modo, os machos podem buscar por parceiras nas flores quando as fêmeas estão coletando recursos florais – pólen, néctar, óleo – para alimentação ou nidificação, podendo apresentar comportamentos distintos a depender da espécie (Alcock *et al.* 1978, Paxton 2005; Danforth *et al.* 2019).

Os machos de *Arhysosage cactorum* Moure, 1999 aguardam as fêmeas dentro das flores (Cerceau *et al.* 2019). Já os machos do gênero *Ancyloscelis* Latreille, 1829 são comumente observados patrulhando flores de Cactaceae, Convolvulaceae e Pontederiaceae que são visitadas por fêmeas (Alves-dos-Santos 1999). No gênero *Tetrapedia* Klug, 1810, os machos coletam óleo em flores de diversas espécies de Malpighiaceae (e.g. *Banisteriopsis argyrophylla* (A.Juss.) B.Gates, *Banisteriopsis gardneriana* (A.Juss.) W.R.Anderson & B.Gates, *Byrsonima coccolobifolia* Kunth, *Heteropterys tomentosa* A.Juss.), onde possivelmente procuram por fêmeas receptivas (Cappellari *et al.* 2012).

Defesa de recursos (Poliginia de defesa de recursos)

Nas espécies em que as fêmeas realizam múltiplas cópulas (poliândricas), não há muita vantagem para os machos investirem tempo e energia patrulhando áreas de nidificação (Alcock *et al.* 2013). Dessa forma, a poliginia de defesa de recursos surge como uma interessante estratégia, em que os machos defendem territórios que possuem recursos importantes para as fêmeas (Emlen & Origin 1977). Em abelhas, é esperado que a defesa de recursos seja vantajosa para o macho quando esses recursos estejam espacialmente agregados (Paxton, 2005). Medeiros & Schlindwein (2003) observaram que na espécie oligolética *Protomeliturga turnerae* (Ducke, 1907), os machos defendiam territórios de *Turnera subulata* (Turneraceae) que compreendiam em média 124 flores por território. As fêmeas de *P. turnerae* coletam pólen apenas em *T. subulata*, onde foram observadas realizando múltiplas cópulas ao longo de um mesmo dia.

Na espécie *Hylaeus alcyoneus* (Erichson, 1842), endêmica da Austrália, machos maiores conseguem defender inflorescências de *Banksia* (Proteaceae), ricas em néctar, que são constantemente visitadas pelas fêmeas da espécie (Paini & Bailey 2002). Machos defendendo recursos também são observados nas espécies oligoléticas *Tetraglossula bigamica* (Strand, 1910) e *Tetraglossula anthracina* (Michener, 1989), que defendem territórios contendo flores de *Ludwigia elegans* (Onagraceae), onde as fêmeas coletam pólen (Gimenes, 1997, 2002). Em *Ptilothrix fructifera* (Holmberg, 1903), espécie oligolética de *Opuntia* (Cactaceae), machos maiores defendem territórios floridos, levando vantagem em relação aos machos menores, que recorrem ao patrulhamento em arbustos, onde a chance de realizar cópulas é consideravelmente menor (Oliveira & Schlindwein 2010).

Atração de fêmeas para sítios de acasalamento (poliginia de lek)

Os machos de algumas espécies podem adotar a estratégia de liberar feromônios ou outras secreções glandulares para atrair fêmeas para locais específicos como, por exemplo, galhos e barrancos (Alcock 2013, Oliveira 2015, Danforth *et al.* 2019). Considerando que esses locais são desprovidos de recursos úteis à nidificação ou forrageamento, é esperado que a presença das fêmeas tenha uma única razão: acasalar (Alcock 2013).

Na espécie *Xylocopa varipuncta* Patton, 1876, os machos pairam sobre arbustos e árvores, onde liberam feromônios para atrair as fêmeas (Alcock & Johnson 1990). Muitos machos podem ser observados no mesmo arbusto e a disputa pelo acesso às fêmeas também acontece em alguns casos (Marshall & Alcock 1981, Alcock & Johnson 1990). A poliginia de *lek* é descrita também para pelos menos duas outras tribos: Bombini e Euglossini (Alcock & Alcock 1983, O'Neill *et al.* 1991, Kindl *et al.* 1999, Baer 2003).

Adaptações morfológicas e comportamentos associados à cópula

Além de estratégias para encontrar fêmeas receptivas, os machos também podem apresentar adaptações morfológicas associadas ao comportamento de cópula. Em abelhas, as diferenças morfológicas observadas entre os sexos estão relacionadas, principalmente, ao comportamento de forrageamento e acasalamento (Micheber 2007, Eickwort &

Ginsberg 1980). Nos machos, tais adaptações estão, geralmente, mais relacionadas ao acasalamento do que aquelas observadas em fêmeas (Bateman 1948).

Adaptações morfológicas nas pernas e metassoma

Machos e fêmeas de abelhas podem apresentar notável dimorfismo sexual nas pernas. Na maior parte das espécies, as fêmeas apresentam estruturas especializadas para a coleta de pólen (corbícula ou escopa), enquanto as adaptações morfológicas dos machos estão relacionadas ao comportamento de cópula (Michener 2007, Danforth *et al.* 2019). O dimorfismo sexual nas pernas de abelhas já foi descrito para diferentes espécies de *Ancyloscelis* spp. e de *Megachile* Latreille, 1802, além de todas as espécies de Euglossini Latreille, 1802, e está relacionada a diferentes aspectos comportamentais (Dressler 1982, Wittmann & Blochtein 1995, Alves-dos-Santos 1999). Os machos de *Ancyloscelis* representam um caso de acentuado dimorfismo sexual nas pernas posteriores, que são consideravelmente dilatadas na coxa, tíbia e fêmur (Alves-dos-Santos 1999). O basitarso posterior apresenta forma de "gancho", sendo um caráter importante para o reconhecimento das espécies do gênero. É esperado que tal estrutura possua importante função durante a cópula, possivelmente servindo como encaixe com as tíbias das pernas posteriores das fêmeas, proporcionando um tempo maior de cópula.

Na tribo Eugossini, os machos apresentam adaptações nas pernas posteriores associadas à coleta de substâncias voláteis odoríferas (fragrâncias) em orquídeas (Dressler1967, Dodson 1969, Dressler 1982). As fragrâncias são armazenadas em fendas localizadas nas tíbias posteriores e posteriormente liberadas em locais de acasalamento (Dressler 1968, Kimsey 1980, Bembé 2004, Eltz *et al.* 2019). Estudos que buscam compreender a função específica dos compostos emitidos pelos machos já foram realizados, sendo possível que estejam relacionados a algum tipo de comunicação intraespecífica (semelhante a um feromônio) que evolui por seleção sexual (Pokorny *et al.* 2017).

Nos machos de algumas espécies do gênero *Megachile* são observadas modificações nas pernas anteriores que podem ser usadas para reter a fêmea durante cópula, aumentando assim suas chances de sucesso reprodutivo (Sihag 1986, Wittmann & Blochtein 1995). Em *Megachile willughbiella* (Kirby, 1802), por exemplo, as cerdas ramificadas no basitarso e tarsômeros (2-4) anteriores dos machos formam uma franja longa e côncava que é utilizada durante a cópula para cobrir os olhos da fêmea, ao mesmo tempo em que as pernas médias e posteriores são utilizadas para segurar o abdome e as asas da parceira (Wittmann & Blochtein 1995).

Adaptações no metassoma dos machos de algumas espécies desempenham importante função no comportamento de acasalamento dos machos. Os machos de algumas espécies de *Anthidium* (Anthidiini) e *Callonychium* (Calliopsini), por exemplo, apresentam projeções nos esternos que auxiliam a fixar-se ao tergo das fêmeas durante a cópula, em um processo de ajuste mecânico (Toro 1985, Toro & Rodriguez 1997). Modificações morfológicas nos tergos de algumas espécies também são observadas e podem estar relacionadas a comportamentos agressivos de machos que defendem territórios. Os machos de *Anthidium manicatum* (Linnaeus, 1758) defendem territórios atacando outros machos, indo em alta velocidade ao encontro deles e atingindo-os com espinhos presentes nos tergos 5-6 (Wirtz et al. 1988), tornando seus territórios mais atraentes para as fêmeas, que não são atacadas, e com as quais podem acasalar.

Sinalização química

O uso de feromônios sexuais ou outros tipos de secreções glandulares para atrair parceiros reprodutivos é descrito para fêmeas e machos de diferentes espécies de abelhas (Alcock *et al.* 1978, Ayasse *et al.* 2001, Paxton, 2005). Em abelhas eussociais como, por exemplo, *Apis mellifera* Linnaeus, 1758 e *Scaptotrigona postica* (Latreille, 1807), as rainhas virgens produzem secreções mandibulares que atraem os machos para a cópula (Butler 1971; Engels *et al.* 1997). As fêmeas de algumas espécies solitárias também produzem feromônios sexuais. Em *Lasioglossum malachurum* (Kirby, 1802), espécie que nidifica em agregações e apresenta curto período receptivo após a emergência, as fêmeas virgens secretam feromônios sexuais que possibilita serem encontradas mais facilmente pelos machos (Ayasse *et al.* 1999). Semelhante ao que ocorre com *Colletes cunicularius* (Linnaeus, 1761), em que os machos, guiados pelo feromônio liberado pelas fêmeas virgens, escavam o solo em busca das parceiras reprodutivas (Cane & Tengö 1981).

Os machos de muitos gêneros (e.g. *Andrena* Fabricius, 1775, *Euglossa* Latreille, 1802, *Bombus* Latreille, 1802, *Nomada* Scopoli, 1770, *Xylocopa* Latreille, 1802) fazem uso de substâncias para demarcar territórios ou atrair fêmeas para locais específicos, como galhos ou barrancos (Raw 1975, Velthuis & Camargo 1975a, 1975b, Svensson & Bergstrom 1977, Tengö & Bergstrom 1977, Tengö 1979, Kimsey 1980). Para Alcock *et al.* (1978), o comportamento de utilizar feromônios pode ter evoluído a partir do momento em que as fêmeas passaram a escolher companheiros que fizessem uso dessas substâncias, de forma a evitar o desperdício de tempo e energia com machos de espécies próximas.

Tengö & Bergström (1977) descreveram uma interessante relação de cleptoparasitismo entre abelhas do gênero *Nomada* (cleptoparasita) e *Andrena* (hospedeiro). As fêmeas de *Nomada* não constroem ninhos para o desenvolvimento da prole, mas depositam os ovos em células construídas e aprovisionadas pelas fêmeas de *Andrena*. Por sua vez, os machos de *Nomada* produzem nas glândulas mandibulares, substâncias químicas específicas de fêmeas do gênero *Andrena* que são utilizadas no processo de nidificação. Tais compostos químicos são pulverizados pelos machos de *Nomada* durante a cópula sobre as suas respectivas parceiras reprodutivas. É esperado que, posteriormente, sirvam como uma espécie de camuflagem química que permita acessar e ovipositar nos ninhos de *Andrena*.

Estratégias pós-inseminação

Os machos de muitos insetos podem adotar estratégias reprodutivas que prolongam a relação com as fêmeas por muito mais tempo além do que seria necessário (Alcock 1994). Esse comportamento tem como objetivo garantir a transferência de um número suficiente de gametas para fertilizar todos os ovos da fêmea. Denominado por Alcock (1994) como associações pós-inseminação, tais comportamentos estão agrupados em quatro categorias: (1) cópula prolongada após a inseminação; (2) doação de *plugs* (um tipo de tampão inserido no trato genital da fêmea) após a inseminação, (3) contato com a fêmea após a cópula e (4) monitoramento da parceria após a cópula.

Em abelhas, a cópula prolongada é descrita, por exemplo, em *Nomadopsis puellae* (Cockerell, 1933) e pode ser observada em espécies onde as fêmeas são poliândricas (Rutowski & Alcock 1980, Wittmann *et al.* 1990). Conforme já havia sido observado por Rozen (1958), os machos do subgênero *Nomadopsis* Ashmead, 1898 podem ficar agarrados nas fêmeas, através da superfície dorsal, por minutos, enquanto elas coletam pólen e néctar ou durante o voo. A estratégia de guardar a parceira, aumentaria as chances de sucesso reprodutivo do macho, especialmente se a fêmea colocar um ovo fertilizado no retorno ao ninho (Rutowski & Alcock 1980). A cópula prolongada é descrita também para outras espécies de Andrenidae como, por exemplo, *Arhysosage biguttulata* Ramos, 2013, *Arhysosage cactorum* Moure, 1999, *Callonychium* (*Callonychium*) *petuniae* Cure & Wittmann, 1990, *Perdita texana* (Cresson) (Barrows *et al.* 1976, Wittmann 1990, Schlindwein & Wittmann 1995, Ramos, 2013 Pimentel 2016).

Os machos de algumas espécies de Diptera e Ledipoptera podem secretar substâncias (*plugs*) que são utilizadas para obstruir ou dificultar o acesso ao trato genital feminino (Drummond 1984, Markow & Ankney 1988). Em abelhas, algumas situações em que os machos utilizam *plugs* também já foram descritas na literatura. Os machos de *Bombus terrestris* (Linnaeus, 1758), por exemplo, transferem uma secreção glandular de aspecto gelatinoso e pegajoso para a fêmea logo após a cópula (Duvoisin *et al.* 1999). Já em *Osmia bicornis* (Linnaeus, 1758), os machos fazem uso de um *plug* que é introduzido na genitália da fêmea e, embora não impeça uma futura cópula com outro macho, evita que o esperma desse seja misturado com a de outros competidores (Seidelmann 2014).

O enigma dos machos que coletam óleo floral

A síndrome de óleo floral foi descrita por Stefan Vogel (1969, 1974) e corresponde a uma relação abelha-planta que envolve aproximadamente 440 espécies de abelhas distribuídas em seis tribos (Ctenoplectrini, Centridini, Tapinostapidini e Tetrapediini (Apidae), Macropidini e Redivivini (Melittidae)) e pelos menos 10 famílias de plantas (Vogel, 1974, Buchmann 1987, Neff & Simpson 2017). O óleo floral é secretado através de estruturas denominadas elaióforos, que foram descritas por Vogel (1971, 1974), e podem ser classificados em epiteliais ou tricomáticos. Segundo Renner & Schaefer (2010), a oferta de óleo floral surgiu pelo menos 28 vezes de forma independente (com pelo menos 36-40 perdas). Nas abelhas, a coleta de óleo teria evoluído de forma independente pelo menos sete vezes (Renner & Schaefer, 2010). Em Apidae, quatro surgimentos independentes teriam ocorrido (Policarová *et al.* 2019).

A coleta de óleo pelas abelhas ocorre através de estruturas especializadas nas pernas e metassoma (Neff & Simpson 1981, Roig-Alsina 1997, Cocucci *et al.* 2000). O óleo coletado pelas fêmeas é utilizado para a alimentação da prole e nidificação (Cane *et al.* 1983, Vinson *et al.* 1997, Pereira *et al.* 1999). Estruturas adaptadas para a coleta de óleo estão presentes apenas nos machos do gênero *Tetrapedia* (Tetrapediini) e da tribo Tapinotaspidini (Vogel & Machado, 1991, Singer & Cocucci, 1999, Cappellari *et al.*, 2012). Contudo, a utilização do óleo coletado pelos machos é desconhecida, uma vez que, como na maior parte das espécies de abelhas solitárias, os machos deixam o ninho após emergirem, não apresentando qualquer tipo de cuidado parental ou comportamento de nidificação, como é observado nas fêmeas (Paxton 2005).

As abelhas das tribos Tapinotaspidini e Tetrapediini são especializadas na coleta de óleos de flores de Iridaceae, Krameriaceae, Malpighiaceae, Plantaginaceae, Calceolariaceae e Orchidaceae (Vogel 1974, Neff & Simpson 1981, Buchmann 1987, Cocucci et al. 2000). O gênero *Tetrapedia* é composto por abelhas solitárias, de tamanho médio, pouco pilosas, restritas às regiões tropicais das Américas, cujas fêmeas nidificam em cavidades pré-existentes (Silveira et al. 2002, Moure 2012). A tribo Tapinotaspidini apresenta abelhas de porte pequeno a médio, elevada diversidade morfológica, com a maior parte das espécies nidificantes no solo (Silveira et al. 2002, Aguiar 2012). Nos machos do gênero *Tetrapedia* (aproximadamente 30 espécies), as pernas anteriores apresentam o aparato coletor de óleo com formato e tamanho semelhantes aos encontrados nas fêmeas, além de uma escopa tergal no metassoma utilizada para armazenar este óleo (Cappellari et al., 2012). As adaptações para coleta de óleo em Tapinotaspidini são muito mais diversas que nas demais tribos, com machos e fêmeas apresentando adaptações morfológicas para coleta de óleo acentuadas (Roig-Alsina 1997, Coccuci et al., 2000; Aguiar & Melo, 2007). Nos machos de Tapinotaspidini também são observadas adaptações para coleta de óleo nas pernas e metasosoma (Fig. 1), e devido à grande diversidade de estruturas especializadas na coleta de óleo nas fêmeas, é possível que haja também uma correspondente diversidade também nos machos (Vogel 1974, Roig-Alsina 1997, Cocucci et al. 2000). Em algumas espécies de Tapinotaspidini as estruturas adaptadas para a coleta de óleo são mais acentuadas nos machos (Aguiar & Melo 2009).

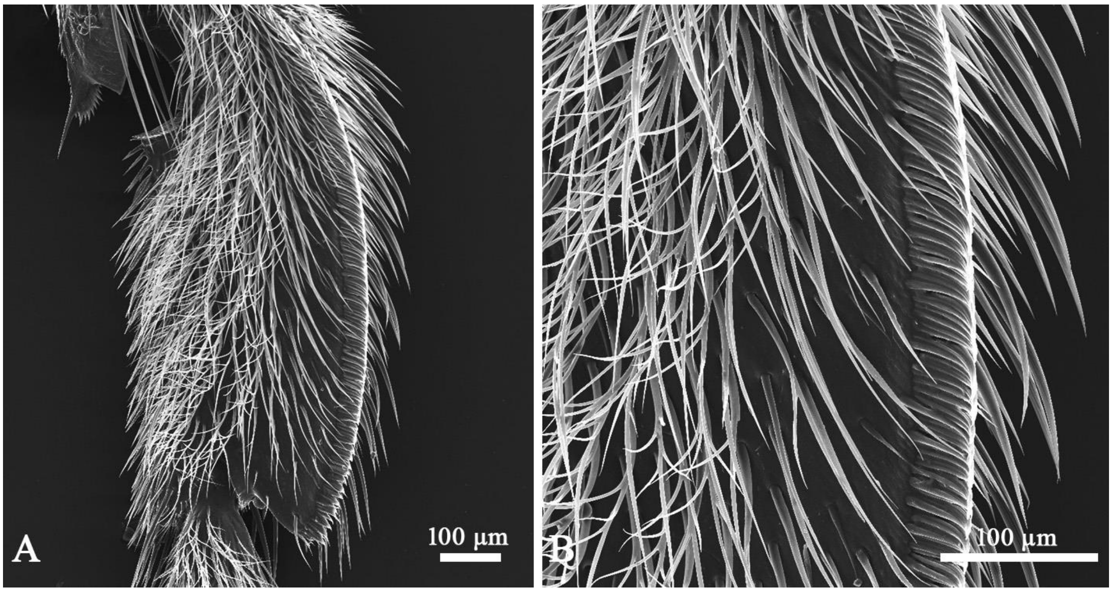


Figura 1. Aparato coletor de óleo na perna direita de macho de *Tropidopedia punctifrons* (Smith, 1879). **A.** Superfície ventral do basitarso. **B.** Detalhe dos pelos plumosos (à esquerda) na superfície ventral do basitarso.

As abelhas Tetrapediini e Tapinostaspidini representam um bom modelo para a investigação da funcionalidade de adaptações morfológicas no processo de seleção sexual de abelhas. Cappellari et al. (2012) observaram que o óleo coletado pelos machos de *Tetrapedia* com o auxílio das pernas anteriores é armazenado na escopa tergal e nas pernas posteriores (Fig. 2). Adaptações morfológicas nas pernas também são observadas nos machos das abelhas da tribo Euglossini, em que compostos aromáticos são coletados, principalmente, em flores de orquídeas, com as pernas anteriores e, posteriormente, transferidos para as pernas médias e posteriores (Eltz *et al.* 1999, Bembé 2004). A coleta de compostos aromáticos por abelhas Tapinotaspidini também foi registrada recentemente na Costa Rica por Etl *et al.* (2017). Os autores observaram que os machos de *Paratetrapedia chocoensis* Aguiar & Melo, 2011 possuem cerdas especializadas no terceiro esterno (E3), que são utilizadas para a coleta de fragrâncias através da raspagem das inflorescências de *Anthurium acutifolium* Engl. (Araceae). Contudo, a compreensão acerca da utilização desse óleo não é clara, embora seja esperado que tenha alguma relação com o comportamento reprodutivo.

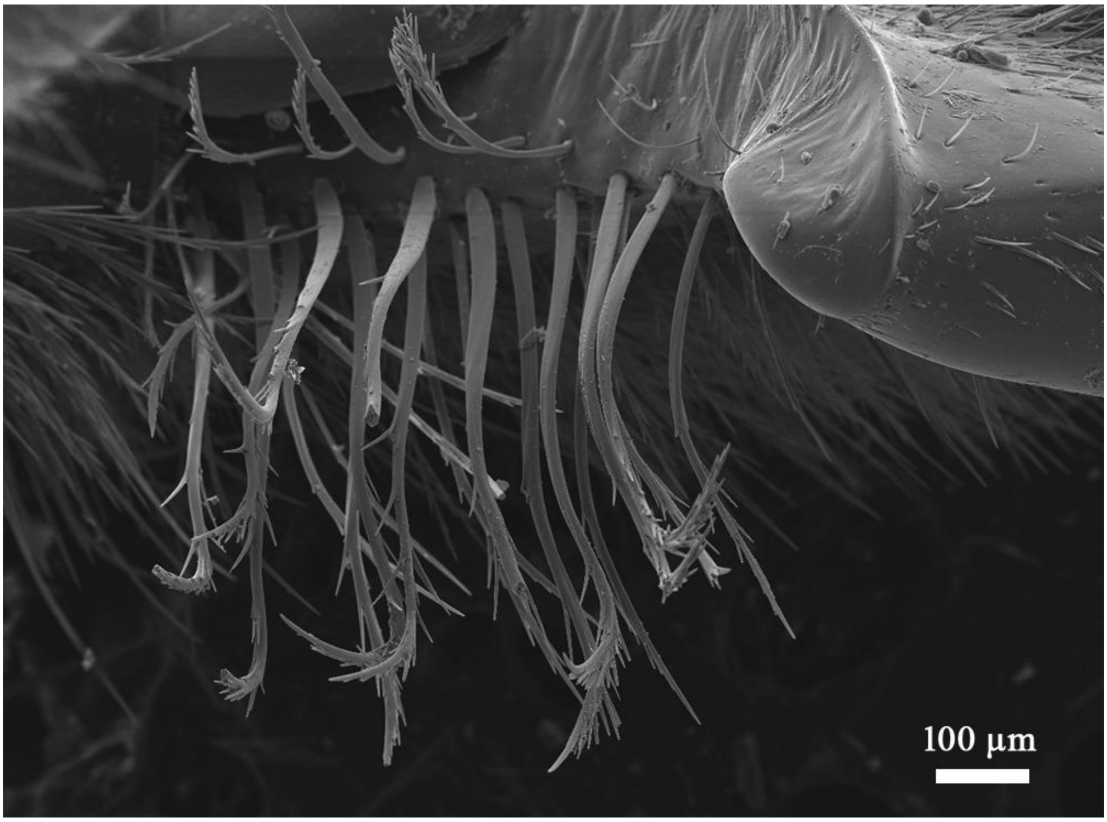


Figura 2. Pelos plumosos presentes na margem ventral do basitarso posterior dos machos de *Tetrapedia peckoltii* Friese, 1899 que podem estar associados a transferência de óleo para a fêmea durante a cópula.

Os machos passam a maior parte do tempo em busca de parceiras, o que possivelmente proporciona uma competição, onde a evolução de uma série de características está envolvida (Thornhill & Alcock 1983). No intuito de atrair parceiras reprodutivas, os machos de algumas espécies fazem uso de benefícios materiais, dentre eles, presentes nupciais (Alcock 2011; Gwynne 2008). Dessa forma, machos que utilizam recursos na forma de presentes nupciais teriam maior chance de obter sucesso reprodutivo (Boggs, 1995, Buzatto *et al.*, 2014). Na literatura, diversos casos de fêmeas recebendo presentes durante a cópula já foram descritos: "dance fly" (Diptera, Empididae); *Leptophyes laticauda* (Frivaldszky, 1868) (Orthopera); *Pieris napi* (Linnaeus, 1758) (Lepidoptera); *Blattella germânica* (Linnaeus, 1767) (Blattaria) (Thornhill & Alcock 1983, Vahed 1998).

Relatos da utilização de presentes nupciais por abelhas não são conhecidos. Entretanto, a utilização do óleo como uma estratégia para acessar fêmeas não pode ser descartado, seja através de um possível presente nupcial, camuflagem química ou demarcação de território. Alguns estudos analisaram as adaptações morfológicas presentes em abelhas coletoras de óleo, ainda que a ênfase seja quase sempre nas fêmeas (Neff & Simpson 1981, Roig-Alsina 1997, Cocucci *et al.*, 2000; Kuhlmann & Hollens 2014). Embora não exista uma total compreensão da funcionalidade das adaptações morfológicas para coleta de óleo nos machos de *Tetrapedia* e Tapinotaspidini, acredita-se que possam estar relacionadas ao comportamento de cópula.

Considerações finais

O conhecimento acerca do comportamento reprodutivo de abelhas, principalmente solitária, ainda é escasso. Embora relevantes informações estejam disponíveis na literatura, muitas lacunas permanecem quanto ao comportamento de acasalamento, inclusive no que diz respeito aos machos. Considerando que boa parte dos estudos têm como foco o comportamento das fêmeas coletando recursos florais e, consequentemente, o seu papel como potenciais polinizadores, avaliar o comportamento dos machos também se faz necessário.

No caso dos machos coletores de óleo (*Tetrapedia* e Tapinotaspidini), investigar o comportamento das fêmeas em relação a receptividade aos machos (monândricas ou poliândricas) podem subsidiar importantes informações, principalmente em relação a utilização do óleo no comportamento de acasalamento e na competição por parceiras reprodutivas.

Estruturação da tese

A tese foi desenvolvida em capítulos independentes com o objetivo de investigar a função do óleo floral nos machos das abelhas *Tetrapedia* e Tapinotaspidini através da caracterização do comportamento e das adaptações morfológicas associadas à coleta de óleo e cópula, visando subsidiar informações sobre a evolução e biologia desses dois grupos de abelhas. Para cada capítulo, informações adicionais são apresentadas nas seções apêndice e anexo. Detalhadamente, os capítulos têm como principais objetivos:

CAPÍTULO 1 – Evolução das estruturas de coleta de óleo nos machos das abelhas: Estudar a evolução das estruturas associadas à coleta, transferência e armazenamento dos machos da tribo Tapinotaspidini.

CAPÍTULO 2 – Coleta de óleos florais por machos de abelhas da tribo Tapinotaspidini e Tetrapediini: fontes florais e comportamento: Estudar os caracteres sexuais secundários dos machos de Tapinotasdini e Tetrapediini, com ênfase no comportamento de coleta de óleo e interação com as plantas hospedeiras.

CAPÍTULO 3 – Agregações e dormitórios: onde e como dormem os machos das abelhas? Analisar e descrever o comportamento de machos de abelhas que dormem em agregações e/ou dormitórios em diferentes áreas do Cerrado do Brasil Central.

CAPÍTULO 4 – Abelhas e vespas que que nidificam em ninhos-armadilha em uma área de Cerrado do Brasil Central. Inventariar as espécies de abelhas e vespas que nidificam em cavidades pré-existentes em uma área de Cerrado do Brasil Central.

Referências bibliográficas

Aguiar, A. J. C. & Melo, G. A. R. 2007. Taxonomic revision, phylogenetic analysis, and biogeography of the bee genus *Tropidopedia* (Hymenoptera, Apidae, Tapinotaspidini). **Zoological Journal of Linnean Society**, 151, 511–554.

Aguiar, A. J. C. &, Melo, G. A. R. 2009. Notes on oil sources for the bee genus *Caenonomada* (Hymenoptera, Apidae, Tapinotaspidini). **Revista Brasileira de Entomologia**, 53(1), 154–156.

Aguiar, A. J. C. 2012. Tapinotaspidini Roig-Alsina & Michener, 1993. In Moure, J. S., Urban, D. & Melo, G. A. R. (Eds.), **Catalogue of Bees (Hymenoptera, Apoidea) in the Neotropical Region - online version**. Disponível em: <http://www.moure.cria.org.br/catalogue>. Acesso em: 22 de agosto de 2020.

Alcock, J. 1978. Notes on male mate locating behavior in some bees and wasps of Arizona. **The Pan-Pacific Entomologist**, 54, 215–525.

Alcock, J. 1980. Natural selection and the mating systems of solitary bees. **American Scientist**, 68, 146–153.

Alcock, J. 1996a. The relation between body size, fighting, and mating success in Dawson’s burrowing bee, *Amegilla dawsoni* (Apidae, Apinae, Anthophorini). **Journal of Zoology**, 239, 663–674.

Alcock, J. 1996b. Site fidelity and homing ability of males of Dawson’s burrowing bee (*Amegilla dawsoni*) (Apidae, Anthophorini). **Journal of the Kansas Entomological Society,** 69, 182–190.

Alcock, J. 1997. Competition from large males and the alternative mating tactics of small males of Dawson's burrowing bee (*Amegilla dawsoni*) (Apidae, Apinae, Anthophorini). **Journal of Insect Behavior**, 10, 99–113.

Alcock, J. 2011. **Comportamento animal: uma abordagem evolutiva**. Artmed, Porto Alegre, p. 606.

Alcock, J. 2013: Sexual selection and the mating behavior of solitary bees. **Advances in the Study of Behavior**, 45, 1–48. DOI: 10.1016/B978-0-12-407186-5.00001-X

Alcock, J. & Alcock, J. P. 1983. Male behavior in two bumblebees, *Bombus nevadensis auricomus* and *Bombus griseicollis* (Hymenoptera, Apidae). **Journal of Zoology**, 200, 561–570.

Alcock, J., Barrows., E. M., Gordh, G., Hubbard, L. J., Kirkendall, L., Pyle, D. W, Ponder, T. L. & Zalom, F. G. 1978. The ecology and evolution of male reprodutive behavior in the bees and wasps. **Zoological Journal of the Linnean Society**, 64, 293–326.

Alcock, J. & Johnson, M. D. 1990. Female mate choice in the carpenter bee *Xylocopa varipuncta* (Hymenoptera; Anthophoridae). **Journal of Zoology**, 221, 195–204.

Alcock, J., Jones, C. E. & Buchmann, S. L. 1976. Location before emergence of the female bee, *Centris pallida*, by its male. **Journal of Zoology**,179, 189–199.

Alves-dos-Santos, I. 1999. Aspectos morfológicos e comportamentais dos machos de *Ancyloscelis* Latreille (Anthophoridae, Apoidea). **Revista Brasileira de Zoologia**, 16(2), 37–43.

Alves-dos-Santos, I., Melo, G. A. R. & Rozen, J. G. 2002. Biology and Imature Stages of the Bee Tribe Tetrapediini (Hymenoptera: Apidae). **American Museum Novitates**, 3377, 1–45.

Ayasse, M., Engels, W., Lubke, G. & Francke, W. 1999. Mating expenditures reduced via female sex. pheromone modulation in the primitively eusocial halictine bee, *Lasioglossum* (*Evylaeus*) *malachurum* (Hymenoptera:Halictidae). **Behavioral Ecology and Sociobiology**, 45, 95–106.

Ayasse, M., Paxton, R. J. & Tengö, J. 2001. Mating behavior and chemical communication in the order Hymenoptera. Annual Review of Entomology, 46, 31–78. DOI: 10.1146/annurev.ento.46.1.31. PMID: 11112163

Baer, B. 2003. Bumblebees as model organisms to study male sexual selection in social insects. **Behavioral Ecology and Sociobiology**, 54, 521–533.

Bateman, A. J. 1948. Intra-sexual selection in *Drosophila*. **Heredity**, 2, 349–368.

Batra, S. W. T. 1978. Aggression, territoriality, mating and nest aggregation of some solitary bees (Hymenoptera, Halictidae, Megachilidae, Colletidae, Anthophoridae). **Journal of the Kansas Entomological Society**, 51, 47–559.

Bembé, B. 2004. Functional morphology in male euglossine bees and their ability to spray fragrances (Hymenoptera, Apidae, Euglossini). **Apidologie**, 35, 283–291.

Boggs, C. L. 1995. Male nuptial gifts: phenotypic consequences and evolutionary implications. In: Leather, S. R., Hardie, J. (Eds). **Insect Reproduction**, pp. 215–242. CRC Press, New York.

Buchmann, S. 1987. The ecology of oil flowers and their bees. **Annual Review of Ecology, Evolution, and Systematics**, 18, 343–369.

Buzatto, B. A., Tomkins, J. L., Simmons, L. W. & Machado, G. 2014. Correlated evolution of sexual dimorphism and male dimorphism in a clade of neotropical harvestmen. **Evolution**, 68, 671–1686.

Butler, C. G. 1971. The mating behavior of the honeybee (*Apis mellifera* L.). **Journal of Entomology**, 46, 1–11.

Cane, J. H., Eickwort, G. C., Wesley, F. R. & Spielholz, J. 1983. Foraging, grooming and mating behaviours of *Macropis nuda* (Hymenoptera, Melittidae) and use of *Lysimachia ciliata* (Primulaceae) oils in larval provisions and cell linings. **American Naturalist**, 110, 257–264.

Cane, J. H., Tengö, J. 1981. Pheromonal cues direct mate-seeking behavior of male *Colletes cunicularius* (Hymenoptera: Colletidae). **Journal of Chemical Ecology**, 7, 427–36.

Cappellari, S. C., Melo, G. A. R., Aguiar, A. J. C. & Neff, J. L. 2012. Floral oil collection by male *Tetrapedia* bees (Hymenoptera: Apidae: Tetrapediini). Apidologie. 43, 39–50. DOI: 10.1007/s13592-011-0072-2

Chapman, T., Arnqvist., G., Bangham, J. & Rowe, L. 2003. Sexual conflict. **Trends in Ecology & Evolution**, 18, 41–47.

Cocucci, A. A., Sérsic, A. & Roig-Alsina, A. 2000. Oil-collecting structures in Tapinotaspidini: their diversity, function and probable origin. **Mitteilungen der Münchner Entomologischen Gesellschaft**, 90, 51–74.

Contreras-Garduño, J. & Lazcano, J. C. 2013. Secondary Sexual Traits,Immune Response, Parasites and Pathogens: The Importance of Studying Neotropical Insects. In: Macedo, R. H. & Machado, G. (Eds.). **Sexual Selection: Perspectives an models from ther Neotropics**, pp. 235–252. Elsevier and Academic Press, London.

Cueva Del Castillo, R. 2007. Sexual selection in tropical insects. In: Del-Claro, K. (Ed.). **International Commission on Tropical Biology and Natural Resources, Encyclopedia of Life Support Systems (EOLSS)**. Developed under the Auspices of the UNESCO, EOLSS Publishers, Oxford.

Cunha, R. & Blochtein, B. 2003. Life history of *Monoeca xanthopyga* Harter-Marques, Cunha & Moure (Hymenoptera, Apidae, Tapinotaspidini) at the Araucária Plateau, Rio Grande do Sul, Brazil. **Revista Brasileira de Zoologia**, 1, 107-113.

Danforth, B., Minckley, R., Neff, J., Fawcett, F. 2019. **The Solitary Bees: Biology, Evolution, Conservation**, p. 488. Princeton University Press.

Darwin, C. 1859. **The Origin of Species by Means of Natural,** University of Pennsylvania Press, Philadelphia, 462 p.

Dodson, C. H., Dressler, R. L., Hills H. G., Adams, R. M. & Williams N. H. 1969. Biologically active compounds in orchid fragrances. **Science**, 164, 1243–1249.

Dressler R. L. 1967. Why do euglossine bees visit orchid flowers? **Atas do Simpósio Sobre a Biota Amazônica**, 5, 171–180.

Dressler R. L. 1968. Pollination by euglossine bees. **Evolution**, 22, 202–210.

Dressler R. L. 1982. Biology of the orchid bees (Euglossini). **Annual Review of Ecology, Evolution, and Systematics**, 13, 373–394.

Drummond, B. A. 1984. Multiple mating and sperm competition in Lepidoptera. In: Smith, R. L. (Ed.). **Sperm Competition and the Evolution of Animal Mating Systems**, pp. 291–370. Academic Press, New York.

Duvoisin, N., Baer, B. & Schmid-Hempel, P. 1999. Sperm transfer and male competition in a bumblebee. **Animal Behaviour**, 58, 743–479.

Eickwort, G. C. & Ginsberg, H. S. 1980. Foraging and mating behavior in Apoidea. **Annual Review of Entomology**, 25, 421–446.

Etl, F., Franschitz, A., Aguiar, A. J. C., Schönenberger, J. & Dötterl, S. 2017. A perfume-collecting male oil bee? Evidences of a novel pollination system involving *Anthurium acutifolium* (Araceae) and *Paratetrapedia chocoensis* (Apidae, Tapinotaspidini). **Flora**, 232, 7–15.

Eltz, T., Whitten, W. M., Roubik, D. W. & Linsenmair, K. E. 1999. Fragrance collection, storage, and accumulation by individual male orchid bees. **Journal of Chemical Ecology**, 25, 157–176.

Emlen, S. T. & Oring, L. W. 1977. Ecology, Sexual Selection, and the Evolution of Mating Systems. **Science**, 197, 215–223.

Engels, W., Engels, E. & Francke, W. 1997. Ontogeny of cephalic volatile patterns in queens and mating biology of the neotropical stingless bee, *Scaptotrigona postica*. **Invertebrate Reproduction & Development**, 30(1-3), 251–56.

Gimenes, M. 1997. Pollinating bees and other visitors of *Ludwigia elegans* (Onagraceae) flowers at a tropical site in Brazil. **Studies on Neotropical Fauna and Environment**, 32(2), 81–88.

Gimenes, M. 2002. Interactions between bees and *Ludwigia elegans* (Camb.) Hara (Onagraceae) flowers at different altitudes in São Paulo, Brazil. **Revista Brasileira de Zoologia**, 19(3), 681–689.

Gwynne, D. T. 2008. Sexual conflict over nuptial gifts in insects. **Annual Review of Entomology**, 53, 83–101. DOI: 10.1186/1471-2148-8-204

Kimsey, L. S. 1980. The behavior of male orchid bees (Apidae, Hymenoptera, Insecta) and the question of leks. **Animal Behaviour**, 28, 996–1004.

Kindl, J., Hovorka, O., Urbanová, K. & Valterová, I. 1999. Scent marking in male premating behavior of *Bombus confusus*. **Journal of Chemical Ecology**, 25, 1489–1500.

Krebs, J. R. & Davies, N. B. 1996. **Introdução à Ecologia Comportamental**. Atheneu, São Paulo, 420 p.

Kuhlmann, M. & Hollens, H. 2014. Morphology of oil-collecting pilosity of female *Rediviva* bees (Hymenoptera: Apoidea: Melittidae) reflects host plant use. **Journal of Natural History**, 49, 561–657.

Markow, T. A. & Ankney, P. F. 1988. Insemination reaction in *Drosophila*: found in species whose males contribute materials to oocytes before fertilization. **Evolution**, 48: 1097–1101.

Marshall, L. D. & Alcock, J. 1981. The evolution of the mating system of the carpenter bee *Xylocopa varipuncta* (Hymenoptera: Anthophoridae). **Journal of Zoology**, 193, 315–324.

Martins, C. F., Neto, V. I. S. & Cruz, R. D. M. 2019. Nesting biology and mating behavior of the solitary bee *Epicharis nigrita* (Apoidea: Centridini). **Journal of Apicultural Research**, (4), 512–521.

Medeiros, P. C. R. & Schlindwein, C. 2003.Territórios de machos, acasalamento, distribuição e relação com plantas em *Protomeliturga turnerae* (Ducke, 1907) (Hymenoptera, Andrenidae). **Revista Brasileira de Entomologia**, 47(4), 589–596.

Michener, C. D. 2007. **The bees of the world**. The Johns Hopkins University Press, Baltimore, p. 953.

Moure, J. S. 2012. Tetrapediini Michener & Moure, 1957. In: MOURE, J. S., URBAN, D.; MELO, G. A. R. (Eds.). **Catalogue of Bees (Hymenoptera, Apoidea) in the Neotropical Region - online version**. Disponível em: <http://www.moure.cria.org.br/catalogue>. Acesso em: 22 de agosto de 2020.

Neff, J. L., & Simpson, B. B. 1981. Oil-collecting structures in the Anthophoridae: Morphology, function and use in systematics. **Journal of the Kansas Entomological Society**, 54, 95–123.

Neff, J. L. & Simpson, B. B. 2017. Vogel's great legacy: The oil flower and oil-collecting bee syndrome. **Flora**, 232, 104–116. DOI: 10.1016/j.flora.2017.01.003

Oliveira, R. 2015. Estratégias de acasalamento em abelhas solitárias no Brasil: o que os machos fazem para se acasalar. In: Aguiar, A. J. C., Gonçalves, R. B. & Ramos, K. S. (Eds.). **Ensaios sobre as abelhas da Região Neotropical**, pp. 439–454. Editora UFPR, Curitiba.

Oliveira, R., Pereira, C. R., Pimentel, A. L. A. F. D. & Schlindwein, C. 2016. The Consequences of Predation Risk on the Male Territorial Behavior in a Solitary Bee. **Ethology**, 122, 632–639. DOI: 10.1111/eth.12509

Oliveira, R. & Schlindwein, C. 2010. Experimental demonstration of alternative mating tactics of male *Ptilothrix fructifera* (Hymenoptera, Apidae). **Animal Behaviour**, 80(2), 241–247. DOI: 10.1016/j.anbehav.2010.04.024

O'Neill, K. M., Evans, H. E., & Bjostad, L. B. 1991. Territorial behaviour in males of three North American species of bumblebees (Hymenoptera: Apidae, Bombus). **Canadian Journal of Zoology**, 69, 604–613.

Paini, D. R. & Bailey, W. J. 2002. Seasonal sex ratios and unbalanced investment sex ratio in the *Banksia* bee *Hylaeus alcyoneus*. **Ecological Entomology**, 27, 713–719.

Parker, G. A. 1979. Sexual selection and sexual conflict. In: Blum, M. S. & Blum, N. A. (Eds.). **Sexual selection and reproductive competition in insects,** pp. 123–166. Academic Press, New York.

Paxton, R. J. 2005. Male mating behaviour and mating systems of bees: an overview. **Apidologie**, 36(2), 145–156.

Pereira, M.; Garófalo, C. A.; Camillo, E.; Serrano, J. C. 1999. Nesting biology of *Centris* (*Hemisiella*) *vittata* Lep. in southeastern Brazil (Hymenoptera, Apidae, Centridini). **Apidologie**, 30, 327–338.

Pimentel, A. L. A. F. D. 2016. **Acasalar demais é uma perda de tempo: custos da cópula para fêmeas de duas espécies de abelhas solitárias.** Dissertação de Mestrado, Programa de Pós-Graduação em Ecologia de Biomas Tropicais. Universidade Federal de Ouro Preto, p. 85.

Pokorny, P., Vogler, I., Losch, R., Schlütting, P., Juarez, P., Bissantz, N., Ramirez, S. R. & Eltz, T. 2017. Blown by the wind: the ecology of male courtship display behavior in orchid bees. **Ecology**, 98, 1140–1152. DOI: 10.1002/ecy.1755

Policarová, J., Cardinal, S., Martins, A. C. & Straka, J. 2019. The role of floral oils in the evolution of apid bees (Hymenoptera: Apidae). Biological Journal of the Linnean Society, 128(2), 486–497. DOI: 10.1093/biolinnean/blz099/5542934Ramos, K. 2013. The bee genus *Arhysosage* Brèthes (Apidae: Andreninae: Calliopsini): new species, taxonomic notes and new distribution records from Brazil. **Journal of Natural History**, 47, 2735–2747. DOI: 10.1080/00222933.2013.791887

Raw, A. 1975. Territoriality and scent marking by *Centris* males in Jamaica. **Behaviour**, 54, 311–321.

Roig-Alsina, A. 1997. A generic study of the bees of the tribe Tapinotaspidini, with notes on the evolution of their oil-collecting structures. **Mitteilungen der Münchner Entomologischen Gesellschaft**, 87, 3–21.

Rozen, Jr., J. G. 1958: Monographic study of the genus *Nomadopsis* Ashmead (Hymenoptera: Andrenidae). **University of California Publications in Entomology**, 15, 1–202.

Rutowski, R., Alcock, J. 1980. Temporal variation in male copulatory behavior in the solitary bee *Nomadopsis puellae* (Hymenoptera: Andrenidae). **Behaviour**, 73, 175–88.

Sabino, W. O, Silva, C. I. & Alves-dos-Santos, I. 2017. Mating System and Sleeping Behaviour of the Male and Female *Centris* (*Paracentris*) *burgdorfi* Friese (Apidae, Centridini). **Journal of Insect Behavior**, 30,103–118. DOI: 10.1007/s10905-017-9600-x

Schlindwein, C. & Wittmann, D. 1995: Specialized solitary bees as effective pollinators of South Brazilian species of *Notocactus* and *Gymnocalycium* (Cactaceae). **Bradleya** 13:25–34.

Searcy, W. A. 1982. The evolutionary effects of mate selection. **Annual Review of Ecology and Systematics**, 13, 57–85.

Seidelmann, K. 2014. Optimal progeny body size in a solitary bee, *Osmia bicornis* (Apoidea: Megachilidae). **Ecological Entomology**, 39, 656–663. DOI: 10.1111/een.12145

Sihag, R. C. 1986. Reproduction of alfalfa pollinating subtropical megachilid bees. II. Reproductive isolating mechanisms. **Zoologischer Anzeiger**, 216, 204–217.

Silveira, F. A., Melo, G. A. R. & Almeida, E. A. B. 2002. **Abelhas brasileiras: sistemática e identificação**. Belo Horizonte, p. 253.

Simmons, L. W., Tomkins, J. L. & Alcock, J. 2000. Can minor males of Dawson's burrowing bee, *Amegilla dawsoni* (Hymenoptera: Anthophorini) compensate for reduced access to virgin females through sperm competition? **Behavioral Ecology**, 11(3) 319–325. DOI: 10.1093/beheco/11.3.319

Singer, R. B. & Cocucci, A. 1999. Pollination mechanisms in four sympatric southern Brazilian Epidendroideae orchids. **Lindleyana**, 14, 47–56.

Svensson, B. G. & Bergstrom, G. 1977. Volatile marking secretions from the labial gland of north European *Pyrobombus* D. T. males. **Insectes Sociaux**, 24, 213–224.

Tengö, J. & Bergström, G. 1977. Cleptoparasitism and odor mimetism in bees: do *Nomada* males imitate the odor of *Andrena* females? **Science**, 196, 1117–1119. DOI: 10.1126/science.196.4294.1117

Tengö, J. 1979. Chemical signals and odour-released behaviour in *Andrena* bees. Abstracts of Uppsala Dissertations, 500, 1–37.

Thornhill, R.; Alcock, J. 1983. **The evolution of insect mating systems**. Harvard University Press, London, p. 548.

Toro, H. 1985. Ajuste mecanico para la copula de *Callonychium chilense* (Hymenoptera, Andrenidae). **Revista Chilena de Entomología**, 12, 153–158.

Toro, H., Chiappa, E., Ruz, L. & Cabezas, V. 1991. Compartamiento reproductivo de *Centris mixta tamarugalis* (Hymenoptera: Anthophoridae). **Acta Entomológica Chilena**, 16, 97–112.

Toro, H. & Rodriguez, S. 1997. Correspondencia estructural para la cópula en *Anthidium* (Hymenoptera: Megachilidae). **Revista Chilena de Entomología**, 24, 61–80.

Turgeon, J. J., Mcneil, J. N. & Roelofs, W. L. 1983. Responsiveness of *Pseudaletia unipuncta* males to the sex pheromone. **Physiological Entomology,** 8, 339–344.

Vahed, K. 1998. The function of nuptial feeding in insects: review of empirical studies. **Ethology**, 113, 105–127.

Velthuis, H. H. W. & Camargo, I. M. F. 1975a. Observations on male territories in a carpenter bee, *Xylocopa* (*Neoxylocopa*) *hirsutissima* Maidl. **Zeitschrift für Tierpsychologie**, 38, 409–418.

Velthuis, H. H. W. & Camargo, I. M. F. 1975b. Further observations on the function of male territories in the Carpenter bee *Xylocopa* (*Neoxylocopa*) *hirsutissima* Maidl. **Netherlands Journal of Zoology**, 25:516–28.

Vinson, S. B., Frankie, G. W. & Williams, H. J. 1996. Chemical ecology of bees of the genus *Centris* (Hymenoptera: Apidae). **Florida Entomologist**, 79(2), 109–129.

Vinson, S. B., Williams, H. J., Frankie, G. W. & Shrum, G. 1997. Floral lipid chemistry of *Byrsonima crassifolia* (Malpighiaceae) and a use of floral lipids by *Centris* bees (Hymenoptera: Apidae). **Biotropica**, 29, 76–83.

Vogel, S. 1969. **Flowers offering fatty oil instead of nectar**. In: Abstracts of the Papers Presented at the XI International Botanical Congress, 2, p. 229.

Vogel, S., 1971. Ölproduzierende Blumen, die durch ölsammelnde Biene bestäubt werden. **Naturwissen**, 58, 58.

Vogel, S. 1974. Ölblumen und ölsammelnde Bienen. Abh. T**ropische Subtropische Pflanzenwelt**, 7, Steiner, Wiesbaden, p. 267.

Vogel, S. & Machado, I. C. 1991. Pollination of four sympatric species of *Angelonia* (Scrophulariaceae) by oil-collecting bees in NE. Brazil. **Plant Systematics and Evolution**, 178,153–178, 1991.

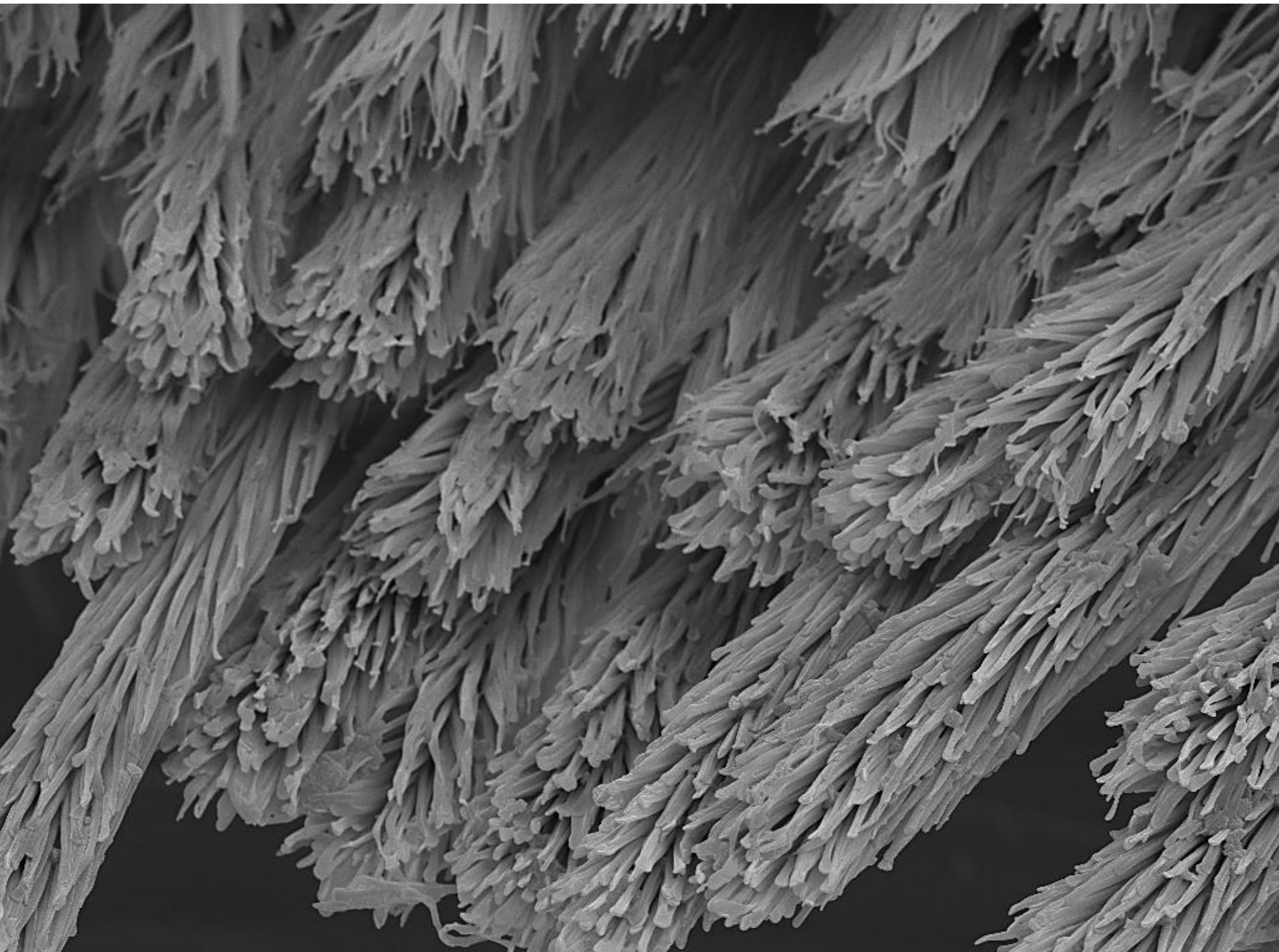
Wittmann, D. & Blochtein, B. 1995. Why males of leafcutter bees hold the females' antennae with their front legs during mating. **Apidologie**, 26(3), 181–196. DOI: 10.1051/apido:19950302

Wittmann, D., Radtke, R., Cure, J. & Schifino-Wittmann, M. 1990. Coevolved reproductive strategies in the oligolectic bee *Callonychium petuniae* (Apoidea, Andrenidae) and three purple flowered Petunia species (Solanaceae). **Journal of Zoology**, 28, 157–165.

CAPÍTULO 1

The evolution of oil-collecting structures in the males of Tapinotaspidini bees (Hymenoptera, Apidae)

Manuscrito formatado segundo as normas da revista Arthropod Structure & Development (Anexo 1)



Highlights

Males of Tapinotaspidini collect floral oil.

The male oil-collecting structures are similar to the female.

The metasoma structures associated with oil storage and transference are exclusive to males.

Many convergences of morphology occurred along the evolution of oil-collecting structures in males of Tapinotaspidini.

The function of the oil for males is unknown but may be associated with mating behavior.

Abstract

Studies with emphasis on the interactions between bees and plants are generally concentrated on females, and few are known about the behavior of males in collecting floral resources. Males exploit floral resources for feeding or for collection of floral fragances, while females exploit various floral resources associated with feeding and nesting. Among the tribes of oil collecting bees only the males of Tapinotaspidini and Tetrapediini present specialized structures, in the legs and metasoma, for the collection of this resource. The morphological characters of the structures of collection, transference and storage of oil present in the males were analyzed for forty species of Tapinotaspidini. These characters were mapped in the phylogeny of Tapinotaspidini tribe. We reconstructed the ancestral states of the structures associated with the collection of oil from the males of the tribe. Our results indicate that all Tapinotaspidini males collect oil. The morphology of the male legs is similar to that of the female legs, although the metasoma structures associated with oil collection are exclusive to males. Our optimizations indicate several cases of convergence of morphology in Tapinotaspidini males. We discuss the possible function of oil and suggest that it can be related to mating behavior.

Keywords: Ancestral states; behavior; mating; morphology; solitary bees.

Resumo

Estudos com ênfase nas interações entre abelhas e plantas são geralmente focados nas fêmeas, sendo que pouco é conhecido sobre o comportamento dos machos na coleta de recursos florais. Os machos exploram recursos florais para a alimentação ou para coleta de essências florais, enquanto as fêmeas exploram diversas fontes florais associadas a alimentação e nidificação. Dentre as tribos de abelhas coletoras de óleo apenas os machos de Tapinotaspidini e Tetrapediini apresentam estruturas especializadas, nas pernas e metassoma, para a coleta desse recurso. Os caracteres morfológicos das estruturas de coleta, transferência e armazenamento de óleo presente nos dos machos foram analisados para quarenta espécies e mapeados na filogenia da tribo Tapinotaspidini. Reconstruímos os estados ancestrais das estruturas associadas a coleta de óleo dos machos da tribo. Os nossos resultados indicam que todos os machos de Tapinotaspidini coletam óleo. A morfologia das pernas dos machos é semelhante a das fêmeas, embora as estruturas do metassoma associadas ao armazeamento de óleo sejam exclusivas dos machos. As nossas otimizações indicam diversos casos de convergência da morfologia nos machos de Tapinotaspidini. Discutimos a possível função do óleo e sugerimos que possa estar relacionada ao comportamento de acasalamento.

Palavras-chave: Abelhas solitárias, acasalamento; comportamento; estados ancestrais

1. Introduction

The study of the interactions between bees and plants is mainly concentrated in females, and comparatively few are known about the differentiated behavior of males in resource collection and their behavior in pollination relations (Smith et al. 2019). In bees, the pollen-female relationship makes the system strongly centered on only one sex, and the role of males in pollination is recognized in a few groups, such as the Euglossini in which they have distinct behaviors from females in interactions with plants (Dressler 1982). The Euglossini represent the exception to this rule, where males represent the most efficient group of visitors and pollinators than females and with a large number of studies on this system (Dodson, 1969; Bembé et al. 2004; Eltz et al. 2015; Eltz et al. 2019). This specialized interaction of males with flowers involves beyond the collection of food resources associated with sexual selection (Eltz et al. 2015; Hetherington-Rauth & Ramirez 2016; Etl et al. 2017).

The collection of floral resources – pollen, nectar, oil, resin – is the result of complex and often specialized interactions between bees and plants (Michener 2007). The males explore floral resources exclusively for their own food, or for collecting floral essences. In turn, females explore a wide range of resources that are used for feeding and nesting behavior (Michener 2007; Danforth et al. 2019).

Among the different groups of solitary bees, the tribes Tapinotaspidini and Tetrapediini tribes by the recent discovery of structures related to the collection of non-food floral resources by males that are possibly related to sexual selection (Cappellari et al. 2012; Etl et al. 2017). Of the six tribes of bees that collect floral oils, only the males of these two tribes have been observed collecting oil, and in the other tribes of Neotropical bees such as Centridini, the males do not present any structure for collecting oil. Although there is no full understanding of the functionality of oil for males, it is believed that it may have some relation to mating behavior, either as a nuptial gift or as camouflage (Cappellari et al. 2012). Described by Vogel (1969, 1974), the oil syndrome represents one of these specialized relationships between plant and pollinator, involving about 440 bees from two families (Apidae and Melittidae) and at least 10 plant families (Buchmann 1987; Michener 2007; Danforth et al. 2019). The collection of oil is carried out through specialized structures, in legs and metasoma, extremely varied and depend on the type and location of the elaiophores (oil secreting glands) explored (Neff and Simpson 1981; Roig-Alsina 1997; Cocucci et al. 2000; Michener 200; Danforth et al. 2019).

The Tapinotaspidini tribe presents small to medium in size bees with high morphological diversity, with about 150 species and distributed in 13 genera, with most species nesting in soil (Silveira et al. 2002; Aguiar 2012). The oil-collecting structures of Neotropical Tapinotaspidini bees have a higher morphological variation than those observed in other tribes, with males and females presenting morphological adaptations for accentuated oil collection (Roig-Alsina 1997; Coccuci et al. 2000; Aguiar & Melo 2007). However, in general, few studies emphasize the manipulation of viscous substances by bees (Sakagami & Camargo 1964; Gastauer et al. 2011).

The majority of studies involving oil-collecting structures in Tapinotaspidini, the description and all knowledge of oil collecting structures are focused on females (Neff & Simpson 1981; Roig-Alsina 1997; Cocucci et al. 2000) and few are known about modifications present in males. However, the structures related to oil collection and transport in males are apparently more diverse than in females and little known among the species of the different genera of the tribe and although there is not a full understanding of the oil's functionality for males, is expected that it may have some relationship with mating behavior.

The knowledge about the structures for collecting odors, resins, oils, nectar and pollen in bees is totally focused on females and little is known about the structural and functional diversity in males of different groups of bees. The understanding of the evolution of biological and morphological characters is only possible in a clear way through the understanding of the distribution of these characters within a phylogenetic tree, and the current tools allow not only to understand the emergence, loss and transformation of characters in the evolution of the group but also the estimation and reconstruction of ancestral states (Wenzel 1992; De Queiroz & Wimberger 1993). Thus, the main objective of the present study is to analyze the evolution of the oil-collecting structures in the Tapinotaspidini males through the transformation of their structures into their phylogeny, and to understand if determined character sets are exclusive or complementary, and their relationships with the plants in terms of adaptation.

2. Material and methods

2.1. Species studied

In order to analyze and survey the characters related to the morphological variation of the oil collecting structures present in males, forty species with more distinct morphologies from all 13 genera of the Tapinotaspidini tribe were analyzed, comprising the largest available variation in terms of lineages and geographical distribution (Table 1). As outgroup four species of the Emphorini tribe were analyzed: *Alepidosceles* sp. Moure, 1947; *Ancyloscelis* sp. Latreille, 1829; *Melitoma* Lepeletier & Serville, 1828 and *Ptilothrix* Smith, 1853 (classification according to Michener 2007). For this study material was obtained through collection in the field, loan or donation from different institutions.

2.2. Study of oil-collecting structures

The collection of oil is carried through specialized structures, in the legs and metasoma, and directly related to the type and location of the elaiophores (oil secreting glands) explored. (Neff and Simpson 1981; Roig-Alsina 1997; Cocucci et al. 2000; Michener 2007; Danforth et al. 2019). In order to evaluate the possible use of different parts of the body in the collection or transport of oil, Roig-Alsina (1997) and Cocucci et al. (2000) inferred the possible adaptation by the presence of oil-like mucilages attached to the hairs of Tapinotaspidini females. In order to analyze the possible adaptations, besides the direct observation of the behavior in the field, we studied the material regarding the presence of oil-like mucilages in different parts of the body such as: fore legs (basitarsus and tarsomeres), middle legs (basitarsus and tarsomeres), hind legs (tibia and basitarsus), sterna (S2, S3, S4 and S5), terga (T2, T3, T4, T5, T6 and T7). These mucilages are potentially oil and distinct from nectar because they are shiny and do not evaporate. The structures studied followed the structure nomenclature proposed by Michener (2007). For the description of the position of the oil collecting structures present in the legs, the following terminology was adopted: internal face (ventral surface) and external face (dorsal surface), as suggested by Neff & Simpson (1981).

The material was studied in Leica stereoscopic microscope (model M125), with magnification up to 100X, in order to analyze general patterns of structures related to oil collection, transport and storage. The difference in size between the different potentially homologous structures between males and females was also evaluated, with the aid of measurements obtained with a micrometric scale coupled to the eyepiece. In order to evaluate and record the structures with the highest level of detail, the specimens were also studied using a scanning electron microscope (SEM) in the Laboratório de Microscopia e Microanálise (LMM), Instituto de Ciências Biológicas da Universidade de Brasília (UnB). The images produced through the SEM are available in SEM are available at MorphoBank (https://morphobank.org/) and can be accessed through the project "P3567: Structures of oil collecting bee males of the tribe Tapinotaspidini and Tetrapediini (Hymenoptera: Apidae)" (Table 2; list of species studied and records of images in Morphobank).

2.3. Reconstruction of ancestral states

The characters used for the reconstruction of the ancestral states were mapped in the phylogeny of the tribe Tapinotaspidini (Aguiar et al. 2020). The optimization of the patterns, decomposing the structures associated with the collection of oil in phylogeny, was realized through the Mesquite software (Maddison & Maddison 2019) by the methods of maximum parsimony (MP) e maximum likelihood (ML).

3. Results

3.1. List of characters

In the analysis of the morphological variation and inferences of the characters of the species studied was used the general principle of similarity (Wiley & Liberman 2011). Each character was proposed based on its special similarity, by position, by possible functional attributes, and by general characteristics of similarity based on its smaller parts. Only characters hypothetically associated with the collection and/or transport of oil in Tapinotaspidini males were implemented. In some conditions the characters listed are compared to those present in females.

The characters have been coded – according to the approach proposed by Sereno (2007) – from the anterior to the posterior region of the body, with the main focus on the legs, sterna and terga. The first tarsomere of each of the legs (fore, middle and hind) of the analyzed species was considered as basitarsus (Michener 2007), and the remaining as tarsomeres 2 to 4. The dense band of plumose hairs on posterior margin of T6-T7 of males was named fimbria according to Michener (2007).

To coding the characters related to the pilosity of the terga (T2-T6) and sterna (S2-S5) of the males, it was adopted the criterion of serial homology (Owen, 1848; Roth, 1984), in such a way that the terga represent conjunct of terms that are repeated and may present some variation about the localization and function of hair.

List of characters observed on males associated to oil collecting, transfer or transport on the species of Tapinotaspidini bees, from anterior to posterior portion of the body:

Fore legs

Character 1 – Fore basitarsus with row of stout setae:

(0) absent; (1) on the inner margin with row of stout setae; (2) on the outer margin with row of stout setae.

Males of *Monoeca* have a conspicuous line of stout simple setae that is distributed throughout the inner margin, from strigilus to basitarsus apice (Fig. 1). Vogel (1974) suggest that this row of setae is more developed on females, with longer setae, than on males. Friese (1925) reported males of the species *Monoeca mexicana* (cited as *Tetrapedia saussurei*) on the flowers of *Malpighia* sp. (Malpighiaceae), but there isn't still complete description of the form and function in males. I observed on females of *Monoeca xanthopyga* Harter-Marques, Cunha & Moure, 2001, that the length of simple setae on comb is much longer than in males (females: setae have about one third of the maximum ventral width of the basitarsus - length of setae 0.15/ 0.75 mm). The set of setae on females is larger than in males, however the shape of the comb is very similar. Males of *Monoeca mourei* Aguiar, 2012, *Monoeca pluricincta* (Vachal, 1909) and *Monoeca xanthopyga*, I observed oil in the plumose hairs of dorsal surface of fore basitarsus. Males and females of *Monoeca* have a strong short setae on the apices of fore basitarsus that is probably associated to cut the epidermis of epithelial elaophores of Malpighiaceae. I observed a conspicuous comb in outer margin of basitarsus (Fig. 2) on the species of the genera *Arhysoceble, Lophopedia, Paratetrapedia, Trigonopedia, Tropidopedia*, *Urbanapis* and *Xanthopedia*. On the males this comb is less developed than in females, the setae of the male comb are about 0.8X longer than in female combs. The species of the group *pallidipennis* of *Tropidopedia* is an exception, were I saw on *Tropidopedia nigrita* Aguiar & Melo, 2007 and *Tropidopedia peruana* Aguiar & Melo, 2007 that this row is much more developed than in females. The shape of the comb of females was already described by Vogel (1974), and Neff & Simpson (1981), who observed a conspicuous comb on the outer margin of the females of the species *Arhysoceble picta* (Friese, 1899), *Paratetrapedia moesta* (Cresson, 1878) and *Tropidopedia punctifrons* (Smith, 1879) (cited as *Paratetrapedia haeckeli*).

Character 2 – Plumose pubescence on the ventral surface of fore basitarsus:

(0) absent; (1) composed by dense fine plumose setae; (2) composed by long scattered plumose setae; (3) composed by very long plumose setae.

All the species of Tapinotaspidini have plumose hairs on ventral surface of fore basitarsus, however this pubescence can vary from dense fine plumose composing pads on *Chalepogenus, Arhysoceble, Caenonomada* and *Lanthanomelissa* (Fig. 3A), and long coarsely plumose setae on *Tapinotaspis, Tapinotaspoides, Paratetrapedia* and *Monoeca* (Fig. 3B). These two patterns were already discussed in Vogel (1974) as part of type alfa and type beta respectively. I considered dense the set of plumose hairs that made it impossible to visualize the integument and scattered the condition in which it was possible to view the integument of the fore basitarsus. The males of genera of *Chalepogenus, Monoeca, Tapinotaspis* and *Tapinotaspoides* have plumose hairs on dorsal surface of fore basitarsus. In *Tropidopedia nigrita* a pattern of pilosity consisting of very long and plumose hair is observed (Fig. 4) as observed in other males of the group *pallidipennis* (Aguiar & Melo 2007). The pads of plumose hairs of fore basitarsus are associated with oil absorption (Vogel 1974).

Character 3 – Pubescence on the dorsal surface of fore tarsomeres 2-4:

(0) composed by stout simple setae; (1) composed by fine simple setae.

Stout simple setae (e.g. *Caenonomada*, Fig. 5; *Lanthanomelissa*, Fig. 6) and fine simple setae are variations of the same archetype of simple setae, and are related to distinct behaviors of oil collection. The stout simple setae are associated with the collection of oil in epithelial elaiophores, through the behavior of the bee scrape the epidermis to have access to oil (Vogel 1974; Cocucci & Vogel 2001). In the other Tapinotaspidini species the males have fine simple setae.

Middle legs

Character 4 – Pubescence on the dorsal surface of middle basitarsus:

(0) composed by only simple hairs; (1) composed by plumose and simple hairs; (2) composed by dense band of long, stout and curved setae to opposite sides at the distal portion of setae.

The dorsal surface of the middle basitarsus of most Tapinotaspidini males have a pilosity consisting of simple and plumose hairs. The absence plumose hairs are noted in the males of *Paratetrapedia duckei* (Friese, 1910), *Paratetrapedia flaveola* Aguiar & Melo, 2011, *Paratetrapedia fervida* (Smith, 1879) and *Trigonopedia ferruginea* (Friese, 1899). In *Tapinotaspis* an exclusive pattern of pilosity is observed (Appendix 1), composed by dense band of long, stout and curved setae to opposite sides at the distal portion of setae (Roig-Alsina 1997, 2003; Cocucci et al. 2000).

Character 5 – Plumose pubescence on the dorsal surface of middle basitarsus:

(0) absent; (1) dense; (2) sparse.

The species of the genera *Chalepogenus*, *Monoeca* and *Tapinotaspoides* present dense plumose pilosity occupying at least 2/3 of the dorsal surface of the middle basitarsus (Appendix 2A), but in the *Caenonomada* males and some species of *Paratetrapedia* are concentrated in the posterior margin (Appendix 2B). Plumose hairs in the middle basitarsus are absent in *Paratetrapedia duckei*, *Paratetrapedia flaveola*, *Paratetrapedia fervida* and *Trigonopedia ferruginea* species.

Hind legs

Character 6 – Tibial scopa:

(0) composed mainly by dense plumose pubescence, with few scattered and longer simple setae;

(1) composed by sparse plumose and simple hairs, some occasions scattered on outer margin;

(2) almost absent, with only a row on outer margin.

Most of the species have the male tibial scopa well developed, however not well developed as females (Appendix 3). The male scopa of some Tapinotaspidini bees are strongly well developed when compared to males of Centridini bees, where the males do not collect oil and almost do not have any scopa on hind legs. Almost all males have dense scopa on hind tibia (e.g. *Chalepogenus* and *Monoeca*), where the hairs cover mostly of tibia. On males of *Lanthanomelissa* and *Lophopedia,* the plumose hairs cover about 2/3 of tibial surface. On males of *Paratetrapedia* the plumose hairs of scopa are very reduced on the outer margin of tibia. *Caenonomada* is the genus with a more pronounced reduction of tibial scopa. The scopa of Tapinotaspidini bees are commonly composed by dense plumose hairs and few scattered simple long setae.

Character 7 – Basitarsal scopa:

(0) composed by dense plumose and simple hairs; (1) composed by sparse simple and plumose hairs; (2) almost absent, with only a row on outer margin.

The basitarsal scopa of males is considerably underdeveloped compared to females, especially for the species of *Caenonomada* and *Monoeca*. Many males have dense scopa (e.g. *Arhysoceble, Chalepogenus, Lanthanomelissa* and *Tropidopedia*) composed by dense plumose and simple hairs. *Paratetrapedia fervida, Paratetrapedia volatilis* and *Monoeca pluricincta* have plumose hairs covering about 2/3 of dorsal surface of basitarsus (Fig. 7). The males of *Caenonomada unicalcarata, Tapinotaspoides serraticornis* and some species of *Paratetrapedia* the plumose hairs are concentrated in the posterior margin of the basitarsus (Appendix 4). The simple hairs of the posterior basitarsus of the males are shorter than those of the females, in the same way that it is observed for the tibial scopa. I observed oil on basitarsal scopa of males of most of genera of Tapinotaspidini (Appendix 5).

Terga

Character 8 – Plumose hairs on posterior margin of the terga:

(0) absent; (1) T2-T6; (2) T3-T6; (3) T4-T6; (4) T5-T6; (5) T6.

The males of the genera *Arhysoceble*, *Chalepogenus*, *Xanthopedia* have plumose hairs on posterior margin of T2-T6. On the lateral margins of T2 of the species of the *Chalepogenus* are observed plumose setae (Roig-Alsina, 1999). I observed oil in the plumose setae on T2 of males of *Chalepogenus caeruleus, Chalepogenus nigripes, Lantanhomelissa clementis.* Males of many of the species studied present plumose setae on the posterior margin of T3. In *Monoeca mourei*, *Monoeca pluricincta*, *Urbanapis danunciae* and *Urbanapis diamatina* the plumose setae occupy the lateral margins of T3. I observed oil in many of the analyzed males, but at *Paratetrapedia connexa* and *Urbanapis diamantina* higher volumes of oil were recorded. In males of many of the species studied have plumose hairs on the posterior margin of T4. In males of *Chalepogenus nigripes* the plumose hairs have marginal band occupying less than third of margin of T4. Most Tapinotaspidini males have plumose pubescence in T5-T6 (Fig. 8A). However, in some species as *Paratetrapedia testacea* (Smith, 1854)*, Paratetrapedia volatilis* and *Trigonopedia ferruginea* the plumose hairs are absent (Fig. 8B). I observed a high quantity of oil in the fimbrias of the T5-T6 of the males of *Paratetrapedia connexa, Tropidopedia flavolineata* and *Urbanapis diamantina.*

Sterna

Character 9 – Plumose hairs on posterior margin of the sterna:

(0) S2-S5; (1) S3; (2) S3-S5; (3) S4-S5; (4) S5.

Almost all the males of the studied species of Tapinotaspidini had a special set of plumose hairs on the sternal margins (Fig. 9). The plumose hairs observed on the sterna of the males vary in relation to density, length and location, usually constituting elongated and convergent fringes (fringes sternal), probably associated with oil storage transference. The males of *Chalepogenus nigripes* and *Lanthanomelissa*, have these fringes more concentrated in the central portion of the S2-S4. The males of *Chalepogenus caeruleus* and *Tapinotaspis chalybaea* the plumose hairs are concentrated in the lateral portions of S2. The males of *Caenonomada unicalcarata* have plumose hairs on posterior margin of S3, longer on the lateral portions. The males of *Lophopedia pygmaea*, *Lophopedia nigrispinis*, *Tropidopedia flavolineata*, *Tropidopedia punctifrons* and some species of *Paratetrapedia* have long, decumbent and plumose hairs in the lateral portions of the S2-S5, almost always presenting viscous resins like oil in the individuals analyzed.

Character 10 – Plumose hairs on posterior margin of the sterna:

(0) dense band of long, fine and plumose hairs, and long foliaceous plumose hairs (1) plumose hairs concentrated in the central or lateral portion; (2) long hairs branched in the distal portion.

The males of *Monoeca* have dense hair bands on the posterior margin of the sterna (S2-S6), consisting of dense band of plumose hairs, with simple and long hair fringes on lateral portions of S2-S5. This pattern is high variable between the species of *Monoeca* and are specially informative for taxonomy delimitation of the species. The dense bands of plumose hairs in *Monoeca* are composed by spiral foliaceus shaped hairs, located under the long fringes of of S2-S5 (Fig. 10). The majority of males have the most concentrated plumose hairs in the lateral (e.g. *Paratetrapedia* and *Tropidopedia*) or central portions (e.g. *Xanthopedia* and *Urbanapis*) of the sterna. In turn, *Arhysoceble* males have long hairs branched in the distal portion of the sterna (Fig. 11). Through SEM analysis I observed that the plumose hairs in the sterna of males do not correspond to those observed in females that have specialized fringes of long, wavy setae along the posterior margins of the sterna (S3-S5). (Fig. 12).

Character 11 – S3, deeply concave with dense plumose pubescence:

(0) absent; (1) present.

The males of *Paratetrapedia fervida* and *Paratetrapedia flaveola* have a deeply concave area in U' shape on posterior margin of S3 (Appendix 6), covered by dense short plumose pubescence. Males of *Paratetrapedia chocoensis* Aguiar and Melo (2011), were observed using this special pubescence on S3 to collect fragrances from inflorescences of *Anthurium acutifolium* (Araceae) (Etl et al. 2017). This deeply concave area isn't observed on the females of Tapinotaspidini, many males of *Paratetrapedia* studied had shiny substances on this pubescence, and some males collected on flowers of *Oncidium* (Orchidaceae) had the pubescence of S3 covered by a shiny viscous substance like oil.

3.2. Reconstruction of adaptations for oil collecting based on parsimony and maximum likelihood

Tapinotaspidini males of all studied species present adaptations for the collection (Fig. 13) and transport of floral oil. The ancestor of Tapinotaspidini males did not have a setae comb on the inner or outer margin of the fore basitarsus (character 1). The comb (external margin) arose only once in the common ancestor (CA) of *Arhysoceble-Paratetrapedia*, occurring a state reversal in *Lanthanomelissa* (Fig. 14). Males of *Monoeca* males have a collecting comb in the outer margin of the basitarsus (fore and middle). This structure arose only once, in the last common ancestor of all *Monoeca* species, representing an autapomorphy of the genus.

All Tapinotaspidini have plumose hairs on the ventral surface of the fore basitarsus (character 2). However, the pilosity pattern presents variations between species (Fig. 15). The common ancestor of all Tapinotaspidini was long and sparse plumose hairs on the ventral surface of the fore basitarsus, as observed in most species (e.g. *Monoeca*, *Paratetrapedia*). The dense, fine and plumose males possibly arose by convergence in three moments of Tapinotaspidini evolution. In the common ancestor of *Arhysoceble-Lanthanomelissa* clade and the genus *Chalepogenus*. However, in *Caenonomada*, this pattern of pilosity would have arisen as a derived state, since the ancestor of the genus had long and sparse plumose hairs (as in the ancestor of the tribe). The third pattern of pilosity on the ventral surface of the fore basitarsus, with extremely elongated plumose hairs, is observed in *Tropidopedia nigrita*. It is expected that this pattern of pilosity represents a derived state, since the ancestor of *T. nigrita* had long and sparse plumose hairs, as occurs with the other species of *Tropidopedia*.

The dorsal surface of the fore tarsomeres (T2-T4) of the common ancestor of all Tapinotaspidini (character 3) was covered with stout simple setae. This pattern of pubescence was maintained in the species of *Caenonomada*, *Monoeca* and *Lanthanomelissa* (except *L. parva*). The rigid and foliaceous bristles, on the margin of the front tarsomers, of *Lanthanomelissa betinae* and *Lanthanomelissa clementis* represent a derived and unique state, since the ancestor of *Arhysoceble-Lanthanomelissa* presented stout simple setae as observed for the other species of the tribe.

Our optimizations suggest that the common ancestor of all Tapinotaspidini presented the dorsal surface of the middle basitarsus (character 4) covered by simple and plumose hairs (Appendix 7). The absence of hair in *Trigonopedia ferruginea* and some species of *Paratetrapedia* represents a reversal of the ancestral state. In *Tapinotaspis*, the presence of a pattern of pilosity distinct from other species (long, stout and curved setae to opposite sides at the distal portion of setae), represents an autapomorphy of the genus. Still in relation to the pilosity in the middle leg, it is expected that the common ancestor of the Tapinotaspidini tribe had dense plumose pilosity on the dorsal surface of the middle basitarso (character 5). This pattern was maintained in most species (Appendix 8).

Most Tapinotaspidini present dense and plumose pilosity in the tibial scopa of the hind legs (character 6). Only in the genera *Caenonomada*, *Paratetrapedia* and *Urbanapis* are differentiated pilosity patterns observed (Appendix 9). In *Paratetrapedia* and *Urbanapis* a sparse scopa, formed by simple and plumose hairs, would have arisen in the common ancestor of both genera. In *Caenonomada unicalcarata* there was a reduced tibial scopa, which is almost absent in males of the species, but not for the ancestor of the genus. In the hind basitarsus (character 7), similar to the pattern observed in the tibia, of most Tapinotaspidini males there is a dense scopa formed by simple and plumose hairs, as expected for the ancestor of the tribe. Most *Paratetrapedia* species have practically no plumose pilosity in the tibia, as was the case in the ancestor of the genus. In some species (e.g. *Monoeca pluricincta*) it is expected that a sparse scopa, formed by simple and plumose hair has arisen independently. While for the *Tapinotaspis* and *Tapinotaspoides* species it is expected that the common ancestor of the clade (Appendix 10).

The ancestor of Tapinotaspidini males had plumose pilosity in the terga (T2-T6), as observed for the genera *Arhysoceble*, *Xanthopedia*, *Chalepogenus*, *Tapinotaspis* and *Tapinotaspoides* (character 8). Our optimizations are ambiguous for *Monoeca* and part of the *Paratetrapedia* lineage (Fig. 16). Based on the maximum likelihood analysis, it expected that ancestor of *Monoeca* ancestor had plumose pilosity in T3-T6; and in *Lophopedia*-*Tropidopedia*-*Paratetrapedia* in T5-T6. Plumose hairs in T5-T6 also arose independently in *Caenonomada unicalcarata*. *Tropidopedia carinata* was the only species of the study presenting by the plumose only in the margins of T6. While the absence of hairs on the tergos is registered for *Paratetrapedia volatilis*, *Paratetrapedia testacaea*, *Trigonopedia ferruginea* and *Tropidopedia nigrita*. The density and location (covering the all the margin or only the lateral portions of the terga) of the plumose hairs can vary depending on the species. In the tergal scopa of males of some genera (e.g. *Paratetrapedia*, *Tropidopedia*) the presence of oil was observed in many evaluated specimens (Appendix 11).

The analyses of parsimony and maximum likelihood present the same result for the evolution of the plumose hairs in the posterior margin of the sterna (character 9) of Tapinotaspidini males (Fig. 17). The common ancestor of the tribe presented plumose hairs in S2-S5, a state currently observed in the genera *Lanthanomelissa*, *Chalepogenus*, *Trigonopedia*, *Monoeca* and in the *Tapinostapis*-*Tapinotaspoides* clade. The *Arhysoceble* and *Paratetrapedia* species have plumose pilosity in S3-S5, which arose independently for both groups. In the males of *Lophopedia* and *Caenonomada*, this character represents an autapomorphy, since the plumose are observed, respectively, only in S4 and S5, of both genera.

Our optimizations suggest that the evolution of the plumose hairs on the posterior margin of the sterna (character 10) of Tapinotaspidini males occurred through an ancestor that presented long pilosity covering the posterior margin of this structure (Appendix 12). In the genus *Arhysoceble* a condition is observed in which the hair is branched in the distal portion and arose only once in the ancestor of the genus. In *Monoeca*, an autapomorphy of the group is also observed, once under the dense band of plumose hairs in the sterna, the presence of an exclusive dense band of long and spiral foliaceous hairs is observed. Tapinotaspidini males present a considerably more complex pilosity in the sterna than that observed in females, which usually have only simple hairs in this region. Through SEM analysis it was possible to visualize in detail the high variety of hair present in the sterna of Tapinotaspidini males (Appendix 13). The presence of a deeply concave area on posterior margin (with dense plumose pubescence) is observed only in *Paratetrapedia fervida* and *Paratetrapedia flaveola* (Appendix 14). Our results suggest that this character arose independently in both species, considering that it was absent in the common ancestor of the genus.

4. Discussion

4.1 Evolution of oil-collecting structures

Our results demonstrate that all Tapinotaspidini males studied have morphological adaptations for the collection, transference and storage of oil. These structures are located in the legs and metasoma, and in some species are more accentuated in males. Previous studies have indicated that the Tapinotaspidini tribe presented the highest diversity of structures specialized in oil collection (Roig-Alsina et al. 1997; Cocucci et al. 2000; Neff & Simpson 2017). However, studies with emphasis on morphological adaptations for oil collection from Tapinotaspidini males had not been carried out, although isolated reports of collection or patrolling behavior in oil flowers are available in the literature (Cocucci 1991, Aguiar & Melo 2009).

The Tapinotaspidini ancestral male had no collector comb in the fore basitarsi. The collection of oil possibly occurred through long plumose and scattered hairs on the ventral surface of the basitarsus, and stout setae on the dorsal surface of the tarsomeres, located on the fore legs. The origin of the collector comb in *Monoeca* (inner margin) occurred in the common ancestor of the genus, in Miocene (ca. 20 Ma), and represents an autapomorphy within the Tapinotaspidini, *Monoeca* males have a similar comb, in relation to position, to that observed in *Centris* and *Epicharis* females (Roig-Alsina 1997; Neff & Simpson 1981). The external comb also appeared only once in the Tapinotaspidini tribe, about 38 Ma. (Eocene), in the common ancestor the lineages *Arhysoceble* + *Lanthanomelissa* and *Paratetrapedia*, although the reversion of the state is observed in the genus *Lanthanomelissa*.

All Tapinotaspidini males have plumose hairs on the ventral surface of the fore basitarsus. According to Vogel (1974), the pilosity that covers the surface of the basitarsus has the function of absorbing the oil secreted by the elaiophores. In Tapinotaspidini males, this character evolves from a common ancestor with pilosity consisting of long and sparse hairs. The diversification occurred, about 37-30 Ma, with the development of a fine and plumose dense pilosity in *Chalepogenus* and *Arhysoceble-Lanthanomelissa* lineage. This state of character occurs in most species of the tribe (e.g. *Monoeca*, *Paratetrapedia*, *Tapinotaspis* and *Tapinotaspoides*) and was described as beta pattern by the german botanist (1974). The males of *Tropidopedia* nigrita and other species that compose the pallidipennis group (e.g. *Tropidopedia peruana*) have much longer hairs than (observed in females) and other Tapinotaspidini species. (Aguiar & Melo 2007). The pattern described as alpha – composed of dense fine and plumose hairs – is observed in males of *Chalepogenus*, *Caenonomada* and *Lanthanomelissa* (Vogel 1974).

The Tapinotaspini tribe has many cases of convergence of morphology, but they are difficult to detect because of subtle differences in the hairs and their shapes. For example, in the independent arising of the dense, fine and plumose pilosity on the ventral surface of the fore basitarsus of *Caenonomada*, *Chalepogenus* and *Arhysoceble-Lanthanomelissa*. When we evaluate the pilosity pattern of these structures in *Arhysoceble* and *Caenonomada*, we perceive similarities in the pattern of distribution and morphology of the hairs. However, *Arhysoceble* is more related to the *Paratetrapedia* genus, with which other states of character are shared (e.g. fringes sternal and comb in outer margin of basitarsus). In this respect, the disposition of the plumose pilosity in the sterna (S3-S5) of the species of *Arhysoceble*, *Lanthanomelissa* and *Paratetrapedia* represent another convergence of state of character. The plumose pilosity in the S3-S5 evolves from the common ancestor of the three genera, which presented by the plumose hairs in S4-S5, in the same way that occurs in the males of *Tropidopedia*, *Xanthopedia* and *Urbanapis*.

Tapinotaspis and *Tapinostaspoides* are the only genera of Tapinotaspidini in which, males and females, do not present morphological adaptations to collect oil in the fore legs (Roig-Alsina 1997; Cocucci et al. 2000; Roig-Alsina 2003; Melo & Gaglianone 2005). The pilosity pattern observed in the middle legs of the *Tapinotaspis* males (long, stout and curved setae to opposite sides at the distal portion of setae) is a peculiar characteristic of the genus. We propose two other cases of autapomorphy, for *Arhysoceble* and *Monoeca*, in relation to the plumose hairs pattern located in the sterna. From the SEM analysis it is possible to verify the difference between long hairs branched in the distal portion of the males of *Arhysoceble*; and the long, foliaceous and plumose hairs of the males of *Monoeca*. However, both structures appear to have the function of oil storage (as will be discussed below).

4.2 Evolution of oil storage structures

The oil collected by the Tapinotaspidini males is subsequently transfered to the scopa (tibia and basitarsus) and the terga (T5-T6). I observed the presence of structures in the hind legs and terga specialized in oil retention and transport. Unlike what occurs in the majority of bee males. Similar structures have been described for males of the genus *Tetrapedia* (Tetrapediini), which also collect floral oil in flowers of Malpighiaceae (Cappellari et al. 2012). Our optimizations indicate that the ancestral common to Tapinotaspidini had the tibia and basitarsi covered by a dense scopa composed of long plumose hairs and few simple hairs as previously proposed by Roig-Alsina (1997). The partial reduction of the scopa occurred (ca. 20 -10 Ma) in males of some genera (e.g. *Caenonomada*, *Monoeca*, *Paratetrapedia*).

The hair pattern observed in the tibial and posterior basitarsi scopa of the males of the different Tapinotaspidini species varies in density and distribution. For Roberts & Vallespir (1978) the type of plumose pubescence observed in the hind legs of oil collector bees has the function of absorption and retention by capillarity, while the simple and rigid hairs would serve as a type of protection for this same dense plumose pilosity. In the tibial scopa of Tapinotaspidini males the simple and plumose hairs are usually shorter when compared to those of females. This difference is well evidenced in the species of the genera *Caenonomada* and *Monoeca*. I observed viscous substances such as oil in the tibial scopa of males of most genera: *Arhysoceble*, *Caenonomada*, *Chalepogenus*, *Lanthanomelissa*, *Lophopedia*, *Monoeca*, *Paratetrapedia*, *Tapinotapis*, *Tropidopedia*, *Urbanapis* and *Xanthopedia*. The basitarsal scopa of the males is markedly underdeveloped compared to females (exception *Tropidopedia peruana* Aguiar & Melo, 2007), especially for the *Caenonomada*. However, a reduced scopa not limit the collection of oil by the males, so we observe oil on the hind leg of the males of most Tapinotaspidini genera.

The tergal scopa (T5-T7), composed of long plumose hairs, present in most Tapinotaspidini males is also associated with the storage of floral oil. Plumose hairs covered the T5-T7 of the Tapinotapidini ancestral male, but throughout the evolution of the group, the reduction of the tergal scopal occurred in some species. Although the location and density of the tergal scopa presented variations among the males of the different Tapinotaspidini species, in many of the analyzed specimens the presence of oil was registered. The behavior surrounding the storage of the oil in the tergal scale is similar to that already described for *Tetrapedia*, in which the males transfer the oil from the hind legs to the T5-T6 (Cappellari et al. 2012).

4.3 Potential role of floral oils for Tapinotaspidini males

The function of the oil collected by the males has aroused the interest of researchers since the first records. Vogel (1974) observed the presence of oil in the hind legs of *Chalepogenus rufipes* Roig-Alsina, 1999 (cited as *Tapinotapis rufipes*) males, but suggested that the presence of the oily substance was related to contact with females after successive attempts at copulation. Rasmussen & Olesen (2000), studying the pollination of *Calceolaria* (Calceolariaceae), observed males of *Chalepogenus rasmusseni* Roig-Alsina, 1999 entering the flowers, and presumed that they were ingesting oil, although he did not visualize such behavior. Neff & Simpson (1981) suggested that although *Tetrapedia* males were constantly observed near oil flowers, in search of reproductive females, there was no evidence that they collected the resource. However, the results presented here indicate that Tapinotaspidini males indubitably collect floral oil (see Chapter 2). And in the absence of records that the males use the oil as a food source or investment in the offspring, it is expected that the oil collected by the Tapinotaspidini males is related to sexual selection.

Based on a detailed analysis of the different Tapinotaspidini species, I observed that there is some correspondence of the collecting structures present in the legs of male and female Tapinotaspidini. According to Neff & Simpson (1981), the oil collecting combs of several species of *Paratetrapedia* are identical to those of their respective females. However, the sexual dimorphism observed in terga and sterna is accentuated. The high complexity of the hairs and the hairs observed in the sterna and terga of Tapinotaspidini males is practically absent in females. Secondary sexual characteristics in males involve different ways of accessing reproductive partners, and signaling potential direct benefits to females (Thornhill & Alcock 1983; Leijs & Hogendoorn 2008). In bees, sexual characters are usually expressed by morphological adaptations in the legs, terga and sterna of some species (Stubblefield & Seger 1994; Wcislo & Buchmann 1995).

We did not register Tapinotaspidini species copulation. However, we cannot ignore that the oil collecting apparatus in the male legs may have some function related to this behavior. The males of different bee species have already been described about morphological variations in the legs which are related to sexual selection. In some species (e.g. *Xylocopa* spp.; *Nomia* spp.), males have adaptations in the legs to hold females during coupling (Wcislo & Buchmann 1995; Wittman et al. 2004; Schindler et al. 2018).In *Megachile willughbiella* (Kirby, 1802) males a dense, concave fringe, consisting of branched bristles, and located on the basitarsi and tarsomeres (2-4) anterior covers the eyes of the female copula and, concomitantly, the middle and hind legs are used to hold the abdomen and wings of the partner (Wittmann & Blochtein 1995).

We describe the morphological variety in the sterna of males, and suggest that the high capacity of some males to retain oil in sterna is related to some reproductive strategy. Secondary sexual characteristics related to sterna are described for males of the genus *Callonychium* (Calliopsini). In which the males present projections in the sterna that contribute to the fixation of the female terga during copulation, in a process of mechanical adjustment (Toro 1985). The pilosity pattern present in the sterna of Tapinotaspidini males does not appear to be related to coupling. However, it is possible that the diversity of fringes sternal (*Paratetrapedia*), the foliaceous (*Monoeca*) and the long hairs branched in the distal portion (*Arhyscoceble*) allow the storage of oil, which can have the function of attracting receptive females. The behavior of males collecting floral resources for this purpose is well documented for the Euglossini tribe. The volatile odoriferous compounds (also referred like terpens, sesquiterpens and aromatics) collected by males, in orchids flowers, are stored in specialized structures located in the hind tibia (see Dressler 1982; Williams & Whithen 1983; Withen et al. 1993; Kaiser 1993; Eltz et al. 1999; Williams & Whitten, 1999). And, later, pulverized in the air, in mating places (e.g., trunks fallen in the undergrowth), as a strategy to attract females (Dodson et al. 1969; Kimsey 1980; Eltz et al. 2003, Bembé 2004; Eltz et al. 2005).

The collection of fragances in Tapinotaspidini has been described for the males of *Paratetrapedia chocoensis* that collect volatile compounds of *Anthurium acutifolium* (Araceae) (Etl et al. 2017). The collection of the fragances in *A. acutifolium* occurs by scraping the inflorescences, with the a brush of hair in sternum 3, similar to that present in the males of *Paratetrapedia fervida* and *Paratetrapedia. flaveola*. The function of the volatile compounds collected by the males of P. chocoensis is also unknown. However, it is expected that it may have some relation with the demarcation of territory or copulation behavior. In some males of *P. flaveola* and *P. fervida* I observed signs of a type of viscous substance in S3. However, we have no way to affirm that these males collected oil from the abdominal scopa.

The tergal scopa of some males analyzed (especially *Caenonomada*, *Tropidopedia*, *Urbanapis*) showed high amounts of oil. Considering that floral oil is a non-volatile compound, and possibly a slow evaporation rate, it is possible that its storage in the tergal scopa is related to female attraction (Buchman 1987; Cappellari et al. 2012). The oil present in the tergal scopa of Tapinotaspidini males may also represent a strategy to facilitate access to females visiting oil flowers. High amounts of oil in the terga (and possibly sterna) could function as a type of chemical camouflage, which would allow males, possibly imperceptible, to attempt copulation with virgin females (or already mating) that are collecting oil or pollen (Cappellari et al. 2012). The behavior of males of different species of *Macropis* oil collector bees (Macropidini) attempting to copulate with flowers has already been described (Rozen & Jacobson 1980; Cane et al. 1983). Tapinotaspidini males patrolling oil flowers in search of females is known for *Arhysoceble* and *Lanthanomelissa* and for Tapinotaspidini (Cocucci 1991). Males using floral resources, possibly as an attempt to access reproductive females has already been described for Meliponini. Boongird &

Michener (2010) described males of *Tetragonula* and *Heterotrigona* entering the nests of their respective species, with pollen or propolis in the hind tibia. Considering that males without resources were prevented from entering the nests, is expected that collecting pollen and propolis is a method to try to deceive workers and have access to virgin queens.

Another possibility would be to use the oil as a nuptial gift for the females. Studies focusing on the use of nuptial gifts in insects are commonly described for Diptera, Lepidoptera and Orthoptera (Boggs 1995; Gwynne 2008). Schindler et al. (2018) suggest that *Nomada* males transfer liquid pheromones, during copulation, through antennal contact with females. For the authors, the function of the substance may be to reduce the attractiveness of the females or to facilitate their access to the nests of *Andrena* (Andrenidae). The transference of chemical substances in *Nomada* had already been described. The females of *Nomada* are cleptoparasites, and oviposites in the nests constructed by the females of *Andrena*. Tengö & Bergström (1977), discovered that males of *Nomada* males produce identical mandibular secretions, those produced in the Dufour glands of female of *Andrena*. Thus, they suggested that the substances produced and released on the females, could act as a kind of chemical camouflage that allows access to the nests of *Andrena*. However, there are no records in bees that describe the transference of substances with the function of nuptial gift (Danforth et al. 2019). Evidence of this behavior would be unique and, consequently, extremely relevant for a greater understanding of the reproductive strategies of male solitary bees.

5. Conclusion

We present relevant contributions on the morphology of Tapinotaspidini males. Our results evidence the collection of oil occurs through specialized structures in the legs and sterna of males. Several of these structures have evolved by convergence and, as with females, are directly related to the types of elaiophores exploited. In the Tapinotaspidini male sterna and terga have a high diversity of exclusive structures are present. We suggest that these structures are associated with the storage of the oil, which is possibly associated with the mating behavior. However, clearly, other studies with emphasis on Tapinotaspidini's mating morphology and behavior are necessary for further elucidation of these open questions.

References

Aguiar, A.J.C. 2012. Tapinotaspidini Roig-Alsina & Michener, 1993. In: Moure, J.S., Urban, D. & Melo, G.A.R. (Eds), Catalogue of Bees (Hymenoptera, Apoidea) in the Neotropical Region - online version. Available at: http://www.moure.cria.org.br/catalogue

Aguiar, A.J.C., Melo, G.A.R. 2007 Taxonomic revision, phylogenetic analysis, and biogeography of the bee genus *Tropidopedia* (Hymenoptera, Apidae, Tapinotaspidini). Zool. J. Linn. Soc. 151, 511-554.

Aguiar, A.J.C., Melo, G.A.R. 2009. Notes on oil sources for the bee genus *Caenonomada* (Hymenoptera, Apidae, Tapinotaspidini). Rev. Bras. Entomol. 53(1), 154-156.

Aguiar, A.J.C., Melo, G.A.R. 2011. Revision and phylogeny of the bee genus *Paratetrapedia* Moure, with description of a new genus from the Andean Cordillera (Hymenoptera, Apidae, Tapinotaspidini). Zool. J. Linn. Soc. 162(2), 351-442. https://doi.org/10.1111/j.1096-3642.2010.00678.x

Aguiar, A.J.C., Melo, G.A.R., Vasconcelos, T.N.C., Gonçalves, R.B., Giuliano, L., Martins, A. C. 2020. Biogeography and early diversification of Tapinotaspidini oil-bees support presence of Paleocene savannas in South America. Mol. Phylogenetics Evol. 143, 106692. https://doi.org/10.1016/j.ympev.2019.106692

Bembé, B. 2004. Functional morphology in male euglossine bees and their ability to spray fragrances (Hymenoptera, Apidae, Euglossini). Apidologie. 35, 283-291.

Boggs, C.L. 1995. Male nuptial gifts: phenotypic consequences and evolutionary implications. In: Leather, S. R., Hardie, J. (Eds), Insect Reproduction, CRC Press, New York, pp. 215-242.

Boongird, S., Michener, C.D. 2010. Pollen and propolis collecting by male stingless bees (Hymenoptera: Apidae). J. Kans. Entomol. Soc. 83, 47-50.

Buchmann, S. 1987. The ecology of oil flowers and their bees. Ann. Rev. Ecol. Syst. 18, 343-369.

Cane, J.H., Eickwort, G.C., Wesley, F.R., Spielholz, J. 1983. Foraging, grooming and mating behaviours of *Macropis nuda* (Hymenoptera, Melittidae) and use of *Lysimachia ciliata* (Primulaceae) oils in larval provisions and cell linings. American Naturalist. 110, 257-264.

Cappellari, S.C., Melo, G.A.R., Aguiar, A.J.C., Neff, J.L. 2012. Floral oil collection by male *Tetrapedia* bees (Hymenoptera: Apidae: Tetrapediini). Apidologie. 43, 39-50. https://doi.org/10.1007/s13592-011-0072-2

Cocucci, A.A., 1991. Pollination biology of *Nierembergia* (Solanaceae). Plant. Syst. Evol. 174, 17-35.

Cocucci, A.A., Vogel, S. 2001. Oil-producing flowers of *Sisyrinchium* species (Iridaceae) and their pollinators in southern South America. Flora. 196(1), 26-46.

Cocucci, A.A., Sérsic, A., Roig-Alsina, A. 2000. Oil-collecting structures in Tapinotaspidini: their diversity, function and probable origin. Mitt. Münch. Entomol. Ges. 90:51-74.

Cunha, R., Blochtein, B. 2003. Life history of *Monoeca xanthopyga* Harter-Marques, Cunha & Moure (Hymenoptera, Apidae, Tapinotaspidini) at the Araucária Plateau, Rio Grande do Sul, Brazil. Rev. Bras. Zool. 1,107-113.

Danforth, B., Minckley, R., Neff, J., Fawcett, F. 2019. The Solitary Bees: Biology, Evolution, Conservation. Princeton University Press.

De Queiroz A. & Wimberger P.H. 1993. The usefulness of behavior for phylogeny estimation: levels of homoplasy in behavioral and morphological characters. Evolution. 47, 46-60.

Dodson, C.H, Dressler, R.L, Hills H.G., Adams, R.M.; Williams, N.H. 1969 Biologically active compounds in orchid fragrances. Science, 164, 1243-1249. https://doi.org/10.1126/science.164.3885.1243

Dressler R.L. 1982. Biology of the orchid bees (Euglossini). Ann. Rev. Ecol. Syst. 13, 373-394.

Etl, F., Franschitz, A., Aguiar, A.J.C., Schönenberger, J., Dötterl, S. 2017. A perfume-collecting male oil bee? Evidences of a novel pollination system involving *Anthurium acutifolium* (Araceae) and *Paratetrapedia chocoensis* (Apidae, Tapinotaspidini). Flora. 232, 7-15.

Eltz, T., Bause, C., Hund, K., Quezada-Euan, J.J.G., Pokorny, T. 2015. Correlates of perfume load in male orchid bees. Chemoecology. 25, 193-199. https://doi.org/10.1007/s00049-015-0190-9

Eltz, T., Josten, S., Mende, T. 2019. Stored perfume dynamics and consequences for signal development in male orchid bees. J. Comp. Physiol. A. 205, 311-320.

Eltz, T., Roubik, D.W., Whitten, M.W. 2003. Fragrances, male display and mating behaviour of *Euglossa hemichlora*: a flight cage experiment. Physiol. Entomol. 28, 251-260.

Eltz, T., Sager, A., Lunau, K. 2005. Juggling with volatiles: exposure of perfumes by displaying male orchid bees. J. Comp. Physiol. A Sens. Neural. Behav. Physiol. 191, 575-581.

Eltz, t., Whitten, W. M., Roubik, D.W., Linsenmair, K.E. 1999. Fragrance collection, storage, and accumulation by individual male orchid bees. J. Chem. Ecol. 25:157-176.

Friese, H. 1925. Neue neotropische Bienenarten, zugleich II. Nachtrag zur Bienenfauna von Costa Rica. (Hym.). Stett. Entomol. Ztg. 86(2), 1-41.

Gastauer, M., Campo, L.A.O.; Wittmann, D. Handling sticky resin by stingless bees (Hymenoptera, Apidae). Rev. Bras. Entomol. 55(2), 234-240. http://dx.doi.org/10.1590/S0085-56262011005000018.

Gwynne, D.T. 2008. Sexual conflict over nuptial gifts in insects. Ann. Rev. Entomol. 53, 83-101. https://doi.org/10.1186/1471-2148-8-204

Hetherington-Rauth, M.C., Ramırez, S.R. 2016. Evolution and diversity of floral scent chemistry in the euglossine bee-pollinated orchid genus *Gongora*. Ann. Bot. 118(1), 135-148.

Kaiser, R. 1993. The scent of orchids. Basel. Editiones Roche, 259 pp.

Kimsey, L.S. 1980. The behavior of male orchid bees (Apidae, Hymenoptera, Insecta) and the question of leks. Anim. Behav. 28, 996-1004.

Kuhlmann, M. 2014. Nest architecture and use of floral oil in the oil-collecting South African solitary bee *Rediviva intermixta* (Cockerell) (Hymenoptera: Apoidea: Melittidae). Nat. Hist. 48, 2633-2644.

Kuhlmann, M., Hollens, H. 2014. Morphology of oil-collecting pilosity of female *Rediviva* bees (Hymenoptera: Apoidea: Melittidae) reflects host plant use. J. Nat. Hist. 49, 561-657.

Leijs, R., Hogendoorn, K. 2008. Correlated evolution of mating behaviour and morphology in large carpenter bees (*Xylocopa*). Apidologie. 39(1), 119-132.

Maddison, W.P., Maddison, D.R. 2019. Mesquite: a modular system for evolutionary analysis. Version 3.61. Available at: http://www.mesquiteproject.org

Melo, G.A.R., Gaglianone, M.C. 2005. Females of *Tapinotaspoides*, a genus in the oil-collecting bee tribe Tapinotaspidini, collect secretions from non-floral trichomes (Hymenoptera, Apidae). Rev. Bras. Entomol. 49(1),167-168.

Michener, C.D. 2007. The bees of the world. The Johns Hopkins University Press, Baltimore.

Neff, J.L., Simpson, B.B. 1981. Oil-collecting structures in the Anthophoridae: Morphology, function and use in systematics. J. Kans. Entomol. Soc. 54, 95-123.

Neff, J.L., Simpson, B.B. 2017. Vogel's great legacy: The oil flower and oil-collecting bee syndrome. Flora. 232, 104-116. https://doi.org/10.1016/j.flora.2017.01.003

Owen, R. 1848. On the Archetype and Homologies of the Vertebrate Skeleton. John Van Voorst, London. https://doi.org/10.5962/bhl.title.118611

Rasmussen, C., Olesen, J.M. 2000. Oil flowers and oil-collecting bees. Norske Vidensk. Akad. I. Mat. Nat. Kl. Skrifter. 39, 23-31.

Roberts, R.B., Vallespir, S.R. 1978. Specialization of hairs bearing pollen and oil on the legs of bees (Apoidea: Hymenoptera). Ann. Entomol. Soc. Am. 71, 619-627. https://doi.org/10.1093/aesa/71.4.619

Roig-Alsina, A. 1997. A generic study of the bees of the tribe Tapinotaspidini, with notes on the evolution of their oil-collecting structures. Mitt. Münch. Entomol. Ges. 87, 3-21.

Roig-Alsina, A. 1999. Revision of the oil-collecting bees of the genus *Chalepogenus* Holmberg (Hymenoptera, Apidae, Tapinotaspidini). Rev. Mus. Argentino Cienc. Nat. 1(1), 67-101.

Roig-Alsina, A. 2003. A revision of the bee genus *Tapinotaspis* Holmberg (Hymenoptera, Apidae, Tapinotaspidini). Mitt. Münch. Entomol. Ges. 93, 45-53.

Roig-Alsina, A., Michener, C.D. 1993. Studies of the phylogeny and classification of long-tongued bees (Hymenoptera: Apoidea). Univ. Kansas Sci. Bull. 55, 123-173.

Roth, V.L. 1984. On homology. Biol. J. Linn. Soc. 22(1), 13-29. https://doi.org/10.1111/j.1095-8312.1984.tb00796.x

Rozen, J.G., Jr., Jacobson, N.R. 1980. Biology and immature stages of *Macropis nuda*, including comparisons to related bees (Apoidea, Melittidae). Am. Mus. Nov. 2702, 1-11.

Sakagami, S.F., J.F.M. Camargo. 1964. Cerumen collection accompanied by thieving and attacking in a stingless bee, *Nannotrigona* (*Scaptotrigona*) *postica* (Latreille), with a consideration on territoriality in social insects. Rev. Biol. Trop. 12, 197-207.

Schindler, M., Hofmann, M.M., Wittmann, D., Renner, S.S. 2018. Courtship behaviour in the genus *Nomada* – antennal grabbing and possible transfer of male secretions. J. Hymenopt. Res. 65,47-59. https://doi.org/10.3897/jhr.65.24947

Sereno, P.C. 2007. Logical basis for morphological characters in phylogenetics. Cladistics. 23, 565-587. https://doi.org/10.1111/j.1096-0031.2007.00161.x

Silveira, F.A., Melo, G.A.R., Almeida, E.A.B. 2002. Abelhas brasileiras: sistemática e identificação. Belo Horizonte.

Smith, G.P., Bronstein, J.L., Papaj, D.R. 2019. Sex differences in pollinator behavior: Patterns across species and consequences for the mutualism. J. Anim. Ecol. 88(7), 971-985.

Stubblefield, J., Seger, J. 1994. Sexual dimorphism in the Hymenoptera. In: Short, R. V., Balaban, E. (Eds). The differences between the sexes. Cambridge University Press.

Tengö, J., Bergström, G. 1977. Cleptoparasitism and odor mimetism in bees: Do *Nomada* males imitate the odor of *Andrena* females? Science. 196, 1117–1119.

Thornhill, R., Alcock, J. 1983. The evolution of insect mating systems. Harvard University Press, London.

Toro, H. 1985. Ajuste mecanico para la copula de *Callonychium chilense* (Hymenoptera, Andrenidae). Rev. Chil. Entomol. 12, 153-158.

Vogel, S. 1969. Flowers offering fatty oil instead of nectar. In: Abstracts of the Papers Presented at the XI International Botanical Congress.

Vogel, S. 1974. Ölblumen und ölsammelnde Bienen. Tropische und Subtropische Pflanzenwelt. Mainz. 7, 285-547.

Vogel, S. 1981. Abdominal oil-mopping—a new type of foraging in bees. Naturwissenschaften. 67,627.

Wenzel, J. 1992. Behavioral homology and phylogeny. Annu. Rev. Ecol. Syst. 23, 361-381.

Wcislo, W.T., Buchmann, S.L. 1995. Mating behaviour in the bees, *Dieunomia heteropoda* and *Nomiatetrazonata*, with a review of courtship in Nomiinae (Hymenoptera: Halictidae), J. Nat. Hist. 29(4), 1015-1027, https://doi.org/10.1080/00222939500770391

Whitten, W.M., Young, A.M., Stern, D.L. 1993. Nonfloral sources of chemicals that attract male euglossine bees (Apidae: Euglossini). J. Chem. Ecol., 19 (12), 3017-3027.

Wiley, E.O., Lieberman, B.S. 2011. Phylogenetics: The Theory and Practice of Phylogenetic Systematics. Wiley-Blackwell, Hoboken, New Jersey.

Williams, N.H., Whitten, W.M. 1983. Orchid floral fragrances and male euglossine bees: methods and advances in last sesquidecade. Biol. Bull., 164, 355-395.

Williams, N.H., Whitten, W.M. 1999. Molecular phylogeny and floral fragrances of male euglossine bee-pollinated orchids: A study of *Stanhopea* (Orchidaceae). Plant Species Biol., 14, 129-136.

Wittmann, D., Blochtein, B. 1995. Why males of leafcutter bees hold the females' antennae with their front legs during mating. Apidologie. 26(3), 181-196. https://doi.org/10.1051/apido:19950302

Wittmann, D., Schindler, M., Blochtein, B., Bahrouz, D. 2004. Mating in bees. How males hug their mates. In: Proceedings of the 8th IBRA International Conference of Tropical Bees, and VI Encontro sobre Abelhas. 374-380.

Table 1. Tapinotaspidini bee species studied, in parentheses number of species estimated according to the Moure's Bee Catalogue (Aguiar, 2012) and complementary literature.

Genera	Species
Arhysoceble (5)	*Arhysoceble huberi* (Ducke, 1908) *Arhysoceble picta* (Friese, 1899) *Arhysoceble* sp. Moure, 1948
Caenonomada (3)	*Caenonomada unicalcarata* (Ducke, 1908)
Chalepogenus (21)	*Chalepogenus caeruleus* (Friese, 1906) *Chalepogenus muelleri* (Friese, 1899) *Chalepogenus nigripes* Holmberg, 1903
Lanthanomelissa (5)	*Lanthanomelissa betinae* Urban, 1995 *Lanthanomelissa clementis* Urban, 1995 *Lanthanomelissa parva*
Lophopedia (13)	*Lophopedia flava* (Smith, 1879) *Lophopedia nigrispinis* (Vachal, 1909) *Lophopedia pygmaea* (Schrottky, 1902)
Monoeca (11)	*Monoeca campestris* Aguiar, 2012 *Monoeca haemorrhoidalis* (Smith, 1854) *Monoeca mourei* Aguiar, 2012 *Monoeca pluricincta* (Vachal, 1909) *Monoeca xanthopyga* Harter-Marques, Cunha & Moure, 2001
Paratetrapedia (32)	*Paratetrapedia connexa* (Vachal, 1909) *Paratetrapedia duckei* (Friese, 1910) *Paratetrapedia fervida* (Smith, 1879) *Paratetrapedia flaveola* Aguiar & Melo, 2011 *Paratetrapedia lugubris* (Cresson, 1878) *Paratetrapedia punctata* Aguiar & Melo, 2011 *Paratetrapedia testacea* (Smith, 1854) *Paratetrapedia volatilis* (Smith, 1879)
Tapinotaspis (4)	*Tapinotaspis chalybaea* (Friese, 1899)
Tapinotaspoides (6)	*Tapinotaspoides serraticornis* (Friese, 1899) *Tapinotaspoides tucumana* (Vachal, 1904)
Trigonopedia (6)	*Trigonopedia ferruginea* (Friese, 1899)
Tropidopedia (17)	*Tropidopedia carinata* Aguiar & Melo, 2007 *Tropidopedia flavolineata* Aguiar & Melo, 2007 *Tropidopedia nigrita* Aguiar & Melo, 2007 *Tropidopedia punctifrons* (Smith, 1879)
Urbanapis (4)	*Urbanapis danunciae* Aguiar & Melo, 2015

	Urbanapis diamantina Aguiar & Melo, 2015
Xanthopedia (8)	*Xanthopedia* aff. *globulosa* (Friese, 1899) *Xanthopedia* aff. *larocai* Moure, 1995

Table 2. List of structures and their respective Tapinotaspidini species available in MorphoBanK through the project "P3567: Structures of oil collecting bee males of the tribe Tapinotaspidini and Tetrapediini (Hymenoptera: Apidae)".

Species	**Structures**
Arhysoceble sp. Moure, 1948	Legs (fore) Metasoma (terga and sterna)
Caenonomada unicalcarata (Ducke, 1908)	Legs (fore, middle and hind) Metasoma (terga and sterna)
Lanthanomelissa betinae Urban, 1995	Legs (fore, middle and hind) Metasoma (terga and sterna)
Lophopedia pygmaea (Schrottky, 1902)	Metasoma (sterna)
Monoeca xanthopyga Harter-Marques, Cunha & Moure, 2001	Legs (fore, middle and hind) Metasoma (terga and sterna)
Paratetrapedia connexa (Vachal, 1909)	Legs (hind) Metasoma (sterna)
Paratetrapedia fervida (Smith, 1879)	Metasoma (sterna)
Paratetrapedia flaveola Aguiar & Melo, 2011	Metasoma (sterna)
Paratetrapedia punctata Aguiar & Melo, 2011	Legs (fore) Metasoma (sterna)
Paratetrapedia testacea (Smith, 1854)	Legs (fore, middle and hind) Metasoma (terga)
Tapinotaspoides serraticornis (Friese, 1899)	Legs (middle and hind) Metasoma (terga and sterna)
Tropidopedia carinata Aguiar & Melo, 2007	Legs (fore)
Tropidopedia flavolineata Aguiar & Melo, 2007	Legs (fore and hind)
Tropidopedia punctifrons (Smith, 1879)	Legs (fore and hind) Metasoma (terga and sterna)

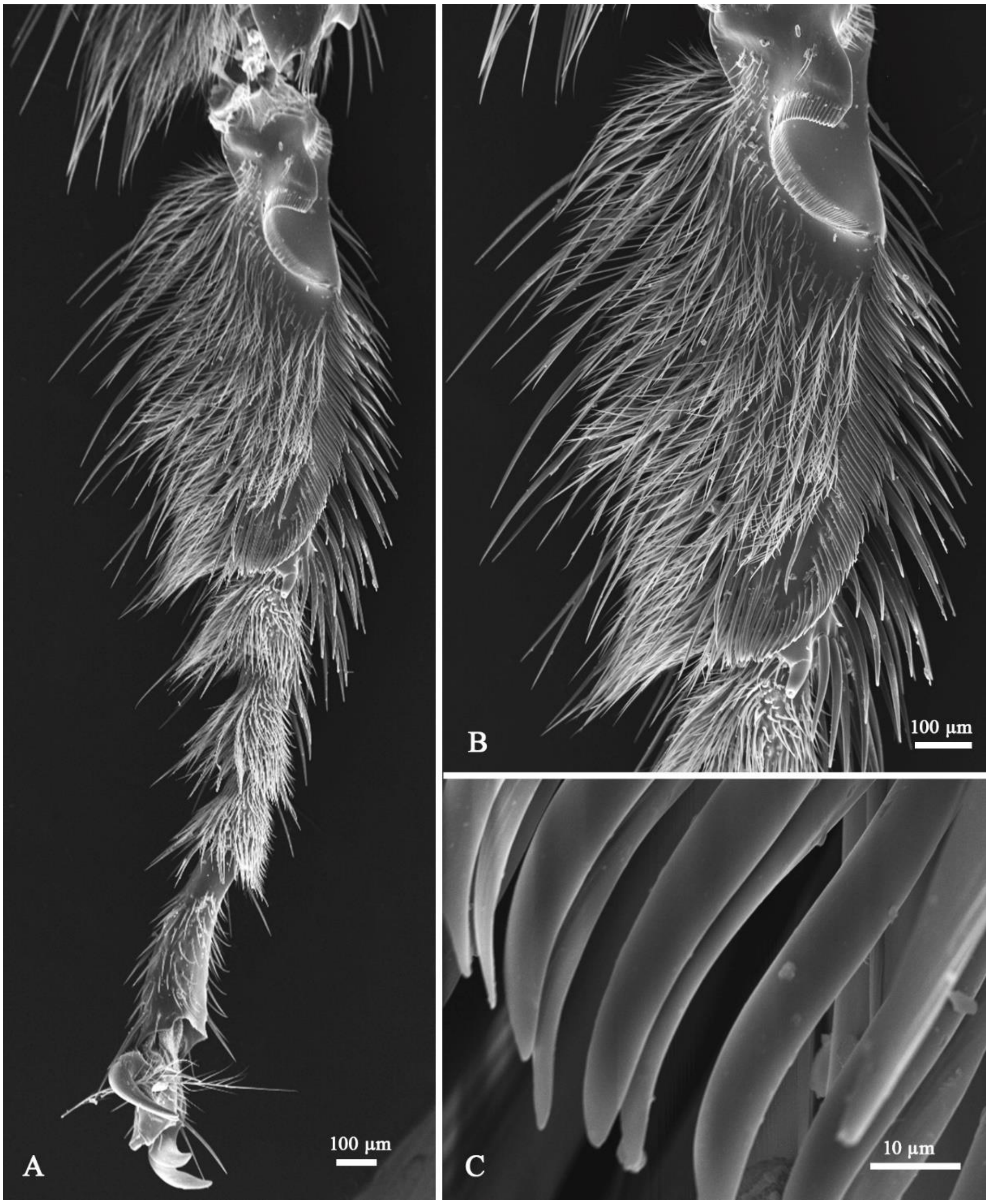


Figura 1. *Monoeca xanthopyga* Harter-Marques, Cunha & Moure, 2001 male. **A.** Ventral surface of the fore leg. **B.** Setae comb on the inner margin of fore basitarsus. **C.** Detail of the setae comb on the inner margin of the fore basitarsus.

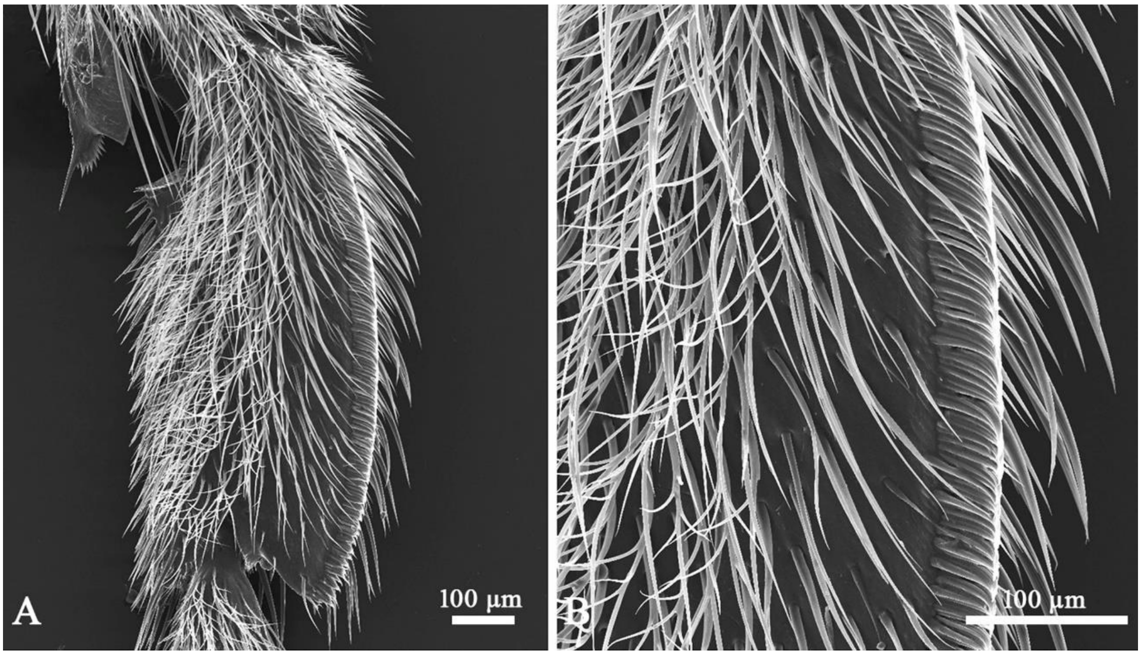


Figure 2. *Tropidopedia punctifrons* male. **A.** Ventral surface of the fore basitarsus. **B.** Detail of the setae comb on the outer margin of the fore basitarsus.

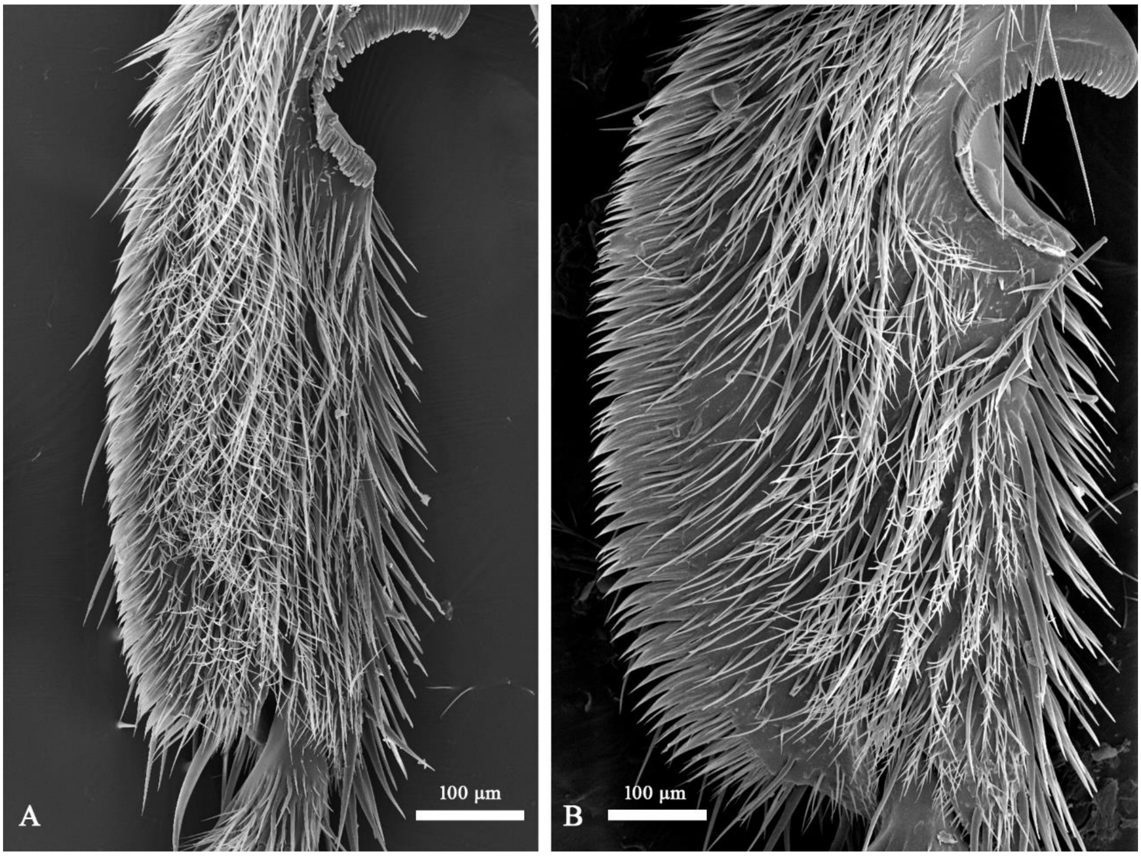


Figure 3. Plumose hairs on the ventral surface of fore basitarsus. **A.** Dense fine and plumose hairs in *Arhysoceble huberi* (Ducke, 1908) male. **B.** Long and sparse plumose hairs in *Tropidopedia carinata* Aguiar & Melo, 2007 male.

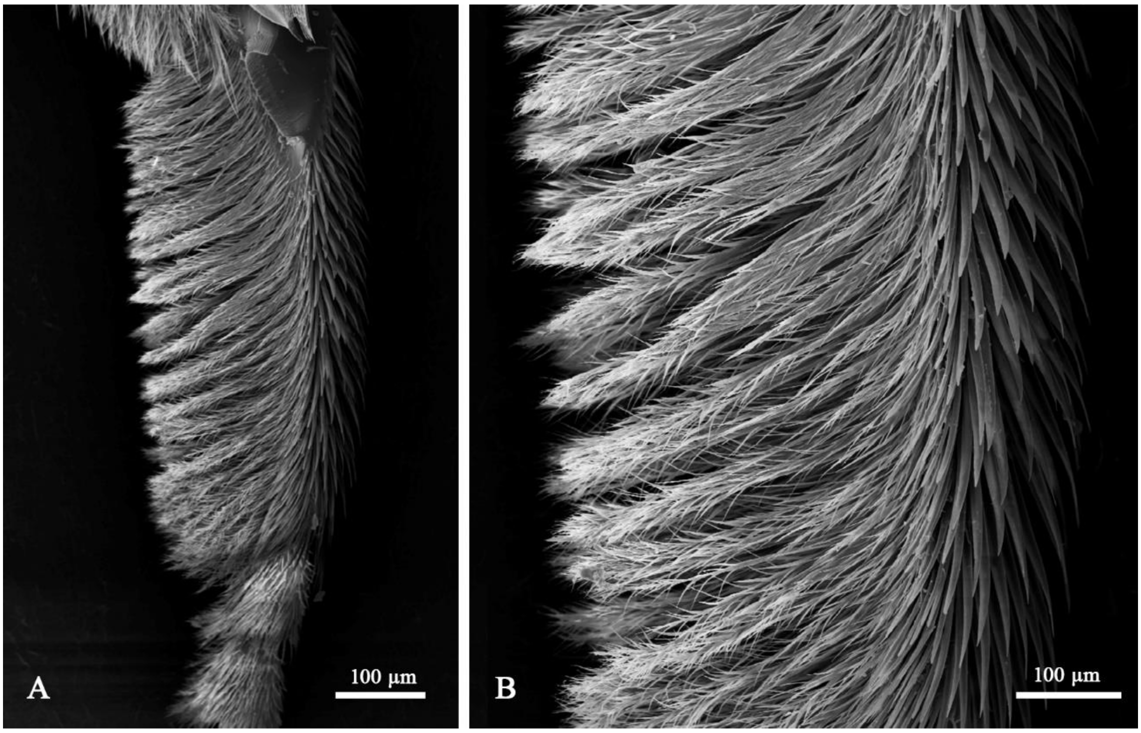


Figure 4. Plumose hairs on the ventral surface of fore basitarsus of *Tropidopedia arcuatilis* (Vachal, 1909) male. **A.** Ventral surface of the fore basitarsus. **B.** Detail of very long and plumose hairs.

Figure 5. *Caenonomada unicalcarata* (Ducke, 1908) male. **A.** Dorsal surface of tarsomeres of the fore leg. **B.** Detail of the plumose hairs and stout setae on the margin of the fore leg.

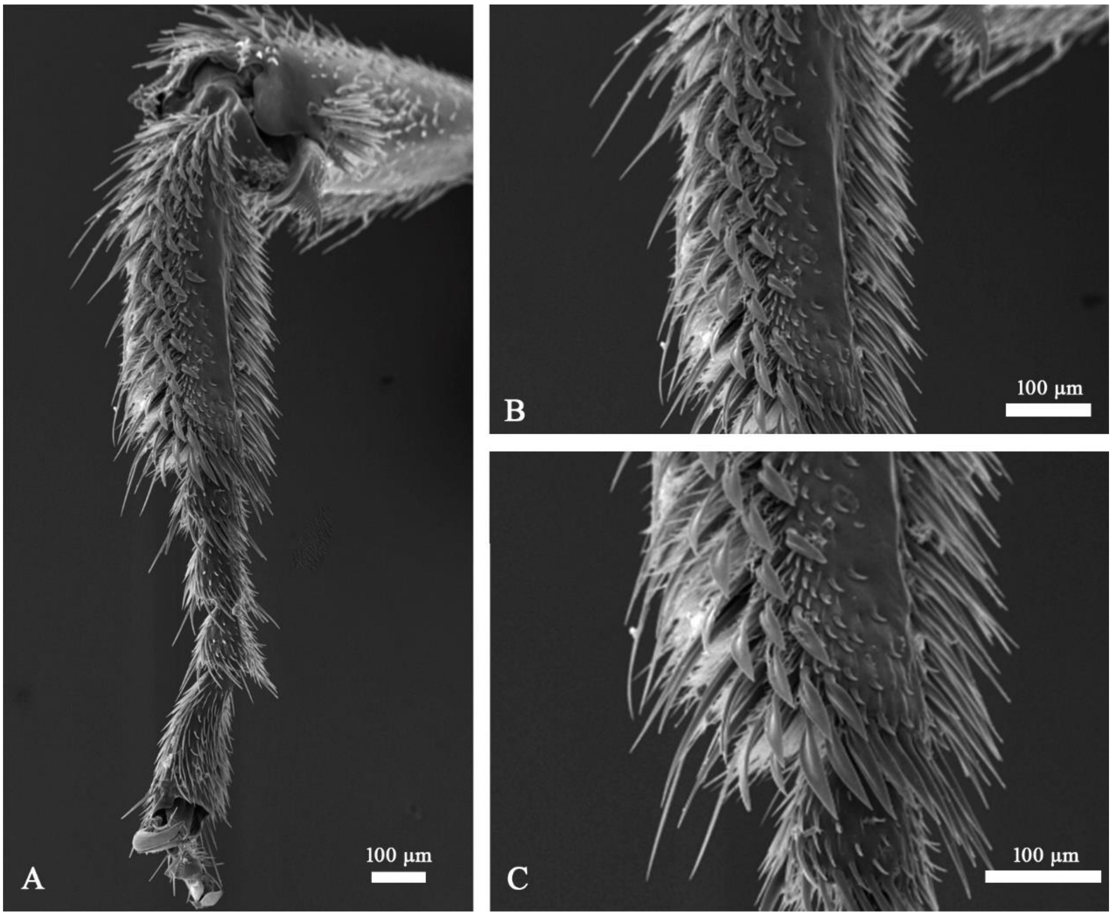


Figure 6. *Lanthanomelissa betinae* Urban, 1995 male. **A.** Ventral surface of the fore leg. **B.** Setae foliaceus located in the posterior margin of the fore basitarsus. **C.** Detail of the setae foliaceous in the posterior margin of the fore basitarsus.

Figure 7. Dorsal surface of the hind leg of *Monoeca xanthopyga* male. **A.** Basitarsal scopa composed by plumose and simple hairs. **B.** Details of the plumose hairs on the basitarsal scopa.

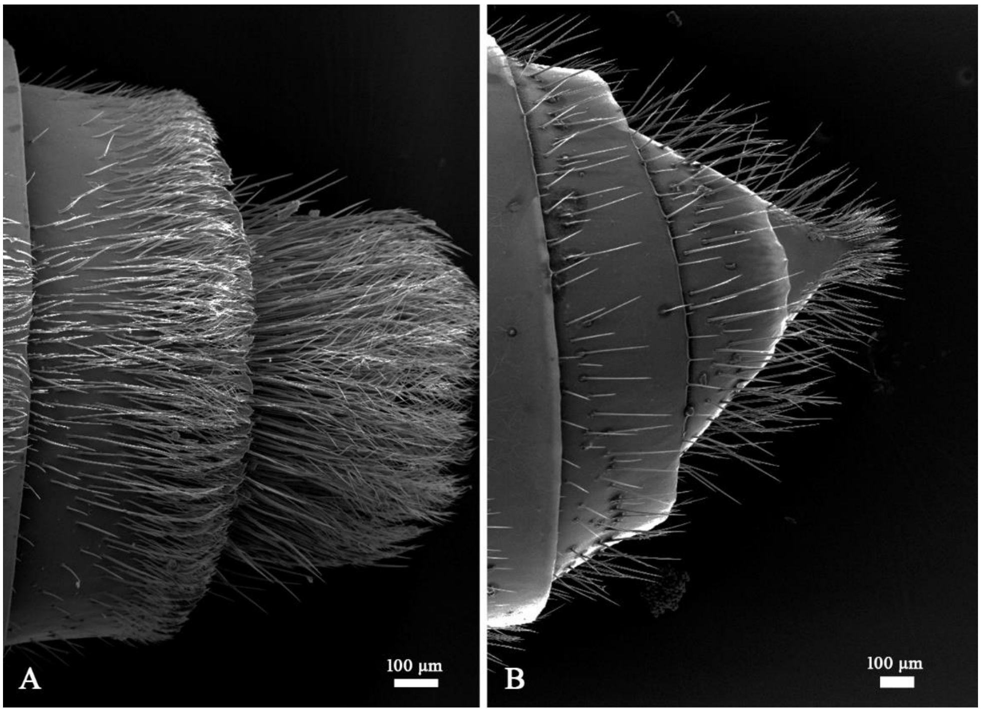


Figure 8. Plumose hairs on posterior margin of the terga. **A.** Plumose hairs on the posterior margin of T5-T6 of *Tropidopedia flavolineata* Aguiar & Melo, 2007 male. **B.** Posterior margin of T5-T6 of a *Paratetrapedia testacea* (Smith, 1854) male with absence of plumose hairs.

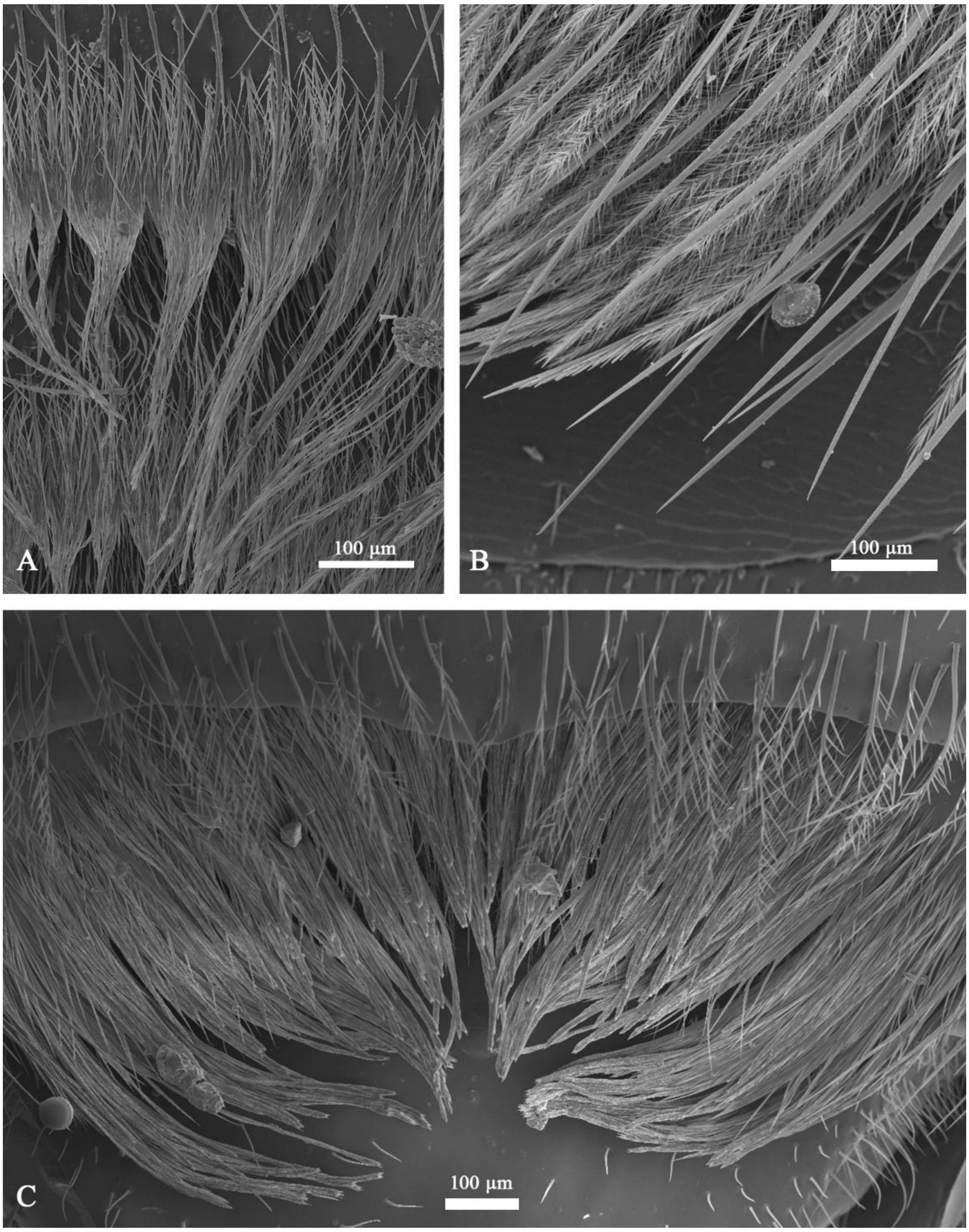


Figure 9. Plumose hairs on posterior margin of the sternum. **A.** Elongated and convergent fringe in the posterior margin of the sternum (S3) of *Arhysoceble* sp. male. **B.** Dense band of plumose and simple long hairs on lateral portions of the posterior margin of the sternum (S2) of *Monoeca xanthopyga* male. **C.** Elongated and convergent fringe in the posterior margin of the sternum (S5) of *Caenonomada unicalcarata* male.

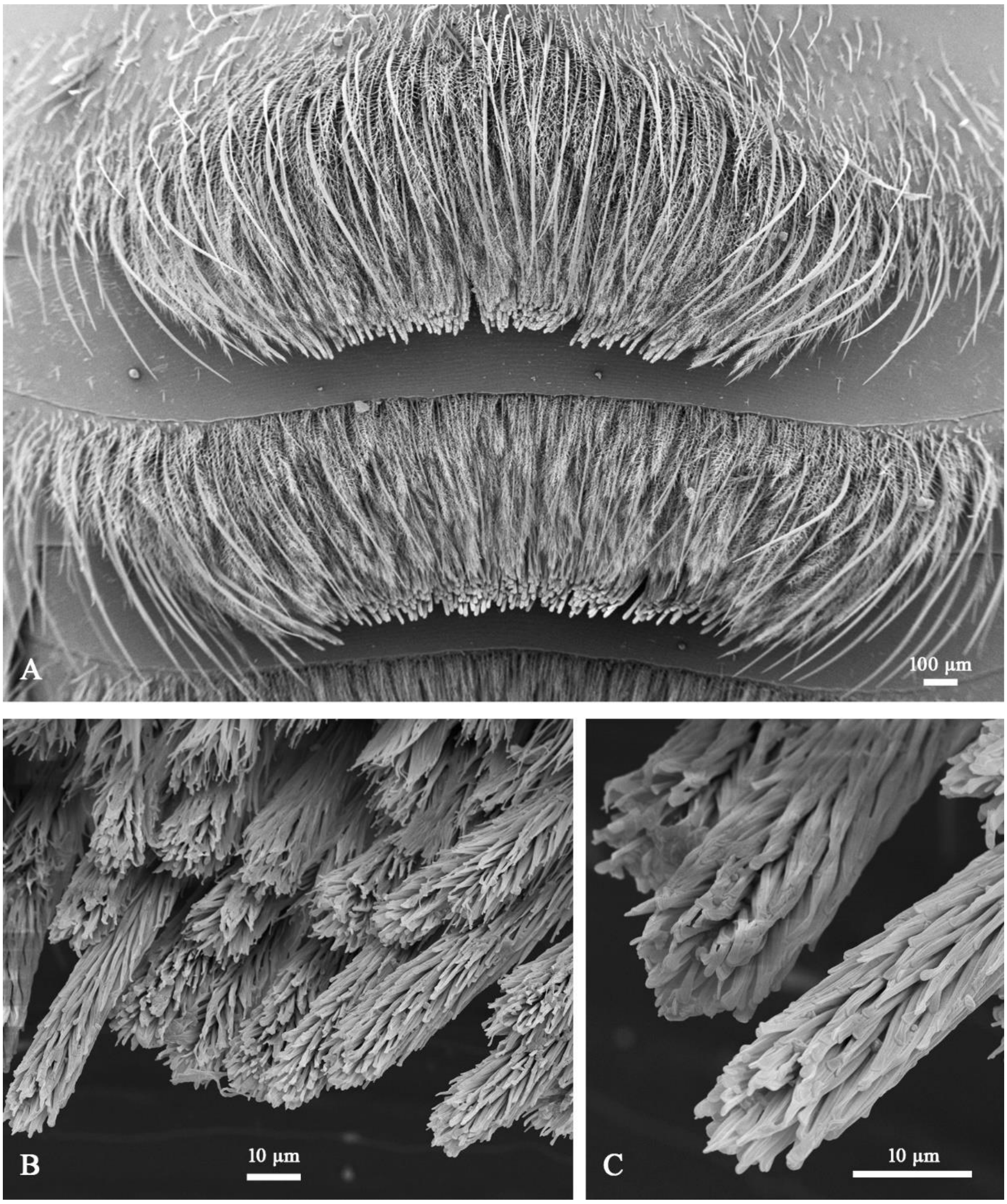


Figure 10. Sterna of *Monoeca xanthopyga* male. **A.** Dense band composed by plumose, foliaceus and simple hairs in the sterna 2-3. **B.** Plumose foliaceous hairs in the sterna 2-3. **C.** Detail of foliaceous hairs.

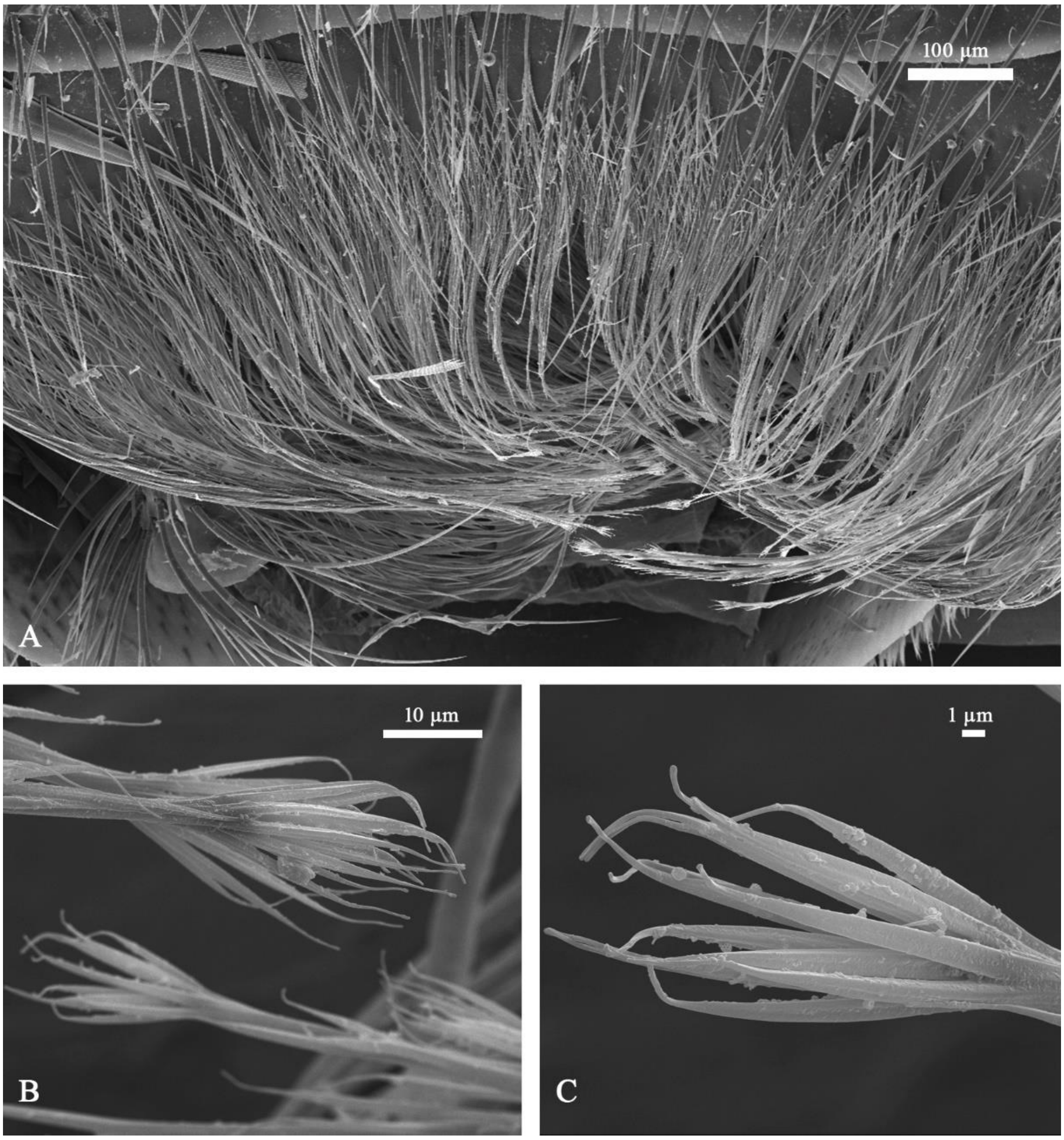


Figure 11. Sternum of *Arhysoceble sp.* male. **A.** Dense band of long and plumose hairs of the sternum (S3). **B.** Long hairs branched in the distal portion of the sternum (S3). **C.** Detail of long and plumose hairs.

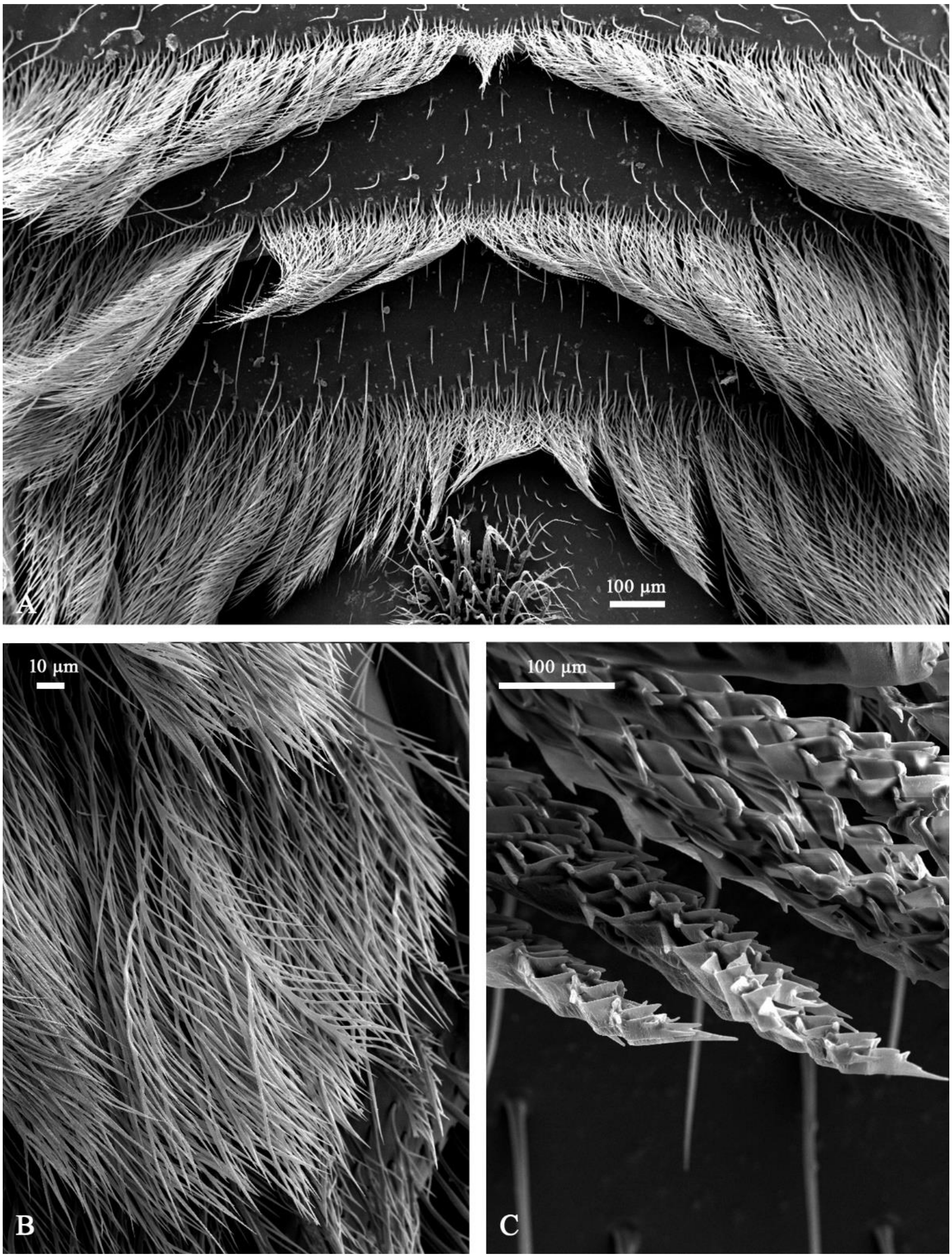


Figure 12. Sterna of *Tapinotaspoides serraticornis* (Friese, 1899). **A.** Plumose hairs on posterior margin of the sterna (S3-S5) of *T. serraticornis* male. **B.** Detail of plumose hairs of *T. serraticornis* male. **C.** Detail of specialized fringes of long wavy setae along the posterior margin of sternum (S2) of *T. serraticornis* female.

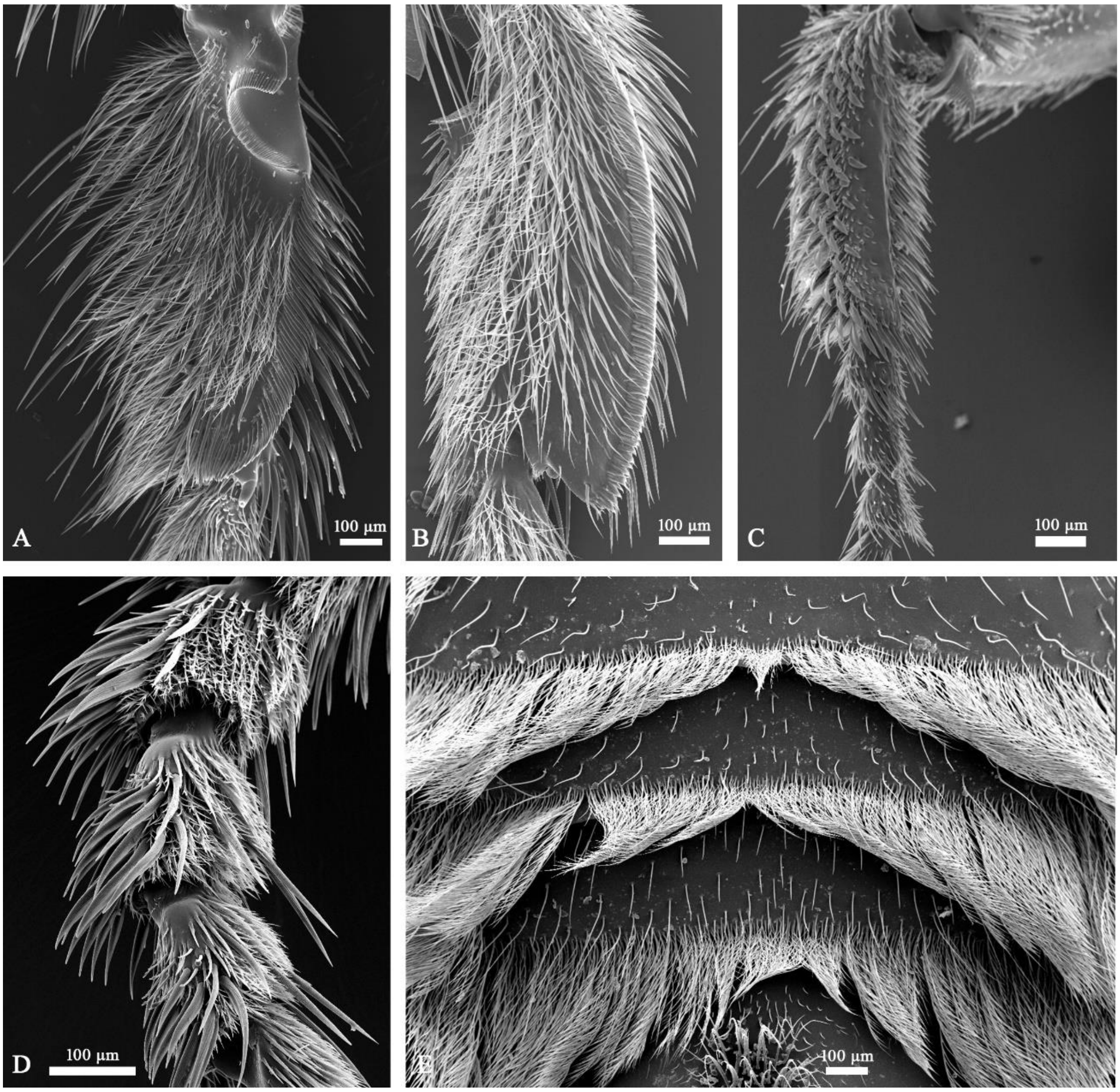


Figure 13. Adaptations for the collection of floral oil in Tapinotaspidini males. **A.** *Monoeca xanthopyga*. **B.** *Tropidopedia punctifrons*. **C.** *Lanthanomelissa betinae*. **D.** *Caenonomada unicalcarata*. **E.** *Tapinotaspoides serraticonis*.

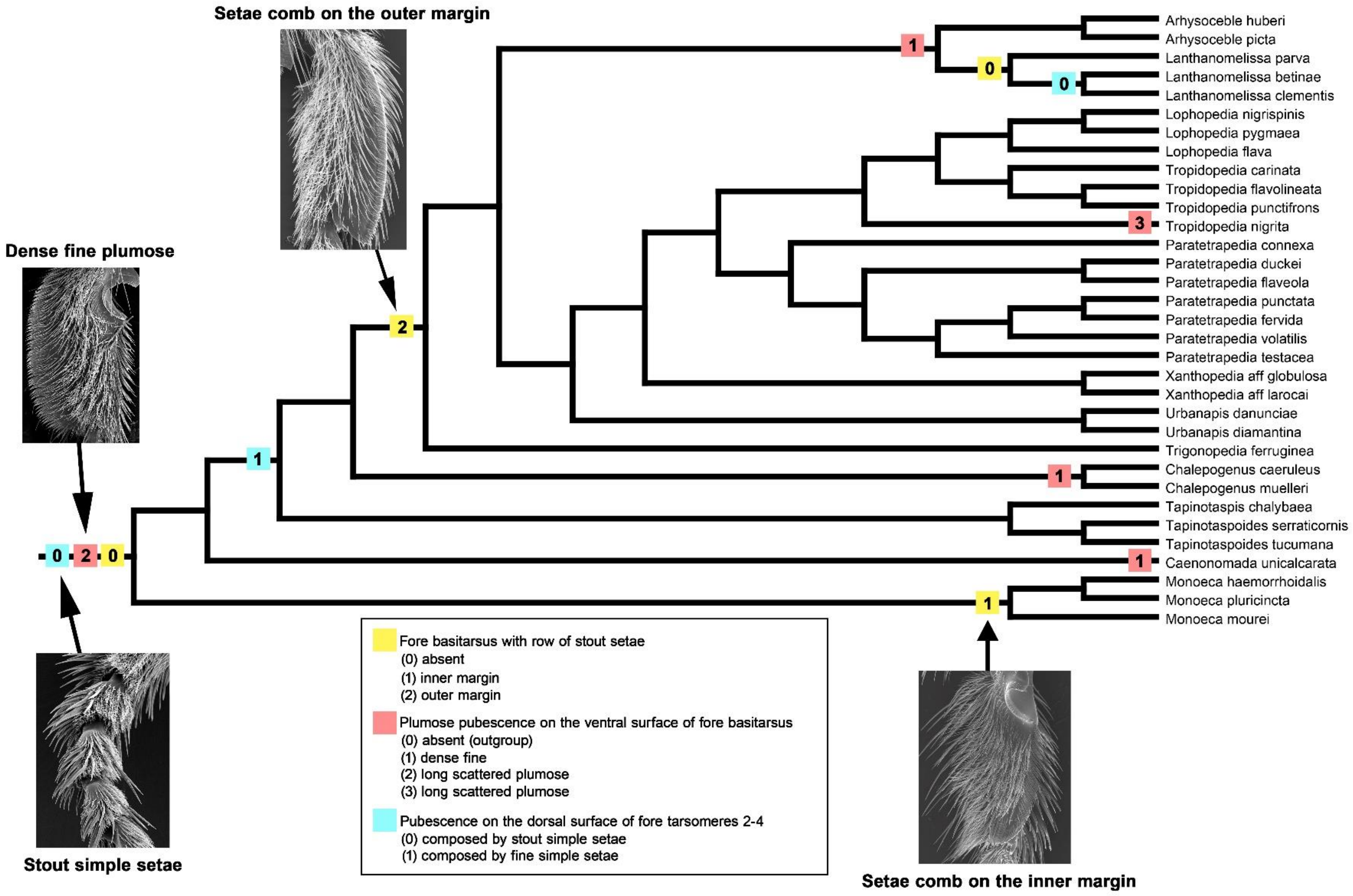


Figure 14. Parsimony optimization the evolution of oil-collecting structures in the fore leg of Tapinotaspidini males.

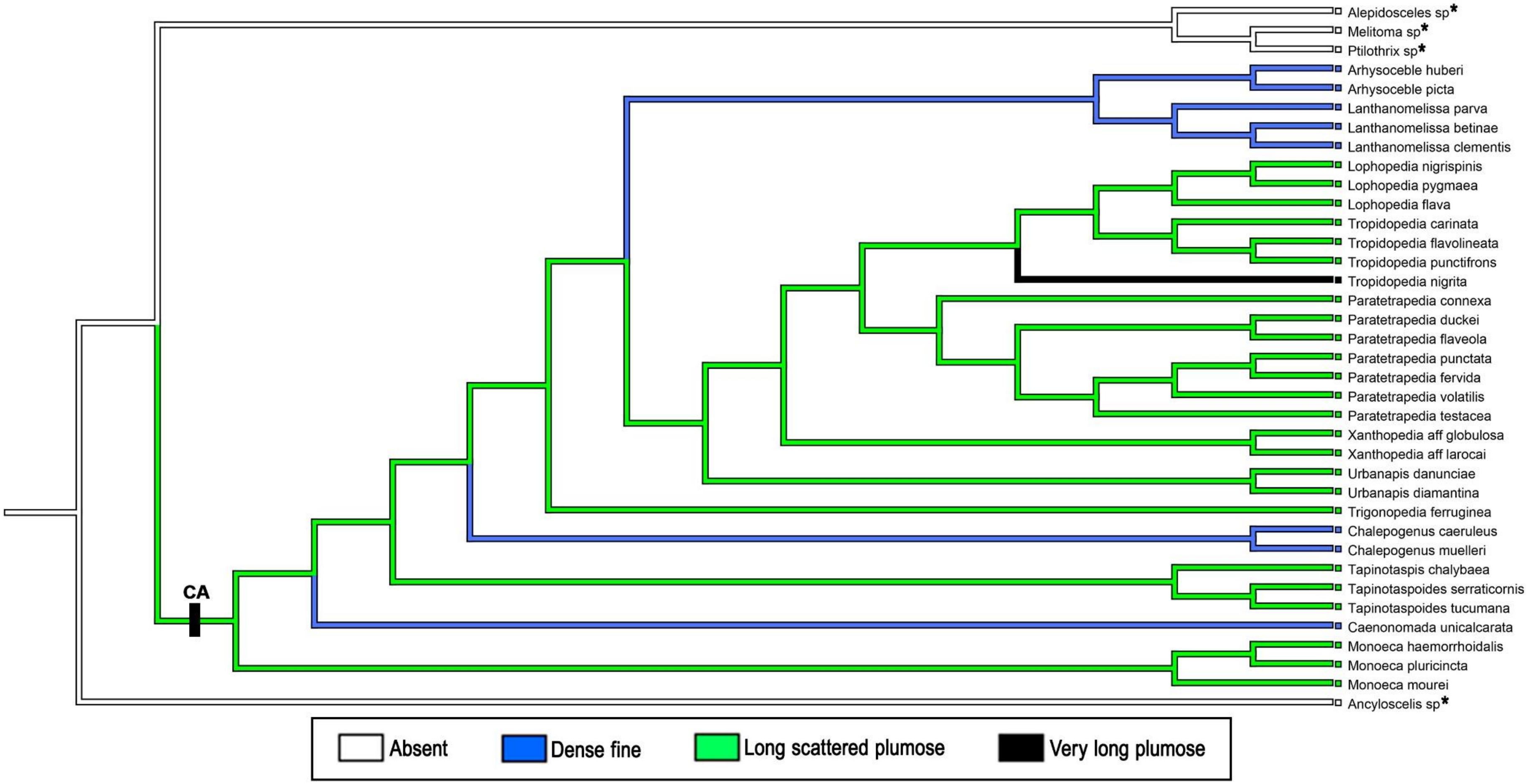


Figure 15. Parsimony optimization the evolution of plumose hairs on the ventral surface of the fore basitarsus of Tapinotaspidini males. (*Outgroup).

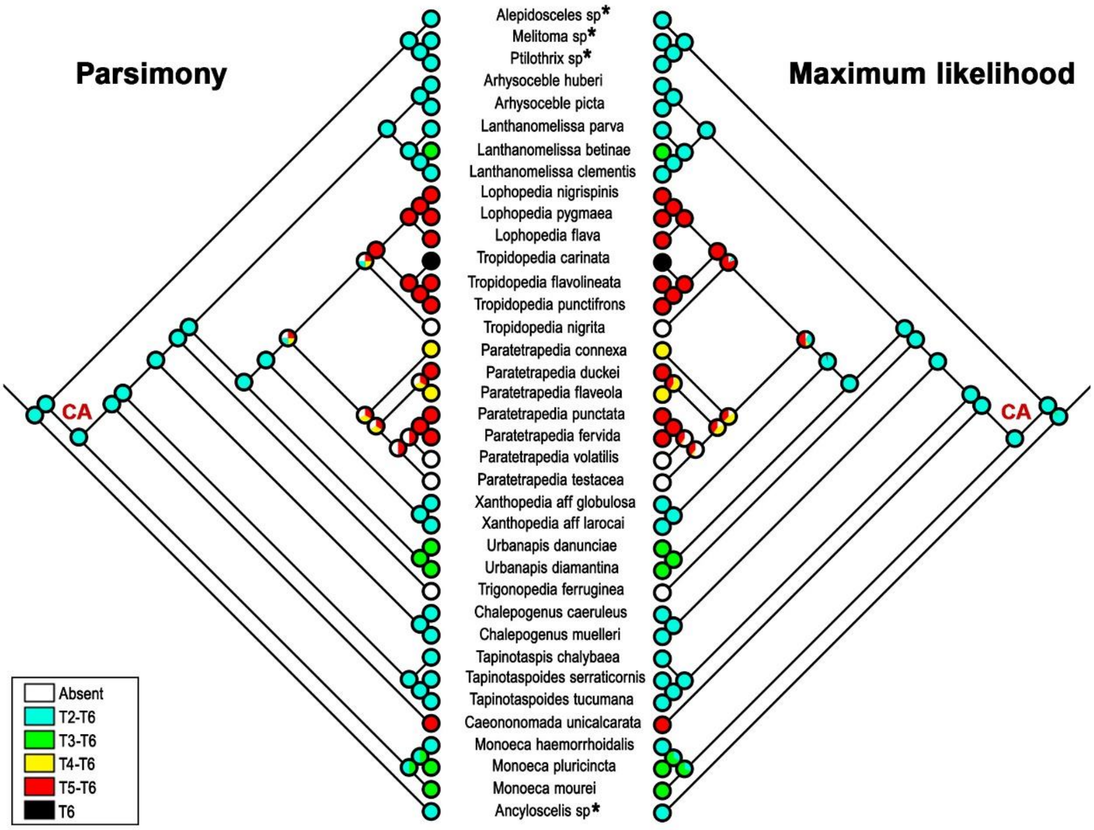


Figure 16. Parsimony and maximum likelihood optimizations the evolution of plumose hairs on the ventral surface of the fore basitarsus of Tapinotaspidini males. (*Outgroup).

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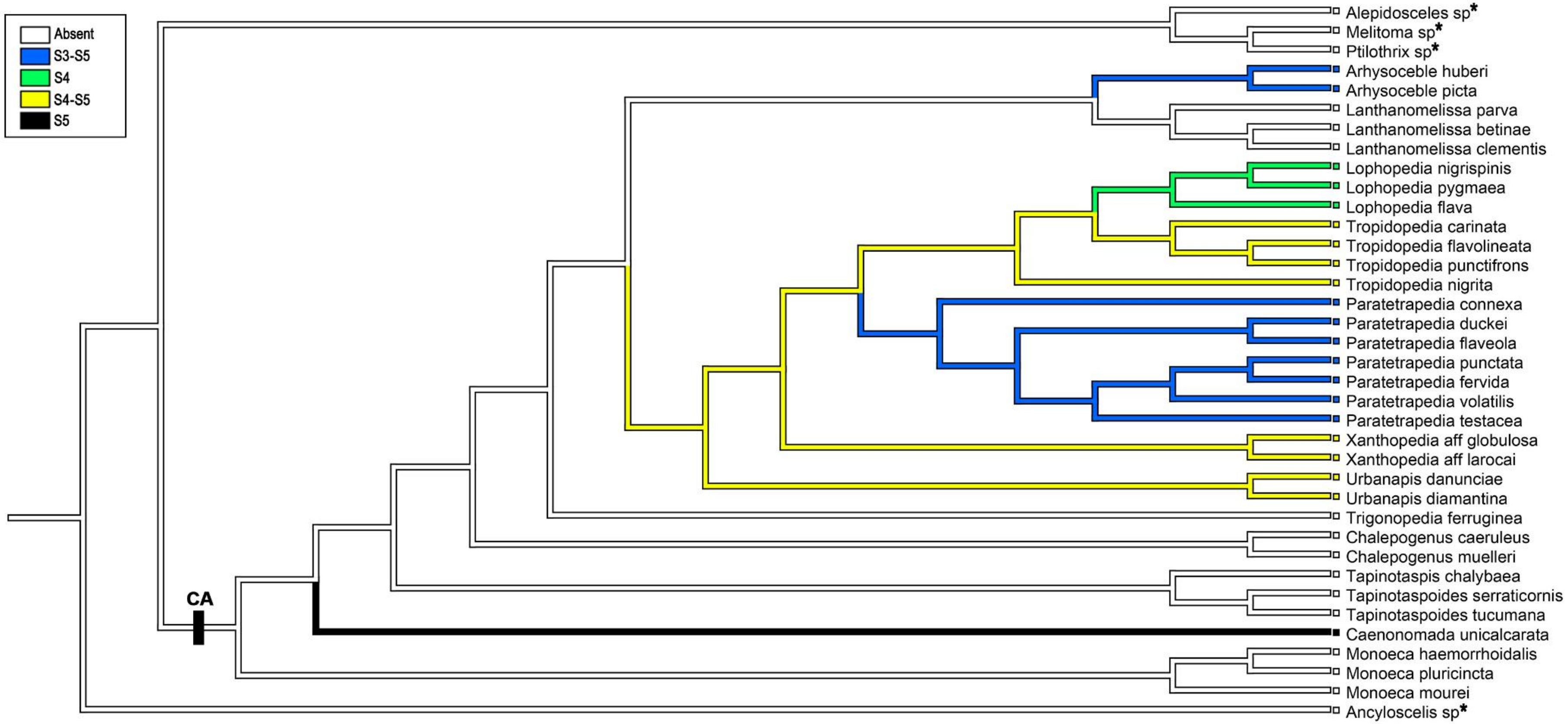


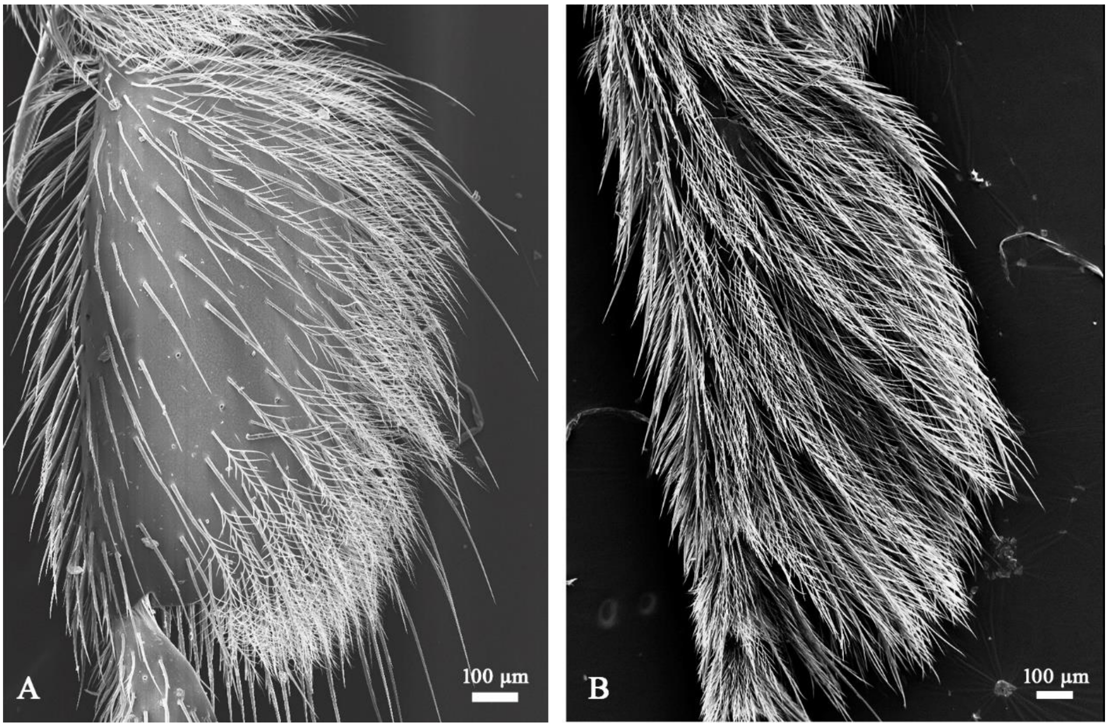
Figure 17. Parsimony optimization the evolution of plumose hairs in the posterior margin of the sterna of Tapinotaspidini males. (*Outgroup).

APPENDIX

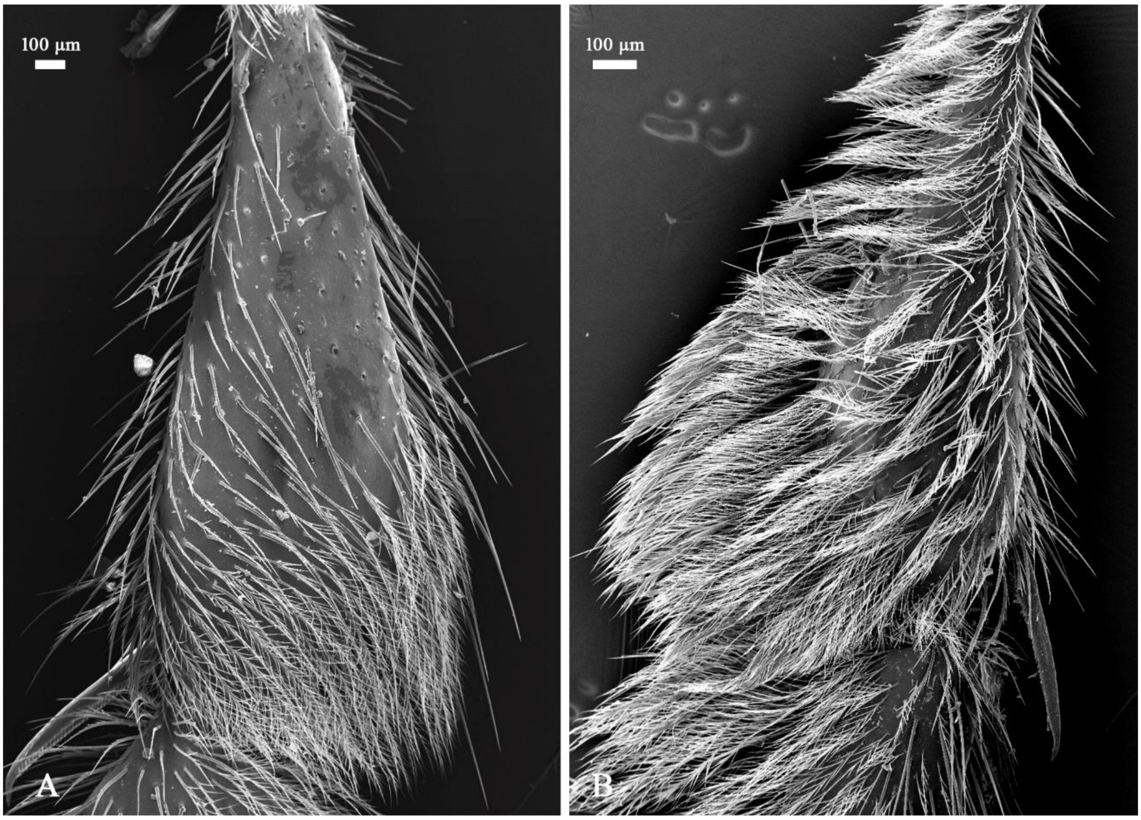
Capítulo 1

The evolution of oil-collecting structures in the male Tapinotaspidini bees (Hymenoptera, Apidae)

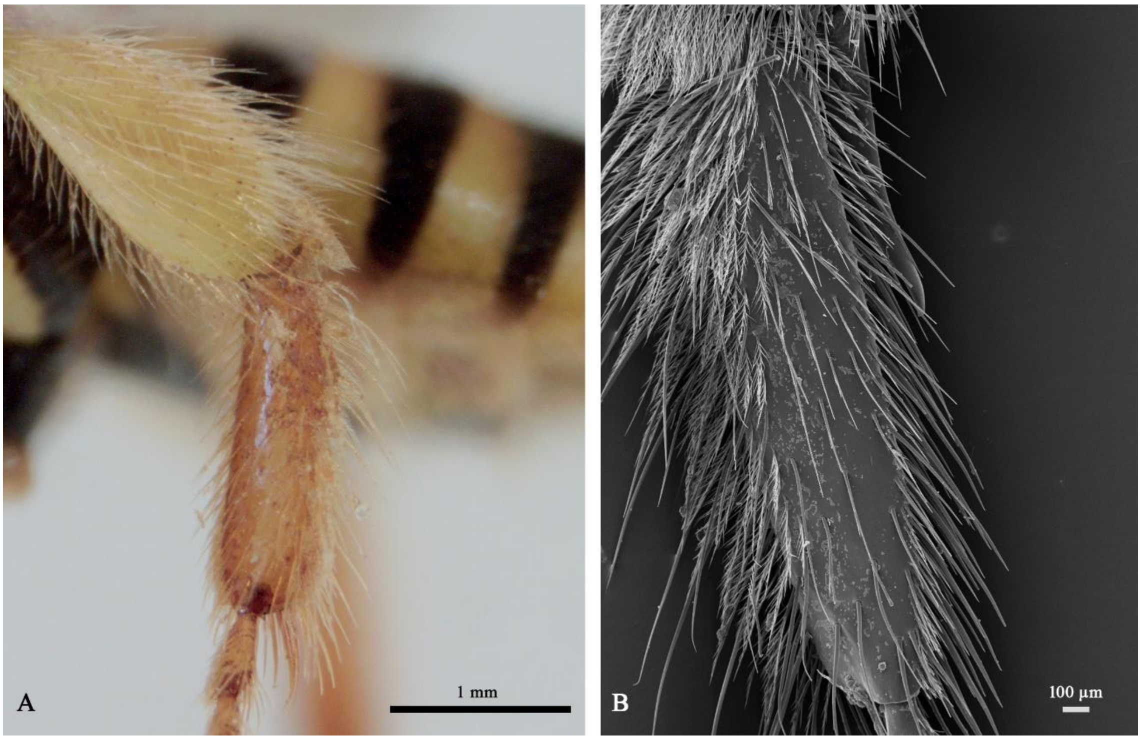
Appendix 1. Long, stout and curved setae in the middle leg tarsomeres of *Tapinotaspis chalybaea* male.



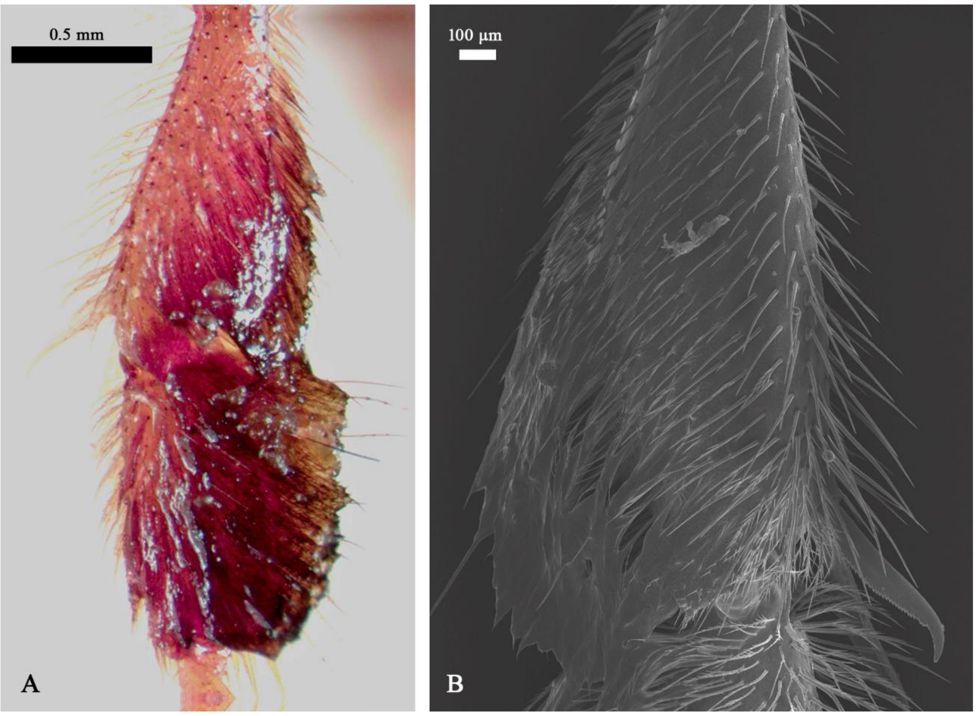
Appendix 2. Plumose hairs on the dorsal surface of middle basitarsus. **A.** Plumose pilosity concentrated in the posterior margin in male of *Paratetrapedia testacea*. **B.** Dense plumose pilosity in male of *Tapinotaspoides serraticornis*.



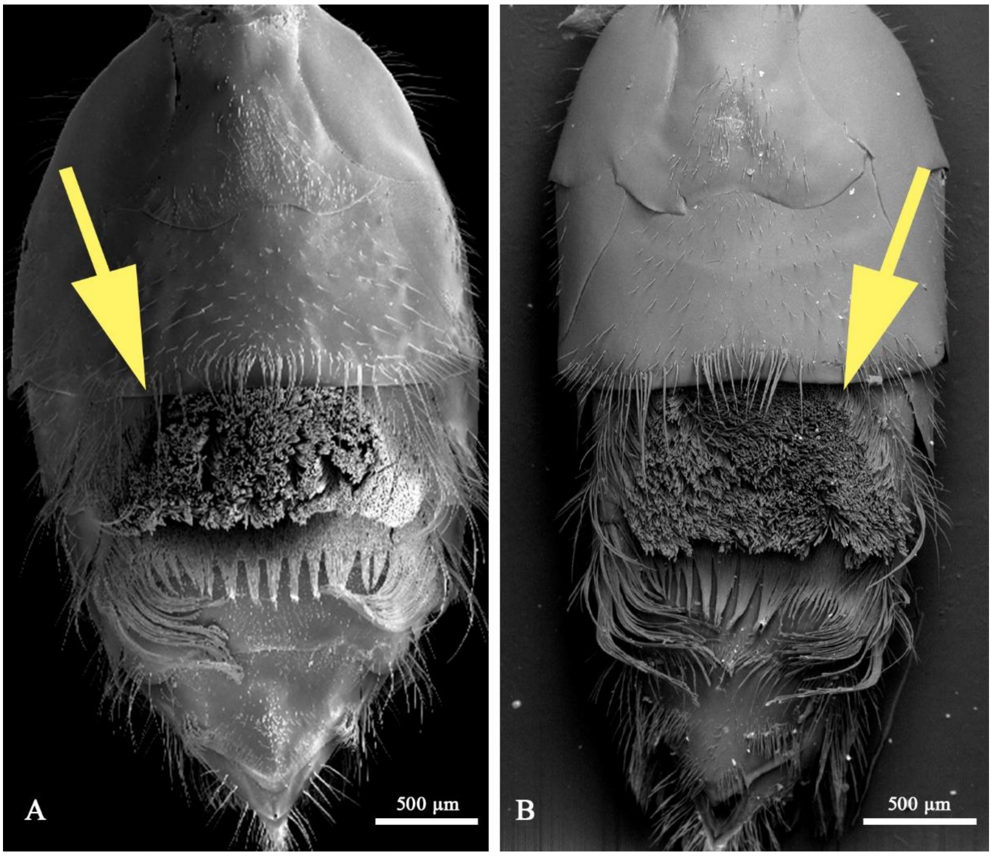
Appendix 3. Tibial scopa of Tapinotaspidini males. **A.** Plumose hairs reduced on the outer margin of *Paratetrapedia testacea*. **B.** Dense pubescence of *Tapinotaspoides serraticornis*.



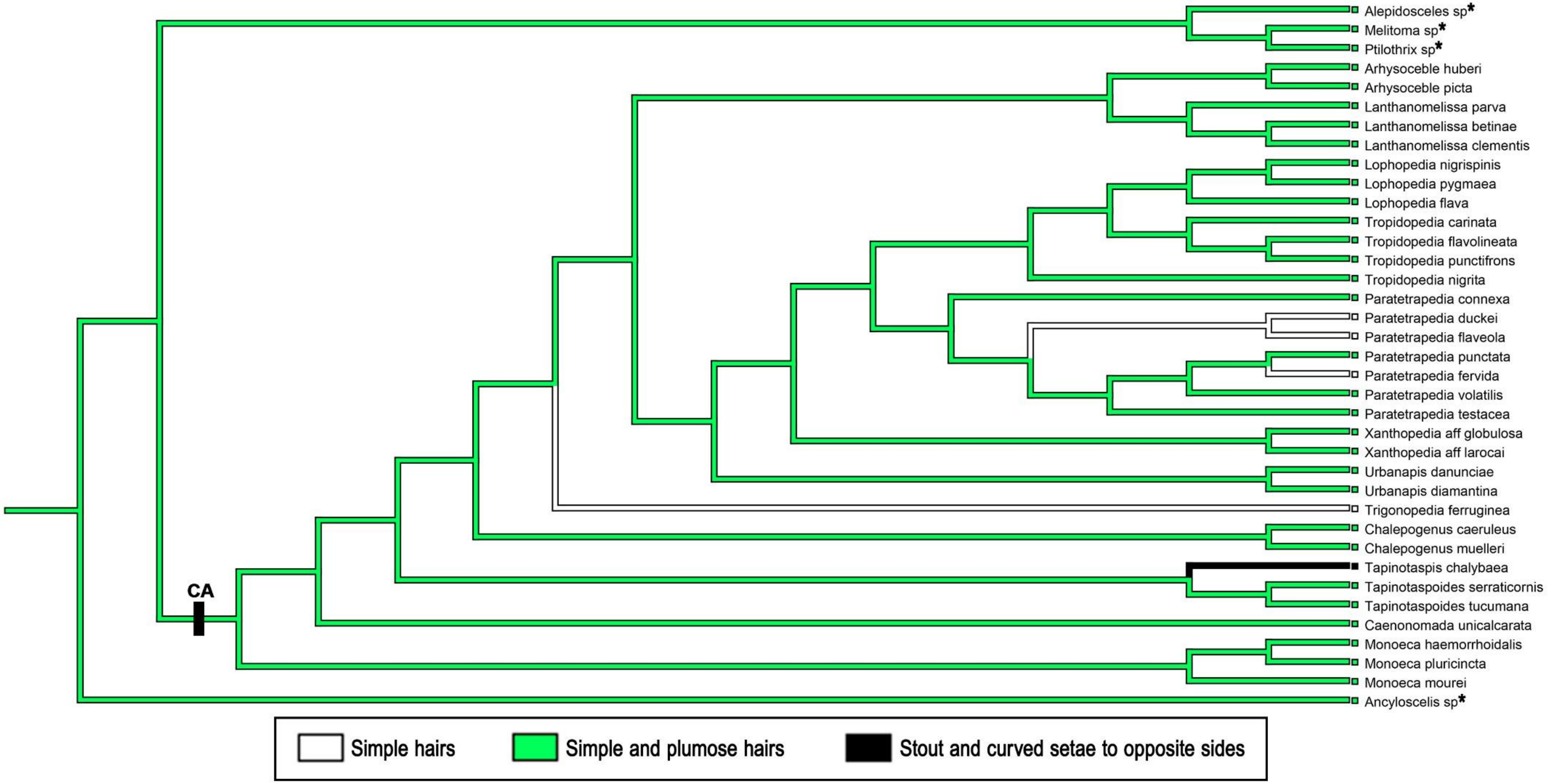
Appendix 4. Reduced basitarsal scopa on the hind leg of *Caenonomada unicalcarata* male. **A.** Pubescence on dorsal surface. **B.** Detail of basitarsal scopa.



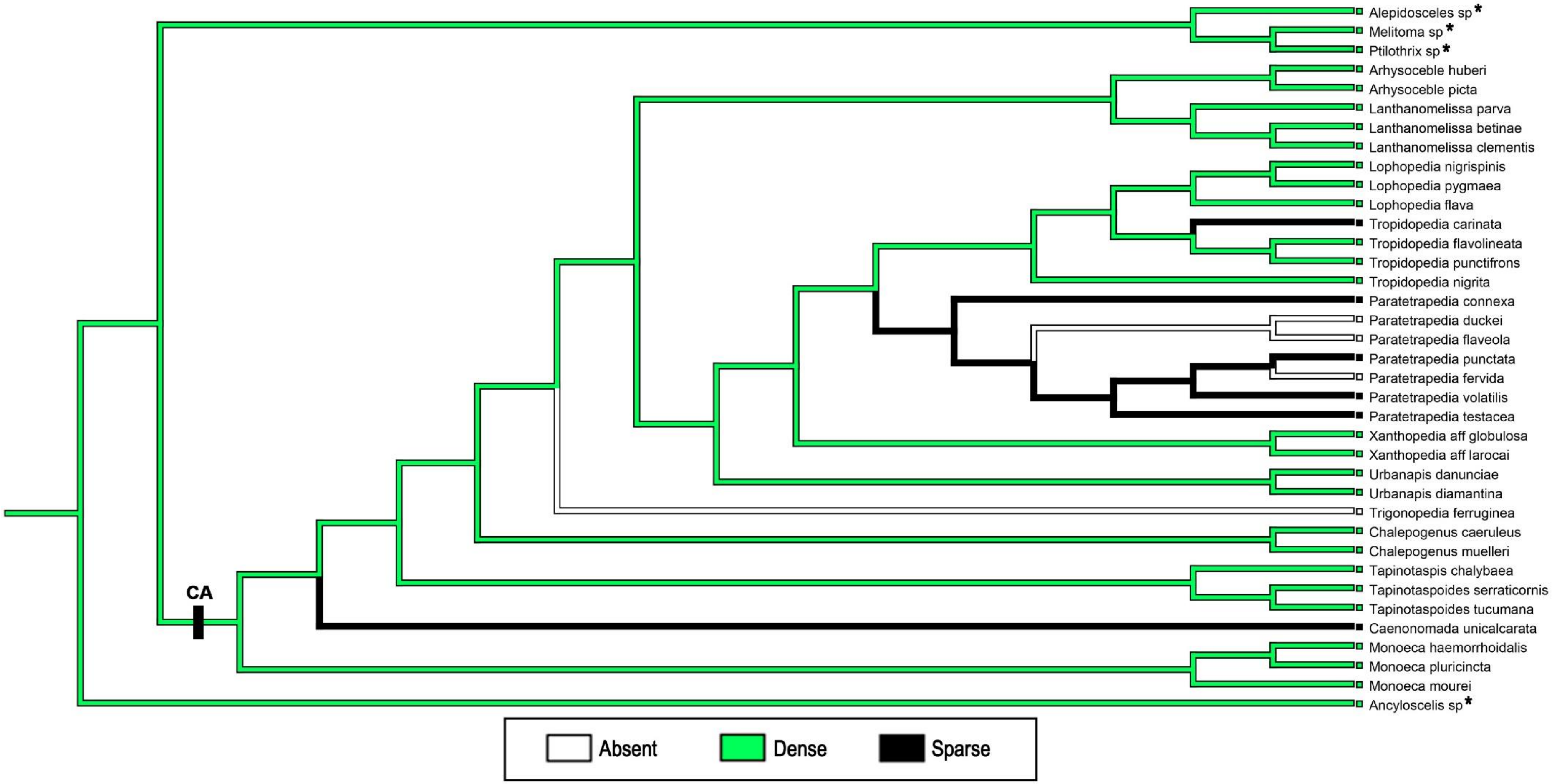
Appendix 5. Basitarsal scopa on the hind leg of Tapinotaspidini males. A. Oil on basitarsal of male *Tropidopedia flavolineata*. B. Oil on basitarsal of male *Paratetrapedia connexa* (Vachal, 1909).



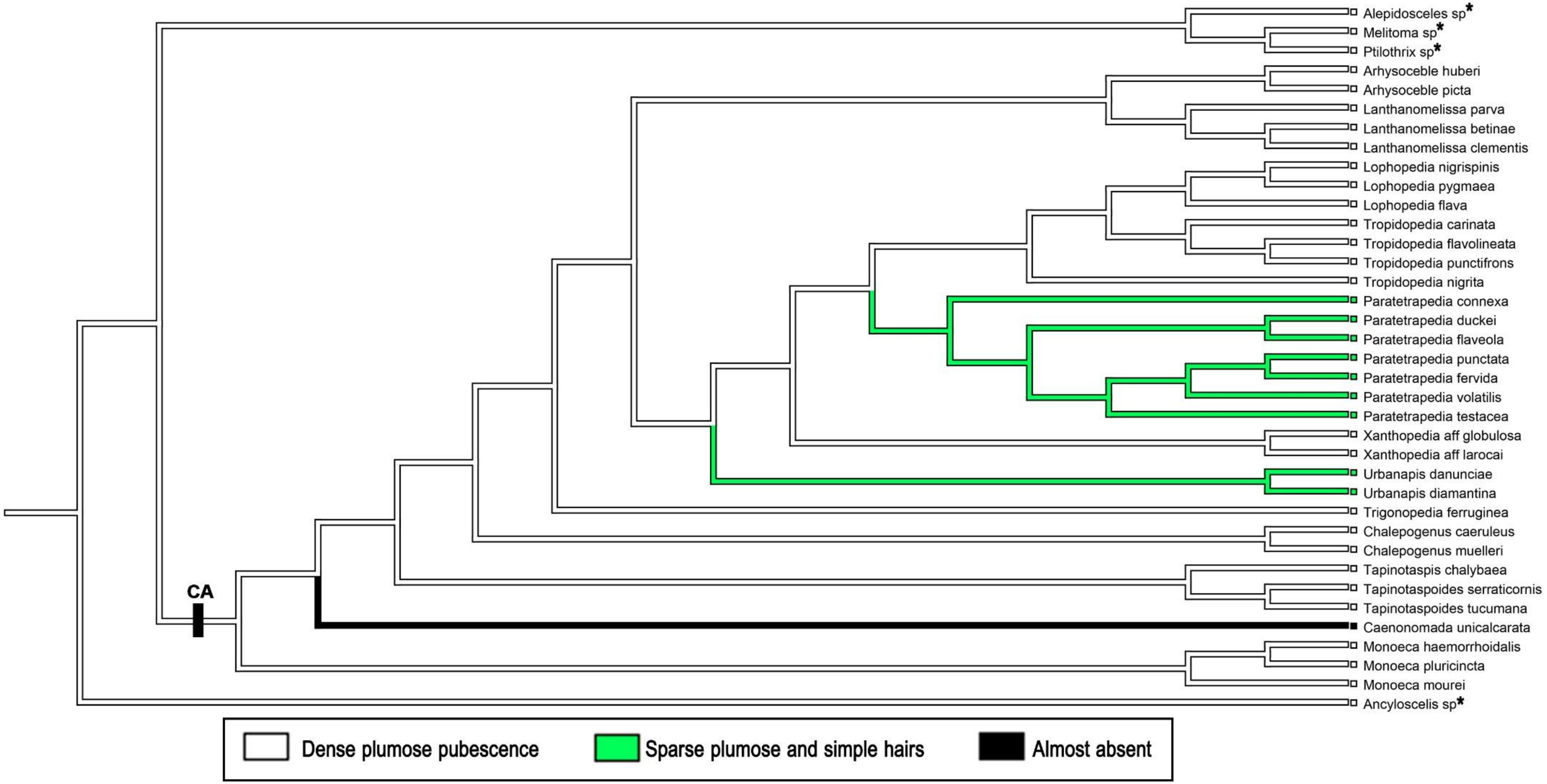
Appendix 6. Deeply concave with dense plumose pubescence on the sternum 3. A. *Paratetrapedia fervida* (Smith, 1879) male. B. *Paratetrapedia flaveola* Aguiar & Melo, 2011 male.



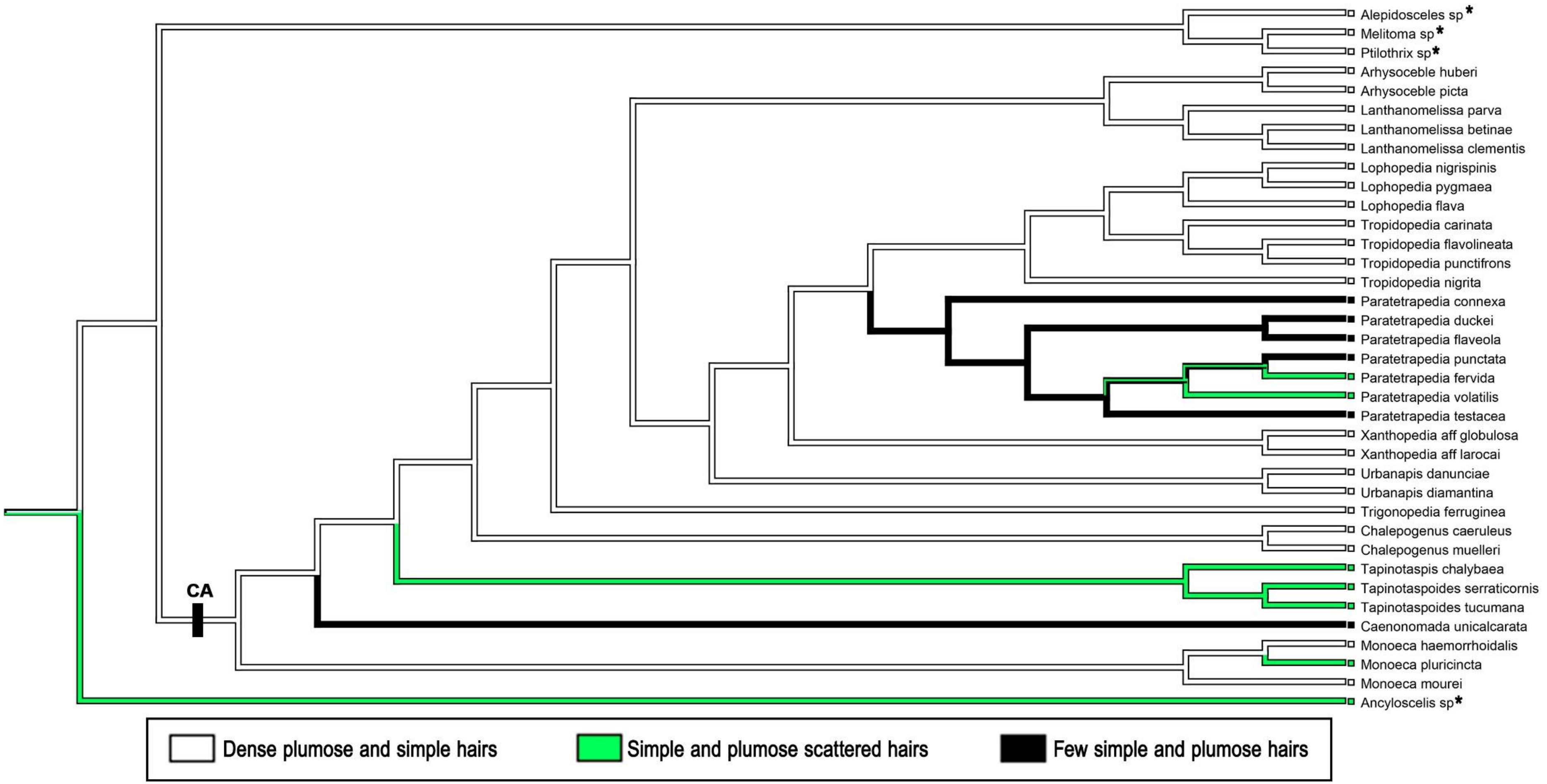
Appendix 7. Parsimony optimization the evolution of pubescence on the dorsal surface of middle basitarsus of Tapinotaspidini males. (*Outgroup).



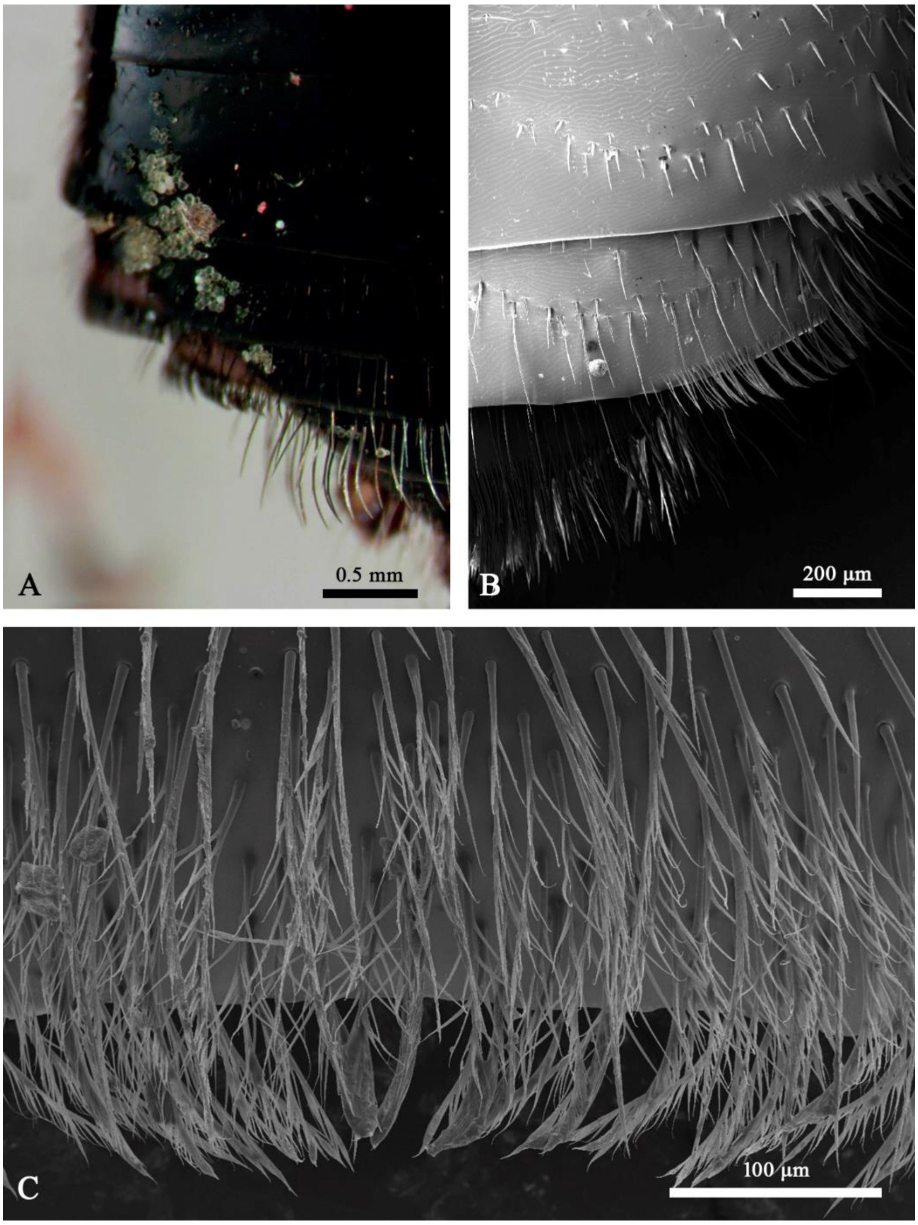
Appendix 8. Parsimony optimization the evolution of plumose pubescence on the dorsal surface of middle basitarsus of Tapinotaspidini males. (*Outgroup).



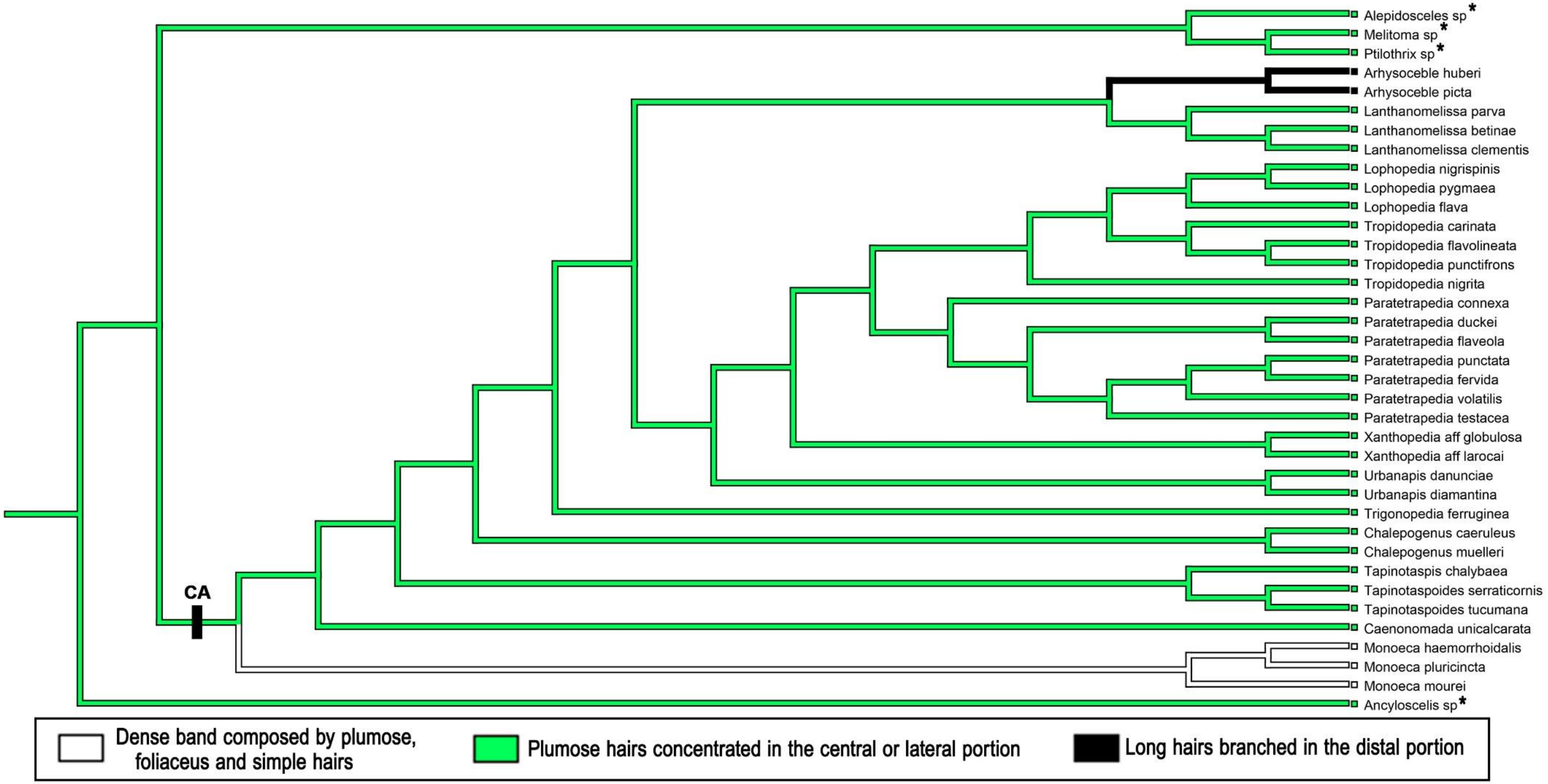
Appendix 9. Parsimony optimization the evolution of tibial scopa of Tapinotaspidini males. (*Outgroup).



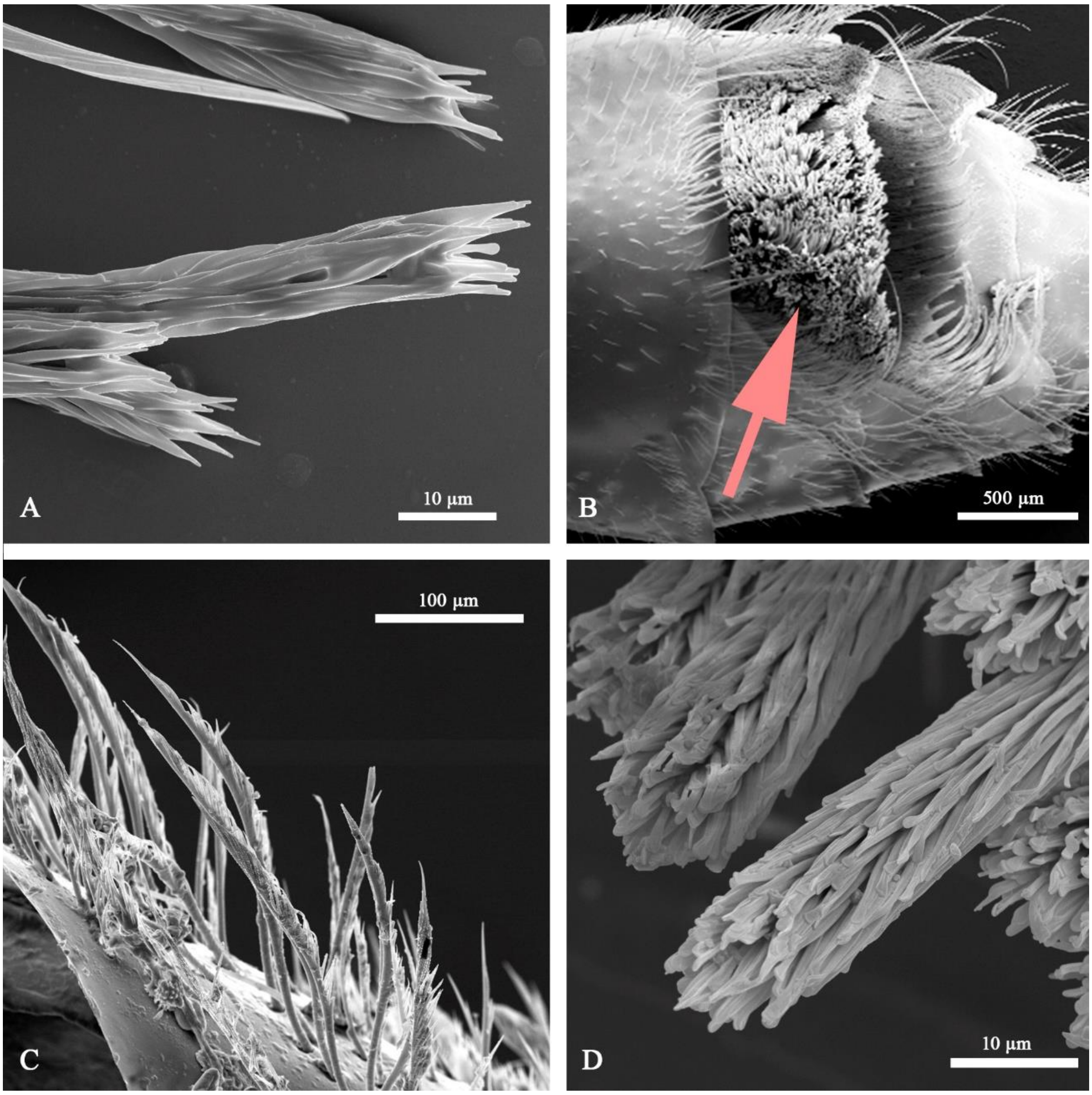
Appendix 10. Parsimony optimization the evolution of basitarsal scopa of Tapinotaspidini males. (*Outgroup).



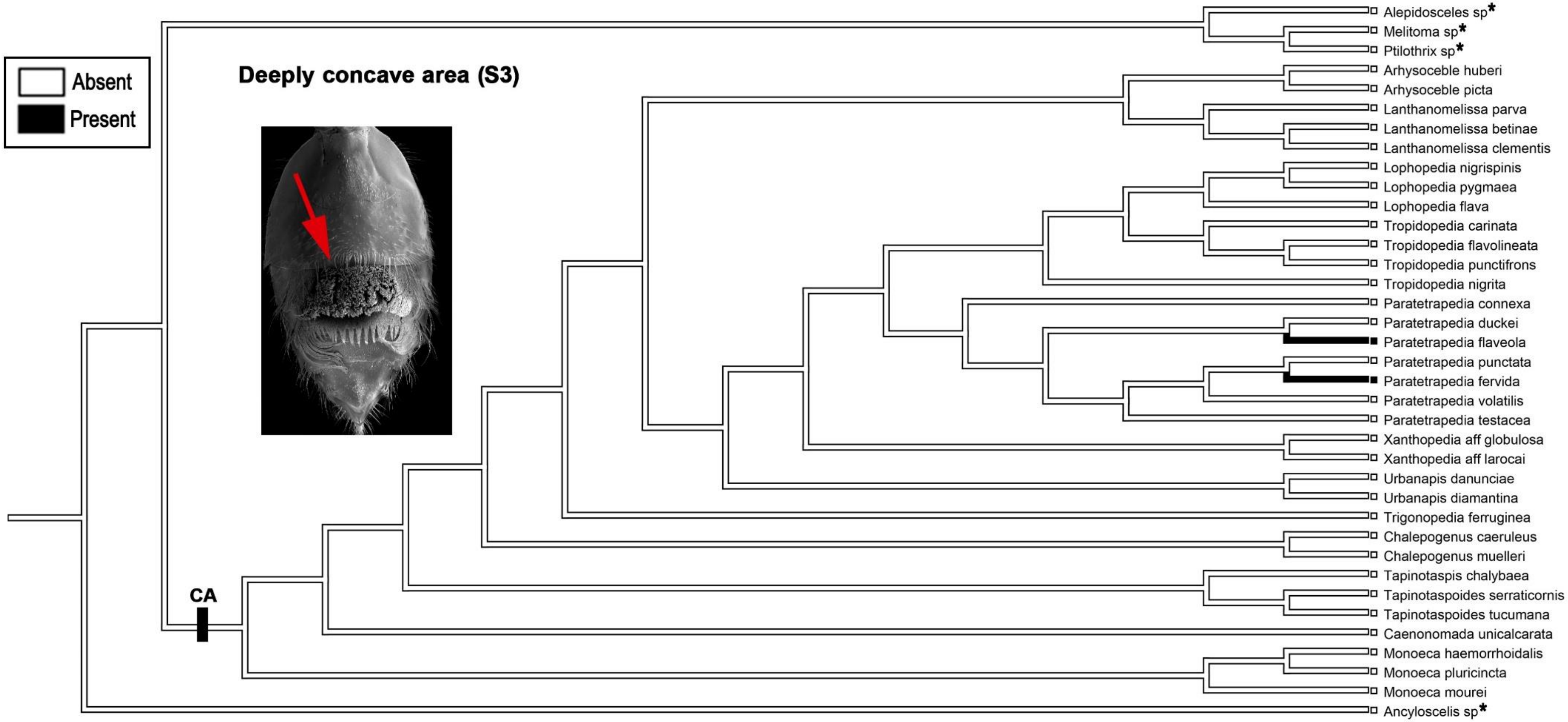
Appendix 11. Tergal scopa of Tapinotaspidini males. **A.** Tergal scopa (T4-T6) of *Paratetrapedia connexa* with presence of oil. **B.** Detail of the plumose hairs in the terga (T4-T6) of *Paratetrapedia connexa*. **C.** Tergal scopa (T5) of *Arhysoceble* sp. with presence of oil.



Appendix 12. Parsimony optimization the evolution of plumose hairs on posterior margin of the sterna of Tapinotaspidini males. (*Outgroup).



Appendix 13. Detail of variety hairs present in the sterna of Tapinotaspidini males. **A.** *Caenonomada unicalcarata*. **B.** *Paratetrapedia fervida*. **C.** *Lanthanomelissa betinae*. **D.** *Monoeca xanthopyga*.



Appendix 14. Parsimony optimization the evolution of deeply concave with dense plumose pubescence on the sternum 3 of Tapinotaspidini males. (*Outgroup).

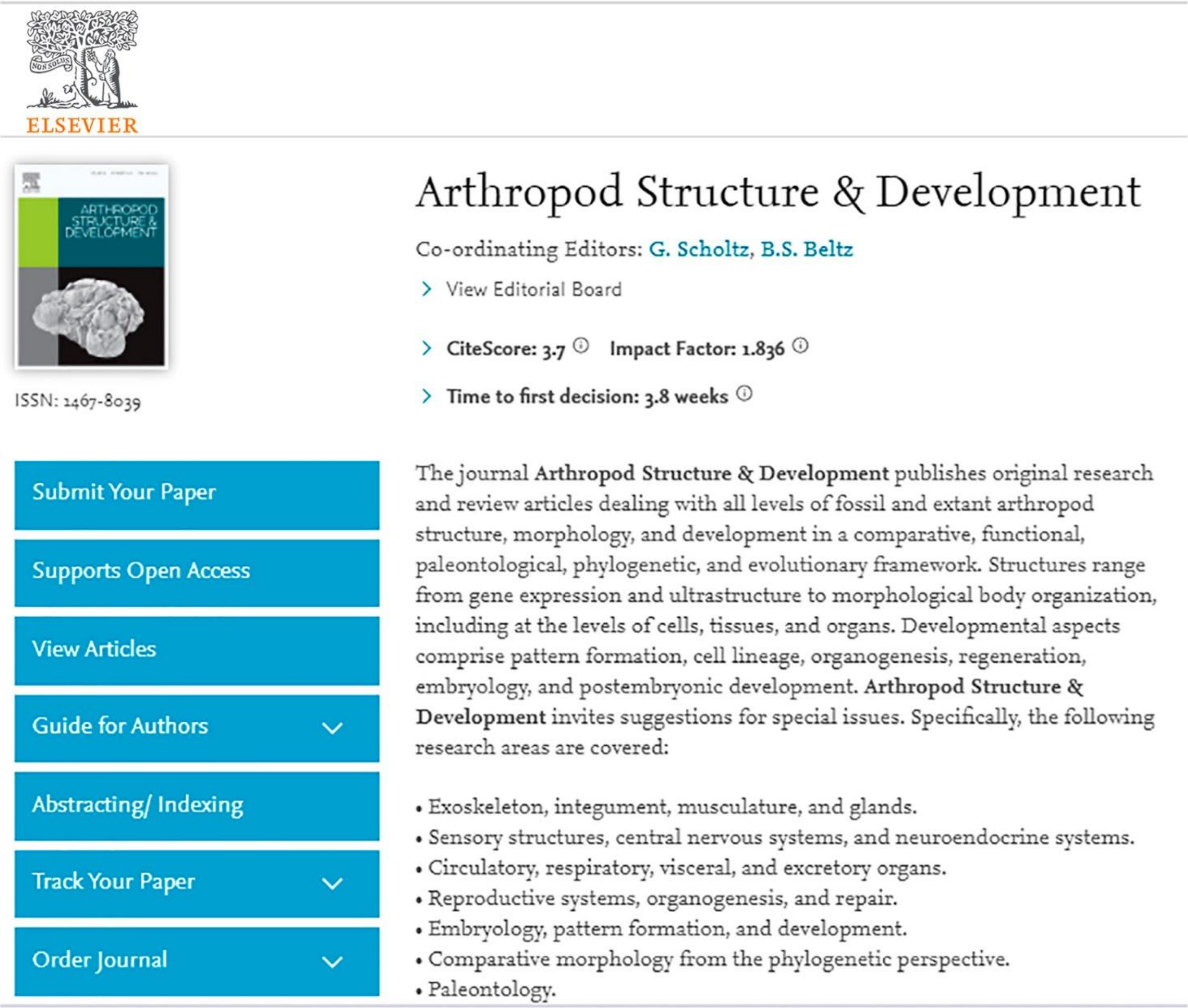
ANNEX

Capítulo 1

The evolution of oil-collecting structures in the male Tapinotaspidini bees (Hymenoptera, Apidae)

Annex 1

Focus and scope of the potential journal to which we intend to submit our manuscript.



Arthropod Structure & Development

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- Exoskeleton, integument, musculature, and glands.
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- Reproductive systems, organogenesis, and repair.
- Embryology, pattern formation, and development.
- Comparative morphology from the phylogenetic perspective.
- Paleontology.

CAPÍTULO 2

Oil collecting males of the Tapinotaspidini and Tetrapediini bee tribes: floral sources and behavior

Coleta de óleos florais por machos de abelhas da tribo Tapinotaspidini e Tetrapediini: fontes florais e comportamento

Manuscrito formatado segundo as normas da revista Iheringia (Anexx 1)

ABSTRACT

The tribes Tapinotaspini and Tetrapediini are the only groups of bees where males are oil collectors. The oil collecting behavior of these bees is potentially related to the courtship. To improve the knowledge of these bees, we performed an explorative and comparative study about the oil plant hosts for these males, their behavior on the flowers and nests, and the adaptative morphology for the oil collecting. We aggregate observations from field observations on Cerrado, and some scattered observations from other biomes and compare them with data from the bibliography. Morphology of males and females from all the genera of Tapipotaspidini and Tetrapediini were studied and compared associated to behavior. Despite the lack of information about copula on these bees, we suggest a complex behavior and morphology related to sexual selection and oil transference to females. All the males of both tribes have developed oil collecting apparatus on fore and or/mid legs, and additional oil transference apparatus on mid leg, related to an oil collecting storage on hind tibial scopa and/or fimbrial terga and sterna. We support the hypothesis that the oil collecting by males was not a precondition for copula, however it was strongly developed through the nesting habitat on wood which difficult the males to search to females on this condition.

KEYWORDS: evolution, courtship, oil collecting bees, Neotropical, pollination.

RESUMO

As tribos Tapinotaspini e Tetrapediini são os únicos grupos de abelhas em que os machos coletam óleo. O comportamento de coleta de óleo destas abelhas está potencialmente relacionado a cópula. Para ampliar o conhecimento destas abelhas, realizamos um estudo exploratório e comparativo sobre os hospedeiros de plantas oleaginosas para estes machos, o seu comportamento nas flores e ninhos, e a morfologia adaptativa para a coleta de óleo. Agregamos observações de campo sobre o Cerrado, e algumas observações dispersas de outros biomas e comparamos com os dados da bibliografia. A morfologia de machos e fêmeas de todos os gêneros de Tapinotaspidini e Tetrapediini foram estudados e comparados associados ao comportamento. Apesar da falta de informação sobre a cópula destas abelhas, sugerimos um comportamento e morfologia complexos relacionados com a seleção sexual e transferência de óleo para as fêmeas. Todos os machos de ambas as tribos desenvolveram aparelhos de coleta de óleo nas pernas dianteiras e ou médias, e aparelhos adicionais de transferência de óleo na perna média, relacionados com o armazenamento de óleo na escopa da tíbia posterior e/ou fímbrias tergais e esternais. Apoiamos a hipótese de que a coleta de óleo pelos machos não era uma condição prévia para a cópula, no entanto, foi fortemente desenvolvida através do hábito de nidificação na madeira, o que dificultou a procura dos machos pelas fêmeas nestas condições.

PALAVRAS-CHAVE: evolução, cópula, abelhas coletoras de óleos, Neotropical. polinização.

INTRODUCTION

Males and females of bees have many differences in morphology, behavior, and physiology (WILLMER & STONE, 2004). While females spend more time in nesting and collecting sources for the larvae, males spend most of time and energy searching for couples (MICHENER, 2007; DANFORTH *et al.*, 2019). However, both sexes have floral dependence as a common aspect (WILLMER & STONE, 2004; ROSWELL *et al.*, 2019). Although males and females may present different behaviours during visitation, both sexes depend on the flowers, even if it is only to collect nectar as a source of nutrition. (ROUBIK, 1989; MICHENER, 2007). Most females of non-parasitic species have specialized structures – scopa and corbicula – for the transport of pollen, which will be used for feeding the larvae (MELO *et al.*, 2002; MICHENER, 2007). In contrast, the relationship of males – who do not collect pollen – with the flowers is usually based on the search for reproductive partners or places for protection and/or rest. Two basic patterns of male searching for females are observed on distinct groups of bees, were males can search for females patrolling over the nesting places or over the flowers (DANFORTH *et al.*, 2019).

Regarding the behavior of males, there are some records of males collecting floral resources – other than nectar – on the genus *Tetrapedia* Klug (Tetrapediini) and in the tribe Tapinostaspidinii (Hymenoptera, Apidae). On the genus *Tetrapedia*, the behavior and structures related to the collection of oil were studied from three species from random records (CAPPELLARI *et al.*, 2012). Most studies on morphological adaptations for oil collection have as emphasis the females (Neff & Simpson, 1981; ROIG-ALSINA, 1997; COCUCCI *et al.*, 2000; KUHLMANN & HOLLENS, 2014). The function of the oil for males of the genus *Tetrapedia* and Tapinotaspidini tribe is uncertain and may be related to mating behavior (CAPPELLARI *et al.*, 2012).

Most of studies neglect the distinct behavior and morphology of males and females as flower visitors and pollinators (SMITH *et al.*, 2019), except for few rare studies (CANE *et al.*, 2011; CAPELLARI *et al.*, 2012; ROSWELL *et al.*, 2019), and taxa that already have a strong knowledge about the male collecting behavior like the orchid bees of Euglossini tribe (DODSON, 1969; BEMBÉ *et al.*, 2004; ELTZ *et al.*, 2019), however related to special resources restricted to males.

On the tribe of Eucerini bees, a close tribe of Tapinotaspidini, we do not have special foraging of males, and CANE *et al.* (2011) saw a very low rate of males on flowers compared to females, mostly related to nectar foraging. However, it is expected that in some bee groups, like oligolectic bees, we can saw higher rates of visitation on flower due to composition of a rendevouz space of numerous males searching for females in the same plant (ALCOCK *et al.*, 1978; EICKWORT & GINSBERG 1980; CANE *et al.*, 2011).

The adaptations for oil collection in females of Tapinotaspidini are much more diverse than in other oil collecting bees tribes (ROIG-ALSINA, 1997; COCUCCI *et al.*, 2000), and some species may present more accentuated structures in males than in females (AGUIAR & MELO, 2007). It is expected that the males have a correspondence in the diversity of morphological apparatus for oil collecting and transport, but also some distinct and unique morphological secondary sexual characters related to sexual selection. The objective of the present study is to review and discuss comparatively the male secondary sexual characters of Tapinotasdini and Tetrapediini, with emphasis on oil collecting behavior, and oil collecting plant hosts.

MATERIAL AND METHODS

We studied the morphology of all the genera of Tapinotaspidini and Tetrapediini tribes, comparing with the behavior observed in the field and data provided in the literature. The material studied was obtained from our collects and from different entomological collections, mainly from the collection of Departamento de Zoologia, Universidade de Brasília. We also evaluated the presence of oil on the body parts of the males, mainly on legs and metasomal hairs. The oil has the characteristic to not volatize and has a viscous appearance in the body parts of the bee. When it was observed viscous substances on other parts of the body of the bee that can suspect that it was dirtied in the collected storage the specimen was not used. Detailed description and observation of the body parts with oil were checked on Scan Microscope of the Laboratório de Microscopia e Microanálise (LMM) from Instituto de Ciências Biológicas da UnB.

To evaluate to male oil collecting behavior and species interactions, we collect data from focused field excursions on Cerrado, and other scattered observations on other biomes. One of us (WPS), took a focused effort collecting numerous observations, during 24 months, totalizing more than one hundred hours of observations. The specimens were observed in the field, and when possible, they were collected to check the identity and recorded with photos and videos.

The morphology of body follows the terminology of Michener (2007), were the terga were treated as T1 to T7, and sterna S1 to S6. The dense bands of hairs on metasoma were treated as premarginal bands on sterna, and tergal fringes. The tarsomeres one to five were treated as basitarsus, tarsomeres 2-4, and distitarsus. The orientation of the structures on the legs follows the system proposed by Neff & Simpson (1981), were the ventral areas of the legs were considered as inner side, and dorsal parts of the legs were treated external side.

It was studied 39 species of the tribe Tapinotaspidini and 8 species of the tribe Tetrapediini, corresponding to all genera of the tribe and about 28% of the total diversity of the tribe Tapinotaspidini. The genus *Tetrapedia* is not revised and based on the current species described, we studied 29% of the diversity of the tribe Tetrapediini. The following species listed were selected based on evidence of the oil collecting behavior (Table 1) or were collected direct on oil flowers (Table 2). The list of genera is present in the sequence of clades observed on the phylogeny of Aguiar et al. (2020). The distribution of the oil on the bodies of the bees were variable, that represents possible distinct oil collecting behaviors. Some genera with restricted distribution of southern grasslands like Pampas and Chaco where not possible to access their behavior in the field, and most of data were inferred based on the study of collection and the distribution of oil over the body of dried specimens. Vouchers of the bee species studied here were deposited on the Coleção Entomológica da Universidade de Brasília, and plant specimens were deposited in the Herbário do Departamento de Botânica da Universidade de Brasília.

On the results, the description of the morphology and behavior related to each genus, follows the phylogeny of the tribe, from the basal lineages with plesiomorphic characters to upper clades with distinct apomorphies of morphology and behavior (vide Fig. 1). Data about the species phylogeny of the genus *Tetrapedia* is presented completely apart from Tapinotaspidini. The tribe and its only genus *Tetrapedia* is included in the subfamily Xylocopinae, and the tribe Tapinotaspidini is included in the subfamily Eucerinae, both in Apidae (Bossert et al. 2019).

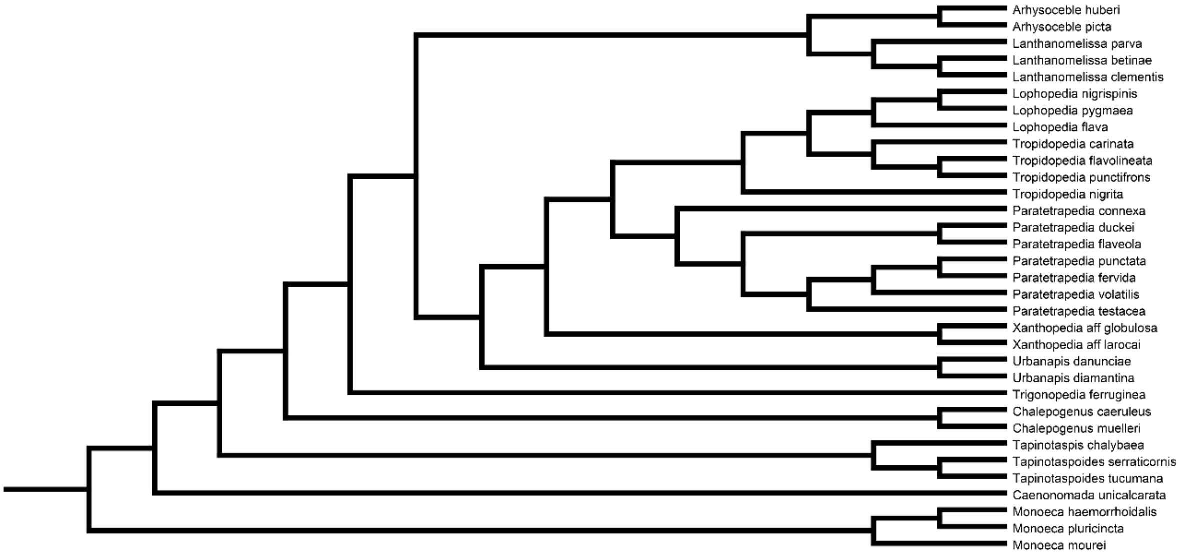


Fig. 1. Phylogeny of the tribe Tapinotaspindini bees, based on molecular data (Aguiar et al. 2020; outgroups were excluded).

RESULTS AND DISCUSSION

Morphology of oil collecting apparatus

Tetrapedia and the distinct genus of Tapinotaspidini are not reciprocally monophyletic (Cardinal et al 2010), and them all the similarities of the oil collecting structures of the males between these groups are convergence of adaptative morphology. *Ctenoplectra* (Ctenoplectrini), is also an oil collecting bee that appeared as sister group to *Tetrapedia*, however do not exist any information about possible oil collecting morphology and behavior of these bees. We also studied at least two species of *Ctenoplectra* in the collection of California Academy of Science, and the males do not have any special morphology related to oil colleting. The closest tribes of Tapinotaspidini on Eucerini line (Emphorini, Eucerini and Exomalopsini) have a complete absence of scopa on hind legs, and tergal fringes composed by similar bands of very short, plumose appressed hairs, while Tapinotaspidini has a complex set of very developed and distinct plumose hairs on the three pairs of legs, tergal and sternal margins (Fig. 2).

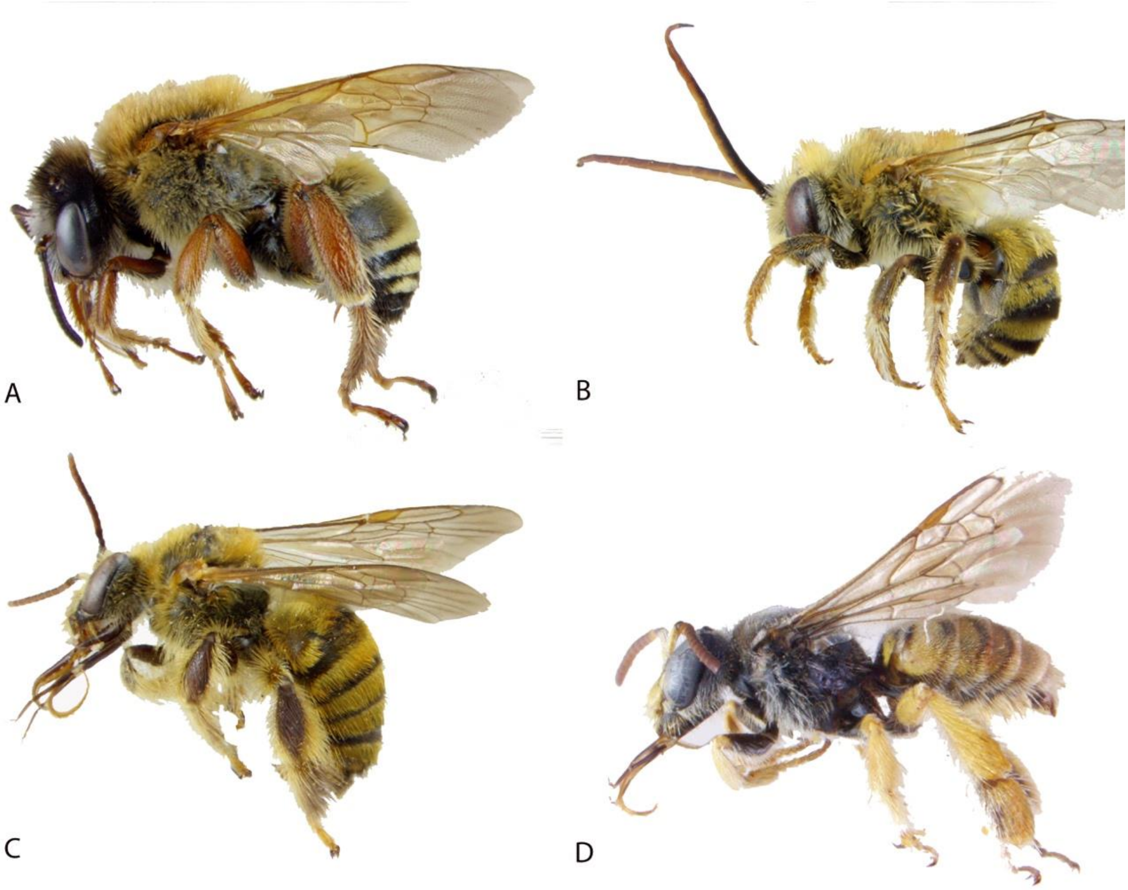


Fig. 2. A. Male of Emphorini, **B.** Male of Eucerini, **C.** Male of Exomalopsini, **D.** Male of Tapinotaspidini.

Despite the tribes Tapinotaspidini and Tetrapediini are not direct related, we compared the oil collecting apparatus of the males to understand the possible convergences and adaptative forms (Table 1). Most of the species studied have oil collecting apparatus developed on the fore legs, especially fore basitarsus, however distinctly smaller than females. This reduction is not very strong in the species of the genus *Tetrapedia*. In the species group pallidipennis of the genus *Tropidopedia*, the oil collecting apparatus on fore legs is strongly more developed than in females. The other structure that is developed almost in all genera and species is the scopa of hind tibia only in the genus *Caenonomada* and *Tetrapedia* we observed a strong reduction compared to females.

Tetrapediini

Tetrapedia

Most of the records of males collecting oil were reported on Malpighiaceae (Table 2), however SINGER & COCUCCI (1999) registered one male of *Tetrapedia diversipes* on oil flowers of *Gomesa paranensoides* (cited as *Oncidium paranaense*; Orchidaceae). The oil collecting behavior of males of *Tetrapedia* on Malpighiaceae flowers is very similar, and almost identical to that performed by the females. After landing the flowers, the males grasp the calices of the flower with mandibles, and holding the flower with the mid legs they brush the glands on the back side with the fore legs. After that, they start the transference of the oil from the fore legs to the scopa and tergal fimbria. They use the mid legs to perform this transference. The transference of the oil to the tergal fimbria is performed by inner margins of the hind legs, specially through the action of special hairs on inner margin of hind basitarsus (Fig. 3, green setae). The oil collecting apparatus of the males of *Tetrapedia* are not reduced in size compared to females like what was observed on Tapinotaspidine bees. The tergal fringes of males of *Tetrapedia* bees are a developed special character, that is composed of very fine branched hairs, almost like cotton, that was usually observed soaked with oil. The possible link related to the transference of these oil from hind legs is the special features observed in the inner margin of the hind basitarsus. On the hind legs, the scopa is reduced to the tibia, and the basitarsus has only fine branched setae on the inner margin. The males also have a special set of fringes on sternal margins, that were usually observed soaked with oil.

The hind basitarsus of males of *Tetrapedia* spp. have a high variation in shape and pubescence (Fig. 4). Most of the species have thin laminas on the margins of hind basitarsus, sometimes forming spines, or plates. The distinct shapes of the inner margin and potential function of these very distinct shapes are related to the holding of metasoma of females during the copula. It was observed in many species of bees, that the males hold the metasoma of females during the copula with posterior legs (WCISLO *et al.*, 1992; SCHLINDER *et al.*, 2018), that are strongly reinforced by flexor muscles (TORO & MAGUNACELAYA, 1987). The inner faces of the basitarsu and tibiae of *Tetrapedia* are strongly concave, suggesting the same pattern observed on *Nomia* bee species, where it adjust and contoured to fit the shape of the female metasoma (WCISLO *et al.*, 1992).

We performed some experimental pairing and copula on the species *Tetrapedia peckoltii*. Males tentatively hold the females with fore and mid legs, and during this action they brush their hind legs over the hind legs of the females (Fig. 5). This behavior, despite of the non-success copula, suggests the transference of the oil from the hind legs of the males to the scopa of females. The special long ramified hairs on the inner margin of basitarsus, are probably the more special set of hairs related to the transference of the oil to the females.

Fig. 3. *Tetrapedia* sp. male, lateral view, and correspondent first to third legs; tergal fringes (red arrows), hind leg scopa (blue arrows), specialized setae on the inner margin of hind basitarsus (green arrow), oil collecting apparatus on forelegs, with row of stout setae in details (black arrows)

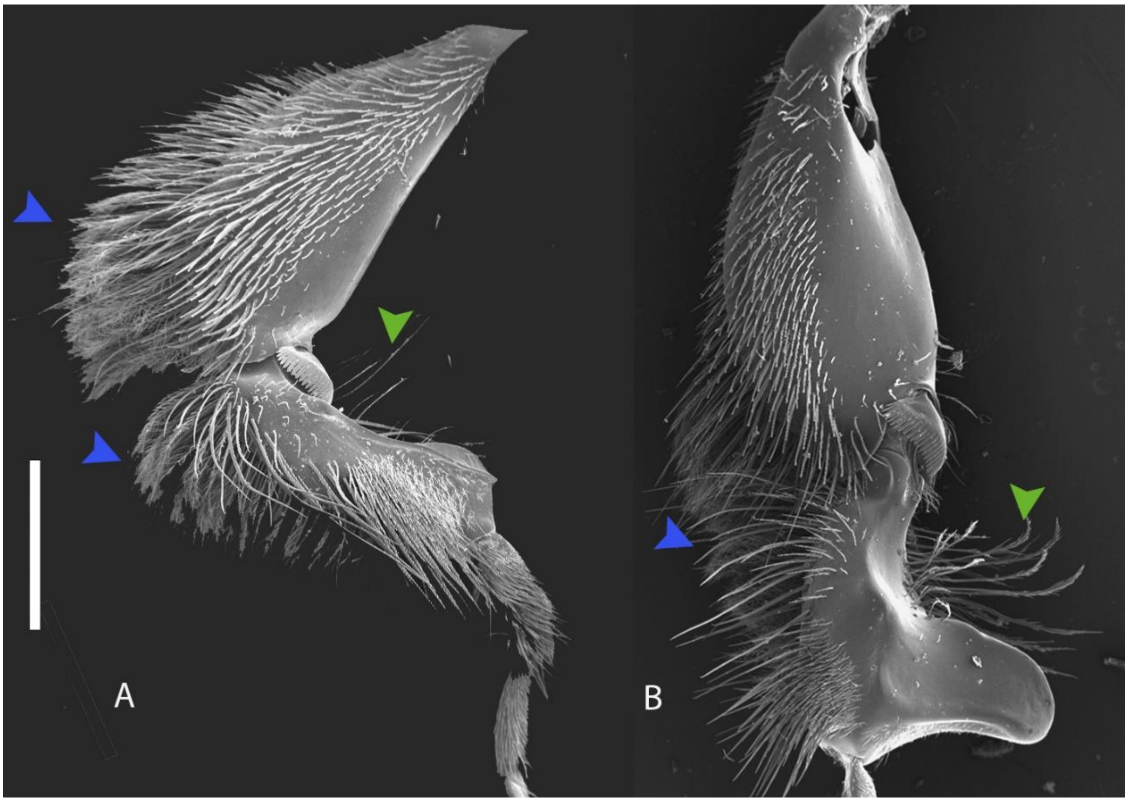


Fig. 4. Hind legs of males of *Tetrapedia peckolti* Friese, 1899. **(A)**, and *Tetrapedia amplitarsis* Friese, 1899. **(B)**, inner view, hind leg scopa (blue arrows), specialized setae on the inner margin of hind basitarsus (green arrows); scale bar 1 mm.

Fig. 5. Experimental pairing and copula on the species *Tetrapedia peckoltii*. **(A)** Male with hind legs elevated (below) before another attempt at copulation. **(B)** Male tentatively hold the female with fore and mid legs, and during this action they brush their hind legs over the hind legs of the female.

Tapinotaspidini

Monoeca

Males of *Monoeca mourei* and *Monoeca pluricincta* were recorded visiting the flowers of *Banisteriopsis* sp. (Malpighiaceae), in the same flowers that many females were observed, mostly on the morning, mainly between 9:00 and 11:00 h. The oil collecting apparatus of the males of *Monoeca* species studied are present on fore and mid legs like the females but are reduced in size and density of the plumose hairs on the concave surface (Fig. 6). The oil collecting behavior of the males is similar to the females of Centridini bee females, where the female grasps the main upper petal with the mandibles, and holding the flower with the fore and mid legs, to brush the oil glands in the calices. The pollen of the flower is deposited on the ventral body, between coxae. The males of *Monoeca* have three sets of setae on posterior margin of between the second and fifth sterna. The set of simple setae is not related to oil collecting, however the other two sets of plumose setae is supposed to be related to oil storage, mopping oil from the fore and mid legs. Males of *Monoeca* have also been observed with viscous substances like oil on scopa of the hind and mid tibia.

The males of *Monoeca* observed in the flowers of *Banisteriopsis* sp. have the "*four-legged"* behavior, exactly the same of females, where the elaiophores were grasped with the rows of stout setae of the fore and mid legs (NEFF & SIMPSON, 1981). The genus *Monoeca* is exclusive to oil collecting in Malpighiaceae flowers, and numerous species have been recorded collecting oil and/or pollen (BUCHMANN, 1987; CUNHA & BLOCHTEIN, 2003; SIGRIST & SAZIMA, 2004; POSSOBOM & MACHADO, 2018; AGUIAR *et al.*, 2020). We do not observed males patrolling the flowers of Malpighiaceae, and the few copula records of the literature were observed on nesting sites (CUNHA & BLOCHTEIN, 2003; ROZEN *et al.*, 2005). This condition can suggest that the oil collecting behavior of males on *Monocea* is not precondition to copula and occurs occasionally, compared to most of the genera of Tapinotaspidini and *Tetrapedia*.

Fig. 6. *Monoeca mourei* Aguiar, 2012 male: hind tibia and mid tibia scopa, blue setae; fore basitarsus, black setae; tergal fimbria, orange setae.

Caenonomada

It was studied all the three species of *Caenonomada*, C. *brunerii*, C. *unicalcarata*, and *C. labrata*. The species studied of the genus *Caenonomada* do not present conspicuous apparatus like pads, combs or rows of setae on the fore basitarsus. It was observed oil on the ventral surface of the tarsomeres 1-4, with numerous fine dense plumose setae covering the surface and a set of foliaceous setae on the outer margin. *Caenonomada* was not mentioned in the study of Vogel (1974) system of oil collecting apparatus, however it seems that the abundance of fine and short ramified setae suggest that these bees may be classified in the alfa type, which is related to oil collecting from trichomatic glands of oil. Three male specimens of *Caenonomada unicalcarata* had the oil on the fore legs. It was observed pollen on the labrum, clypeus, supraclypeal area, frons and mesoscutum of these bees, suggesting possible oil collecting from species of Plantaginaceae, Iridaceae or Krameriaceae, which are the some of oil plant sources for the females (AGUIAR & MELO, 2008; COCUCCI *et al.*, 2001, CARNEIRO *et al.*, 2019). The species of *Caeonomada* represent the group with more reduced hind tibial scopa in the tribe, however they are much larger than in Centridini bees that do not collect oil by males, and is almost equivalent to the scopa of *Tetrapedia* species which the males collect oil very commonly (Fig. 7). The oil collecting apparatus of the females were briefly described for *Caenonomada bruneri* and *Caenonomada* affs. *unicalcarata* from specimens collected on flowers of Iridaceae and Plantaginaceae (COCUCCI *et al.*, 2001, MARTINS *et al.*, 2013). Despite that we do observed males of *Caenonomada* on oil flowers, the morphology, size and position of pollen deposition on males suggest that they act as pollinators of these plants exactly as females. We do not also observed males patrolling the flowers, wich suggest that these bees also has a mating behavior related to nesting sites.

Fig. 7. *Caenonomada unicalcarata* (Ducke, 1908) male, lateral view; blue arrows, incipient scopa of hind legs; red arrow, tergal fimbria; small box, S5 with sternal fimbria.

Tapinotaspis lineage

The *Tapinotaspis* lineage is composed by two genera, *Tapinotaspoides* and *Tapinotaspis* (MICHENER & MOURE, 1957; AGUIAR *et al.*, 2020). The oil collecting behavior of *Tapinotaspis* is well studied and the females has a long and very hairy mid legs, that allow these bees to brush the trichomatic glands of *Nierembergia* (COCUCCI, 1991).

Females of *Tapinotaspoides* have very distinct fringes of long, ramified and very specialized setae on the margins of sterna (COCUCCI *et al.*, 2000). These bees collects oil or resins from extrafloral glands and also floral glands (MELO & GAGLIANONE, 2005), brushing these dense fringes over the oil trichomes. The males of *Tapinotaspoides* do not have these fringes on sternal margins, and we do not observed males collecting oil.

However, we observed oil mopping on the sternal fringes, on tergal fringes, on fore and mid basitarsus, and also on the hind tibial scopa of numerous male specimens of *Tapinotaspoides tucumana* and *T. serraticornis*. Males of *Tapinotaspoides* have long plumose hairs on the margins of sterna, but these hairs are completely distinct from the females (Fig.8). Males and females have also dense pads of very long hairs on fore and mid basitarsus, however these long branched hairs are completely distinct from the hairs from the legs of *Tapinotaspis*. It was not registered males of *Tapinotaspoides* on oil flowers, or on bibliography. ROZEN (1984) described the nesting habitus of *Tapinotaspoides serraticornis* however he did not mention any special aspect related to male patrolling for mating. The species of *Tapinotaspoides* is generalist exploring oil from extrafloral glands and floral glands (MELO & GAGLIANONE 2005; our data). We observed males and females of *Tapinotaspoides serraticornis* on the same areas performing same flight routes, suggesting that this species do not have a special patrolling behavior over the flowers or over the nesting sites.

Fig. 8. Margins of sterna of *Tapinotaspoides serraticornis* (Friese, 1899). **A.** Long plumose hairs on the margins of sterna of *T. serraticornis* male. **B.** Fringes of long, ramified and very specialized setae on the margins of sterna. of *T. serraticornis* female.

Most of the species of *Tapinotaspis* are poorly known, except for the species *Tapinotaspis chalybaea*, which has numerous studies about its oil collecting behavior on *Nieremebergia* (Solanaceae). On Cerrado, it was not reported any species of *Tapinotaspis*, however through loans from some collections, it was possible to study some female specimens and one male of *T. chalybaea.* The males of *Tapinotaspis chalybaea* are extremally rare in the collections, and we got the loan of one male from

Instituto Miguel Lilo (Argentina, Tucuman). The male of *T. chalybaeae* has a long mid leg with a dense tuft of special hairs related to oil collecting, a very conspicuous scopa with dense plumose pubescence (Fig. 9), and a dense fringe of plumose appressed hairs on tergal margins, between T4-T7. The pubescence on sternal margins is very similar to males of *Tapinotaspoides,* without special fringes or hairs. Despite the absence of oil on legs, scopa or tergal fringes, these conspicuous pubescence strongly suggest that they collect and store oil on this plumose pubescence. COCUCCI (1991) observed numerous species of *Nierembergia,* without records of males of *T. chalybaea* on its flowers. This species is not very common and the potential protandry may difficult to find the males.

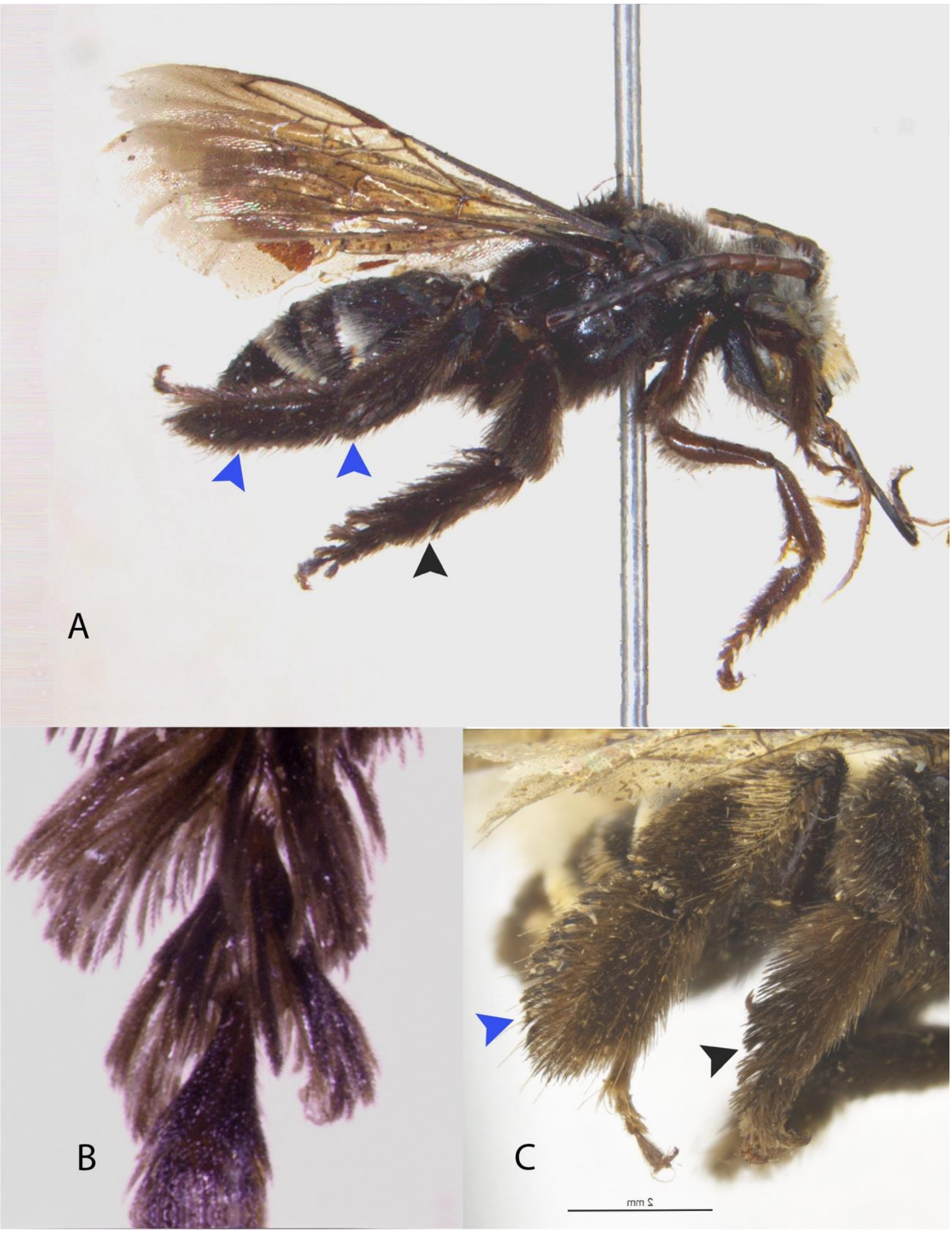


Fig. 9. *Tapinotaspis chalybaea*, (Friese, 1899) male: **(A)** lateral habitus, **(B)** mid tarsi, **(C)**, mid and hind legs; blue setae, scopa of hind leg; black setae, oil collecting apparatus of mid legs.

Chalepogenus lineage

The genus *Chalepogenus* include the type species *Chalepogenus muelleri* and related species from at least two or three clades according to the main classification of the genus (ROIG-ALSINA, 1999; COCUCCI *et al.*, 2000, AGUIAR *et al.*, 2020). The first reports about males of *Chalepogenus* collecting oil was performed by COCUCCI (1991) who first reported that the males frequent the flowers of *Nierembergia* for copulation and sleeping places. He also list males with hind legs soaked with oil, with the more common one the species *Chalepogenus cocucci* (cited as *Tapinotaspis* (*Tapinorhina*) spec. 1). On the review of ROIG-ALSINA (1999), he reported males of nine species of *Chalepogenus* on oil flowers of *Calceolaria*, *Nierembergia, Cypella,* and *Sysinrichium*. We reported an male of *Chalepogenus calceolariae* on oil flowers of *Calceolaria* (Fig. 10), and the shiny scopa suggests strongly that he was collecting oil. The male has the mesoscutum very charged of pollen, also suggesting that the bee perform the same behavior of females that provide pollination on these plants. It is not uncommon to observe males with mesoscutum and frons loaded with pollen, collected on *Calceolaria* flowers, suggesting that these bees not also collect the oil as are also pollinators. An further study about the identity of the pollen from numerous specimens with loads of pollen on mesoscutum and frons can reinforce the function of these bees as pollinators. Similar to females, *Chalepogenus* has a dense plumose pubescence on the fore and mid basitarsi, and a very dense and developed scopa (Fig. 11).

Fig. 10. *Chalepogenus* sp., male, on *Calceolaria* sp. (Calceolariaceae), **(A)** with the mesoscutum very charged of pollen (the left) and **(B)** leaving the flower (the right).

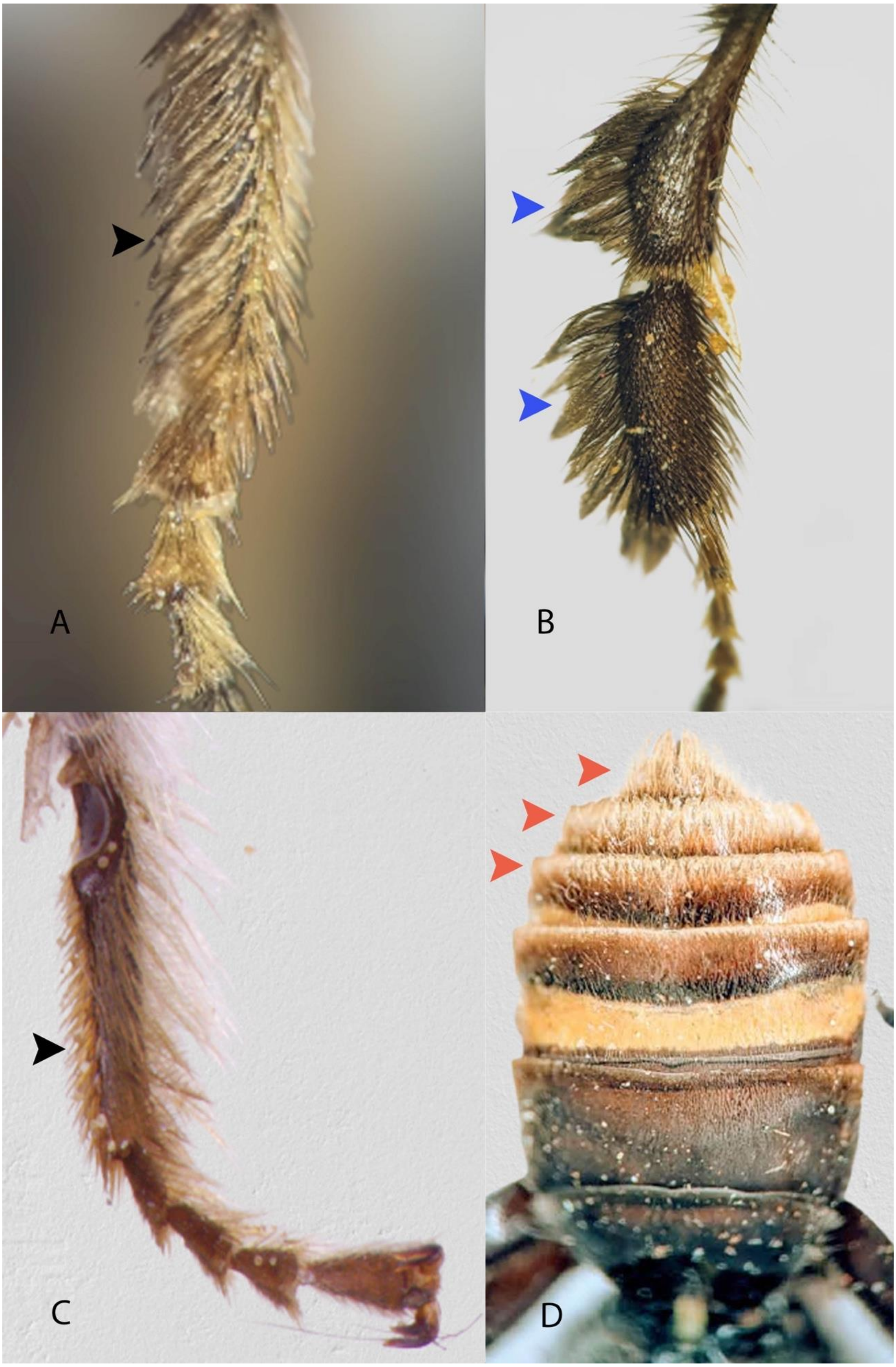


Fig. 11. (A) *Chalepogenus cocucci* Roig-Alsina, 1999 male, fore basitarsus; **(B)** and **(D)**, *Chalepogenus muelleri* (Friese, 1899) male, hind leg and tergal fringes; **(C)**, *Chalepogenus herbstii* (Friese, 1906) male, fore basitarsus.

Lanthanomellissa lineage

We studied the males of four species of *Arhysoceble,* two species of *Lanthanomelissa* and one species of *Lanthanella.* The lineage *Lanthanomelissa* comprehends the genera *Lanthanomelissa*, *Arhysoceble* and *Lanthanella.* These bees have a strong relation with the oil flowers species of the family Iridaceae and Solanaceae (COCUCCI *et al.,* 2000), and the males are commonly observed patrolling, mating and collecting oil over the oil flowers (COCUCCI, 1991). The short and foliaceous setae of *Lanthanomelissa* are similar to those seen in females of *Rediviva albifasciata* Whitehead & Steiner 1994 and *Redivivia intermixta* (Cockerell, 1934), endemic to South Africa and Lesotho, which belong to the family Melittidae (Kuhlmann 2014; KUHLMANN & HOLLENS, 2014). In *R. albifasciata,* the foliaceous setae, in the shape of a plate, is used to rupture the elaiophores of *Colpias mollis* E. Mey. ex. Benth. or different species of *Hemimeris*, both from the Scrophulariaceae family (STEINER & WHITEHEAD, 2002; KUHLMANN & HOLLENS, 2014).

Males of *Lanthanomelissa* have the same oil collecting apparatus as females, however smaller in size, which also allow them to oil collecting behavior. We observed males with oil on tergal and sternal fringes, however interesting some few males also possess pollen mixed with the oil on tergal fringes. This pollen could be considered as an additional resource storage to attract the females. On stingless bees, tribe Meliponini, it is not rare to observe males with pollen to access the females in the colony (BOONGIRD & MICHENER, 2010). These authors observed males of distinct species of the tribe Meliponini (*Tetragonula collina, Tetragonula fuscobalteata*, *Tetragonula pagdenina* e *Heterotrigona apicalis*) resting and flying over the colonies, allowing entrance only the males that have pollen or propolis on their tibia. These males propably colletc these resources on flowers and have more chance to access the new emerged virgin queens.

The oil collecting behavior of the females of *Lathanomelissa* on the flowers of *Sisyrinchium* was described by COCUCCI & VOGEL (2001). The genus is specilized on the flowers of this Iridaceae, and is common to observe males patrolling the flowers searching for females with aggressive behavior between them. Males and females have the same oil collecting apparatus, however distinctly smaller on males. As the same way that was observed on *Chalepogenus*, males of *Lanthanomellissa* use the flowers of *Sisyrinchium* as sleeping shelters (PINHEIRO *et al.*, 2017).

The species of the genus *Arhysoceble* are specially abundant in the grassland fields of Cerrado, with strong interactions with numerous oil plant families, like Malpighiaceae, Kramericeae, and Plantaginaceae, however only in the flowers of oil genera of Iridaceae, we were able to record males commonly collecting oil. The males of *Arhysoceble* present the same pattern of oil collecting apparatus of the females, with a strong comb on outer margin of fore basitarsus, and a dense brush of plumose setae on inner concave surface. We observed numerous males of distinct species (three species) of *Arhyscoceble* collecting oil from trichomatic elaiophores of *Cipura xanthomelas* and *Cipura* sp. (Iridaceae). The males perform the same female behavior on the flowers, landing over the inner tepal and grasping the oil trichomes with the fore legs. However the transference of the oil is clearly distinc, where the males, transfer the oil for the terga and sterna fringes (Fig. 12, A-E). The male observed display the same behavior described by Cappellari et al. (2012) for *Tetrapedia* males in the transference of the oil from the fore legs to tergal and sternal scopa. The males have abundant pollen over the frons and mesossoma, and act as pollinators on *Cipura xanthomellas* and *Cipura* sp., however the bees commonly clean their bodies after each visit and eliminate most of pollen. The males patrol the flowers searching for females, and potentially have the copula on flowers.

Fig. 12. *Arhysoceble* sp., male, on *Cipura xanthomellas* Klatt (Iridaceae), performing oil collecting **(A)** and transference from hind legs to sternal and tergal fringes **(B-F)**.

Paratetrapedia lineage

The *Paratetrapedia* lineage is composed by numerous genera, at least seven, with a conserved pattern of morphology of the oil collecting apparatus of the fore legs and a variable density of plumose hairs related to oil collecting on mid legs, hind legs, and fringes of terga and sterna. The variation of the oil collecting apparatus of these bees is poorly known and underestimated. According to VOGEL (1974) and ROIG-ALSINA (1997), these bees collect oil with a strong comb on the outer margin of the fore basitarsus. The records of females of *Paratetrapedia* and *Tropidopedia* visiting flowers of Malpighiaceae are very common (NEFF & SIMPSON, 1981; SIGRIST & SAZIMA, 2004; AGUIAR *et al.*, 2017; POSSOBOM & MACHADO, 2018), however these bees are not considered pollinators (SIGRIST & SAZIMA, 2004; POSSOBOM & MACHADO, 2018). Females and males access the elaiophores of Malpighiaceae through the abaxial part of the flowers, without any contact to reproductive parts. We observed numerous males of *Paratetrapedia punctata* patrolling and visiting flowers of *Banisteriopsis campestris,* in the same period when numerous females were also observed. The males of *Paratetrapedia* and disctinct lineages must be more closely related to oil glands of numerous genera of Oncidinae (Orchidaceae) which are very common in forested areas where these bees are abundant.

The scopa of hind legs on *Paratetrapedia* and *Tropidopedia* are very developed, with numerous dense plumose hairs (Fig. 13, eg. male of *Tropidopedia peruana*). The males of the species of *Tropidopedia* group *flavipennis* have oil collecting apparatus, including hind tibial scopa, much more enlarged than in females. The special structure observed that is not developed on females, is the high density of plumose hairs on mid basitarsi composing an developed scopa on the mid leg.

Fig. 13. Male of *Tropidopedia peruana* Aguiar & Melo, 2007 showing the very developed hind tibia scopa (blue arrows), with dense, fine plumose hairs (red arrows).

Some oil was observed on the fringes of sterna of *Paratetrapedia punctata* (S5-S6), *Tropidopedia nigrocarinata* (S5) and *Tropidopedia punctifrons* (S2-S4). Morphology and behavior work together to allow distinct species to access the better fitness during the source search and collect. The oil collecting morphology of males have the exactly the distinct direction of the development compared to females, due to a smaller pressure to perform the oil collecting behavior.

The few observations about mating on the species of *Paratetrapedia* lineage, suggests that the males mate with the females on the flowers. The males of *Xanthopedia swainsonae* were not observed on nesting aggregations and the authors suggest that the matting place were on flowers (ROZEN & MICHENER, 1988). We observed an tentatively couple of *Lophopedia* sp. on flowers of *Byrsonima verbasifolia*, and numerous males of

Paratetrapedia punctata patrolling over the flowers of different species of Malpighiaceae.

CONCLUSION REMARKS

The current knowledge about the oil collecting behavior of males do not have a consensus for which purpose the males collect oil. CAPELLARI *et al.* (2012), registered males of six species of *Tetrapedia*, on Malpighiaceae flowers on Cerrado and strongly suggested that the oil must be used in matting behavior, but was not consensus if it can be used as a fragrance to access the females or a nuptial gift. One distinct hypothesis was not evaluated by the authors, which was the use of the oil as anti-predacious. The oil could act as anti-predacious as it can act as anti-herbivory. Our observations support this possible hypothesis, because on many dormitories observed with males of distinct genera of Tapinotaspidini and Tetrapediini, we observed the scopa and terga of males full of oil (see also chapter about male aggregations).

The oil collecting behavior and specialized morphology represent covergences of behavior and also morphology of some characters betweeen the tribes Tapinotaspidini and Tetrapediini, however we can observe numerous synapomorphic characters on distinct genera and lineages. The more common convergence is the hind tibial scopa that is developed in all species of both tribes, and is not observed any similar development in any of the remaining tribes of oil collecting bees.

References

AGUIAR, A. J. C.; MELO, G. A. R. 2007. Taxonomic revision, phylogenetic analysis, and biogeography of the bee genus *Tropidopedia* (Hymenoptera, Apidae, Tapinotaspidini). **Zoological Journal of Linnean Society.** 151:511-554.

AGUIAR, A. J. C. 2012. New bee species of the genus *Monoeca* (Hymenoptera, Apidae, Tapinotaspidini) from Brazil. **Zootaxa**. 3564(3564):17-32.

AGUIAR, A. J. C.; MELO, G. A. R.; VASCONCELOS, T. N. C.; GONÇALVES, R. B., GIULIANO, L. & MARTINS, A. C. 2020. Biogeography and early diversification of Tapinotaspidini oil-bees support presence of Paleocene savannas in South America. Molecular **Phylogenetics and Evolution**. 143:106692. https://doi.org/10.1016/j.ympev.2019.106692

ALCOCK, J.; BARROWS, E. M.; GORDH, G.; HUBBARD, L. J.; KIRKENDALL, L.; PYLE, D. W.; PONDER, T. L. & ZALOM, F. G. 1978. The ecology and Evolution of male reprodutive behavior in the bees and wasps. **Zoological Journal of the Linnean Society**. 64:293-326.

BEMBÉ, B. 2004. Functional morphology in male euglossine bees and their ability to spray fragrances (Hymenoptera, Apidae, Euglossini). **Apidologie**. 35: 283-291.

BOONGIRD, S. & MICHENER, C. D. 2010. Pollen and Propolis Collecting by Male Stingless Bees (Hymenoptera: Apidae). **Journal of the Kansas Entomological Society.** 83:47-50.

BOSSERT, S.; MURRAY, E.; ALMEIDA, E.A.; BRADY, S.; BLAIMER, B.B. & DANFORTH, B. 2019. Combining transcriptomes and ultraconserved elements to illuminate the phylogeny of Apidae. **Molecular phylogenetics and evolution.** 131:121-131.

BUCHMANN, S. 1987. The ecology of oil flowers and their bees. **Annual Review of Ecology, Evolution, and Systematics.** 18:343-369.

Cane, J. H.; Eickwort, G. C.; Wesley, F. R. & Spielholz, J. 1983. Foraging, grooming and mating behaviours of *Macropis nuda* (Hymenoptera, Melittidae) and use of *Lysimachia ciliata* (Primulaceae) oils in larval provisions and cell linings. **American Naturalist**. 110:257-264.

Cane, J. H.; Sampson, B. J. & Miller, S. A. 2011. Pollination Value of Male Bees: The Specialist Bee *Peponapis pruinosa* (Apidae) at Summer Squash (*Cucurbita pepo*). **Environmental Entomology**. 40(3):614-620. https://doi.org/10.1603/EN10084

CAPPELLARI, S, C.; MELO, G. A. R.; AGUIAR, A. J. C. & NEFF, J. L. 2012. Floral oil collection by male *Tetrapedia* bees (Hymenoptera: Apidae: Tetrapediini). **Apidologie**. 43:39-50.

CARDINAL, S., STRAKA, J., & DANFORTH, B.N. 2010. Comprehensive phylogeny of apid bees reveals the evolutionary origins and antiquity of cleptoparasitism. **Proceedings of the National Academy of Sciences**. 107:16207-16211.

CARNEIRO, L. T.; ANDRÉ, C. B. D. S.; TAKAHASI, A. & ALVES-DOS-SANTOS, I. 2019. Interactions between oil-collecting bees and *Krameria grandiflora* (Krameriaceae) with emphasis on the role of specialized floral traits in the mutual fit. **Arthropod-Plant Interactions**. 13(2):213-226. https://doi.org/10.1007/s11829-019-0989-w

COCUCCI, A. 1991. Pollination biology of *Nierembergia* (Solanaceae). **Plant Systematics and Evolution**. 174(1/2), 17-35, 1991.

COCUCCI, A. A., SÉRSIC, A., ROIG-ALSINA, A. 2000. Oil-collecting structures in Tapinotaspidini: their diversity, function and probable origin. **Mitteilungen der Münchner Entomologischen Gesellschaft.** 90:51-74.

COCUCCI A. A., VOGEL, S. Oil producing flowers of *Sisyrinchium* species (Iridaceae) and their pollinators in southern South America. **Flora**, 196:26-46, 2001. https://doi.org/10.1016/S0367-2530(17)30010-5

CUNHA, R.; BLOCHTEIN, B. Life history of *Monoeca xanthopyga* Harter-Marques, Cunha & Moure (Hymenoptera, Apidae, Tapinotaspidini) at the Araucária Plateau, Rio Grande do Sul, Brazil. **Revista Brasileira de Zoologia.** 1:107-113, 2003.

DANFORTH, B.; MINCKLEY, R.; NEFF, J. & FAWCETT, F. 2019. **The Solitary Bees: Biology, Evolution, Conservation**. Princeton University Press, 488 pp.

DODSON, C. H; DRESSLER, R. L; HILLS, H. G., ADAMS, R. M. & WILLIAMS, N. H. 1969. Biologically active compounds in orchid fragrances. **Science**. 164, 1243–1249.

EICKWORT, G. C. & GINSBERG, H. S. 1980. Foraging and mating behavior in Apoidea. **Annual Review of Entomology**. 25:421-446.

ELTZ, T; JOSTEN, S. & MENDE, T. 2019. Stored perfume dynamics and consequences for signal development in male orchid bees. **Journal of Comparative Physiology A**. 205:311-320.

KUHLMANN, M. 2014. Nest architecture and use of floral oil in the oil-collecting South African solitary bee *Rediviva intermixta* (Cockerell) (Hymenoptera: Apoidea: Melittidae). **Journal of Natural History**. 48:2633-2644.

KUHLMANN, M., HOLLENS, H. 2014. Morphology of oil-collecting pilosity of female *Rediviva* bees (Hymenoptera: Apoidea: Melittidae) reflects host plant use. **Journal of Natural History**. 49:561-657.

MICHENER, C. D. **The bees of the world**. The Johns Hopkins University Press, Baltimore, 953 pp, 2007.

NEFF, J. L. & SIMPSON, B. B. 1981. Oil-collecting structures in the Anthophoridae: Morphology, function and use in systematics. **Journal of the Kansas Entomological Society**. 54:95-123.

NEFF, J. L. & SIMPSON, B. B. 2017. Vogel's great legacy: The oil flower and oil-collecting bee syndrome. **Flora**, 232:104-116. https://doi.org/10.1016/j.flora.2017.01.003

PEREIRA, M.; GARÓFALO, C. A.; CAMILLO, E. & SERRANO, J. C. 1999. Nesting biology of *Centris (Hemisiella) vittata* Lep. in southeastern Brazil (Hymenoptera, Apidae, Centridini). **Apidologie**. 30:327-338.

PINHEIRO M.; ALVES-DOS-SANTOS, I. & SAZIMA, M. 2017. Flowers as sleeping places for male bees: somehow the males know which flowers their females prefer. **Arthropod Plant Interact**. 11:329-337. https://doi.org/10.1007/s11829-017-9532-6

POSSOBOM, C. C. F. & MACHADO, S. R. 2018. Elaiophores in three Neotropical Malpighiaceae species: a comparative study. **Plant Systematics and Evolution**. 304:15-32.

ROIG-ALSINA, A. A. 1997. generic study of the bees of the tribe Tapinotaspidini, with notes on the evolution of their oil-collecting structures. **Mitteilungen der Münchner Entomologischen Gesellschaft**. 87:3-21.

ROSWELL, M.; DUSHOFFI, J. & WINFREE, R. 2019. Male and female bees show large diferences in floral preference. **PloS One**. 4;14(6):e0217714.

ROUBIK, D. M. 1989. **Ecology and natural history of tropical bees**. Cambridge University Press, Cambridge, 514 pp.

SCHINDLER, M., HOFMANN, M., WITTMANN, D., & RENNER, S.S. 2018. Courtship behaviour in the genus *Nomada* - antennal grabbing and possible transfer of male secretions. **Journal of Hymenoptera Research**. 65:47-59.

SIGRIST, M. R.; SAZIMA, M. 2004. Pollination and Reproductive Biology of Twelve Species of Neotropical Malpighiaceae: Stigma Morphology and its Implications for the Breeding System. **Annals of Botany**, 94:33-34.

SINGER, R.B., COCUCCI, A. 1999. Pollination mechanisms in four sympatric southern Brazilian Epidendroideae orchids. **Lindleyana**. 14,47-56.

SMITH GP, BRONSTEIN JL, PAPAJ DR. 2019. Sex differences in pollinator behavior: Patterns across species and consequences for the mutualism. **Journal of Animal Ecology**. 88(7):971-985.

STEINER KE, WHITEHEAD VB. 2002. Oil secretion and the pollination of *Colpias mollis* (Scrophulariaceae). **Plant Systematics and Evolution**. 235:53-66.

TORO, H. & MAGUNACELAYA, J.C. 1987. Estructura muscular femoral de Xeromelissinae (Hymenoptera, Colletidae). **Acta Entomológica Chilena**. 14:13-24.

VINSON, S. B.; WILLIAMS, H. J.; FRANKIE, G. W.; SHRUM, G. 1997. Floral lipid chemistry of *Byrsonima crassifolia* (Malpighiaceae) and a use of floral lipids by *Centris* bees (Hymenoptera: Apidae). **Biotropica**, 29:76-83.

VOGEL, S. 1971. Olproduzierende Ölproduzierende Blumen, die durch ölsammelnde Bienen bestäubt werden. **Naturwissenschaften**. 58:58-59.

VOGEL, S. 1974. Ölblumen und ölsammelnde Bienen. **Tropische und Subtropische Pflanzenwelt**. 7:283-547.

WATTS, S.; SAPIR, Y.; SEGAL, B.; DAFNI, A. 2013. The endangered *Iris atropurpurea* (Iridaceae) in Israel: honey-bees, night-sheltering male bees and female solitary bees as pollinators. **Annals of Botany**. 111:395-407.

WCISLO, W., MINCKLEY, R.L., & SPANGLER, H.C. 1992. Pre-copulatory courtship behavior in a solitary bee, *Nomia triangulifera* Vachal (Hymenoptera: Halictidae). **Apidologie**. 23: 431-442.

WILLMER, P. G.; STONE, G. N. 2004. Behavioral, Ecological, and Physiological Determinants of the Activity Patterns of Bees. **Advances in The Study of Behavior**. 34:347-466.

WHITEHEAD VB, STEINER KE. 1994. A new *Rediviva* bee (Hymenoptera, Melittidae) from the north-Western Cape Province, South Africa. **Annals of the South African Museum**. 104:1-11

Table 1. Species of Tapinotaspidini and Tetrapediini that have their males studied, and respective structures related to oil collecting and transport. On metasoma, we used S1-S6 to refer to distinct elements of sterna, and T1-T6 to distinct elements of terga.

Species	**Legs**	**Sterna**	**Terga**
Tapinotaspidini			
Arhysoceble huberi	fore, mid, hind	S2, S3, S4, S5	T6
Arhysoceble picta	fore, hind	S3, S4, S5, S6S6	T3
***Arhysoceble* sp.**	hind	-	-
Caenonomada unicalcarata	fore, mid, hind	S5	T6, T7
Chalepogenus caeruleus	fore, mid, hind	S3, S4, S5	T2, T3, T4
Chalepogenus muelleri	fore, mid, hind	S2, S3, S4, S5	T5, T6, T7
Chalepogenus nigripes	fore, mid, hind	S3, S4, S5	T2, T3, T4, T5, T6
Lanthanomelissa parva	mid, hind	S4	
Lanthanomelissa betinae	fore, mid, hind	S2, S3, S4, S5	T2, T3, T4, T5, T6
Lanthanomelissa clementis	fore, mid, hind	S2, S3, S4, S5, S6S6	T2, T3, T4, T5, T6, T7
Lophopedia flava	fore, hind	-	-
Lophopedia nigrispinis	mid, hind	S3, S4, S5	-
Lophopedia pygmaea	fore, mid, hind	S4, S5, S6S6	T5, T6
Monoeca haemorrhoidalis	fore, mid	S3, S4, S5	T2, T3, T6
Monoeca mourei	fore, mid, hind	S2, S3, S4	T3, T4, T5, T6, T7
Monoeca pluricincta	fore, mid, hind	S2, S3, S4, S5, S6S6	T3, T4, T5, T6, T7
Paratetrapedia connexa	fore, mid, hind	S3, S4, S5	T3, T4, T5, T6
Paratetrapedia duckei	hind	S4	T6, T7
Paratetrapedia fervida	fore, mid, hind	S2, S3, S4, S5, S6S6	T2, T3, T5, T6, T7
Paratetrapedia flaveola	fore, mid, hind	S2, S3, S4, S5, S6S6	T4, T5, T6
Paratetrapedia lugubris	fore, mid, hind	S3, S4, S5, S6S6	T4, T5
Paratetrapedia punctata	fore, mid, hind	S2, S3, S4, S5, S6S6	-
Paratetrapedia testacea	fore, mid, hind	S2, S3, S4, S5, S6S6	-
Paratetrapedia volatilis	fore, mid, hind	S3, S4, S5, S6S6	-
Tapinotaspis chalybaea	fore, mid, hind	S3, S4, S5	T2, T3, T4, T5, T6
Tapinotaspoides serraticornis	fore	S2, S3, S4, S5	T5, T6, T7
Tapinotaspoides tucumana	fore, hind	S2, S3, S4, S5	T6
Trigonopedia ferruginea	fore	S2, S3, S4, S5	T7
Tropidopedia carinata	fore, mid, hind	S2	T5, T6, T7
Tropidopedia flavolineata	fore, mid, hind	S3, S6S6	T5, T6, T7
Tropidopedia punctifrons	fore, mid, hind	S4	T5, T6
Urbanapis danunciae	fore, mid, hind	S2, S3, S4	T2, T3, T4, T5, T6
Urbanapis diamantina	fore, mid, hind	S2, S3, S4, S5, S6S6	T2, T3, T4, T5, T6, T7
Xanthopedia* cf. *larocai	fore, mid, hind	S5, S6S6	T4, T5, T6, T7
Tetrapediini			
Tetrapedia amplitarsis	fore, hind	-	T5, T6, T7
Tetrapedia diversipes	fore, hind	-	T5, T6, T7
Tetrapedia peckoltii	fore, mid, hind	-	T5, T6, T7
Tetrapedia* aff. *peckoltii	fore, mid, hind	S3, S4	T5, T6, T7
***Tetrapedia* sp. 1**	fore, mid, hind	S3, S4	T5, T6, T7
***Tetrapedia* sp. 2**	hind	-	T5, T6, T7
***Tetrapedia* sp. 3**	fore, hind	-	

Table 2. Records of males of the tribes Tapinotaspidini and Tetrapediini collected on oil flowers.

Species	Specimens	Oil records	Locality	Reference
Tapinotaspidini				
Arhysoceble huberi	1	*Angelonia arguta* (Plantaginaceae)	Flores de Goiás (GO)	DZUB
***Arhysoceble* spp.**	-	*Cipura xanthomelas* (Iridaceae)	Cavalcante (GO)	Present study
	-	*Cipura xanthomelas* (Iridaceae)	Flores de Goiás (GO)	Present study
	-	*Cipura* sp. (Iridaceae)	Cavalcante (GO)	Present study
Caenonomada bruneri	-	*Angelonia salicariifolia* (Plantaginaceae)	Porto Murtinho (MS)	Aguiar & Melo (2009)
	-	*Monopera perennis* (Plantaginaceae)	Porto Murtinho (MS)	Aguiar & Melo (2009)
Caenonomada unicalcarata	-	*Angelonia salicariifolia* (Plantaginaceae)	Porto Murtinho (MS)	Aguiar & Melo (2009)
	-	*Monopera perennis* (Plantaginaceae)	Porto Murtinho (MS)	Aguiar & Melo (2009)
	3	*Angelonia* sp. (Plantaginaceae)	Flores de Goiás (GO)	Present study
Chalepogenus muelleri		*Cypella herbertii* (Iridaceae)	-	Roig-Asina (1999)
Chalepogenus cocucci	1	*Nierembergia aristata* (Solanaceae)	Tucuman (Argentina)	Cocucci (1991)
	12	*Nierembergia hippomanica* (Solanaceae)	Tucuman (Argentina)	Cocucci (1991)
	7	*Nierembergia stricta* (Solanaceae)	Tucuman (Argentina)	Cocucci (1991)
	11	*Nierembergia gracilis* (Solanaceae)	Tucuman (Argentina)	
Chalepogenus vogeli	2	*Calceolaria schickendantziana* (Calceolariaceae)	Tucuman, Salta (Argentina)	Roig-Alsina (1999)
Chalepogenus rufipes	1	*Calceolaria ruiz-pavonii* (Calceolariaceae)	Argentina	Roig-Alsina (1999)
	1	*Calceolaria schickendantziana* (Calceolariaceae)	Argentina	Roig-Alsina (1999)
Chalepogenus calceolariae	3	*Calceolaria* sp. (Calceolariaceae)	Peru	Roig-Alsina (1999)
Chalepogenus rasmusseni	3	*Calceolaria* cf. *tripartida* (Calceolariaceae)	Hunuco (Peru)	Roig-Alsina (1999)
Chalepogenus herbsti	-	*Sisyrinchium arenarium* (Iridaceae)	-	Roig-Alsina (1999)
Chalepogenus rozeni	29	*Calceolaria thyrsiflora* (Calceolariaceae)	Coquimbo (Chile)	Roig-Alsina (1999)
Chalepogenus perimelaena	2	*Calceolaria* sp. (Calceolariaceae)	Mendonza (Argentina)	Roig-Alsina (1999)
Lanthanomelissa betinae	4	*Sisyrinchium* sp. (Iridaceae)	Curitiba (PR)	DZUP

Lanthanomelissa clementis	1	*Nierembergia* sp. (Solanaceae)	Pinheiro Machado (RS)	RPSP
Lanthanomelissa	1	*Sisyrinchium micranthum* (Iridaceae)	Caçapava (RS)	UFFS*
discrepans	2	*Sisyrinchium ostenianum* (Iridaceae)	Caçapava (RS)	UFFS*
	4	*Sisyrinchium scariosum* (Iridaceae)	Caçapava (RS)	UFFS*
Lanthanomelissa magaliae	2	*Sisyrinchium micranthum* (Iridaceae)	Caçapava (RS)	UFFS*
	2	*Sisyrinchium setaceum* (Iridaceae)	Caçapava (RS)	UFFS*
Lanthanella neffi	-	*Calceolaria* sp. (Calceolariaceae)	Argentina	Roig-Alsina (1999)
	-	*Nierembergia* sp. (Solanaecae)	Argentina	Roig-Alsina (1999)
Monoeca mourei	3	*Banisteriopsis* sp. (Malpighiaceae)	Serra do Salitre (MG)	Present study
Monoeca pluricincta	1	*Banisteriopsis* sp. (Malpighiaceae)	Gama (DF)	Present study
Monoeca xanthopyga.	-	*Oncidium* sp. (Orchidaceae)	São Francisco de Paula (RS)	Cunha & Blochtein (2003)
			Cambará do Sul (RS)	Cunha & Blochtein (2003)
Paratetrapedia connexa	1	*Banisteriopsis* sp. (Malpighiaceae)	Sobradinho	Present study
Paratetrapedia punctata	1	*Banisteriopsis argyrophylla*	Cavalcante (GO)	Present study
	2	(Malpighiaceae)	Brasília (DF)	Present study
	1	*Banisteriopsis campestris* (Malpighiaceae)	Brasília (DF)	DZUB
	1	*Banisteriopsis stellaris* (Malpighiaceae)	Brasília (DF)	DZUB
	1	*Banisteriopsis* sp. (Malpighiaceae)	Brasília (DF)	DZUB
	1	*Byrsonima coccolobifolia* (Malpighiaceae)	Brasília (DF)	DZUB
	1	*Byrsonima guilleminiana* (Malpighiaceae)	Brasília (DF)	DZUB
	1	*Byrsonima rigida* (Malpighiaceae)	Brasília (DF)	DZUB
	1	*Byrsonima subterranea* (Malpighiaceae)	Brasília (DF)	DZUB
	2	*Byrsonima verbascifolia* (Malpighiaceae)	Brasília (DF)	DZUB
		Camarea affinis (Malpighiaceae)		
Tropidopedia flavolineata	1	*Banisteriopsis argyrophylla*	Brasília (DF)	Present study
	2	(Malpighiaceae)	Cavalcante (GO)	Present study
		Banisteriopsis gardneriana (Malpighiaceae)		
Tropidopedia nigrocarinata	1	*Byrsonima laxiflora* (Malpighiaceae)	Brasília (DF)	DZUB
Tropidopedia punctifrons	3	*Banisteriopsis* sp. (Malpighiaceae)	Brasília (DF)	DZUB
	1	*Banisteriopsis* sp. (Malpighiaceae)	Serra do Salitre (MG)	DZUB
	1	*Byrsonima crassa* (Malpighiaceae)	Brasília (DF)	Present study

	1	*Byrsonima pachyphylla* (Malpighiaceae)	Brasília (DF)	DZUB
	1	*Byrsonima verbascifolia* (Malpighiaceae)	Brasília (DF)	DZUB
	1	*Diplopterys pubipetala* (Malpighiaceae)	Brasília (DF)	DZUB
	1	*Peixotoa goiana* (Malpighiaceae)	Brasília (DF)	DZUB
	5	*Peixotoa* sp. (Malpighiaceae)	Brasília (DF)	DZUB
Urbanapis diamantina	3	*Byrsonima* sp. (Malpighiaceae)	Palmeiras (BA)	DZUB
Xanthopedia* cf. *larocai	1	*Byrsonima sericea* (Malpighiaceae)	Serra do Salitre (MG)	Present study
Tetrapediini				
Tetrapedia amplitarsis	2	*Byrsonima* sp. (Malpighiaceae)	Palmeiras (BA)	DZUB
Tetrapedia curvitarsis	2	*Byrsonima brachybotrya* (Malpighiaceae)	Tibagi (PR)	Cappellari et al (2012)
Tetrapedia diversipes	1	*Banisteriopsis campestris* (Malpighiaceae)	Brasília (DF)	Cappellari et al (2012)
	1	*Heteropterys pteropetala* (Malpighiaceae)	Brasília (DF)	Cappellari et al (2012)
	4	*Heteropterys tomentosa* (Malpighiaceae)	Brasília (DF)	Cappellari et al (2012)
	1	*Peixotoa goiana* (Malpighiaceae)	Brasília (DF)	Cappellari et al (2012)
	2	*Banisteriopsis malifolia* (Malpighiaceae)	Jaboticatubas (MG)	DZUB
	1	*Oncidium paranaense* (Orchidaceae)	Curitiba (PR)	Singer & Cocucci (1999)
Tetrapedia peckoltii	2	*Banisteriopsis argyrophylla* (Malpighiaceae)	Cavalcante (GO)	Present study
	2	*Banisteriopsis malifolia* (Malpighiaceae)	Jaboticatubas (MG)	DZUB
Tetrapedia* aff. *peckoltii	1	*Byrsonima coccolobifolia* (Malpighiaceae)	Serra do Salitre (MG)	Cappellari et al (2012)
	2	*Banisteriopsis campestris* (Malpighiaceae)	Brasília (DF)	Cappellari et al (2012)
	1	*Byrsonima basiloba* (Malpighiaceae)	Brasília (DF)	Cappellari et al (2012)
	1	*Heteropterys tomentosa* (Malpighiaceae)	Brasília (DF)	Cappellari et al (2012)
	1	*Banisteriopsis gardneriana* (Malpighiaceae)	Brasília (DF)	Present study
	3	*Banisteriopsis argyrophylla* (Malpighiaceae)	Cavalcante (GO)	Present study
***Tetrapedia* sp. 1**	1	*Banisteriopsis gardneriana* (Malpighiaceae)	Brasília (DF)	Present study
	3	*Banisteriopsis malifolia* (Malpighiaceae)	Jaboticatubas (MG)	DZUB
***Tetrapedia* sp. 2**	1	*Heteropterys tomentosa* (Malpighiaceae)	Brasília (DF)	Cappellari et al (2012)
	1	*Banisteriopsis gardneriana* (Malpighiaceae)	Brasília (DF)	Present study

ANNEX

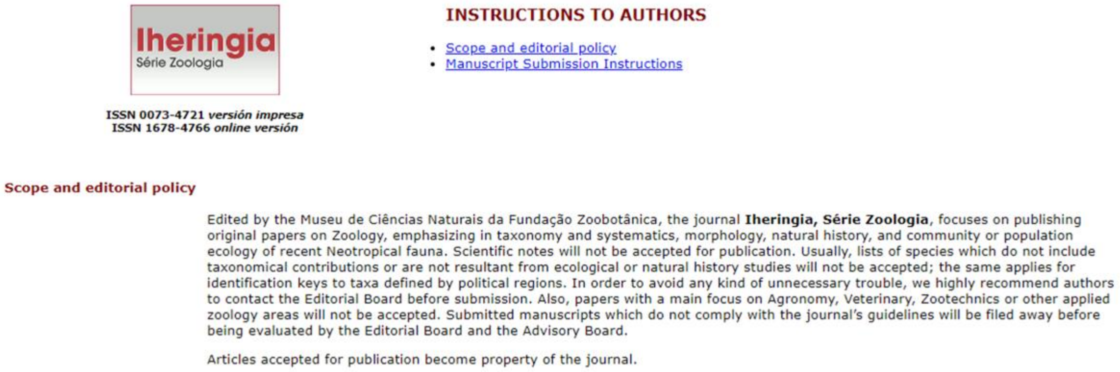
Capítulo 2

Oil collecting males of the Tapinotaspidini and

Tetrapediini bee tribes: morphology and behavior

Annex 1

Focus and scope of the potential journal to which we intend to submit our manuscript.



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INSTRUCTIONS TO AUTHORS

- Scope and editorial policy
- Manuscript Submission Instructions

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CAPÍTULO 3

Agregações e dormitórios: onde e como dormem os machos das abelhas?

Manuscrito formatado segundo as normas da revista Arthropod-Plant Interaction (Anexo 1)

RESUMO

As fêmeas de abelhas solitárias geralmente passam a noite nos ninhos que constroem ao longo do dia. Por sua vez, os machos podem ser encontrados dormindo em agregações temporárias, dentro de flores ou cavidades. A estrutura das agregações pode variar muito em relação ao substrato, número de indivíduos e duração, enquanto o comportamento dos machos dentro das flores pode definir o seu papel como polinizadores efetivos ou potenciais, ou como visitantes florais. No presente estudo apresentamos dados originais acerca do comportamento dos machos de nove tribos de abelhas solitárias (Apidae e Megachilidae) que dormem em agregações ou em flores no Cerrado do Brasil Central, e discutimos as possíveis razões pelas quais os machos apresentam tais comportamentos.

Palavras-chave: abelhas solitárias, abrigo noturno, Tapinotaspidini, *Tetrapedia*.

ABSTRACT

Female solitary bees usually spend the night in the nests they build throughout the day. However, males can be found sleeping in aggregations, inside flowers or cavities. The structure of the aggregations can vary greatly with respect to substrate, number of individuals and duration. While the behaviour of males within flowers may define their role as effective or potential pollinators or flowers visitors only. In this study we present original data on the behavior of males from nine bee tribes (Apidae and Megachilidae) that sleep in aggregations or flowers in the Central Brazilian Cerrado. We discuss the possible reasons why males present such behaviors.

Keywords: night shelter, solitary bees, Tapinotaspidini, *Tetrapedia*.

INTRODUÇÃO

A maior parte das fêmeas de abelhas solitárias passam a noite dentro dos ninhos que constroem ao longo do dia (Evans & Linsley 1960; Alcock 1998; Danforth et al. 2019). No entanto, de maneira oposta, os machos podem ser encontrados dormindo em agregações temporárias, dentro de flores ou cavidades (Evans & Linsley 1960; Linsley 1962; Dafni et al. 1981; Sapir et al. 2005; Pinheiro et al. 2017). Na literatura, a maioria dos registros de machos observados dormindo foram realizados em flores. Embora as flores sejam dormitórios de machos, outros locais, como folhas, ninhos abandonados, cavidades pré-existentes também podem servir de abrigo durante à noite (Danforth et al. 2019). A formação de agregações representa um fenômeno interessante, uma vez que durante o dia os machos não apresentam qualquer comportamento social, pelo contrário, competem entre si para ter acesso às fêmeas (Hurd & Linsley 1976; Alcock 1998). As agregações geralmente são monoespecíficas, embora agregações mistas também já tenham sido descritas, podendo durar semanas, meses ou até mesmo anos (Banks 1902; Rau & Rau 1916; Evans & Linsley 1960; Linsley 1962).

Ao contrário das fêmeas, que coletam recursos florais por conta do investimento na prole (construção dos ninhos e provisão das células), os machos podem recorrer às flores não apenas para alimentação ou reprodução, mas, também como uma possibilidade de abrigo temporário (Eickwort & Ginsberg 1980; Paxton 2005; Sapir et al. 2005; Michener 2007). Assim, os machos de algumas espécies de abelhas solitárias podem ser observados entre as flores, buscando por um local ideal para passar a noite (Danforth et al. 2019). Nas flores escolhidas para pernoitar (dormitórios), a quantidade de machos pode variar de um único individuo até dezenas deles (Krug et al. 2010; Watts et al. 2013). E a depender do comportamento adotado, os machos, podem ser considerados polinizadores das espécies onde dormem (Sapir et al. 2005; Monty et al. 2006; Watts et al. 2013).

Registros de abelhas dormindo associados a plantas são realizados a mais de um século (Friese 1899; Green 1899; Banks 1902; Bradley 1908; Rau & Rau 1916). Friese (1899) descreveu o comportamento de machos de *Tetrapedia peckoltii* Friese, 1899 que sempre pousavam no galho de um mesmo arbusto ao pôr-do-sol, e dormiam nos ramos das flores agarrados pelas mandíbulas. Na mesma época, Green (1899) observou um indivíduo de “abelha-cuco” *Thyreus ramosus* (Lepeletier, 1841) dormindo durante várias noites em um galho morto. Na literatura, grande parte dos primeiros registros de machos formando agregações foram relacionados às abelhas da tribo Halictini (Halictidae). Schremmer (1956) descreveu informações de machos de várias espécies de Halictini formando agregações em caules ou ramos. Já Linsley (1962). observou machos e fêmeas de *Agapostemon angelicus* Cockerell, 1924 dormindo em flores secas de *Heterotheca subaxillaris* (Lam.) Britton & Rusby (Asteraceae) por noites sucessivas. Enquanto os machos de *A. angelicus* compartilhavam a mesma flor com até seis indivíduos, as fêmeas dormiam sempre sozinhas. Com o passar do tempo, os machos de muitas outras tribos também foram registrados formando agregações, com destaque para espécies das tribos Anthoporini (Yokoi & Watanabe 2015; Yokoi et al. 2016), Centridini (Gottsberger et al. 1988, Star &Vélez 2009; Martins et al. 2018), Eucerini (LaBerge & Ribble 1966), Megachilini (Linsley 1962; Thoenes 1994) e Tetrapediini (Alves-dos-Santos et al. 2002).

Há registros de agregações e dormitórios de machos para os diferentes biomas e regiões do mundo. Na região Neártica os estudos estão concentrados nos Estados Unidos (Mathewson & Daly 1955; Linsley & MacSwain 1958; Evans & Linsley 1960; Alcock 1998). Na região Neotropical, as abelhas Apidae, principalmente das tribos

Centridini (Frankie et al. 1980; Starr & Vélez 2009; Sabino et al. 2017; Martins et al. 2018) e Tapinotaspidini (Rozen et al. 2006; Alves-dos-Santos et al. 2009; Pinheiro et al. 2017) estão entre os mais descritos na literatura específica. Os demais registros estão distribuídos entre estudos realizados nas regiões Paleártica (Dafni et al. 1981; Sapir et al. 2005; Vereecken et al. 2012), Australiana (Houston 1991; Kubiak 2013), Oriental (Miyanaga & Maeta 1998; Yokoi & Watanabe 2015; Yokoi et al. 2016) e Etiópica (Whitehead 1984; Peter & Johnson 2008).

Registros e descrições de agregações existem para diversos grupos de abelhas solitárias. Contudo, ainda assim não há uma total compreensão das razões desse fenômeno (Evans & Linsley 1960), mesmo com alguns estudos sugerindo possíveis razões (Linsley & Cazier 1972). Para Grassé (1942), as agregações poderiam representar uma etapa da evolução do comportamento social em abelhas. Evans & Linsley (1960) e Alcock (1998) hipotetizam que a formação de agregações representaria uma tentativa de diminuir os riscos de predação noturna. Por fim, alguns autores sugerem que a formação de agregações poderia elevar a capacidade de termorregulação dos indivíduos (Evans & Gillaspy 1964; Linsley & Cazier 1972).

Nesse estudo apresentamos dados originais de machos de abelhas solitárias das famílias Apidae e Megachilidae que dormem em agregações e/ou dormitórios no Cerrado do Brasil Central. Relatamos os comportamentos observados em campo e discutimos acerca das hipóteses que influenciam no estabelecimento de agregações temporárias de machos ou de dormitórios em flores.

MATERIAL E MÉTODOS

O presente estudo foi conduzido no Cerrado do Brasil Central. As áreas de estudo incluem: Brasília (n=6), Gama (n =1), Região Administrativa de Fercal (Fercal) (n=1) e Planaltina (n=1), todas situadas no Distrito Federal e descritas na Tabela 1. A vegetação original da região é caracterizada por formações savânicas com árvores de poucos metros de altura, sub-bosque arbustivo e formação campestre periodicamente sujeita ao fogo (Ribeiro & Walter 1998). O clima da região é o tropical de savana (Aw) de Köppen, sazonal com uma estação seca e fria (maio a setembro) e outra úmida e quente (outubro a abril) (Alvares et al. 2013). A temperatura média é 22° C, com precipitação média anual de 1.453 mm e altitude entre 1.045-146 m (Brasil 2004).

Agregações e dormitórios de machos de abelhas solitárias foram encontrados, acompanhados e observados em campo entre março de 2018 e fevereiro de 2020, no Brasil Central, totalizando mais de 100 horas de observação. Assim, para cada agregação acompanhada, os machos (e as fêmeas, eventualmente, observadas dormindo em agregação ou flores) foram contabilizados e anotações acerca do comportamento foram realizadas. Para classificar o comportamento dos machos em relação ao local onde passaram a noite, duas categorias foram estabelecidas: i) *agregações*: condição em que duas ou mais abelhas passaram a noite juntas em um determinado substrato (e.g. ramo ou galho seco); ii) *dormitórios*: ocasiões em que as abelhas dormiam sozinhas ou acompanhadas no interior de flores. As abelhas foram identificadas com auxílio de chaves taxonômicas e por comparação com o material de referência. Os espécimes foram coletados e, posteriormente, depositados na Coleção Entomológica da Universidade de Brasília (UnB), Departamento de Zoologia.

RESULTADOS

Registramos 20 espécies de machos de abelhas pertencentes às famílias Apidae (integrantes de seis tribos: Bombini, Emphorini, Ericrocidini, Eucerini, Tapinotaspidini e Tetrapediini) e Megachilidae (oriundos de três tribos: Anthidiini, Lithurgini e Megachilini) dormindo em agregações ou flores fechadas (Tabela 1). No total, foram registradas 205 abelhas durante o período de estudo, sendo 83.4% (n=171) machos. As famílias botânicas Asteraceae, Malvaceae e Poaceae destacam-se como os substratos mais utilizados pelos machos para repousar.

Agregações de machos de *Tetrapedia* e Tapinotaspidini

Duas agregações de machos de abelhas coletoras de óleo foram observadas em duas regiões distintas de Brasília. Na Fazenda Água Limpa (FAL), uma agregação de 13 machos de *Tetrapedia peckoltii* Friese, 1899 foi acompanhada entre maio e junho de 2018 (Figura 1). Os primeiros machos chegaram à agregação, geralmente, a partir das 15:30 h, quando a temperatura média era 26 °C. Após chegar à agregação, os machos esfregavam as pernas posteriores que, em alguns casos, eram também esfregadas nos esternos ou tergos. Posteriormente, prendiam-se a ramos de capim seco pela mandíbula, ficando imóveis logo em seguida. A presença de óleo nas pernas posteriores de alguns indivíduos foi registrada, o que pode sugerir que os machos visitavam flores de diferentes espécies de Malpighiaceae (e.g. *Banisteriopsis* spp. e *Byrsonima* spp.). Estas plantas ocorriam nas adjacências da agregação, onde já havia sido registrada a presença de fêmeas da mesma espécie. No dia seguinte, os primeiros comportamentos observados, entre 8:00 - 9:00 h, eram de esfregar pernas e esternos, bem como movimentar as asas. Porém, os primeiros machos deixaram as agregações apenas a partir das 9:00 h, com a temperatura superior a 19 °C. Não foram observados machos voando à temperatura inferior a 19.2 °C, mesmo quando o sol já incidia sobre a agregação.

No Centro Olímpico (CO) da UnB, uma segunda agregação com 34 machos de abelhas coletoras de óleo, pertencentes a *Tetrapedia diversipes* Klug, 1810; *Tetrapedia* sp. 1; *Tropidopedia punctifrons* (Smith, 1879) e *Xanthopedia* cf. *larocai* Moure, 1995 foi observada em novembro de 2018, formando agregação em uma inflorescência seca de Asteraceae sp. (Figuras 2 e 3). Os primeiros machos chegavam à agregação por volta das 16:00 h, mas também podiam ser observados no local um pouco antes, geralmente, em tardes nubladas ou pouco tempo antes da precipitação. Os machos de Tapinotaspidini e *Tetrapedia* prendiam-se a inflorescência seca com o auxílio das mandíbulas e pernas anteriores e médias, em alguns instantes disputando posição na agregação, mas não registramos qualquer outro tipo de comportamento agonístico. Foi verificada a presença de óleo nas pernas posteriores de alguns machos de *Tetrapedia e* Tapinotaspidini, possivelmente coletados em flores de *Byrsonima coccolobifolia* (Malpighiaceae), que apresentava muitos indivíduos floridos nos arredores da agregação. Em *B. coccolobifolia*, visualizamos a visitação de machos e fêmeas de abelhas coletoras de óleo *Tetrapedia* sp. 1, *Tetrapedia diversipes* e *Tropidopedia flavolineata* Aguiar & Melo, 2007.

Às margens do Rio Maranhão, em Planaltina (DF), em maio de 2019, foram coletados três machos de *Tetrapedia* cf. *peckoltii* formando uma agregação, às 15:32, em uma folha de *Cissus rhombifolia* M. Vahl (Vitaceae). Os machos estavam imóveis e não foi registrado nenhum tipo de competição por substrato. Já na Reserva Ecológica do IBGE (Lago Sul, Brasília, DF), em abril de 2019, foram registrados machos *Monoeca pluricincta* (Vachal, 1909), às 08:58 h, dormindo no pedicelo e em folhas de uma Asteraceae sp. (Figura 4). Todavia, informações mais detalhadas acerca do comportamento dos machos nas agregações do Rio Maranhão e IBGE não foram realizadas.

Agregações de machos de Eucerini

Duas agregações de machos de *Melissodes* (*Ecplectica*) *nigroaenea* (Smith, 1854) foram observadas por aproximadamente 15 dias (março e abril de 2018), em uma área de agrofloresta do *campus* Darcy Ribeiro da Universidade de Brasília (UnB). No local são mantidas espécies de plantas nativas e cultivadas num solo rico em matéria orgânica exógena e adubo verde. No total, as duas agregações de *M. nigroaenea* apresentavam aproximadamente 15 machos, registradas a cerca de 40 e 60 cm acima do solo, em inflorescências secas de *Bidens pilosa* L. (Asteraceae). Os primeiros machos chegaram nas agregações por volta de 16:00 h e se estabeleciam em algum local entre as inflorescências (Figura 5).

Os machos de *M. nigroaenea* utilizaram a mandíbula para fixar-se em inflorescências secas de *B. pilosa*, da mesma forma que o observado para os machos de *Tetrapedia* e Tapinotaspidini. Após fixar-se nas inflorescências de *B. pilosa*, as pernas anteriores e médias eram utilizadas para encontrar a melhor posição para se estabelecer na agregação e passar a noite. Em seguida, alguns machos friccionavam as pernas posteriores, concomitantemente, algumas vezes esfregando-as nos esternos ou tergos. Interações entre os machos de *M. nigroaenea* foram observadas, em algumas situações, quase sempre devido a chegada de um novo indivíduo à agregação, que esbarrava em outro macho, estabelecendo por alguns poucos segundos algum tipo de contato, quase sempre em relação à disputa por uma posição na inflorescência. Os últimos machos chegaram nas agregações por volta das 17:00 h, quando os demais indivíduos já se encontravam em repouso.

Os machos de *M. nigroaenea* deixavam as agregações bem cedo, não sendo observados indivíduos repousando após às 7:00 h. Entretanto, quando o céu estava nublado ou chovia, ao longo do dia, os machos muitas vezes reestabeleciam as agregações. As fêmeas de *M. nigroaenea* foram observadas com relativa frequência coletando pólen, principalmente entre 10:00 - 16:00 h, em flores de *Cosmus sulphureus* Cav. (Asteraceae) próximas das agregações. Os machos de *M. nigroaenea* eram frequentemente observados bebendo néctar nas flores de *C. sulphureus* e *Ipomoea* sp. (Convolvulaceae), porém, nenhum registro de cópula foi realizado (Figura 6).

Agregação de machos de *Thygater*

Uma agregação de machos de *Thygater* sp. 1 Holmberg, 1884 foi registrada em janeiro de 2020, no Jardim Botânico de Brasília (JBB). No momento do registro, a agregação era constituída de aproximadamente 12 machos e estava reunida em uma raiz, dentro de uma cavidade no solo, muito semelhante aos buracos escavados e/ou utilizados por tatus. No local, também foram visualizadas algumas dezenas de lepidópteros que, aparentemente, também repousavam na cavidade (Apêndice 1).

Abelhas dormindo isoladamente

Abelhas dormindo isoladamente também foram registradas (Apêndice 2), principalmente no JBB. Abelhas dos gêneros *Ctenioschelus* sp. 1, *Mesonychium coerulescens* Lepeletier & Serville, 1825 e *Megachile* (*Pseudocentron*) sp. Mitchell, 1934 foram observadas dormindo sozinhos em indivíduos secos de *Axonopus* sp. Beauv. (Poaceae). Entetranto, em algumas situações, indivíduos de *Mesonychium coerulescens* e *Ctenioschelus* sp. foram observados compartilhando diferentes ramos secos de um mesmo indíviduo de *Axonopus* sp.

Em *C. sulphureus* também foram registrados machos de *Bombus* (*Fervidobombus*) *pauloensis* Friese, 1913 e *Epanthidium* sp. Moure, 1947. No CO também foi registrado um macho de *Tetrapedia* sp. 1 dormindo sozinho em um ramo de capim seco, preso pelas mandíbulas, por volta das 17:00 h. Na maior parte dos registros, essas abelhas eram visualizadas chegando aos locais de repouso a partir das 16:00 h.

Dormitórios em flores de Malvaceae

Abelhas das tribos Emphorini, Eucerini (Apidae) e Lithurgini (Megachilidae) foram observadas dormindo dentro de flores de duas espécies de Malvaceae: *Cienfuegosia affinis* (Kunth) e *Hibiscus capitalensis* Krapov. & Fryxell (Figura 7). Entre os meses de maio de 2018 e junho de 2019, 105 abelhas foram coletadas dormindo em flores de *C. affinis* (n=62) e *H. capitalensis* (n=43), sendo a grande maioria machos (n= 75). As espécies mais abundantes foram *Diadasina riparia* (Ducke, 1907) (n=40), *Ptilothrix plumata* Smith, 1853 (n=33) e *Lithurgus huberi* Ducke, 1907 (n=22). Em menor proporção, machos e fêmeas de Eucerini (n=6) também utilizaram as flores de Malvaceae como dormitório. Além disso, um macho de óleo *Monoeca pluricincta* foi coletado, às 14:32, em uma flor de *H. capitalensis* que estava praticamente fechada. Na escopa (basitarso e tíbia posterior) e esternos havia grande quantidade de pólen de *H. capitalensis* (Figura 8).

Na maior parte dos registros um único macho foi encontrado dormindo por flor, mas, ocasionalmente, dois indivíduos foram observados compartilhando a mesma flor. Os machos foram coletados, em sua maioria, entre 14:00 - 15:30 h, quando as flores de *C. affinis* e *H. capitalensis* estavam fechadas. Pela manhã e início da tarde, as fêmeas eram observadas coletando pólen nas flores de Malvaceae, onde os machos constantemente passam a noite (Apêndice 3). Em poucas ocasiões visualizamos machos de *Lithurgus huberi* repousando (10:00 - 12:00 h) dentro de flores de *H. capitalensis*. Nas proximidades das flores de Malvaceae, alguns machos de *L. huberi* foram observados dormindo em flores de *Cuspidaria sceptrum* (Cham.) (Bignoneaceae) e de *Distimake macrocalyx* (Ruiz & Pav.) (Convolvulaceae).

Dormitório em flores de Curcubitaceae

Machos de *Peponapis fervens* (Smith, 1879) foram coletados em flores de *Curcubita pepo* L. (Curcubitacee) em uma agrofloresta cultivada em uma área urbana de Brasília. Machos de *P. fervens* foram registrados pernoitando em flores fechadas de *C. pepo.* Pela manhã, machos e fêmeas de *Peponapis fervens* foram coletados visitando flores, entre 6:30 - 7:30 h, muitas vezes quando estavam a ingerir néctar (Apêndice 4).

DISCUSSÃO

Durante os 24 meses de estudo registramos diferentes grupos de abelhas dormindo em agregações ou flores. O número de abelhas dormindo dentro de uma flor ou em outra estrutura vegetal pode variar muito. Do mesmo modo que um único indivíduo pode ser visualizado dormindo em flor ou ramo seco, como foi registrado para *Bombus* (*Fervidobombus*) *pauloensis* e *Epanthidium* sp., agregações formadas por dezenas (e.g. *Amegilla florea urens* (Cockerell, 1911) e *Megachile* (*Chelostomoides*) *chilopsidis*) ou até mesmo centenas de abelhas (e.g. *Idiomelissodes duplocincta* Cockerell), 1905) já foram descritas na literatura (Thoenes 1994; Alcock 1998; Yokoi et al. 2016).

Os comportamentos observados para os machos que formaram as agregações (Eucerini, Tapinotaspidini e Tetrapediini) apresentaram muitas semelhanças. Os primeiros indivíduos chegavam ao local geralmente nos mesmos horários (15:30 - 16:00 h) e estabeleciam um local para fixar-se aos ramos secos com o auxílio das mandíbulas e, em alguns casos, com as pernas anteriores ou médias. Logo em seguida, alguns machos de *Melissodes nigroaenea*, *Tetrapedia* spp. e Tapinotaspidini friccionavam as pernas posteriores, esfregando-as nos esternos ou tergos machos os esternos em alguns casos. O hábito de fixar-se no substrato através das mandíbulas possivelmente confere maior estabilidade para passar a noite presa ao substrato (Rayment 1935), e representa um comportamento característico de muitas espécies de abelhas solitárias como, por exemplo, *Coelioxys deplanata* Cresson, 1878, *Melissoptila* aff. *bonaerensis* Holmberg, 1903, *Centris* (*Paracentris) xanthomelaena* Moure & Castro, 2001 (Linsley 1962; Mahlmann et al., 2014; Martins et al., 2018). Entretanto, nem sempre os machos fazem uso da mandíbula ao formar à agregação. Eickwort & Eickwort (1971), na Costa Rica, observaram que mesmo em meio a uma tempestade, às 7:40 h, machos de *Lasioglossum* (*Dialictus*) *umbripenne* (Ellis, 1913) mantiveram se presos em uma pteridófita com o auxílio das pernas, porém, sem utilizar a mandíbula. Por sua vez, o comportamento de esfregar as pernas e os esternos ou tergos já foi descrito para os machos de *Tetrapedia diversipes* (Alves-dos-Santos et al. 2009). Os autores sugerem que esse comportamento poderia representar algum tipo de sinalização química do grupo, envolvendo a secreção de compostos químicos através de possíveis poros glandulares localizados na superfície interna do basitarso traseiro dos machos.

O comportamento de chegada e saída das agregações, respectivamente, ao entardecer e início da manhã, refletem o que já foi descrito para os machos de diferentes espécies. LaBerge & Ribble (1966) relatam que machos *Florilegus* (*Florilegus*) *condignus* Cresson 1878 que passavam a noite em racemos de alfafa geralmente deixavam a agregação entre 6:30-7:30 h. Estudando o ciclo de vida da abelha *Augochlora pura* (Say, 1837), em campo e laboratório, Stockhammer (1966) percebeu que os machos retornavam as agregações entre 16:00-17:00h (no campo) e 17:00-18:00 (laboratório).

De modo geral, machos de *Melissodes nigroaenea* deixaram as agregações mais cedo do que os de *Tetrapedia* e Tapinotaspidini. O fato das agregações de *Tetrapedia* e Tapinotaspidini estarem situados sob um local sombreado, onde os primeiros raios solares incidiam mais tarde, pode ter influenciado no início mais tardio das atividades de voo. Por sua vez, a agregação de *M. nigroaenea* estava localizada em um área sombreada onde, logo cedo, a luminosidade incidia sobre as abelhas. A temperatura e intensidade luminosa exercem grande influência sobre o início das atividades das abelhas solitárias (Wilmer & Stone 2004). Os machos de *Tetrapedia* não iniciaram as atividades em temperaturas inferiores aos 19 ºC, o que talvez possa estar relacionado a um valor de temperatura limitante ao voo dessas abelhas, tendo em vista que, em algumas situações, alguns indivíduos passavam alguns minutos movimentando asas e pernas, mas só deixavam a agregação quando a temperatura média era superior a 19.2 ºC. Segundo Wilmer et al. (2005), muitas espécies de abelhas necessitam aquecer os músculos para alcançar uma potência adequada (cerca de 100 W) e alçar voo.

A agregação mista formada por machos de abelhas coletoras de óleo Tapinoteaspidni e *Tetrapedia* representa um importante registro considerando que não existem relatos na literatura para diferentes espécies de abelhas coletoras de óleo dormindo juntas. Entretanto, esse tipo de comportamento já foi descrito para agrupamentos formados por espécies de diferentes tribos, como, por exemplo, Epeolini (Banks 1902), Megachilini (Evans & Linsley 1960) e Anthoporini (Linsley 1962). Os registros de machos de *Monoeca pluricincta*, *Tropidopedia punctifrons* e *Xanthopedia* cf. *larocai* representam novas informações acerca de agregações formadas por abelhas Tapinotaspidini. Outros estudos já haviam descrito machos de Tapinotaspidini dormindo em caules, folhas ou flores (Rozen et al. 2006; Rocha-Filho 2007; Alves-dos-Santos et al. 2009; Pinheiro et al. 2017; Maubecin et al. 2020).

Machos de *T. diversipes* e *T. peckoltii* também já foram registrados dormindo em agregações (Alves-dos-Santos et al. 2002; Alves-dos-Santos et al. 2009). A função do óleo coletado pelos machos de Tapinotaspidini e *Tetrapedia* é desconhecida, embora seja esperado alguma relação com o comportamento reprodutivo (ver Capítulo 1). Todavia, a possibilidade de que o óleo possa ser usado como um possível composto químico utilizado para inibir a ação de predadores noturnos não pode ser descartada, embora não tenha sido testada. Estudos indicam que lipídios secretados por diferentes espécies de plantas podem atuar como defesa contra herbívoros (Eigenbrode & Espelie 1995, Igenbrode, 2004). Assim, a complexa estruturação química e provável toxicidade do óleo floral exigiria do predador dessas abelhas o desenvolvimento de metabólitos específicos (Policarová et al. 2019).

No presente estudo encontramos machos de Eucerini dormindo tanto em agregações (*Melissodes nigroaenea* e *Thygater* sp. 1) como dentro de flores (*Melissodes* spp. e *Melissoptila* spp.). Na literatura, muitos registros de machos de Eucerini passando a noite em agregações estão disponíveis (ver Bradley 1908; Mathewson & Daly 1955; Linsley 1962; LaBerge e Ribble1966; Mahlmann et al. 2014) ou dentro de flores (Hurd & Linsley; 1964; Dafni et al. 1981; Willis & Kevan 1995; Vereecken et al. 2012; Watts et al. 2013).

De modo geral, observamos machos dormindo dentro de flores de Malvaceae, embora registros de machos em Bignoniaceae, Convolvulaceae e Curcubitaceae também tenham sido observados. Estudos descrevendo machos dormindo dentro de flores tornaram-se mais frequentes a partir da década de 80 (Dafni et al. 1981; Willis & Kevan 1995; Gaglianone 2000; Sapir et al. 2005; Monty et al. 2006; Watts et al. 2013; Pinheiro et al. 2017). Os machos de *Diadasina riparia, Ptilothrix plumata e Lithurgus huberi*, os mais frequentes em flores de Malvaceae, geralmente dormem sozinhos, embora em algumas poucas oportunidades tenham sido coletados mais de um indivíduo na mesma flor. As informações disponíveis acerca de machos que utilizam flores como dormitórios sugerem que a quantidade de machos por flor pode variar de um a dezenas de indivíduos. Watts et al. (2013), estudando *Iris atropurpurea* Baker, uma espécie de Iridaceae ameaçada de extinção em Israel, que serve de dormitório para machos de Eucerini, registraram até 31 indivíduos de *Eucera* (*Synhalonia*) *spectabilis* Morawitz 1875 dormindo em uma única flor. Já Monty et al. (2006), estudando a polinização de *Oncocyclus* (Siems.) (Iridaceae), um complexo de plantas endêmica do Líbano, relataram até 25 machos de *Eucera* spp. pernoitando em uma mesma flor.

O comportamento que os machos de algumas espécies apresentam de dormir dentro de flores pode determinar o potencial deles como possíveis polinizadores (Sapir et al. 2005; Monty et al. 2006). No presente estudo, alguns machos foram verificados com alguma quantidade de pólen nas pernas posteriores e/ou esternos. Todavia, a maior quantidade de pólen foi observada num macho de *Monoeca pluricincta* coletado em uma flor de *H. capitalensis*. Os machos de *M. pluricinta* são coletores de óleo floral (ver Capítulo 2), e outro indivíduo do mesmo sexo já havia sido coletado em *Banisteriopsis* sp., nas adjacências das populações de *H. capitalensis* que eram utilizadas como dormitório. O comportamento que alguns machos apresentam de entrar em mais de uma flor antes de definir aquela que servirá como dormitório pode implicar na dispersão de pólen e favorecer a polinização. Segundo Dafni et al. (1981), a polinização de *S. vomeracea* é realizada, na maioria dos casos, por machos de *Andrena* (Andrenidae), *Eucera*, *Tetralonia* (Apidae) e *Osmia* (Megachilidae), que acabam por transportar polínias ao visitar diferentes flores em busca do lugar adequado para pernoitar. Já os machos de *Eucera* spp. e *Xylocopa* spp. foram descritos como polinizadores de *Oncocyclus* devido ao comportamento dentro das flores e carga de pólen carregada (Monty et al. 2006). Entretanto, nem sempre o comportamento dos machos é benéfico à planta. Machos de *Hexantheda missionica* Ogloblin, 1948, por exemplo, cortam e removem os estiletes e anteras das flores masculinas de *Calibrachoa* Cerv. (Solanaceae) para obter mais espaço para dormir, consequentemente, tornando as estéreis (Danforth et al. 2019).

A maior parte dos machos observados, e isso independe de dormir em agregação ou dormitório, durante o dia visitaram flores onde as fêmeas coletam pólen, néctar e/ou óleo. Machos de *Melissodes nigroaenea* e *Peponapis fervens*, por exemplo, foram registrados ingerindo néctar, respectivamente, em flores de *Cosmos sulphureus* e *Curcubita pepo*, nas mesmas flores em que as fêmeas coletam recursos durante o dia. Outro comportamento comum aos machos de algumas espécies de abelhas solitárias é o de patrulhar recursos florais visitados pelas fêmeas, como foi observado par os machos de *Lithurgus huberi*, *Peponapis fervens* e *Melissodes nigroaenea* (Alcock et al. 1978; Paxton 2005). Machos de *Tetrapedia* já haviam sido registrados coletando óleo floral, através de estruturas especializadas nas pernas anteriores, em flores de Malpighiaceae (Cappellari et al. 2012). O uso do óleo coletado pelos machos de *Tetrapedia* e Tapinotaspidini ainda não é bem esclarecido, mas é esperado que esteja associado à reprodução (ver também Capítulo 2), seja como um possível presente nupcial para as fêmeas, ou como uma camuflagem química que serviria para facilitar o acesso as fêmeas.

É esperado que estabelecer agregações ou dormir em flores visitadas pelas fêmeas também represente uma estratégia para facilitar o acesso às parceiras reprodutivas no dia seguinte (Alves-dos-Santos et al., 2009; Pinheiro et al., 2017). Segundo Chemsak & Thorp (1962), os machos de *Melissodes robustior* Cockerell, 1915, outra espécie de Eucerini, demonstram alguma preferência por dormir em flores de *Cosmos* sp., onde as fêmeas coletam pólen durante o dia. As possíveis razões pelas quais machos formam agregações ou dormem dentro de flores parecem mais bem esclarecidas para o segundo caso. Enquanto as fêmeas de abelhas solitárias dormem dentro dos ninhos, os machos buscam por lugares para passar a noite (Alcock 1998; Danforth et al. 2019). Dessa forma, um refúgio que proporcione proteção e calor parecem uma boa justificativa para a utilização das flores como dormitório (Sapir et al. 2005). Considerando que, sempre que possível, os machos de muitas espécies de Hymenoptera dormem em cavidades, é possível que, em alguns casos flores, principalmente as de cores escuras e/ou tubulares, sejam percebidas pelas abelhas como cavidades que forneçam abrigo (Dafni et al. 1981; Sapir et al. 2005). Contudo, os motivos que levem os machos a formarem agregações temporárias para passar a noite seguem sem respostas conclusivas, embora algumas hipóteses tenham sido propostas. Em geral, associadas à estratégias anti-predação, ainda mais quando considerado o fato de que muitas delas são iniciadas durante o dia, quando muitos predadores ainda estão ativos (Evans e Linsley 1960; Alcock 1998). Dessa forma, o efeito diluição favoreceria os machos de uma agregação ao diminuir a chance de serem predados durante a noite (Bertram 1978; Delm 1990). Segundo Alcock (1998), os machos de *Idiomelissodes duplocincta* que dormiam em agregações em folhas e caulas, no Arizona, sofreram poucos ataques de *Apiomerus flaviventris* (Hemiptera, Reduviidae). Para o autor, é provável que o comportamento gregário tenha favorecido a sobrevivência dos machos de *I. duplocincta* devido ao efeito diluição, uma vez que quanto maior a agregação, menor o número de abelhas predadas. Outra possível vantagem de dormir em agregações estaria relacionada à termorregulação. Assim, diante da ação de um predador noturno, as abelhas poderiam alcançar mais rapidamente a temperatura mínima para ativar a musculatura e fugir, tendo em vista que a temperatura do agrupamento é maior em agregações mais densas (Rayment 1935).

CONCLUSÃO

O comportamento dos machos que dormem em agregações ou dormitórios é bastante variável, ainda mais quando considerados aspectos taxonômicos das abelhas. Embora diversos relatos estejam disponíveis, ainda restam muitos questionamentos acerca das razões que levam os machos a dormir em ramos, folhas ou flores. Os nossos resultados sugerem, de alguma forma, que o comportamento dos machos formar agregações ou dormitórios podem ter alguma relação com a tentativa de potencializar o sucesso reprodutivo, seja através do patrulhamento das flores ou estabelecendo sítios de repouso nas proximidades dos recursos utilizados pelas fêmeas. Estudos comportamentais que avaliem o potencial reprodutivo desses machos são necessários para maior entendimento desses processos. Ademais, investigar diferentes aspectos como, por exemplo, o potencial dos machos como polinizadores e a evolução desses comportamentos são necessários para o maior entendimento desse fenômeno.

REFERÊNCIAS BIBLIOGRÁFICAS

Alcock J (1998) Sleeping aggregations of the bee Idiomelissodes duplocincta (Cockerell) (Hymenoptera: Anthophorini) and their possible function. J Kansas Entomol Soc 71:1984. https://doi.org/10.2307/25085817

Alcock J, Barrows EM, Gordh G, Hubbard LJ, Kirkendall L, Pyle DW, Ponder TL, Zalom FG (1978) The ecology and evolution of male reproductive behavior in the bees and wasps. Zool J Linn Soc 64:293–326. https://doi.org/10.1111/j.1096-3642.1978.tb01075.x

Alvares CA, Stape JL, Sentelhas PC, Gonçalves JLM, Sparovek G, (2013) Köppen's climate classification map for Brazil. Meteorol Zeitschrift 22:711–728.

Alves-dos-Santos I, Gaglianone MC, Naxara SRC, Engel MS (2009) Male sleeping aggregations of solitary oil-collecting bees in Brazil (Centridini, Tapinotaspidini, and Tetrapediini; Hymenoptera: Apidae). Genet Mol Res 8:515–524. https://doi.org/10.4238/vol8-2kerr003

Alves-dos-Santos I, Melo, GAR, Rozen JG (2002) Biology and Immature Stages of the Bee Tribe Tetrapediini (Hymenoptera: Apidae). Am Museum Novit 3377:1-45. https://doi.org/10.1206/0003-0082(2002)377<0001:BAISOT>2.0.CO;2

Banks N (1902) Sleeping habits of certain Hymenoptera. J New York Entomol Soc 10:209–214

Bertram BCR (1978) Living in groups: predators and prey. In: Krebs JR, Davies NB (eds.), Behavioural Ecology: An Evolutionary Approach. Blackwell Scientific, Oxford

Bradley JC (1908) The entomological society of america. Can Entomol 40:69–71. https://doi.org/10.4039/Ent4069-2

Brasil (2004) Ministério do Planejamento, Orçamento e Gestão. Instituto Brasileiro de Geografia e Estatística (IBGE). Reserva Ecológica do IBGE: ambiente e plantas vasculares. Estudos e Pesquisas Informação Geográfica, n 3, Brasil, Rio de Janeiro, 70p

Cappellari SC, Melo GAR, Aguiar AJC, Neff JL (2012) Floral oil collection by male *Tetrapedia* bees (Hymenoptera: Apidae: Tetrapediini). Apidologie, 43:39–50

Chemsak JA, Thorp RW (1962) Note on the sleeping habits of males of *Melissodes robustior* Cockerell. Pan-Pac Ent 38:53–55.

Dafni A, Ivri Y, Brantjes NBM (1981) Pollination of Serapias Vomeracea Briq. (Orchidaceae) by Imitation of Holes for Sleeping Solitary Male Bees (Hymenoptera). Acta Bot Neerl 30:69–73. https://doi.org/10.1111/j.1438-8677.1981.tb00388.x

Danforth BN, Minckley RL, Neff JL (2019) The Solitary Bees. Princeton University Press

Delm MM (1990) Vigilance for predators: detection and dilution effects. Behav Ecol Sociobiol 26:337–342. https://doi.org/10.1007/BF00171099

Eickwort G, Eickwort K. (1971) Aspects of the Biology of Costa Rican Halictine Bees, II. *Dialictus umbripennis* and Adaptations of Its Caste Structure to Different Climates. J Kans Entomol Soc 44(3):343–373

Eickwort GC, Ginsberg HS (1980) Foraging and mating behavior in Apoidea. Annu Rev Entomol 25:421–446 https://doi.org/10.1146/annurev.en.25.010180.002225

Eigenbrode SD (2004) The effects of plant epicuticular waxy blooms on attachment and effectiveness of predatory insects. Arthropod Struct Dev. 33(1):91–102.

Eigenbrode, SD, Espelie, KE (1995) Effects of plant epicuticular lipids on insect herbivores. Annu Rev Entomol 40:171–194.

Evans HE, Gillaspy JE (1964) Observations on the Ethology of Digger Wasps of the Genus Steniolia (Hymenoptera: Sphecidae: Bembicini). Am Midl Nat 72:257–280

Evans HE, Linsley EG (1960) Notes on a Sleeping Aggregation of Solitary Bees and Wasps. Bull South Calif Acad Sci 59:30–37

Frankie GW, Vinson SB, Coville RE (1980) Territorial behavior of Centris adani and its reproductive function in the Costa Rican dry forest (Hymenoptera: Anthophoridae). J Kansas Entomol Soc 53:837–857

Friese H (1899) Monographie der Bienengattungen Exomalopsis, Ptilothrix, Melitoma und Tetrapedia. Ann k.k Naturhist Hofmus 14:247–304

Gaglianone MC (2000) Behavior on flowers, structures associated to pollen transport and nesting biology of Perditomorpha brunerii and Cephalurgus anomalus (Hymenoptera: Colletidae, Andrenidae). Rev Biol Trop 48:89–99

Gottsberger G, Camargo JMF, Silberbauer-Gottsberger I (1988) A bee-pollinated tropical community: the beach dune vegetation of Ilha de São Luís, Maranhão, Brazil. Bot. Jahrb. Syst 109:469–500

Grassé PP (1942). Les rassemblements de sommeil des Hyménoptères et leur interprétation. Bull Soc Entomol France 47:142–148

Green EE (1899) Sleeping habits of a Bee, Crocisa ramose. Ent Mon Mag 35:214

Houston TF (1991) Ecology and behaviour of the bee Amegilla (Asaropoda) dawsoni (Rayment) with notes on a related species (Hymenoptera: Anthophoridae). Rec West Aust Museum 15:535–553

Hurd PD, Linsley EG (1976) The bee family Oxaeidae with a revision of the North American species (Hymenoptera, Apoidea). Smithson Contrib to Zool 1–75. https://doi.org/10.5479/si.00810282.220

Hurd PD, Linsley EG (1964). The squash and gourd bees—genera *Peponapis* Robertson and *Xenoglossa* Smith—inhabiting America north of Mexico (Hymenoptera: Apoidea). Hilgardia 35(15):375–477 DOI:10.3733/hilg.v35n15p375

Kubiak PJ (2013) Sleeping aggregations of bees in relation to the risk of fire at their roosting sites in a forested, suburban landscape in eastern Australia. Vic Nat 130:22–36

Krug C, Alves-dos-Santos, Cane, J (2010) Visiting bees of *Cucurbita* flowers (Cucurbitaceae) with emphasis on the presence of *Peponapis fervens* Smith (Eucerini – Apidae) - Santa Catarina, Southern Brazil. Oecol Aust 14(1):128-139 https://doi.org/10.4257/oeco.2010.1401.06

LaBerge WE, Ribble DW (1966) Biology of *Florilegus condignus* (Hymenoptera: Anthophoridae), with a Description of Its Larva, and Remarks on Its Importance in Alfalfa Pollination. Ann. Entomol. Soc. Am 59:5

Linsley EG (1962) Sleeping Aggregations of Aculeate Hymenoptera—II. Ann Entomol Soc Am 55:148–164. https://doi.org/10.1093/aesa/55.2.148

Linsley EG, Cazier M (1972) Diurnal and seasonal behavior patterns among adults of *Protoxaea gloriosa* (Hymenoptera, Oxaeidae). Am Museum Novit 2509:1–25

Linsley EG, MacSwain JW (1958) Sleeping habits of males of *Hesperapis* (Hymenoptera: Melittidae). J Kansas Entomol Soc 31:273–274

Mahlmann T, Hipólito J, Oliveira FF. (2014). Male sleeping aggregation of multiple Eucerini bee genera (Hymenoptera: Apidae) in Chapada Diamantina, Bahia, Brazil. Biodivers Data J 2:15–56. https://doi.org/10.3897/BDJ.2.e1556

Martins HOJ, Oliveira-Rebouças P, Ferreira VS (2018) Sleeping behaviour of an oil-collecting bee, *Centris* (*Paracentris*) *xanthomelaena* Moure & Castro (Hymenoptera: Apidae: Centridini). Sociobiology 65:770–772. https://doi.org/10.13102/sociobiology.v65i4.3452

Mathewson JA, Daly H V. (1955) A brief note on the sleep of male *Melissodes* (Hymenoptera: Apidae). J Kansas Entomol Soc 28:120

Maubecin CC, Boero L, Sérsic NA (2020) Specialisation in pollen collection, pollination interactions and phenotypic variation of the oil-collecting bee *Chalepogenus cocuccii*. Apidologie, 51:710–723. https://doi.org/10.1007/s13592-020-00755-4

Michener CD (2007) The bees of the world. The Johns Hopkins University Press.

Miyanaga R, Maeta Y (1998) Notes on a Male Sleeping Aggregation of *Lasioglossum* (*Ctenonomia*) *kumejimense* (Hymenoptera: Halictidae). Entomol Sci 1:357–358

Monty A, Saad L, Mahy G (2006) Bimodal pollination system in rare endemic Oncocyclus irises (Iridaceae) of Lebanon. Can J Bot 84:1327–1338. https://doi.org/10.1139/B06-081

Paxton RJ (2005) Male mating behaviour and mating systems of bees: an overview. Apidologie 36(2):145–156. https://doi.org/10.1051/apido:2005007

Peter CI, Johnson SD (2008) Mimics and magnets: The importance of color and ecological facilitation in floral deception. Ecology 89:1583–1595. https://doi.org/10.1890/07-1098.1

Pinheiro M, Alves-dos-Santos I, Sazima M (2017) Flowers as sleeping places for male bees: somehow the males know which flowers their females prefer. Arthropod Plant Interact 11:329–337. https://doi.org/10.1007/s11829-017-9532-6

Policarová J, Cardinal S, Martins AC, Straka, J (2019) The role of floral oils in the evolution of apid bees (Hymenoptera: Apidae). Biol J Linn. Soc, 128(2): 486–497.

Rau P, Rau N (1916) The sleep of insects; an ecological study. Ann Entomol Soc Am 9:227–274

Rayment T (1935) A cluster of bees. Sydney, Austrália, Endeavour Press

Ribeiro JF, Walter BMP (1998) Fitofisionomias do bioma Cerrado. In: Sano SM, Almeida SP (eds.). Cerrado: ambiente e flora. Planaltina: EMBRAPA-CPAC, 89-116

Roig-Alsina A (1997) A generic study of the bees of the tribe Tapinotaspidini, with notes on the evolution of their oil-collecting structures (Hymenoptera, Apidae). Mitteilungen der Münchner Entomol Gesellschaft 87:3–21

Rozen JG, Melo GAR, Aguiar AJC, Alves-dos-Santos I (2006) Nesting Biologies and Immature Stages of the Tapinotaspidine Bee Genera Monoeca and Lanthanomelissa and of Their Osirine Cleptoparasites Protosiris and Parepeolus (Hymenoptera: Apidae: Apinae). Am Museum Novit 3501:1. https://doi.org/10.1206/0003-0082(2006)501[0001:nbaiso]2.0.co;2

Sabino WO, da Silva CI, Alves-dos-Santos I (2017) Mating System and Sleeping Behaviour of the Male and Female Centris (Paracentris) burgdorfi Friese (Apidae, Centridini). J Insect Behav 30:103–118. https://doi.org/10.1007/s10905-017-9600-x

Sapir Y, Shmida A, Ne'eman G (2005) Pollination of Oncocyclus irises (Iris: Iridaceae) by night-sheltering male bees. Plant Biol 7:417–424. https://doi.org/10.1055/s-2005-837709

Schremmer F (1956) Beobachtungen über die Nachtruhe bei Hymenopteren, insbesondere die Männchenschlafgesellschaften von *Halictus*. Oesterr. Österreichische Zool Z 6: 70–89

Starr CK, Vélez D (2009) A dense daytime aggregation of solitary bees (Hymenoptera: Apidae: Centridini) in the lesser antilles. J Hymenopt Res 18:175–177

Thoenes SC (1994). Description of a sleeping aggregation of male *Chalicodoma chilopsis* (Cockerell) (Hymenoptera: Megachilidae). Pan-Pac Ent 70:328–330

Vereecken NJ, Wilson CA, Hötling S, et al (2012) Pre-adaptations and the evolution of pollination by sexual deception: Cope's rule of specialization revisited. Proc R Soc B Biol Sci 279:4786–4794. https://doi.org/10.1098/rspb.2012.1804

Whitehead VB (1984) Distribution, biology and flower relationships of fideliid bees of southern Africa (Hymenoptera, Apoidea, Fideliidae). South African J Zool 19:87–90. https://doi.org/10.1080/02541858.1984.11447863

Watts SS, Sapir Y, Segal B, Dafni, A. (2013). The endangered *Iris atropurpurea* (Iridaceae) in Israel: honey-bees. Annals of Botany 111:395–407 DOI: 10.1093/aob/mcs292

Willis DS, Kevan PG (1995) Foraging Dynamics of Peponapis Pruinosa (Hymenoptera: Anthophoridae) on Pumpkin (Cucurbita Pepo) in Southern Ontario. Can Entomol 127:167–175. https://doi.org/10.4039/Ent127167-2

Willmer PG, Stone GN (2004) Behavioral, ecological, and physiological determinants of the activity patterns of bees. Advances in the Study of Behavior, 34: 347-466

Wilmer PG, Stone GN, Johnston I (2005) Environmental Physiology of Animals. Wiley-Blackwell

Yokoi T, Idogawa N, Konagaya T, Watanabe M (2016) The Non-Use of Sleeping Substrate by the Sympatric Bees Amegilla florea urens and A. senahai senahai (Hymenoptera: Apoidea). Entomol News 126:138–143. https://doi.org/10.3157/021.126.0210

Yokoi T, Watanabe M (2015) Female-biased sleeping aggregations of Amegilla florea urens (Hymenoptera: Apidae). Entomol Sci 18:274–277. https://doi.org/10.1111/ens.12112

Tabela 1. Registro das abelhas em agregações e dormitórios em localidades do Distrito Federal, entre março de 2018 e fevereiro de 2020.

Espécie	Planta	Machos	Fêmeas	Hábito	Localidade
APIDAE					
Bombini					
***Bombus* (*Fervidobombus*) *pauloensis* Friese, 1913**	*Cosmos sulphureus* (Asteraceae)	2	0	Solitário	Campus Darcy Ribeiro (UnB), Brasília, DF
Emphorini					
***Diadasina riparia* (Ducke, 1907)**	*Cienfuegosia affinis* (Malvaceae)	37	3	Dormitório	Núcleo Rural Palmeira, Fercal, DF
***Melitoma segmentaria* (Fabricius, 1804)**	*Hibiscus capitalensis* (Malvaceae)	1	0	Dormitório	Campus Gama (UnB), Gama, DF
***Ptilothrix plumata* Smith, 1853**	*Cienfuegosia affinis* (Malvaceae)	6	9	Dormitório	Núcleo Rural Palmeira, Fercal, DF
	Hibiscus capitalensis (Malvaceae)	10	8	Dormitório	Campus Gama (UnB), Gama, DF
Ericrocidini					
***Ctenioschelus* sp. 1 Romand, 1840**	*Axonopus* sp. (Poaceae)	2	2	Agregação	Jardim Botânico de Brasília, Brasília, DF
***Mesonychium coerulescens* Lepeletier & Serville, 1825**	*Axonopus* sp. (Poaceae)	0	1	Agregação	Jardim Botânico de Brasília, Brasília, DF
Eucerini					
***Melissodes* (*Ecplectica*) *nigroaenea* (Smith, 1854)**	*Cosmos sulphureus* (Asteraceae)	15	0	Agregação	Campus Darcy Ribeiro (UnB), Brasília, DF
***Melissodes* sp. 1 Latreille, 1829**	*Hibiscus capitalensis* (Malvaceae)	3	0	Dormitório	Campus Gama (UnB), Gama, DF
***Melissoptila* sp. 1 Holmberg, 1884**	*Cienfuegosia affinis* (Malvaceae)	1	4	Dormitório	Núcleo Rural Palmeira, Fercal, DF
***Melissoptila* sp. 2 Holmberg, 1884**	*Hibiscus capitalensis* (Malvaceae)	0	1	Dormitório	Campus Gama (UnB), Gama, DF
***Peponapis fervens* (Smith, 1879)**	*Curcubita pepo* (Curcubitaceae)	3	0	Dormitório	Asa Norte, Brasília, DF
***Thygater sp. 1* Holmberg, 1884**	Raiz (planta não indentificada)	12	0	Agregação	Jardim Botânico de Brasília, Brasília, DF
Tapinotaspidini					
***Monoeca pluricincta* (Vachal, 1909)**	Asteraceae sp.	4	0	Agregação	Reserva Ecológica do IBGE, Brasília, DF

***Tropidopedia punctifrons* (Smith, 1879)**	Asteraceae sp.	1	0	Agregação	Centro Olímpico (UnB), Brasília, DF
***Xanthopedia* cf. *larocai* Moure, 1995**	Asteraceae sp.	20	0	Agregação	Centro Olímpico (UnB), Brasília, DF
Tetrapediini					
***Tetrapedia diversipes* Klug, 1810**	Asteraceae sp.	10	0	Agregação	Centro Olímpico (UnB), Brasília, DF
***Tetrapedia peckoltii* Friese, 1899**	Poaceae sp.	13	0	Agregação	Fazenda Água Limpa (UnB), Brasília, DF
***Tetrapedia* cf. *peckoltii* Friese, 1899**	*Cissus rhombifolia* (Vitaceae)	3	0	Agregação	Rio Maranhão, Planaltina, DF
***Tetrapedia* sp. 1 Klug, 1810**	Asteraceae sp.	3	0	Agregação	Centro Olímpico (UnB), Brasília, DF
	Axonopus sp. (Poaceae)	1	0	Solitário	Centro Olímpico (UnB), Brasília, DF
MEGACHILIDAE					
Anthidiini					
***Epanthidium* sp. 1 Moure, 1947**	*Cosmos sulphureus* (Asteraceae)	2	0	Solitário	Campus Darcy Ribeiro (UnB), Brasília, DF
Lithurgini					
***Lithurgus huberi* Ducke, 1907**	*Cienfuegosia affinis* (Malvaceae)	2	0	Dormitório	Núcleo Rural Palmeira, Fercal, DF
	Cuspidaria sceptrum (Bignoneaceae)	1	0	Dormitório	Campus Gama (UnB), Gama, DF
	Distimake macrocalyx (Convolvulaceae)	3	0	Dormitório	Campus Gama (UnB), Brasília, DF
	Hibiscus capitalensis (Malvaceae)	1	0	Dormitório	Campus Gama (UnB), Brasília, DF
	Hibiscus capitalensis (Malvaceae)	14	5	Dormitório	Campus Gama (UnB), Gama, DF
Megachilini					
***Megachile* (*Pseudocentron*) sp. 1 Mitchell, 1934**	*Axonopus* sp. (Poaceae)	1	1	Agregação	Jardim Botânico de Brasília, Brasília, DF

Figura 1. Agregação de machos de *Tetrapedia peckoltii*, em ramos secos de uma gramínea, na Fazenda Água Limpa (FAL), em 2018.

Figura 2. Agregação de machos de abelhas Tapinotaspidini e *Tetrapedia*. **A.** Machos de *Tetrapedia diversipes* em repouso na agregação. **B.** Detalhe dos machos de *Tetrapedia peckoltii*. **C.** Machos de *Tetrapedia diversipes* e *Tetrapedia* sp. 1 dormindo próximos.

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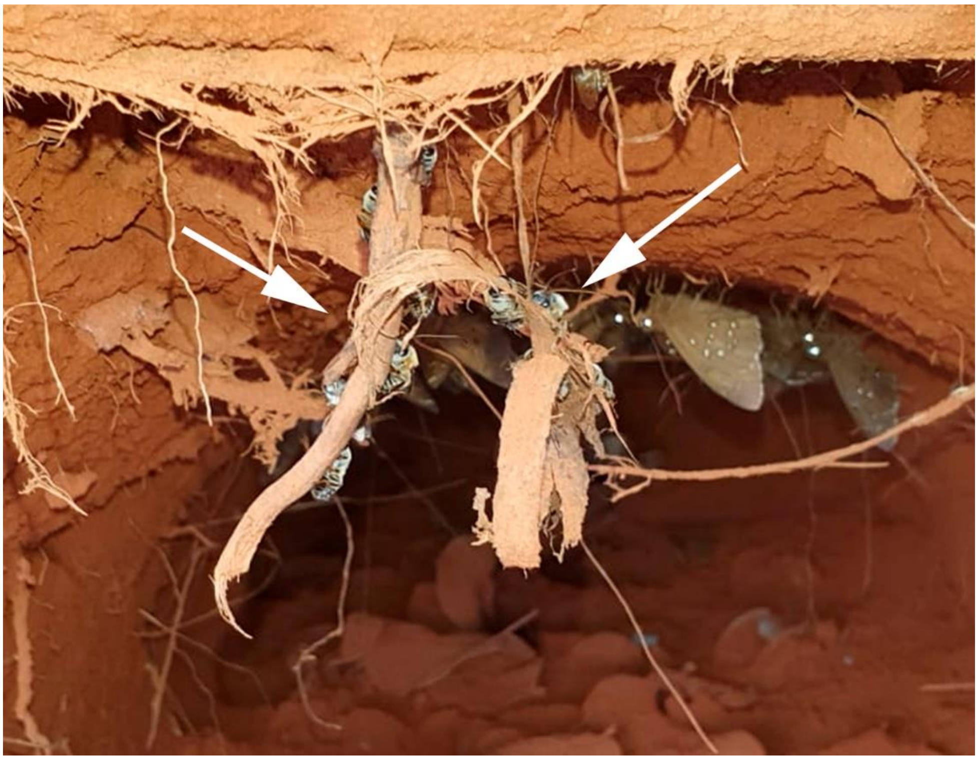
APÊNDICE

Capítulo3

Agregações e dormitórios: onde e como dormem os machos das abelhas?



Apêndice 1. Agregação de machos de *Thygater* sp. 1 localizada em uma raiz, dentro de cavidade, no Jardim Botânico de Brasília. Foto: Jair Faria.

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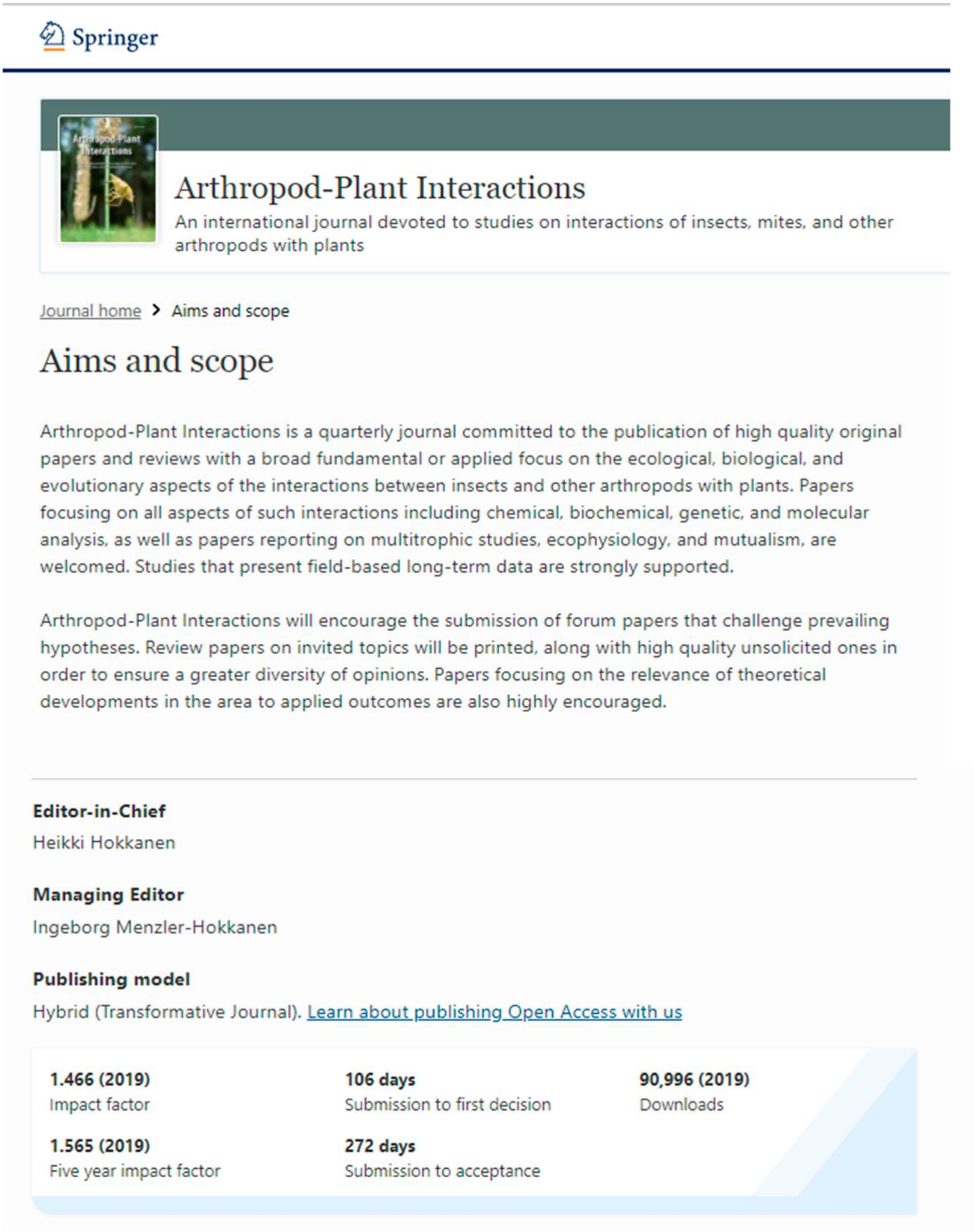
ANEXO

Capítulo 3

Agregações e dormitórios: onde e como dormem os machos das abelhas?

Anexo 1

Foco e escopo do periódico potencial ao qual pretendemos submeter o nosso manuscrito.



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CAPÍTULO 4

Trap-nesting bees and wasps in a Cerrado savanna area of Central Brazil

Manuscrito formatado segundo as normas da revista Papéis Avulsos de Zoologia (Anexo 1)

RESUMO

A utilização de ninhos-armadilha consiste numa importante técnica para inventários e monitoramento de abelhas e vespas solitárias que nidificam em cavidades pré-existentes. A técnica é especialmente importante quando incorporado dados de fenologia de suas espécies e a rica diversidade de parasitas e parasitóides associada. O presente estudo teve como objetivo realizar o inventário preliminar das espécies de abelhas e vespas que nidificam em cavidades pré-existentes na Fazenda Água Limpa (Distrito Federal), Estação de Pesquisa da Universidade de Brasília que abrange distintas formações de Cerrado. O estudo foi realizado durante dois anos, onde oitocentos e dez ninhos-armadilha foram instalados em três formações vegetacionais: mata de galeria, cerrado *stricto sensu* e área de transição entre mata de galeria e cerrado. Um total de 94 ninhos de dez espécies de abelhas e vespas foram coletados, além de oito espécies cleptoparasitas. As espécies mais abundantes foram *Tetrapedia peckoltii*, *Tetrapedia diversipes* e *Trypoxylon albitarse*. A área de transição apresentou maior número de nidificações (n=48) e índices de diversidade. Foi observada uma correlação positiva e significativa para o número mensal de nidificações e os valores de precipitação média mensal. As frequências de nidificação mais altas ocorreram na estação chuvosa (74,4%). *Tetrapedia peckoltii* foi a espécie com mais ninhos parasitados (n=10) e a espécie parasita mais abundante foi o besouro *Nemognatha* sp. A riqueza de espécies e abundância de ninhos são semelhantes a outros estudos realizados no Cerrado. Os resultados sugerem que a precipitação influencia no comportamento de nidificação das abelhas e vespas.

Keywords: Apoidea; inventory; nesting biology; *Tetrapedia.*

ABSTRACT

The use of trap-nests is an important technique for the inventory and monitoring of solitary bees and wasps that nesting in pre-existing cavities. The technique is particularly important when incorporating phenology data from its species, and the rich diversity of parasites and parasitoids associated. The present study aimed to conduct a preliminary inventory of bee and wasp species that nest in pre-existing cavities at Água Limpa Farm (Federal District), Research Station of the University of Brasilia, which involves different formations of Cerrado. The study was conducted over two years, where eight hundred and ten trap nests were installed in three vegetational formations: gallery forest, Cerrado stricto sensu and a transition area between gallery forest and Cerrado. A total of 94 nests of ten species of bees and nesting wasps were collected, in addition to eight cleptoparasite species. The most abundant species were *Tetrapedia peckoltii*, *Tetrapedia diversipes* and *Trypoxylon albitarse*. The transition area had the highest number of nidification (n=48) and diversity indexes. A positive and significant correlation was observed for the monthly number of nests and the mean monthly precipitation values. The highest nesting frequencies occurred in the rainy season (74.4%). *Tetrapedia peckoltii* was the species with the most parasitic nests (n=10) and the most abundant parasitic species was *Nemognatha* sp. The species richness and abundance of nests are similar to other studies conducted in the Cerrado. The results suggest that precipitation influences the nesting behaviour of bees and wasps.

Keywords: Apoidea; inventory; nesting biology; *Tetrapedia*.

INTRODUCTION

In the last 25 years, studies have been conducted in the Neotropical Region (see Costa & Gonçalves, 2019) through the trap-nests method (Camillo et al., 1995; Assis & Camillo, 1997; Jesus & Garófalo, 2000; Morato & Campos, 2000; Aguiar & Martins, 2002; Aguiar et al., 2005; Camillo, 2005; Melo & Zanella; 2012; Nascimento & Garófalo 2014; Oliveira & Gonçalves, 2017; Rocha-Filho et al., 2017; Cordeiro et al. 2019). The use of nests made of different materials and sizes allow the capture of bees and wasps nests in pre-existing cavities (e.g. tree trunks, dry branches, wall holes or abandoned nests) in order to generate information about the diversity, biology, and behaviour of these insects (Batra, 1984; Michener, 2007). Besides subsidizing data on the richness and structure of local communities, the trap-nest method enables the use of species nesting in pre-existing cavities as bioindicators of environmental quality such as, for example, the effect of habitat loss on these insects (Frankie et al. 1998; Tscharntke et al., 1998; Morato & Campos, 2000; Stangler et al., 2014). The information subsidized through the use of trap-nests also allows the study of the interaction networks between solitary bees and wasps and their natural enemies (Steffan-Dewenter, 2002; Lima et al., 2018; Torné-Noguera et al., 2020), and the communities of these insects in urban environments (Tommasi et al., 2004; Loyola & Martins, 2006).

The Cerrado is the second largest biome in Brazil, representing about 23% of the territory, with high biodiversity and a high degree of endemism (Ribeiro & Walter, 2008). However, the biological diversity of the Cerrado is threatened by the recurrent process of fragmentation in recent decades, making the biome one of the 35 world hotspots for biodiversity conservation (Mittermeier et al., 2011).

Considering the high number of studies based on trap-nests published in Brazil, subsidizing the basis for monitoring and expanding knowledge about the diversity of bees and wasps that nest in pre-existing cavities in the Cerrado is of extreme importance in the face of climate change and global extinction of insects. This study aimed to inventory the species of solitary bees and wasps that nest in pre-existing cavities in an area of Cerrado in the Federal District. The specific objectives were: to evaluate the temporal distribution of nesting activity and general aspects of parasitism incidence.

MATERIAL AND METHODS

The study was conducted in a Cerrado area (Fig. 1), at Fazenda Água Limpa (FAL) which belongs to the University of Brasilia (UnB) and is located in the Rural Nucleus of Vargem Bonita, southern of the Federal District. The sample area is inserted in two conservation units: Area of Relevant Ecological Interest (ARIE) Capetinga-Taquara and Environmental Protection Area (APA) Ribeirões Gama and Cabeça de Veado. Both are part of the São Bartolomeu River polygon, which is of high ecological importance (MMA, 2016). The Cerrado biome is characterized by the presence of two well-defined seasons, dry (April to September) and rainy (October to March), classified according to Köppen as tropical rain (Alvares et al., 2013). Occurring at altitudes ranging from 300 to more than 1600 m, the Cerrado has an average annual rainfall of 1500 mm, ranging from 750 to 2000 mm and range of temperature (Ribeiro & Walter, 2008).

The trap-nests were constructed with black cardboard in three diameters (0.6, 0.8 and 1.0 cm) and inserted in holes made of 10 cm deep wooden blocks (*Pinus* sp.). The trap nests were installed in three areas (28.1 ha) with distinct phytophysiognomies: gallery forest (15°56’49.4” S, 47°56'16.7" W), Cerrado stricto sensu (15°57'13.9" S, 47°56'20.7" W) and a transition area between gallery forest and Cerrado (15°57'23.1" S, 47°55'59.7" W). Three sample units were installed in each area, totaling nine sample points. Each sample unit consisted of 90 trap nests: 30 of 0.6 cm, 30 of 0.8 cm and 30 of 1.0 cm. In total 270 trap-nests were installed per area and 810 nests in total. The nests were inspected monthly between June 2016 and May 2018, totaling 24 months of sampling. The nests that were occupied or showed signs of nesting process were removed from the wooden blocks and replaced with others of the same diameter.

The occupied nests were taken to the Entomology Laboratory (Department of Zoology at UnB), where they were kept until the emergence of the individuals. In the laboratory the nests were kept at room temperature. The emerging individuals were sacrificed with ethyl acetate, pinned, labeled and deposited in the Entomological Collection of the University of Brasilia. The climatic data of the study area were obtained through the Automatic Meteorological Station of Água Limpa Farm. The bees was were identified with taxonomic keys and by comparison with reference material with specimens from the Entomological Collection.

The normality of data for the number of nests collected over the months (p=0.001635) was evaluated using the Shapiro-Wilk test. Correlations between abiotic factors (temperature and precipitation) and temporal distribution of nesting activities of the most abundant species were calculated using Spearman's correlation. The Shannon-Wiener (H') (Pielou, 1975) and Simpson (D) (Magurran, 1988) indices were used to estimate the diversity of the communities that nested in the three sample study areas. The Jaccard similarity index (SJ) was applied to estimate the degree of similarity between the species composition of the environments. The occupation rate (%) of species in each of the areas was calculated using the equation: total founded nests / (total available nests) x 100. The analyses were conducted in R software version 3.3.2 (R Development Core Team, 2016).

RESULTS

Trap-nests occupation, species richness and abundance

During the two years, 94 nests were collected in the three sample areas distributed for ten species: 69 (73.4%) of bees and 25 (26.6%) of wasps. The nesting species were distributed in five families: Apidae, Crabronidae, Megachilidae, Pompilidae and Sphecidae (Tables 1 and 2). The Apidae family presented highest species richness (n=5), corresponding to 71.3% of the nests collected. The solitary bee species *Tetrapedia peckoltii* Friese, 1899 was the species that most built nests (n = 36). In the 94 nests collected, 203 individuals (18 species) emerged, including the parasites (eight species)

(Appendix 1). Of the total number of individuals, bees were represented by 62.1% (n=126), wasps 27.1% (n=55), beetles 5.9% (n=12) and flies 4.9% (n=10). *Tetrapedia diversipes* Klug, 1810 was the most abundant species with 55 individuals sampled, followed by *Tetrapedia peckoltii* (n=53) and the wasp *Trypoxylon albitarse* (Fabricius,1804) (n=32).

Bees and wasps nested in all nest diameters provided in the three study areas. The 0.6 cm diameter nests were the most used (47.8%), followed 0.8 cm (27.7%) and 1.0 cm (24.5%). Among the ten species that nested in the trap-nests, only *Centris (Hemisiella) tarsata* (Smith,1874), *T. diversipes, T. peckoltii* and *T. albitarse* used more than one diameter. Regarding the number of individuals per diameter, the smalle trap nests (0.6 cm) were those with the largest number of emerging insects (n=98), followed the large (1.0 cm) and medium (0.8 cm) nests, respectively, with 58 and 47 individuals.

In relation to presence-absence of species in the three areas, the similarity between transition area and Cerrado was the highest (SJ=0.3333); with less similarity between gallery forest and transition area (SJ=0.2143) and between gallery forest and Cerrado (SJ=0.1538). The gallery forest area (n=6) presented a much lower number of collected nests when comparing the transition area (n=48) and Cerrado (n=40). The transition (H'= 1.7724; D=0.7666) and Cerrado (H'= 1.4856; D=0.6326) areas also presented a higher diversity index than forest area (H'= 0.8982; D=0.5612). In the forest area only the wasp species *Priochilus* sp. Banks, 1944 and *T. albitarse* nested in the trap nests. *Tetrapedia peckoltii* and *T. diversipes* were collected only in the transition and Cerrado areas. *Tetrapedia albitarse* was the only species that nested in the three areas. The other species were only observed nesting in one of the study areas.

Considered the sex ratio of *T. peckoltii* and *T. diversipes* bees. Of the total *T. peckoltii* bees, males represented 56.2% (n=27) and females 44.8% (n=26) of the individuals, resulting in a sex ratio of 1F:1M (χ^2=0.019). More females (n=32) than males (n=21) emerged in the nests of *T. diversipes*, showing a sex ratio of 1F:0.7M and a significant difference (χ^2=2.283) of 1:1 as expected for *Tetrapedia* species.

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Temporal distribution of nesting activity

The nesting activity was more intense in the rainy season (October - March), with 67 nests collected (74.4%) in this period (Fig. 2). When related to climatic factors, a positive and significant correlation (r=0.42; p=0.041) was found for the monthly number of nests and the mean monthly precipitation values. However, no significant correlation (r=0.33; p=0.118) was found between the monthly number of nidifications and the monthly mean temperature values. The months with the highest nidification rate were December/16 (n=13) and December/17 (n=12), representing 26.6% of the nests collected.

In order to understand the relationship between nest abundance and rainfall, the temporal distribution of activities for the three most nesting species was analyzed: *T. peckoltii* (n=36), *T. diversipes* (n=22) and *T. albitarse* (n=18) (Fig. 3). No significant correlation was found between the mean monthly precipitation values and monthly number of nests of *T. peckoltii* (r=0.32; p=0.125), *T. diversipes* (r=0.11; p=0.612) and *T. albitarse* (r=0.25; p=0.224). The period between the nesting to emergence of adults of *T. diversipes* and *T. peckoltii* varied from four to seven months.

Nine Centridini bee nests were collected: *Centris analis* (Fabricius, 1804) (n=4), *C. tarsata* (n=4) and *Centris (Hemisiella) trigonoides* Lepeletier, 1841 (n=1). *Centris tarsata* nests were collected in both seasons (50% dry; 50% rainy), while *C. trigonoides* nests nested only in the rainy season.

Parasitic nests

Parasitized nests represent 16% (n=15) of the 94 collected nests. A total of 28 parasites were sampled, representing 13.8% of all individuals collected (Table 3). The bee nests were the most parasitic (n=10), *T. peckoltii* being the species that had more nests occupied by parasites (n=6). The beetle *Nemognatha* sp. Illiger, 1807 was the most frequent parasite, occurring in the transition and Cerrado areas*. Tetrapedia peckoltti* was the most parasitized species by *Nemognatha* sp., representing 75% of the nests parasitized by beetles. In one of these nests, besides an individual of *Nemognatha* sp., an individual of the parasitic bee *Coelioxoides waltheriae* Ducke, 1908 was observed. Two wasp species from the Ichneumonidae family were observed parasitizing wasp nests *Priochilus* sp. and *T. albitarse*. Two other wasp species of the genus *Leucospis* Fabricius, 1775 also parasitized *C. tarsata* and *T. albitarse* nests. From the only nest of *T. diversipes* parasitized, emerged a diptera of the genus *Anthrax* cf. *irrotatus* Say, 1823, the only record in which host and parasite completed development. A nest of *T. albitarse* was parasitized by nine individuals of Miltogramminae sp. that did not emerge, although they were in the adult stage.

DISCUSSION

The number of nests collected (n=94) at Água Limpa Farm, during the two years of sampling, is similar to that observed for other recent studies in the Cerrado area (Mesquita & Augusto, 2011; Araújo et al., 2016). Mesquita & Augusto (2011) recorded 96 nests in an area composed of typical Cerrado and forest vegetation, in the Triângulo Mineiro region in the State of Minas Gerais (MG). Araújo et al. (2016), in a campo rupestre area of Diamantina (MG), recorded 97 nests over 15 months of study. However, when compared to studies in Caatinga - another open area biome - the number of nests is generally lower: arboreal Caatinga (n=121; n=248; n=368), Restinga (n=193) (Aguiar et al., 2005; Melo & Zanella, 2010; Melo & Zanella, 2012).

The higher rate of nesting by bees than wasps has also been reported for other studies in Cerrado and Atlantic Forest (Camillo et al., 1995; Aguiar & Martins, 2002). However, other studies in the Amazon (Morato & Campos, 2000; Morato, 2001a) and in a remaining urban forest in Belo Horizonte (MG) (Loyola & Martins, 2006) registered wasps with a higher occupation rate than bees. This difference can be related to the environment, as well as to method variations, where different substrates attract different species (Staab et al., 2018).

Ten species nested in the trap-nests, but it is important to consider that the three most abundant species, two species of bees *T. peckoltii, T. diversipes,* and one species of wasp *Trypoxylon albitarse*, accounted for 70.2% of the nests established. This pattern, with few species representing higher nesting rates, is already reported in other studies with trap-nests (Assis & Camillo, 1997; Viana et al., 2001; Aguiar & Martins, 2002; Nascimento & Garófalo, 2014; Oliveira & Gonçalves, 2017).

This may be related to the capacity of some species to have a higher degree of specificity with the resources available for feeding and nesting in the study area (Mendes & Regô, 2007).

Several factors interfere with species sampling in trap-nests studies, for example, the type of trap used, the duration of the study, the local vegetation, the diameter and length of the nests available (Staab et al., 2018). Some species use different substrates for nesting – *C. analis*, *C. tarsata*, *Tetrapedia curvitarsis* Friese, 1899, *T. diversipes*, *Tetrapedia rugulosa* Friese, 1899 – such as bamboo and cardboard (Aguiar & Garófalo, 2004; Camillo, 2005, Garófalo, 2008). In this study, the use of trap-nests of only one substrate (cardboard) possibly determined the absence of commonly sampled species in studies with trap-nests as *Euglossa* spp., *Xylocopa* spp., and *Megachile* spp., which nest preferentially in bamboo or wood (Augusto & Garófalo, 2004; Marchi & Melo, 2010; Pereira & Garófalo, 2010).

The highest number of nests in the rainy season (69.5%) is similar to that already reported in the literature (Camillo et al., 1995; Pereira et al., 1999; Aguiar & Martins, 2002). For Melo & Zanella (2012), the highest nesting rates in the rainy season could be related to greater availability of resources for nest development and construction. In FAL, *T. diversipes* (59.1%) and *T. peckoltii* (77.8%) presented higher nesting rates in the rainy season, as already observed in other studies for species of *Tetrapedia*

(Camillo, 2005; Cavalcante et al., 2019). *Tetrapedia* females are commonly observed collecting floral oil from Malpighiaceae (Neff & Simpson, 1981), and pollen from *Dalechampia* (Euphorbiaceae) and *Ludwigia* (Onagraceae) species (Menezes et al., 2012; Neves et al., 2014). Malpighiaceae, *Dalechampia* and *Ludwigia* species are found in the study area and in the rainy season *Tetrapedia* bees are frequently observed foraging on these flowers. Changes in nest frequency over the months could also be influenced by parasites, pathogens and/or predators that can cause mortality of bees in the larval or pupal stage (Frankie et al., 1988).

The number of nests in the Transition (n=48) and Cerrado (n=40) areas were similar. However, in the Forest area only six nests were collected. For Macarthur & Macarthur (1961), environments with higher structural complexity present greater availability of resources and, consequently, larger biodiversity. Thus the Transition area may have provided the highest abundance of bees and wasps due to a greater availability of substrates for nesting and floral resources. According to Fye (1972), open areas provide higher light penetration, favoring a greater diversity of plant cover and, consequently, food availability for bees and wasps.

The species richness (n=10) sampled in the FAL is similar to that observed for other studies conducted in the Cerrado: preserved Cerrado (n=10), Cerrado and forest area (n=8), campo rupestre (n=8) (Mesquita & Augusto, 2011; Araújo et al., 2016). If compared to studies on Caatinga phytophysiology, our results are higher than those recorded for Restinga areas in Salvador (n=7) and Mata de São João (n=5), both in the state of Bahia (Viana et al. 2001; Oliveira-Rebouças et al., 2018), but smaller than those observed in an area of herbaceous caatinga (n=18; n=17), in the State of Rio Grande do Norte (Melo & Zanella 2010, 2012). *Tetrapedia diversipes* and *T. peckoltti*, the species with the highest number of nests collected, are often listed in several studies with trap-nests (Aguiar & Martins, 2002; Alves-dos-Santos, 2003; Garófalo, 2008; Gazola & Garófalo, 2009; Rocha-Filho et al., 2017; Oliveira-Rebouças et al., 2018). According to Santos et al. (2020), *T. diversipes* presents philopatric behaviour, which may be one of the factors that favor the formation of aggregations of females nesting in the same area and, consequently, being sampled frequently in trap-nests.

Tripoxylon wasps are widely distributed and are also commonly collected in trap-nest studies conducted in different regions and biomes of Brazil (Camillo et al., 1995; Assis & Camillo, 1997; Morato & Campos, 2000; Viana et al., 2006; Melo & Zanella, 2010). Although not sampled with high frequency as in other studies, *C. tarsata* and *C. analis* are also common species in studies with trap-nests (Jesus & Garófalo, 2000; Loyola & Martins, 2006; Garófalo, 2008; Gazola & Garófalo, 2009; Rocha-Filho et al., 2017; Oliveira-Rebouças et al., 2018). In recent studies, in the Cerrado, both species were among the most abundant in relation to the number of nests founded (Mesquita & Augusto, 2011; Araújo et al., 2016). Still considering open areas, studies conducted in arboreal caatinga (Aguiar et al., 2005), herbaceous caatinga (Melo & Zanella, 2010; Melo & Zanella; 2012) and restinga (Viana et al., 2001; Oliveira-Rebouças et al., 2018), presented *C. tarsata* as the species with the highest nest occupation rate. In this study, *C. tarsata* nests were collected in the same proportion in dry and rainy seasons. In the literature, the highest nesting activity pattern of *C. tarsata* has been recorded for both the dry season (Aguiar & Martins, 2002; Mendes & Rego, 2007) and the rainy season (Aguiar & Garófalo, 2004). Studies evaluating the availability of floral resources in the dry and rainy periods may help to better understand the nesting behaviour of *C. tarsata* (Aguiar & Garófalo, 2004; Melo & Zanella, 2012).

When compared to the three areas the lowest species richness is clearly observed for Forest area (n=2). Although *T. diversipes* and *C. tarsata* are frequent in studies in forest areas such as semi-deciduous forest (Garófalo, 2008; Gazola & Garófalo, 2009), none of the bee species nested in the forest area. Surrounded by a stream or small rivers, the gallery forests are composed of trees that vary between 20 and 30 meters in height, providing a shaded perimeter and quite humid that can comprise 70 to 95% of the area (Ribeiro & Walter, 2008). Thus, one of the reasons for the reduced nesting rate may be the high humidity inside the forest, which may provide the proliferation of fungi, termites or ants in the nesting sites (Mendes & Regô, 2007). Studies with solitary bees in forest areas have shown high mortality rates caused by fungi, which may imply a greater difficulty in colonizing these environments (Morato 2001 b; Camarotti-de-Lima & Martins, 2005). Although the occurrence of species in the forest area has been low, forests in tropical regions represent a favorable environment for bee and wasp nesting (Morato & Martins 2006). Mainly in preserved forests, which have more trees that die standing, providing cavities that can be used by different animals (Moorman et al. 1999; Morato & Martins 2006).

The higher nesting rate in the rainy season could also be related to physiological aspects such as diapause. *Tetrapedia diversipes*, for example, a bivoltine bee oil-collecting endemic to the Neotropical region, usually produces the first generation in the rainy season (G1) (Santos et al., 2018). Thus, while the first generation of individuals shows complete development (egg to adult) more quickly (74.4 days average), the second generation (G2), which occurs in the dry season, takes longer to complete development (224.6 days average), presenting a drastic reduction in activities associated with nesting (Alves-dos-Santos et al., 2002; Alves-dos-Santos et al., 2006). Our results suggest that *T. diversipes* and *T. peckoltii* bees originated from nests collected in the dry season and with longer development (varying between four and seven months) presented diapause, similar to that observed in other studies with *T. diversipes* (Alves-dos-Santos et al., 2002; Camilo, 2005; Rocha-Filho & Garófalo, 2015). Thus, in solitary bees that nest in pre-existing cavities, diapause could represent an important strategy to survive unfavorable conditions of a certain period (e.g., dry season) such as environmental and climatic factors (Danforth, 1999; Aguiar & Garófalo, 2004).

The occurrence of *Nemognatha, Leucospis, Anthrax* and *Coelioxoides* parasitoids is commonly reported in studies with trap nests representing important elements of monitoring (Krombein, 1967; Garófalo 2008, Gazola & Garófalo, 2009; Mesquita & Augusto, 2011; Araújo et al., 2016; Rocha-Filho et al., 2017; Lima et al., 2018, Cordeiro et al., 2019). In the present study, the parasitoids attacked mostly a single nesting species, with the exception of *Nemognatha* sp. which was observed in nests of *C. analis* and also of *T. peckoltii.* Evaluating the structure of the bee-parasitic interaction network in a

Cerrado and Atlantic Forest areas, Lima et al. (2018) reported that few host species were parasitized, although many of these parasitoids are specialized in attacking different species of bees and insects. *Nemognatha* sp. was the most abundant parasite, mainly in the nests of *C. analis* and *T. peckoltii*. Meloidae beetles are often reported in the literature parasitizing bee and wasp nests (Aguiar & Martins, 2002; Pereira & Garófalo, 2010). Among the parasitic wasps, individuals from the family Ichneumonidae were observed (Peruquetti & Del Lama, 2003; Loyola & Martins, 2006; Oliveira & Gonçalves, 2017) and from the genus *Leucospis* (Cooperband et al., 1999; Gazola & Garófalo, 2003; Oliveira & Gonçalves, 2017), also commonly found in other studies. Only one individual of *Anthrax* cf. *irrotatus* was observed parasitizing a *T. diversipes* nest, however, other authors have reported the presence of different flies of the genus in *T. albitarse*, *C. tarsata* and *T. diversipes* nests (Assis & Camillo, 1997; Cavalcante et al., 2019). A nest of *T. albitarse* was parasitized by Miltogramminae sp., a species of diptera that develops in cells of bees and wasps that nest in cavities, feeding on provisions and in some cases on eggs and larvae (Rafael et al., 2012). For Tscharntke et al. (1998), the interactions between the hymenopterous community that nest in pre-existing cavities and their respective parasites represent important bioindicators of ecological change and habitat quality. Thus, a better understanding of biological aspects of insects that parasitize bee and wasp nests may provide greater possibilities for monitoring and conservation of local entomofauna.

CONCLUSION

Ten nesting species were recorded in the FAL, representing a diversity similar to studies conducted in the Cerrado. The correlation observed between the rainy season and nesting rate may be an indication that not only the availability of resources, but also abiotic factors and physiological attributes such as diapause directly interfere in the behaviour of these insects. Studies that focus on behavioural and physiological aspects of bees and wasps that nest in pre-existing cavities can provide important data for a better understanding of the biology of these organisms, including in regions with well-defined dry and rainy seasons, such as the Cerrado. Investigating the effect of man-made actions on bee and wasp communities is also necessary in order to measure the impact on these insects, which are of relevant importance in ecological processes.

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REFERENCES

Aguiar, C.M.L. & Garófalo, C.A. 2004. Nesting biology of *Centris (Hemisiella) tarsata* Smith (Hymenoptera, Apidae, Centridini). *Revista Brasileira de Zoologia*, 21(3):477-486. Available at: https://doi.org/10.1590/S0101-81752004000300009

Aguiar, C.M.L.; Garófalo, C.A. & Almeida, G.F. 2005. Trap-nesting bees (Hymenoptera, Apoidea) in areas of dry semideciduous forest and Caatinga, Bahia, Brazil. *Revista Brasileira de Zoologia*, 22:1030-1038. Available at: https://doi.org/10.1590/S0101-81752005000400031

Aguiar, A.J.C. & Martins, C.F. 2002. Abelhas e vespas solitárias em ninhos armadilha na Reserva Biológica Guaribas (Mamanguape, Paraíba, Brasil). *Revista Brasileira de Zoologia*, 19:101-116. Available at: http://dx.doi.org/10.1590/S0101-81752002000500005

Alvares, C.A.; Stape, J.L.; Sentelhas, P.C.; Gonçalves, J.L.M. & Sparovek, G. 2013. Köppen's climate classification map for Brazil. *Meteorologische Zeitschrift*, 22:711-728.

Alves-dos-Santos, I.; Melo, G.A.R. & Rozen, J.G.J. 2002. Biology and immature stages of the bee tribe Tetrapediini (Hymenoptera, Apidae). *American Museum Novitates*, 3377:1-45. Available at: http://hdl.handle.net/2246/2866

Alves-dos-Santos, I. 2003. Trap-nesting bees and wasps on the university campus in São Paulo, Southeastern Brazil (Hymenoptera: Aculeata). *Journal of the Kansas Entomological Society*, 76: 328-334. Available at: https://www.jstor.org/stable/25086119

Alves-dos-Santos, I.; Naxara, S.R.C. & Patrício, E.F.L.R.A. 2006. Notes on the morphology of *Tetrapedia diversipes* Klug 1810 (Tetrapediini, Apidae), an oil-collecting bee. Brazilian *Journal of Morphological Sciences*, 23:425-430. Available at: http://www.jms.periodikos.com.br/article/587cb4647f8c9d0d058b4649

Araújo, P.C.S.; Lourenço, A.P. & Raw, A. 2016. Trap-nesting bees in Montane Grassland (Campo Rupestre) and Cerrado in Brazil: collecting generalist or specialist nesters. *Neotropical Entomology*, 45: 482-489. Available at: https://doi.org/10.1007/s13744-016-0395-9

Assis, J.M.F. & Camillo, E. 1997. Diversity, seasonality and nesting biology of solitary wasps (Hymenoptera: Sphecidae: Vespidae) in Trap-Nests in the Ituiutaba Region, MG. *Anais da Sociedade Entomológica do Brasil*, 26:335-347. Available at: https://doi.org/10.1590/S0301-80591997000200016

Augusto, S.C. & Garófalo, C.A. 2004. Nesting biology and social structure of *Euglossa (Euglossa) townsendi* Cockerell (Hymenoptera, Apidae, Euglossini). *Insectes Sociaux*, 51: 400-409. Available at: https://doi.org/10.1007/s00040-004-0760-2

Batra, S.W.T. 1984. Solitary bees. *Scientific American*, 250:120-127.

Buschini, M.L.T. 2006. Species diversity and community structure in trapnesting bees in Southern Brazil. *Apidologie*, 37: 58-66. Available at: https://doi.org/10.1051/apido:2005059

Buschini, M.L.T. & Woiski, T.D. 2008. Alpha–beta diversity in trap-nesting wasps (Hymenoptera: Aculeata) in Southern Brazil. *Acta Zoologica (Stockholm)*, 89:351-358. Available at: https://doi.org/10.1111/j.1463-6395.2008.00325.x

Camarotti-de-Lima, M.F. & Martins, C.F. 2005. Biologia de nidificação e aspectos ecológicos de *Anthodioctes lunatus* (Smith) (Hymenoptera: Megachilidae, Anthidiini) em área de tabuleiro nordestino, PB. *Neotropical Entomology*, 34(3):375-380. Available at: http://dx.doi.org/10.1590/S1519-566X2005000300003

Camillo, E. 2005. Nesting biology of four *Tetrapedia* species in trap-nests (Hymenoptera: Apidae: Tetrapediini). *Revista de Biologia Tropical*, 53:175-186.

Camillo, E.; Garófalo, C.A; Serrano, J.C.& Muccillo, G. 1995. Diversidade e abundância sazonal de abelhas e vespas solitárias em ninhos armadilhas (Hymenoptera, Apocrita, Aculeata). *Revista Brasileira de Entomologia*, 39:459-470. Available at: http://dx.doi.org/10.1590/S1519-566X2001000200006

Cavalcante, A.M.; Silva, C.I.; Gomes, A.M.S.; Pinto, G.S.; Bonfim, I.G.A. & Freitas, B.M. 2019. Bionomic Aspects of the Solitary Bee *Tetrapedia diversipes* Klug, 1810 (Hymenoptera: Apidae: Tetrapediini). *Sociobiology*, 66(1): 113-119. Available at: http://dx.doi.org/10.13102/sociobiology.v66i1.3338

Cooperband, M.F.; Wharton, R.A.; Frankie, G.W. & Vinson, S.B. 1999. New host and distribution records for *Leucospis* (Hymenoptera: Leucospidae) associated primarily with nests of *Centris* (Hymenotpera: Anthophoridae) in the dry forests of Costa Rica. *Journal of Hymenoptera Research*, 8:154-164.

Cordeiro, G.D.; Boff, S. & Alves-dos-Santos, I. 2019. Trap-nesting Bees from Protected Areas of Atlantic Forest, Southeastern Brazil. Sociobiology, 66(2):306-315. Available at: http://dx.doi.org/10.13102/sociobiology.v66i2.3448

Costa, C.C. & Gonçalves, R. 2019. What do we know about Neotropical trap-nesting bees? Synopsis about their nest biology and taxonomy. *Papéis Avulsos de Zoologia*, 59, e20195926. Available at: https://doi.org/10.11606/1807-0205/2019.59.26

Danforth, B.N. 1999. Emergence dynamics and bet hedging in a desert bee, *Perdita portalis*. *Proceedings of the Royal Society of London. Series B, Biological Sciences*, 266(1432):1985-1994. Available at: https://doi.org/10.1098/rspb.1999.0876

Frankie, G.W.; Vinson, S.B.; Newstrom, L.E. & Barthell J.F. 1988. Nest site and habitat preferences of *Centris* bees in Costa Rican dry forest. *Biotropica*, 20:301-310. Available at: https://www.jstor.org/stable/2388320

Frankie, G.W.; Thorp, R.W.; Newstrom-Lloyd, L.E.; Rizzardi, M.A.; Barthell, J.F.; Griswold, T.L.; Kim, J.Y. & Kappagoda, S. 1998. Monitoring solitary bees in modified wildland habitats: implications for bee ecology and conservation. *Environmental Entomology*. 27:1137-1148. Available at: https://doi.org/10.1093/ee/27.5.1137

Fye, R.E. 1972. The effect of forest disturbances on populations of wasps and bees in Northwestern Ontario (Hymenoptera: Aculeata). *Canadian Entomology*, 104:1623-1633. Available at: https://doi.org/10.4039/Ent1041623-10

Garófalo, C.A. 2008. Abelhas (Hymenoptera, Apoidea) Nidificando em Ninhos-Armadilha na Estação Ecológica dos Caetetus, Gália, SP. *Anais do VIII Encontro sobre Abelhas-Ribeirão Preto, SP*, p.208-217.

Gazola, A.L. & Garófalo, C.A. 2003. Parasitic behavior of *Leucospis cayennensis* Westwood (Hymenoptera, Leucospidae) and rates of parasitism in populations of *Centris (Heterocentris)* *analis* (Fabricius 1804) (Hymenoptera: Apidae: Centridini). *Journal of the Kansas Entomological Society*, 76:131-142. Available at: https://www.jstor.org/stable/25086098

Gazola, A.L. & Garófalo, C.A. 2009. Trap-nesting bees (Hymenoptera: Apoidea) in forest fragments of the State of São Paulo, Brazil. *Genetics and Molecular Research*, 8: 607-662.

Jesus, B.M.V. & Garófalo, C.A. 2000. Nesting behaviour of *Centris (Heterocentris) analis* (Fabricius) in southeastern Brazil (Hymenoptera, Apidae, Centridini). *Apidologie*, 31:503-515. Available at: https://doi.org/10.1051/apido:2000142

Krombein, K. 1967. *Trap-nesting wasps and bees. Life-histories, nests and associates*. Smithsonian Press, Washington.

Krug, C. & Alves-dos-Santos, I.O. 2008. Uso de diferentes métodos para amostragem da fauna de abelhas (Hymenoptera: Apoidea), um estudo em Floresta Ombrófila Mista em Santa Catarina. *Neotropical Entomology*, 37:265-278. Available at: http://dx.doi.org/10.1590/S1519-566X2008000300005

Lima, R.; Moure-Oliveira, D. & Garófalo, C.A. 2018. Interaction Network and Niche Analysis of Natural Enemy Communities and their Host Bees (Hymenoptera: Apoidea) in fragments of Cerrado and Atlantic Forest. *Sociobiology*, 65(4): 591-602. Available at: http://dx.doi.org/10.13102/sociobiology.v65i4.3386

Loyola, R. & Martins, R.P. 2006. Trap-nest occupation by solitary wasps and bees (Hymenoptera: Aculeata) in a Forest Urban Remanent. *Neotropical Entomology*, 35:41-48. Available at: http://dx.doi.org/10.1590/S1519-566X2006000100006

Macarthur, R.H. & Macarthur, J.W. 1961. On bird species diversity. *Ecology*, 42:594-598. Available at: https://doi.org/10.2307/1932254

Marchi, P. & Melo, G.A.R. 2010. Biologia de nidificação de *Xylocopa (Neoxylocopa) frontalis* (Olivier) (Hymenoptera, Apidae, Xylocopini) em Morretes, Paraná. *Oecologia Australis*, 14:210-231. Available at: doi:10.4257/oeco.2010.1401.12

Melo, R.R. & Zanella, S.C.V. 2010. Sazonalidade na nidificação de abelhas e vespas solitárias em ninhos-armadilha em área de Caatinga. *Sitentibus Série Ciências Biológicas*, 10: 258-266.

Melo, R.R. & Zanella, F.C.V. 2012. Dinâmica de Fundação de Ninhos por Abelhas e Vespas Solitárias (Hymenoptera, Aculeta) em Área de Caatinga na Estação Ecológica do Seridó. *Revista Brasileira de Ciências Agrárias*, 7(4):657-662. Available at: DOI:10.5039/agraria.v7i4a1966

Mendes, F.N. & Rego, M.M.C. 2007. Nidificação de *Centris (Hemisiella) tarsata* Smith (Hymenoptera, Apidae, Centridini) em ninhos-armadilha no Nordeste do Maranhão, Brasil. *Revista Brasileira de Entomologia*, 51:382-388. Available at: http://dx.doi.org/10.1590/S0085-56262007000300017

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. Available at: http://dx.doi.org/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.

Michener, C.D. 2007. *The Bees of The world*. John Hopkins University Press, Baltimore.

Mittermeier, R.A.; Turner, W.R.; Larsen, F.W.; Brooks, T.M.& Gascon, C. 2011. *Global Biodiversity Conservation: The Critical Role of Hotspots.* In: Zachos, F.E. & Habel, J.C. (Eds.). Biodiversity Hotspots: Distribution and protection of conservation priority areas, Springer, Berlin Heidelberg. Available at: https://doi.org/10.1007/978-3-642-20992-5_1

MMA. 2016. *Ministério do Meio Ambiente*. Available at: http://www.mma.gov.br/informma/item/10724-resultados-da-2%C2%B0-atualiza%C3%A7%C3%A3o-das-%C3%A1reas-priorit%C3%A1rias.html. Access in: 14/01/2020.

Moorman, C.E.; Russell, K.R.; Sabin, G.R. & Guynn Jr, D.C. 1999. Snag dynamics and cavity occurrence in the South Carolina Piedmont. *Forest Ecology and Management*, 118:37-48. Available at: http://dx.doi.org/10.1016/S0378-1127(98)00482-4

Morato, E.F. 2001a. Efeitos da fragmentação florestal sobre abelhas e vespas solitárias na Amazônia Central. II. Estratificação vertical. *Revista Brasileira de Zoologia*, 18:737-747. Available at: https://doi.org/10.1590/S0101-81752001000300010

Morato, E.F. 2001b. Biologia e ecologia de *Anthodioctes moratoi* Urban (Hymenoptera, Megachilidae, Anthidiini) em matas contínuas e fragmentos na Amazônia Central, Brasil. *Revista Brasileira de Zoologia*, 18(3):729-736. Available at: https://doi.org/10.1590/S0101-81752001000300009

Morato, E.F. & Martins, R.P. 2006. An Overview of Proximate Factors Affecting the Nesting Behavior of Solitary Wasps and Bees (Hymenoptera: Aculeata) in Preexisting Cavities in Wood. *Neotropical Entomology*, 35(3):285-298. Available at: https://doi.org/10.1590/S1519-566X2006000300001

Morato, E.F.& Campos, L.A.O. 2000. Efeitos da fragmentação florestal sobre vespas e abelhas solitárias em uma área da Amazônia Central. *Revista Brasileira de Zoologia*, 17(2):429-444. Available at: https://doi.org/10.1590/S0101-81752000000200014

Nascimento, A.L.O. & Garófalo, C.A. 2014. Trap nesting solitary wasps (Hymenoptera: Aculeata) in an insular landscape: mortality rates for immature wasps, parasitism, and sex ratios. *Sociobiology*, 61:207-217. Available at: http://dx.doi.org/10.13102/sociobiology.v61i2.207-217

Neff, J.L. & B.B. Simpson. 1981. Oil-colleting structures in the Anthophoridae (Hymenoptera): Morphology, function, and use in systematics. *Journal of the Kansas Entomological Society*, 54: 95-123. Available at: https://www.jstor.org/stable/25084137

Nether, M.C.; Dudek, J.; Buschini, M.L.T. 2019. Trophic interaction and diversity of cavity-nesting bees and wasps (Hymenoptera: Aculeata) in Atlantic forest fragments and in adjacent matrices. *Apidologie*, 50:104-115. Available at: https://doi.org/10.1007/s13592-018-0623-x

Neves, C.M.L.; Carvalho, C.A.L.; Machado, C.S.; Aguiar, C.M.L. & Sousa, F.S.M. 2014. Pollen consumed by the solitary bee *Tetrapedia diversipes* (Apidae: Tetrapediini) in a tropical agroecosystem. *Grana*, 53:302-308. Available at: https://doi.org/10.1080/00173134.2014.931455

Oliveira, P.S. & Gonçalves, R.B. 2017. Trap-nesting bees and wasps (Hymenoptera, Aculeata) in a Semidecidual Seasonal Forest fragment, southern Brazil. *Papéis Avulsos de Zoologia*, 57(13):149-156. Available at: https://doi.org/10.11606/0031-1049.2017.57.13

Oliveira-Rebouças, P.; Aguiar, C.M.L.; Ferreira, V.S.; Sodré, G.S.; Carvalho, C.A.L & Gimenes, M. 2018. The Cavity-Nesting Bee Guild (Apoidea) in a Neotropical Sandy Coastal Plain. *Sociobiology*, 65(4): 706-713. Available at: http://dx.doi.org/10.13102/sociobiology.v65i4.3339

Pereira, M. & Garófalo, C.A. 2010. Biologia da nidificação de *Xylocopa frontalis* e *Xylocopa grisescens* (Hymenoptera, Apidae, Xylocopini) em ninhos-armadilha. *Oecologia Australis*, 14(1):193-209. Available at: doi:10.4257/oeco.2010.1401.11

Pereira, M.; Garófalo, C.A.; Camillo, E. & Serrano, J.C. 1999. Nesting biology of *Centris (Hemisiella) vittata* Lepeletier in Southern Brazil (Hymenoptera, Apidae, Centridini). *Apidologie*, 30:327-338. Available at: http://dx.doi.org/10.1590/S0101-81752004000300009

Peruquetti, R.C. & del lama, M.A. 2003. Notas sobre a socialidade e a biologia de nidificação de *Trypoxylon (Trypoxylon) asuncicola* Strand, 1910 (Hymenoptera, Sphecidae). *Revista Brasileira* *de Entomologia*, 47(2):297-301. Available at: http://dx.doi.org/10.1590/S0085-56262003000200018

R Development Core Team. 2016. *R: A Language and Environment for Statistical Computing.* R Foundation for Statistical Computing, Vienna. Disponível em: www.R-project.org.

Rafael, J.A.; Melo, G.A.R.; Carvalho, C.J.B.; Casari, S.A. & Constantino, R. 2012. *Insetos do Brasil: diversidade e taxonomia.* Holos, Ribeirão Preto.

Ribeiro, J.F. & Walter, B.M.T. 2008. *As fitofisionomias do bioma Cerrado*. In: Sano, S.M.; Almeida, S.P. & Ribeiro, J.F. (Eds.). Cerrado: ecologia e flora, Embrapa Cerrados, Planaltina. Rocha-Filho, L.C. da & Garófalo, C.A. 2015. Natural History of *Tetrapedia diversipes* (Hymenoptera: Apidae) in an Atlantic Semideciduous Forest Remnant Surrounded by Coffee Crops, Coffea arabica (Rubiaceae). *Annals of the Entomological Society of America*, 109(2): 183-197. Available at: https://doi.org/10.1093/aesa/sav153

Rocha-Filho, L.C. da, Rabelo, L.S.; Augusto, S.C. & Garófalo, C.A. 2017. Cavity nesting bees and wasps (Hymenoptera: Aculeata) in a semi-deciduous Atlantic forest fragment immersed in a matrix of agricultural land. Journal of Insect Conservation, 21(4): 727-736. Available at: https://doi.org/10.1007/s10841-017-0016-x

Santos, P.K.F.; Araujo, N.S.; Françoso, E.; Zuntini, A.R. & Arias, M.C. 2018. Diapause in a tropical oil-collecting bee: molecular basis unveiledby RNA-Seq. *BMC Genomics*, 19:305. Available at: doi: 10.1186/s12864-018-4694-x

Santos, P.K.F.; Araujo, N.S.; Françoso, E.; Cordeiro, G.D.; Alves-dos-Santos, I. & Arias, M.C. 2020. Genetic analyses reveal female philopatric behavior and nest usage by multiple females of the solitary oil-collecting bee Tetrapedia diversipes (Hymenoptera: Apidae). Available at: https://doi.org/10.1007/s13592-020-00763-4

Silveira, F.A.; Melo, G.A.R. & Almeida, E.A.B. 2002. Abelhas brasileiras: Sistemática e identificação. Belo Horizonte.

Staab, M.; Pufal, G.; Tscharntke, T. & Klein, A. 2018. Trap nests for bees and wasps to analyse trophic interactions in changing environments — A systematic overview and user guide. *Methods in Ecology and Evolution*, 9:2226–2239. Available at: https://doi.org/10.1111/2041-210X.13070

Stangler, E.S.; Hanson, P.E. & Steffan-Dewenter, I. 2014. Interactive effects of habitat fragmentation and microclimate on trap-nesting Hymenoptera and their trophic interactions in small secondary rainforest remnants. Biodiversity and Conservation, 24: 563-578. Available at: https://doi.org/10.1007/s10531-014-0836-x

Steffan-Dewenter, I. 2002. Landscape context affects trap-nesting bees, wasps, and their natural enemies. *Ecological Entomology*, 27:631-637. Available at: https://doi.org/10.1046/j.1365-2311.2002.00437.x

Tommasi, D.; Miro, A.; Higo, H.A. & Winston, M.L. 2004. Bee diversity and abundance in an urban setting. *The Canadian Entomologist*, 136:851-869. Available at: https://doi.org/10.4039/n04-010

Tscharntke, T.; Gathmann, A. & Steffan-Dewenter, I. 1998. Bioindication using trap-nesting bees and wasps and their natural enemies: community structure and interactions. *Journal of Applied Ecology*, 35:708-719. Available at: https://doi.org/10.1046/j.1365-2664.1998.355343.x

Torné-Noguera, A.; Arnan, X.; Rodrigo, A. & Bosch, J. 2020. Spatial variability of hosts, parasitoids and their interactions across a homogeneous landscape. Ecology and Evolution, 10(8): 3696-3705. Available at: https://doi.org/10.1002/ece3.6158

Viana, B.F.; Silva, F.O. & Kleinert, A.M.P. 2001. Diversidade e sazonalidade de abelhas solitárias (Hymenoptera: Apoidea) em dunas litorâneas no nordeste do Brasil. *Neotropical Entomology*, 30(2):245-251. Available at: https://doi.org/10.1590/S1519-566X2001000200006

Viana, B.F.; Melo, A.M.C. & Drumond, P.D. 2006. Variação na estrutura do habitat afetando a composição de abelhas e vespas solitárias em remanescentes florestais urbanos de Mata Atlântica no Nordeste do Brasil. *Sitientibus, Série Ciências Biológicas*, 6:282-295.

TABLES

TABLE 1. Number of nests and emerged adults of trap-nesting bees and wasps in Água Limpa Farm from June 2016 to May 2018.

Species	Nests collected (n)			Diameter of trap-nests (cm)	Emerged adults (n)
	Gallery forest	Transition	Cerrado		
Apidae					
Centris (Hemisiella) trigonoides Lepeletier, 1841	–	–	1	1.0	1
Centris (Hemisiella) tarsata (Smith,1874)	–	1	3	0.8/1.0	7
Centris (Heterocentris) analis (Fabricius, 1804)		3	1	1.0	7
Tetrapedia diversipes Klug,1810	–	19	3	0.6/0.8	55
Tetrapedia peckoltii Friese, 1899	–	10	26	0.6/0.8/1.0	53
Megachilidae					
Epanthidium tigrinum (Schrottky, 1905)	–	2	–	0.6	2
Crabronidae					
Trypoxylon albitarse (Fabricius,1804)	3	11	4	0.8/1.0	32
Trypoxylon sp. Latreille, 1796	–	–	2	0.6	7
Pompilidae					
Priochilus sp. Banks, 1944	3	1	–	0.8	9
Sphecidae					
Sphecini sp.	–	1	–	0.8	2
Total*	**6**	**48**	**40**		**203**

TABLE 2. Studies on the occurrence of trap-nesting bees and wasps in different Brazilian regions in the last 20 years.

Phytophysiognomy	Locality	Study group	Sampling (months)	Nests collected (n)	Nesting species (n)	Reference
Cerrado						
Cerrado *stricto sensu*; Galley forest	Brasília (DF)	Both	24	94	10	Present study
Cerrado undisturbed	Triângulo Mineiro (MG)	Bees	18	182	10	Mesquita & Augusto (2011)
Cerrado and forest	Triângulo Mineiro (MG)	Bees	18	96	8	Mesquita & Augusto (2011)
Campo rupestre	Diamantina (MG)	Bees	15	97	8	Araújo et al. (2016)
Caatinga						
Arboreal Caatinga	Ipirá (BA)	Bees	18	121	7	Aguiar et al. (2005)
Herbaceous Caatinga	Serra Negra do Norte (RN)	Both	24	368	18	Melo & Zanella (2010)
Herbaceous Caatinga	Serra Negra do Norte (RN)	Both	24	248	17	Melo & Zanella (2012)
Restinga	Salvador (BA)	Bees	24	62	7	Viana et al. (2001)
Restinga	Mata de São João (BA)	Bees	12	193	5	Oliveira-Rebouças et al. (2018
Atlantic Forest						
Agricultural area	Alhandra (PB)	Both	12	1857	14	Martins et al. (2012)
Agricultural area (Tabuleiro)	João Pessoa (PB)	Both	12	489	14	Martins et al. (2012)
Araucaria forest	Guarapuava (PR)	Bees	24	10	4	Buschini (2006)
Araucaria forest	Guarapuava (PR)	Wasps	36	523	12	Buschini & Woiski (2008)
Araucaria forest	Porto União (SC)	Bees	12	244	5	Krug & Alves-dos-Santos (2008

Araucaria forest	Guarapuava (PR)	Both	10	257	8	Nether et al. (2019)
Araucaria forest (forest removed)	Turvo (PR)	Both	10	296	12	Nether et al. (2019)
Forest urban remanent	Belo Horizonte (MG)	Both	10	137	11	Loyola & Martins (2006)
Grassland	Guarapuava (PR)	Bees	24	47	7	Buschini (2006)
Grassland	Guarapuava (PR)	Wasps	36	332	12	Buschini & Woiski (2008)
Ombrophilous dense forest	Salesópolis (SP)	Bees	24	847	17	Cordeiro et al. (2019)
Semideciduous forest; Tabuleiro forest	Mamanguape (PB)	Both	12	176	14	Aguiar & Martins (2002)
Semideciduous forest	Baixa Grande (BA)	Bees	28	146	11	Aguiar et al. (2005)
Semideciduous forest	Gália (SP)	Bees	18	218	9	Garófalo (2008)
Semideciduous forest	Paulo de Faria (SP)	Bees	24	507	16	Gazola & Garófalo (2009)
Semideciduous forest	Gália (SP)	Both	24	942	24	Rocha-Filho et al. (2017)
Semideciduous forest	Palotina (PR)	Both	24	80	10	Oliveira & Gonçalves (2017)
Semideciduous forest	São Paulo (SP)	Both	36	326	15	Alves-dos-Santos (2003)
Secondary forest	Ubatuba (SP)	Wasps	24	254	14	Nascimento & Garófalo (2014)
Swamp	Guarapuava (PR)	Bees	24	63	7	Buschini (2006)
Swamp	Guarapuava (PR)	Wasps	36	242	10	Buschini & Woiski (2008)
Urban fragments of forest	Salvador (BA)	Both	14	248	5	Viana et al. (2006)
Amazonia						
Primary forest	Manaus (AM)	Both	24	1934	38	Morato & Campos (2000)
Primary forest	Manaus (AM)	Both	24	1327	33	Morato (2001)

TABLE 3. Parasites species, number of parasitized nests and their respective host species in trap-nests at Água Limpa Farm from June 2016 to May 2018.

Parasitic species	Parasitic nests (n)	Emerging parasites (n)	Host species
Hymenoptera			
Coelioxoides waltheriae Ducke, 1908 (Apidae)	1*	1	*Tetrapedia peckoltii*
Leucospis sp. 1 Fabricius, 1775 (Leucospidae)	1	1	*Centris tarsata*
Leucospis sp. 2 Fabricius, 1775 (Leucospidae)	1	1	*Trypoxylon albitarse*
Orthocentrinae sp. (Ichneumonidae)	2	2	*Trypoxylon albitarse*
Tryphoninae sp. (Ichneumonidae)	1	1	*Priochilus* sp.
Diptera			
Anthrax cf. *irrotatus* Say, 1823 (Bombyliidae)	1	1	*Tetrapedia diversipes*
Miltogramminae sp. (Sarcophagidae)	1	9	*Trypoxylon albitarse*
Coleoptera			
Nemognatha sp. Illiger, 1807 (Meloidae)	2	3	*Centris analis*
	6*	9	*Tetrapedia peckoltii*
Total	**15**	**28**	

* In one nest of *Tetrapedia peckoltii* was parasitized concomitantly by *Coelioxoides waltheriae* and *Nemognatha* sp.

FIGURES

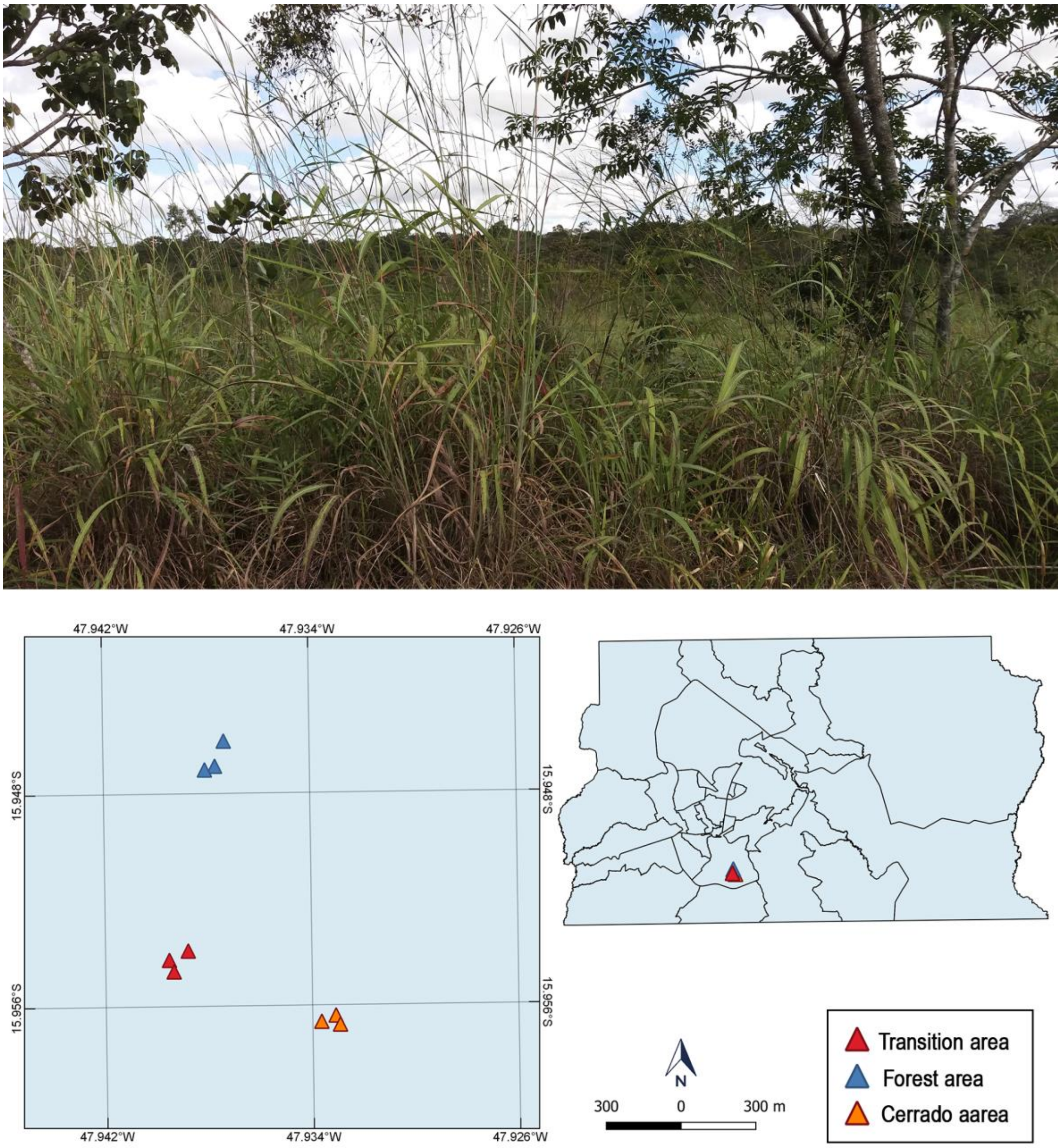


FIGURE 1. Study area at the Água Limpa Farm where the trap-nests were installed.

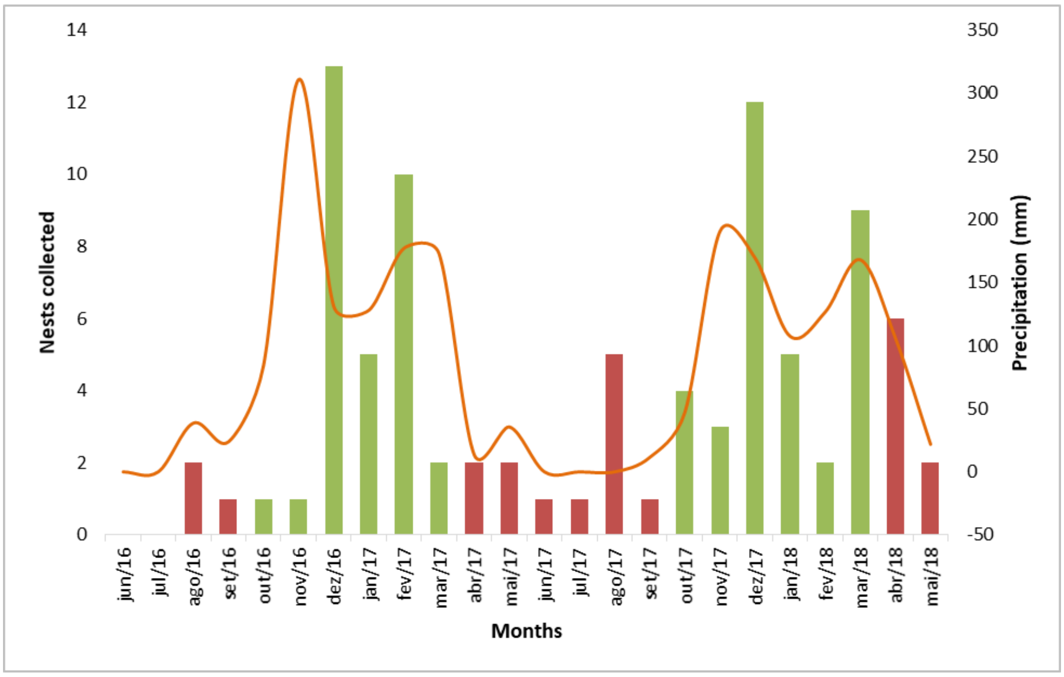


FIGURE 2. Number of nests collected monthly at the Água Limpa Farm related to the monthly precipitation values in the dry (red) and rainy (green) seasons from June 2016 to May 2018.

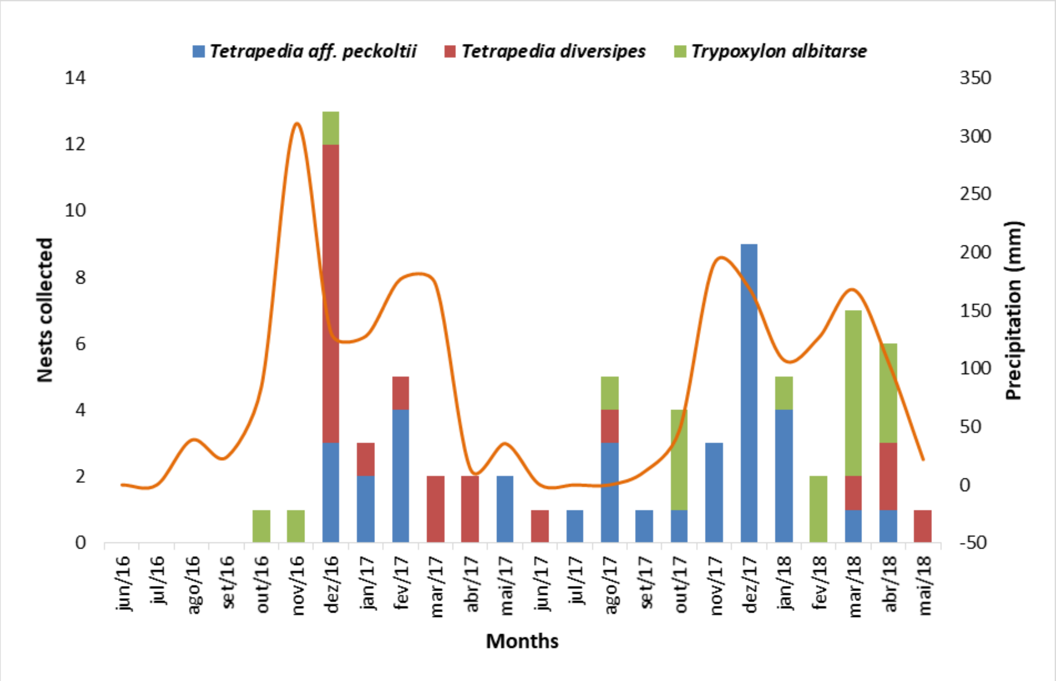


FIGURE 3. Number of nests collected from the most abundant species *Tetrapedia peckoltii*, *Tetrapedia diversipes* and *Trypoxylon albitarse* at Água Limpa Farm related to monthly precipitation values from June 2016 to May 2018.

APPENDIX

Capítulo 4

Trap-nesting bees and wasps in a Cerrado savanna area of Central Brazil

APPENDIX

APPENDIX 1. List of individuals sampled and deposited in the Entomological Collection of the University of Brasilia, with their respective voucher numbers.

***Anthrax* cf. *irrotatus* Say, 1823:** UNB063764.

***Centris (Hemisiella) trigonoides* Lepeletier, 1841:** UNB063836.

***Centris (Hemisiella) tarsata* (Smith,1874):** UNB065143, UNB065144, UNB065148, UNB065150, UNB065172, UNB065177, UNB065388, UNB065389.

***Centris (Heterocentris) analis* (Fabricius, 1804):** UNB064033, UNB064034, UNB064047, UNB064048, UNB064049, UNB064050, UNB064065.

***Coelioxoides waltheriae* Ducke, 1908:** UNB065379.

***Epanthidium tigrinum* (Schrottky, 1905):** UNB060085, UNB065667.

***Leucospis* sp. 1 Fabricius, 1775:** UNB065172.

***Leucospis* sp. 2 Fabricius, 1775:** UNB063924.

Miltogramminae sp.: UNB065394, UNB065395, UNB065396, UNB065397, UNB065398, UNB065399, UNB065400, UNB065401, UNB065402.

***Nemognatha* sp. Illiger, 1807:** UNB060086, UNB060087, UNB060089, UNB065378, UNB065380, UNB065381, UNB065382, UNB065383, UNB065384, UNB065385, UNB065391, UNB065392.

Orthocentrinae sp.: UNB063950, UNB065393.

***Priochilus* sp. Banks, 1944:** UNB065668, UNB065669.

Sphecini sp.: UNB065157, UNB065176.

***Tetrapedia diversipes* Klug,1810:** UNB063793, UNB063794, UNB063822, UNB063823, UNB063824, UNB063825, UNB063826, UNB063827, UNB063828, UNB063829, UNB064036, UNB064038, UNB064083, UNB064084, UNB064085, UNB065140, UNB065147, UNB065149, UNB065151, UNB065152, UNB065153, UNB065165, UNB065166, UNB065167, UNB065168, UNB065169, UNB065170, UNB065173, UNB065175, UNB065178, UNB065180, UNB065183, UNB065184, UNB065187, UNB065194, UNB065195, UNB065196, UNB065197, UNB065201, UNB065215, UNB065216, UNB065217, UNB065218, UNB065219, UNB065282, UNB065283, UNB065284, UNB065285, UNB065374, UNB065375, UNB065386, UNB065387, UNB065390, UNB065665, UNB065666.

***Tetrapedia peckoltii* Friese, 1899:** UNB063743, UNB063753, UNB063757, UNB063758, UNB063835, UNB063837, UNB063838, UNB063851, UNB063852, UNB063853, UNB063854, UNB063855, UNB063856, UNB063857, UNB063860, UNB063861, UNB063862, UNB063872, UNB063873, UNB063874, UNB063900, UNB063901, UNB063902, UNB063903, UNB063904, UNB063905, UNB063906, UNB063910, UNB063918, UNB063921, UNB063971, UNB064037, UNB065154, UNB065155, UNB065158, UNB065171, UNB065174, UNB065179, UNB065181, UNB065186, UNB065202, UNB065255, UNB065256, UNB065257, UNB065258, UNB065259, UNB065260, UNB065261, UNB065262, UNB065263, UNB065264, UNB065376, UNB065377.

Tryphoninae sp.: UNB063736.

***Trypoxylon albitarse* (Fabricius,1804):** UNB060103, UNB063907, UNB063908, UNB063909, UNB063927, UNB063928, UNB063929, UNB063930, UNB063931, UNB063970, UNB063983, UNB063984, UNB064035, UNB064043, UNB064044, UNB064045, UNB064046, UNB064051, UNB064052, UNB064053, UNB064054, UNB064076, UNB064077, UNB064078, UNB064079, UNB064080, UNB064081, UNB064082, UNB065162, UNB065163, UNB065188, UNB065189.

***Trypoxylon* sp. Latreille, 1796:** UNB063728, UNB063740, UNB063747, UNB063750, UNB065190, UNB065191, UNB065192.

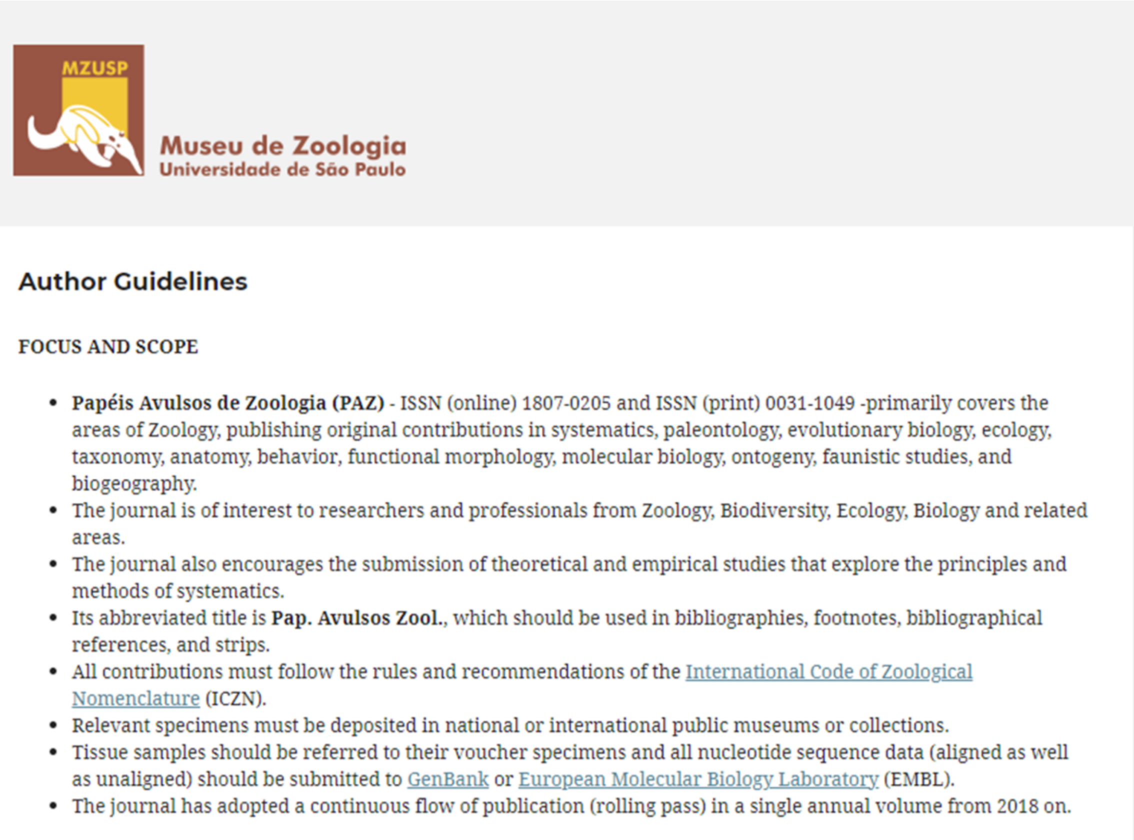
ANNEX

Capítulo 4

Trap-nesting bees and wasps in a Cerrado savanna area of Central Brazil

Annex 1

Focus and scope of the potential journal to which we intend to submit our manuscript.



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Author Guidelines

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- All contributions must follow the rules and recommendations of the International Code of Zoological Nomenclature (ICZN).
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- The journal has adopted a continuous flow of publication (rolling pass) in a single annual volume from 2018 on.

CONSIDERAÇÕES FINAIS

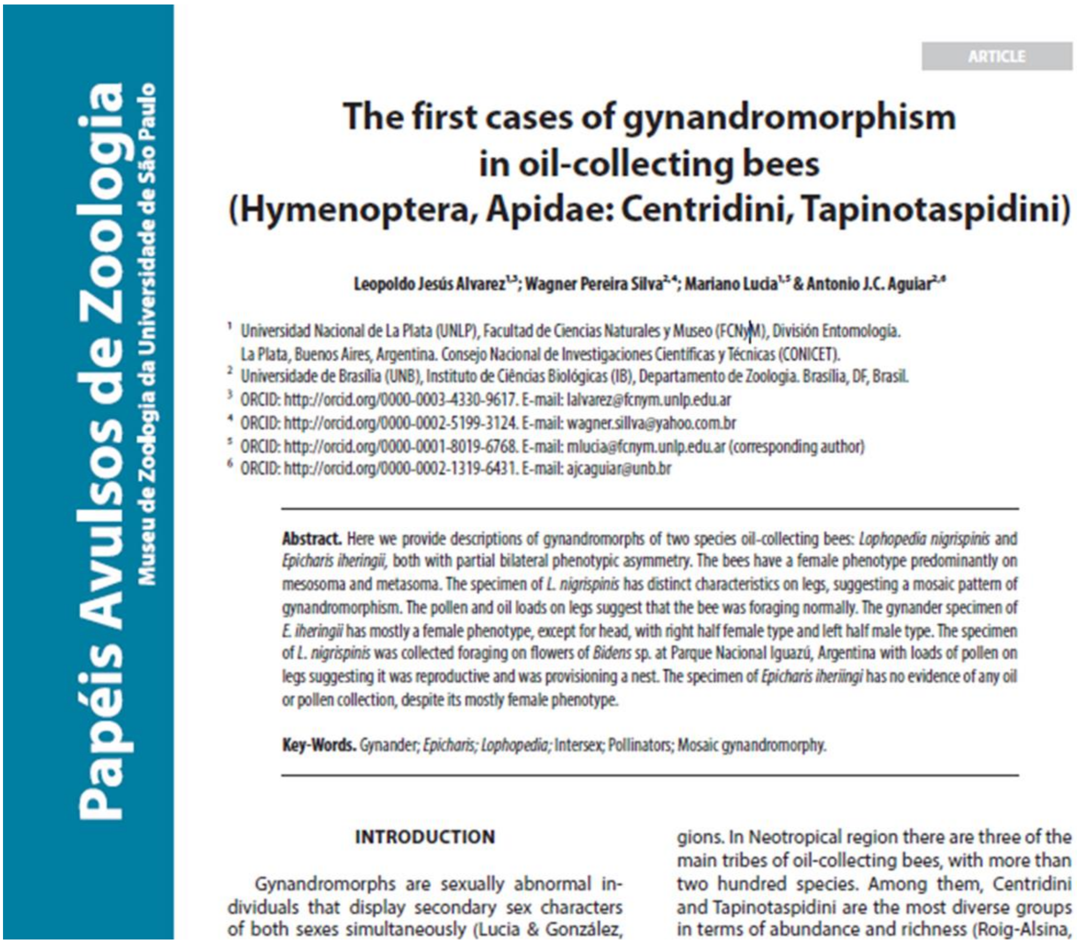
Os machos de abelhas Tapinotaspidini e *Tetrapedia* coletam óleo floral. Esse comportamento é único e ocorre através de adaptações morfológica especializadas e que apresentam elevada diversidade. Embora não existam mais questionamentos acerca da coleta do óleo pelos machos, os esclarecimentos acerca da funcionalidade do óleo permanecem. Contudo, a partir dos resultados obtidos, sugerimos que o óleo esteja diretamente relacionado ao comportamento de acasalamento dessas abelhas.

A capacidade de armazenamento do óleo nas escopas (pernas, esternos e tergos) que os machos de *Tetrapedia* e Tapinotaspidini apresentam estão possivelmente associados a algum tipo de estratégia que potencialize o acesso a parceiras reprodutivas como, por exemplo, na utilização de presentes nupciais, camuflagem química ou demarcação de território. Em contraste, a observação de machos dessas tribos dormindo em agregações (muitos deles com óleo nas pernas e escopas) podem indicar a possibilidade de o óleo fazer parte de uma estratégia de defesa contra predadores noturnos. Por fim, os resultados obtidos através do inventário de abelhas e vespas que nidificam em ninhos-armadilha corroboram informações já disponíveis na literatura, acerca da correlação entre o número de nidificações e a estação chuvosa.

Esta tese amplia o conhecimento acerca da diversidade morfológica dos machos de abelhas coletoras de óleo e propõe a discussão (e possíveis questionamentos) sobre as estratégias reprodutivas desses grupos. Estudos filogenéticos são fortemente incentivados, principalmente com a inclusão de aspectos comportamentais, e podem proporcionar maior compreensão acerca da evolução de Tetrapediini, Tapinotaspidini e demais abelhas coletoras de óleo.

APÊNDICE GERAL 1

Artigo publicado durante o doutorado



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ARTICLE

The first cases of gynandromorphism in oil-collecting bees (Hymenoptera, Apidae: Centridini, Tapinotaspidini)

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Abstract. Here we provide descriptions of gynandromorphs of two species oil-collecting bees: *Lophopedia nigrispinis* and *Epicharis iheringii,* both with partial bilateral phenotypic asymmetry. The bees have a female phenotype predominantly on mesosoma and metasoma. The specimen of *L. nigrispinis* has distinct characteristics on legs, suggesting a mosaic pattern of gynandromorphism. The pollen and oil loads on legs suggest that the bee was foraging normally. The gynander specimen of *E. iheringii* has mostly a female phenotype, except for head, with right half female type and left half male type. The specimen of *L. nigrispinis* was collected foraging on flowers of *Bidens* sp. at Parque Nacional Iguazú, Argentina with loads of pollen on legs suggesting it was reproductive and was provisioning a nest. The specimen of *Epicharis iheriingi* has no evidence of any oil or pollen collection, despite its mostly female phenotype.

Key-Words. Gynander; *Epicharis; Lophopedia;* Intersex; Pollinators; Mosaic gynandromorphy.

INTRODUCTION

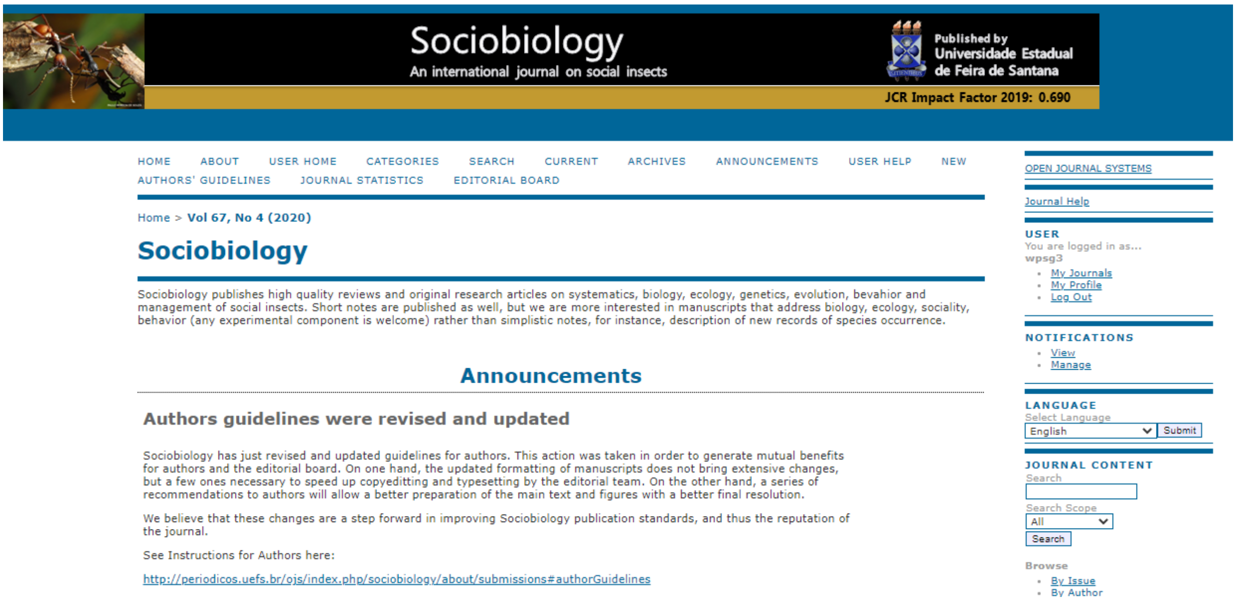
Gynandromorphs are sexually abnormal individuals that display secondary sex characters of both sexes simultaneously (Lucia & González,

gions. In Neotropical region there are three of the main tribes of oil-collecting bees, with more than two hundred species. Among them, Centridini and Tapinotaspidini are the most diverse groups in terms of abundance and richness (Roig-Alsina,

Versão completa disponível em: https://doi.org/10.11606/1807-0205/2019.59.36

APÊNDICE GERAL 2

Comunicação curta aceita durante o doutorado (na primeira rodada de revisão)



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Sociobiology

Sociobiology publishes high quality reviews and original research articles on systematics, biology, ecology, genetics, evolution, bevahior and management of social insects. Short notes are published as well, but we are more interested in manuscripts that address biology, ecology, sociality, behavior (any experimental component is welcome) rather than simplistic notes, for instance, description of new records of species occurrence.

Announcements

Authors guidelines were revised and updated

Sociobiology has just revised and updated guidelines for authors. This action was taken in order to generate mutual benefits for authors and the editorial board. On one hand, the updated formatting of manuscripts does not bring extensive changes, but a few ones necessary to speed up copyeditting and typesetting by the editorial team. On the other hand, a series of recommendations to authors will allow a better preparation of the main text and figures with a better final resolution.

We believe that these changes are a step forward in improving Sociobiology publication standards, and thus the reputation of the journal.

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Male Sleeping Aggregation of *Melissodes* (*Ecplectica*) *nigroaenea* (Smith, 1854) (Hymenoptera, Apidae, Eucerini) in Cerrado, DF

WP Silva[1] & RR Andrade[1]

Keywords: bees, dormitory, night shelter, solitary bees.

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ABSTRACT - Bee males are usually found forming aggregations on stems of bushes or trees to sleep at night, although there is not a complete understanding of the reasons for this behaviour. This note described the behavior of *Melissodes* (*Ecplectica*) *nigroaenea* (Smith, 1854) males forming temporary aggregations in dry inflorescences of *Bidens pilosa* L. The aggregations of *M. nigroaenea* were observed for approximately 15 days in an area of Cerrado, Brasília, DF. During the day the of *M. nigroaenea* males visit flowers of *Cosmos sulphureus* Cav. near the aggregations, where the females collect pollen. In the late afternoon, the males return to the aggregations, where they sleep at night. These data subsidize new information about the behavior of the *M. nigroaenea* males.

The behavior of forming aggregations on stems of bushes and trees or in the nesting site to sleep at night is characteristic of many species of bees and wasps (Evans & Linsley, 1960; Linsley, 1962; Alves-dos-Santos et al., 2002). Records of these insects forming aggregations have been made for more than a century, but there is not full understanding of the reasons for this phenomenon (Banks, 1902; Bradley, 1908; Rau & Rau, 1916). However, studies suggest that such behavior may be related to thermoregulation (Evans & Gillaspy, 1964; Linsley & Cazier, 1972), anti-predation defense (Evans & Linsley, 1960; Alcock, 1998) or the evolution of social behavior (Grassé, 1942).

Bees aggregations are usually composed of males because females usually sleep inside the nests they build along the day (Alcock, 1998). Aggregations can last weeks, months (Evans & Linsley, 1960) or even years when shared by individuals of different generations (Linsley, 1962). Record of aggregations of the tribe Eucerini (Apidae) has already been reported for different species (Table 1). Here we describe general aspects of the behavior of *Melissodes* (*Ecplectica*) *nigroaenea* males in an aggregation in Central Brazil.

The aggregations of *M. nigroaenea* males were observed for approximately 15 days (between March and April 2018), in an agroforestry of campus Darcy Ribeiro of the University of Brasília (UnB), Federal District, Brazil (15°45'51.1" S and 47°52'05.4" W), where native plant species are maintained and cultivated in a soil rich in exogenous organic matter and green manure. Two species of Asteraceae – *Bidens pilosa* L. and *Cosmos sulphureus* Cav. – stand out in the place, by the occurrence of different species of bees using the reproductive structures to sleep, for example, *Bombus (Fervidobombus) pauloensis* Friese, 1913; *Epanthidium tigrinum* (Schrottky, 1905); *Megachile* spp. Latreille, 1802. Two aggregations of *M. nigroaenea* were recorded at about 40 and 60 cm above the ground. On the day with higher frequency of individuals 15 males of *M.*

nigroaenea were recorded. The collected specimens were assembled, identified and deposited in the Entomological Collection of UnB (Department of Zoology).

The first males arrived at the aggregations around 16:00 h (Figure 1). *M. nigroaenea* males used the mandibles to fix on dried inflorescences of *B. pilosa*. This behavior of attaching to the substrate through the mandibles is common for many – *Coelioxys deplanata* Cresson, 1878; *Melissoptila* aff. *bonaerensis* Holmberg, 1903; *Centris* (*Paracentris*) *xanthomelaena* Moure & Castro, 2001 – of bees (Linsley, 1962; Mahlmann et al., 2014; Martins et al., 2018). After fixing on the inflorescences, the fore and middle legs were used to find the ideal position to establish in the aggregation and spend the night (Figure 2). Some males then frictioned the hind legs, concomitantly, sometimes scrubbing them in the sterna or terga. This behavior of scrubbing the legs and the sterna or terga may be related to chemical signaling, being observed also in *Tetrapedia* oil-collecting bee species (Alves-dos-Santos et al., 2009).

Interactions between the *M. nigroaenea* males were registered in some situations, usually due to the arrival of a new individual the aggregation, which collided with another male, establishing for a few seconds some contact. The last males arrived at the aggregations around 17:00 h, when the other individuals were already resting. *M. nigroaenea* males left the aggregations early, not being observed individuals resting after 07:00 h. However, sometimes aggregations were observed during the day when the sky was cloudy or raining. *M. nigroaenea* females were observed with relative frequency collecting pollen, mainly between 10:00 and 16:00 h, in flowers of *C. sulphureus* near the aggregations. *M. nigroaenea* males were often observed ingesting nectar from the flowers of *C. sulphureus*, although no record of copulation has observed (Figure 3). According to Chemsak & Thorp (1962), *Melissodes robustior* Cockerell, 1915 males, another Eucerini species, seem to present a preference for sleeping in *Cosmos* sp. flowers, where females collect pollen during the day. Some authors suggest that establishing aggregations near flowers used by females as a source of floral resources may represent a strategy adopted by males in the search for reproductive partners the next day (Alves-dos-Santos et al., 2009; Pinheiro et al., 2017).

Mahlmann et al. (2014) also recorded an aggregation formed by *M. nigroaenea* males. The individuals of two Eucerini species – *M. nigroaenea* and *Melissoptila* aff. *bonaerensis* – formed an aggregation where the individuals remained fixed through the mandibles in dry inflorescences of *Hyptis* sp. (Lamiaceae). Other studies report aggregations formed by individuals of both sexes (Evans & Linsley,1960; Starr & Vélez, 2009; Yokoi et al. 2016, 2017). However, in the present study only *M. nigroaenea* males were observed in both aggregations.

Many tries to explain the function of aggregations already have been realized, but are still inconclusive. Aggregations could represent a strategy to reduce the risk of nocturnal predation, although it could also represent a greater risk of predation and parasitism as has already been described for stingless bees (Evans & Linsley, 1960; Brown, 1997; Alcock, 1998). Another benefit that aggregations could provide would be the possibility of elevating the capacity of individuals to thermoregulate (Linsley & Cazier, 1972). Thus, in the face of the action of a predator, the bees could present a minimum temperature to activate the musculature associated with the flight and escape. The hypothesis that males form aggregations near sources of pollen visited by possible reproductive partners cannot be discarded. Thus, *Cosmos* flowers could serve not only as a source of floral resource but also as a mating site for Eucerini species (Chemsak & Thorp, 1962). Besides sleeping in aggregations, some studies have reported Eucerini males sleeping inside flowers mainly from Cucurbitaceae and Orchidaceae families (Hurd

& Linsley, 1964; Dafni et al., 1981; Willis & Kevan, 1995; Vereecken et al. et al., 2012; Watts et al., 2013).

There is not yet full understanding of which factors are determinant for the establishment of aggregations. However, the results obtained subsidize new information about the behavior of Eucerini males. Future studies, addressing how this behavior may be related to a possible strategy of defense or sexual selection in solitary bees are needed.

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REFERENCES

Alcock, J. (1998). Sleeping aggregations of the bee *Idiomelissodes duplocincta* (Cockerell) (Hymenoptera: Anthophorini) and their possible function. **Journal of the Kansas Entomological Society**, 71: 74-84.

Alves-dos-Santos, I., Melo, G.A.R., & Rozen, J.G. Biology and Immature Stages of the Bee Tribe Tetrapediini (Hymenoptera: Apidae). **American Museum Novitates**, 3377: 1-45.

Alves-dos-Santos, I., Gaglianone, M.C., Naxara, S.R.C. & Engel, M.S. (2009). Male sleeping aggregations of solitary oil-collecting bees in Brazil (Centridini, Tapinotaspidini, and Tetrapediini; Hymenoptera: Apidae). **Genetics and Molecular Research**, 8: 515-524. doi: 10.4238/vol8-2kerr003

Banks, N. (1902). Sleeping Habits of Certain Hymenoptera. **Journal of the New York Entomological Society**, 10: 209-214.

Bradley, J.C. (1908). A case of gregarious sleeping habits among aculeate Hymenoptera. **Entomological Society of America**, 1: 22-27. doi: 10.1093/aesa/1.2.127

Brown, B.V. (1997). Parasitic Phorid Flies: A Previously Unrecognized Cost to Aggregation Behavior of Male Stingless Bees. **Biotropica**, 29(3): 370-372. doi: 10.1111/j.1744-7429.1997.tb00439.x

Chemsak, J.A. & Thorp, R.W. (1962). Note on the sleeping habits of males of *Melissodes robustior* Cockerell. **The Pan-Pacific Entomologist**, 38: 53-55.

Cockerell, T.D.A. (1915). Habits of *Xenoglossa brevicornis* (Cresson) (Hym.). **Entomological News**, 26: 364.

Dafni, A., Ivri, Y. & Brantjes, N.B.M. (1981) Pollination of *Serapias vomeracea* Briq. (Orchidaceae) by imitation of holes for sleeping solitary male bees (Hymenoptera). **Acta Botanica Neerlandica**, 10: 69-73. doi: 10.1111/j.1438-8677.1981.tb00388.x

Evans, H.E. & Linsley, E.G. (1960). Notes on the Sleeping Aggregation of Solitary Bees and Wasps. **Bulletin of the South California Academy of Sciences**, 59: 30–37.

Evans, H.E. & Gillaspy, J.E. (1964). Observations on the ethology of digger wasps of the genus *Steniolia* (Hymenoptera: Sphecidae: Bembicini). **American Midland Naturalist**, 72: 257-280. doi: 10.2307/2423504

Grassé, P.P. (1942). Les rassemblements de sommeil des Hyménoptères et leur interprétation. **Bulletin de la Société entomologique de France**, 47: 142-148.

Hurd, P.D. & Linsley, E.G. (1964). The squash and gourd bees—genera *Peponapis* Robertson and *Xenoglossa* Smith—inhabiting America north of Mexico (Hymenoptera: Apoidea). **Hilgardia**, 35(15):375-477. doi:10.3733/hilg.v35n15p375

Laberge, W.E. & Ribble, D.W. (1966). Biology of *Florilegus condignus* (Hymenoptera: Anthophoridae), with a Description of Its Larva, and Remarks on Its Importance in Alfalfa Pollination. **Annals of the Entomological Society of America**, 59(5):944-950.

Linsley, E.G. (1962). Sleeping aggregations of aculeate Hymenoptera – II. **Annals of the Entomological Society of America**, 55: 148-164. doi: 10.1093/aesa/55.2.148

Linsley, E.G. & Cazier, M.A. (1962). Diurnal and Seasonal Behavior Patterns Among Adults of *Protoxaea gloriosa* (Hymenoptera, Oxaeidae). **American Museum Novitates**, 2509: 1-25.

Mahlmann, T., Hipólito, J. & Oliveira, F.F. (2014). Male sleeping aggregation of multiple Eucerini bee genera (Hymenoptera: Apidae) in Chapada Diamantina, Bahia, Brazil. **Biodiversity Data Journal 2**, 2: 15-56. doi: 10.3897/BDJ.2.e1556

Martins, H.O.J., Oliveira-Rebouças, P. & Ferreira, V.S. (2018). Sleeping behaviour of an oil-collecting bee, *Centris (Paracentris) xanthomelaena* Moure & Castro (Hymenoptera: Apidae: Centridini). **Sociobiology**, 65(4): 770–772. doi: 10.13102/sociobiology.v65i4.3452

Mathewson, J. & Daly, H. (1955). A brief note on the sleep of male *Melissodes* (Hymenoptera: Apidae). **Journal of the Kansas Entomological Society**, 28: 120.

Pinheiro, M., Alves-dos-Santos, I. & Sazima, M. (2017). Flowers as sleeping places for male bees: somehow the males know which flowers their females prefer. **Arthropod-Plant Interactions**, 11: 329-337. doi: 10.1007/s11829-017-9532-6

Rau, P. (1938) Additional Observations on the Sleep of Insects. **Annals of the Entomological Society of America**, 31: 540–556. doi: 10.1093/aesa/31.4.540

Rau, P. & Rau, N. (1916). The sleep of insects; an ecological study. **Annals of the Entomological Society of America**, 9: 227–274. doi: 10.1093/aesa/9.3.227

Starr, C.K. & Vélez, D. (2009). A Dense Daytime Aggregation of Solitary Bees (Hymenoptera: Apidae: Centridini) in the Lesser Antilles. **Journal of Hymenoptera Research**, 18(2): 175-177.

Vereecken, N.J.; Wilson, C.A., Hötling, S., Schulz, S., Banketov, S.A. & Mardulyn, P. (2012). Pre-adaptations and the evolution of pollination by sexual deception: Cope's rule of specialization revisited. **Proceedings of the Royal Society B: Biological Sciences**, 279: 4786-4794. doi: 10.1098/rspb.2012.1804

Watts, S., Sapir, Y., Segal, B. & Dafni, A. (2013). The endangered *Iris atropurpurea* (Iridaceae) in Israel: honey-bees. **Annals of Botany**, 111: 395–407. doi: 10.1093/aob/mcs292

Willis, D.S. & Kevan, P.G. (1995). Foraging Dynamics of *Peponapis pruinosa* (Hymenoptera: Anthophoridae) on Pumpkin (*Cucurbita pepo*) in Southern Ontario. **The Canadian Entomologist**, 127: 167–175. doi.org/10.4039/Ent127167-2

Yokoi, T., Idogawa, N., Konagaya, T. & Watanabe, M. (2016). The Non-Use of Sleeping Substrate by the Sympatric Bees *Amegilla florea urens* and *A. senahai senahai* (Hymenoptera: Apoidea). **Entomology News**, 126: 138-143. doi: 10.3157/021.126.0210

Yokoi, T., Idogawa, N., Kandori, I., Nikkeshi, A. & Watanabe, M. (2017). The choosing of sleeping position in the overnight aggregation by the solitary bees *Amegilla florea urens* in Iriomote Island of Japan. **The Science of Nature**, 104: 1-8. doi: 10.1007/s00114-017-1438-8

TABLES

Table 1. Aggregation records of Eucerini available in the literature.

Species	**Substrates**	**Sex**	**Reference**
Florilegus (*Florilegus*) *condignus* Cresson 1878	On racemes of *Medicago sativa* (Fabaceae)	Male	LaBerge & Ribble (1966)
Gaesochira obscura (Smith, 1879)	Stems of an unidentified species	Unknow	Rau & Rau (1916)
Melissodes (*Ecplectica*) *nigroaenea* (Smith, 1854)	Dried inflorescence of *Hyptis* sp. (Lamiaceae)	Male	Mahlmann et al. (2014)
	Dried stems of *Bidens pilosa* L. (Asteraceae)	Male	Present study
Melissodes (*Eumelissodes*) *agilis* Cress 1878	Stems of an unidentified species	Unknow	Bradley (1908)
	In sunflowers (*Helianthus* sp., Asteraceae)	Unknow	Rau & Rau (1916)
Melissodes (*Eumelissodes*) *denticulata* Smith, 1854	*Verbena stricta* (Verbenaceae)	Male	Mathewson & Daly (1955)
Melissodes (*Eumelissodes*) *robustior* Cockerell, 1915	Inside flower of *Cosmos* sp. (Asteraceae)	Male	Chemsak & Thorp (1962)
Melissodes (*Eumelissodes*) *vernoniae* Robertson, 1902	*Verbena stricta* (Verbenaceae)	Male	Mathewson & Daly (1955)
Melissodes (*Melissodes*) *bimaculata* (Lepeletier, 1825)	Stems of an unidentified species	Unknow	Banks (1902)
	Stems of an unidentified species	Unknow	Rau & Rau (1916)
	Melilotus sp. (Fabaceae)	Unknow	Rau (1938)
Melissodes verroniana Robt.	Stems of an unidentified species	Unknow	Rau & Rau (1916)
Melissoptila aff. *bonaerensis* Holmberg, 1903	Dried inflorescence of *Hyptis* sp. (Lamiaceae)	Both	Mahlmann et al. (2014)
Svastra (*Brachymelissodes*) *cressonii* (Dalla Torre, 1896)	Petioles of the leaves an unidentified species	Male	Cockerell (1915)
Svastra (*Epimelissodes*) *obliqua* (Say, 1837)	Stems of an unidentified species	Male	Rau & Rau (1916)
Svastra (*Idiomelissodes*) *duplocincta* (Cockerell, 1905)	Leaf or stem of *Encelia farinosa* (Asteraceae)	Male	Alcock (1998)
	Leaf or stem of *Calliandra eriophylla* (Fabaceae)	Male	Alcock (1998)
	Leaf or stem of *Ruellia peninsulares* (Acanthaceae)	Male	Alcock (1998)
	Leaf or stem of *Justicia californica* (Acanthaceae)	Male	Alcock (1998)

FIGURE LEGENDS

Figure 1. Male sleeping aggregation on dried inflorescence of *Bidens pilosa*. **(A)** *Melissodes nigroaenea* males arriving at the aggregation. **(B)** *M. nigroaenea* males trying to find a position in the aggregation

Figure 2. *Melissodes nigroaenea* males fixed in *Bidens pilosa* through the mandibles.

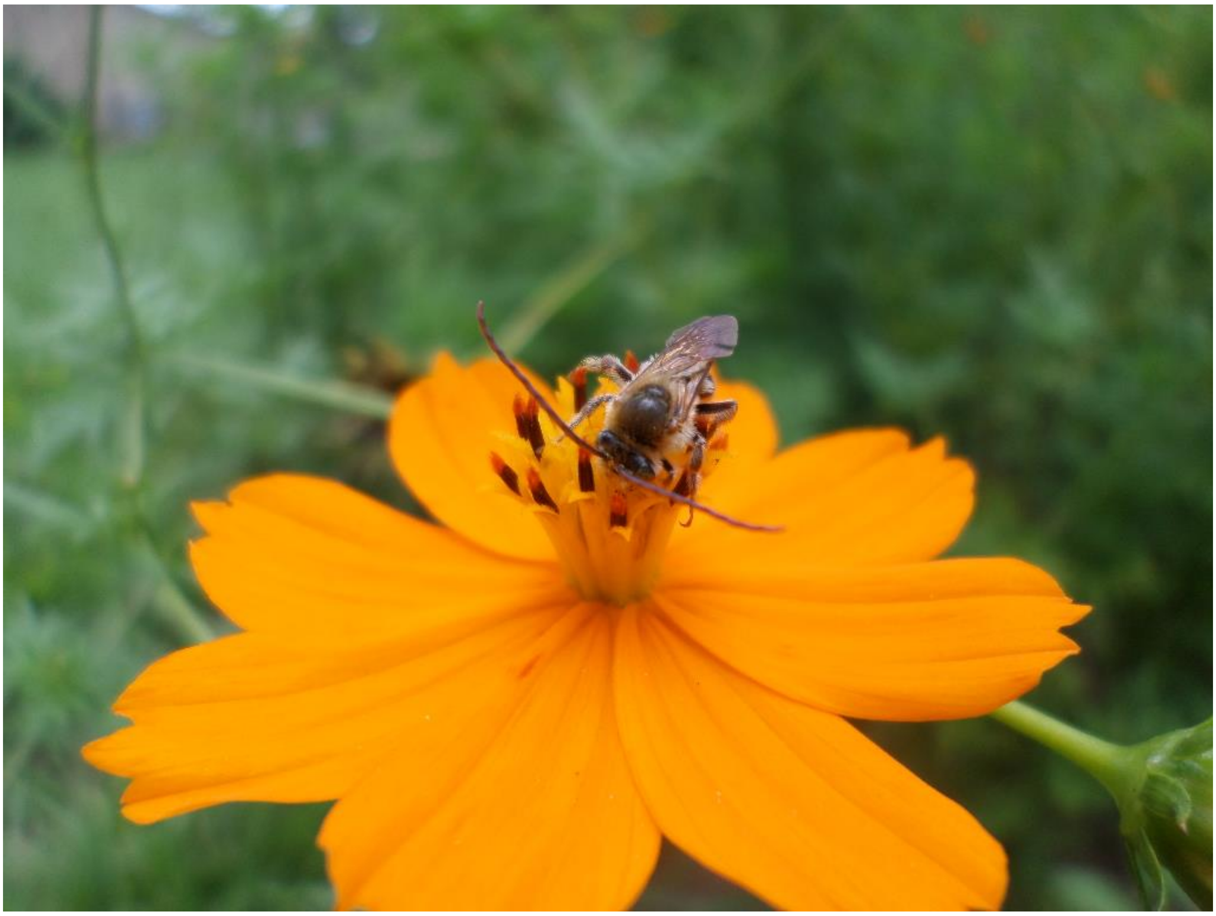
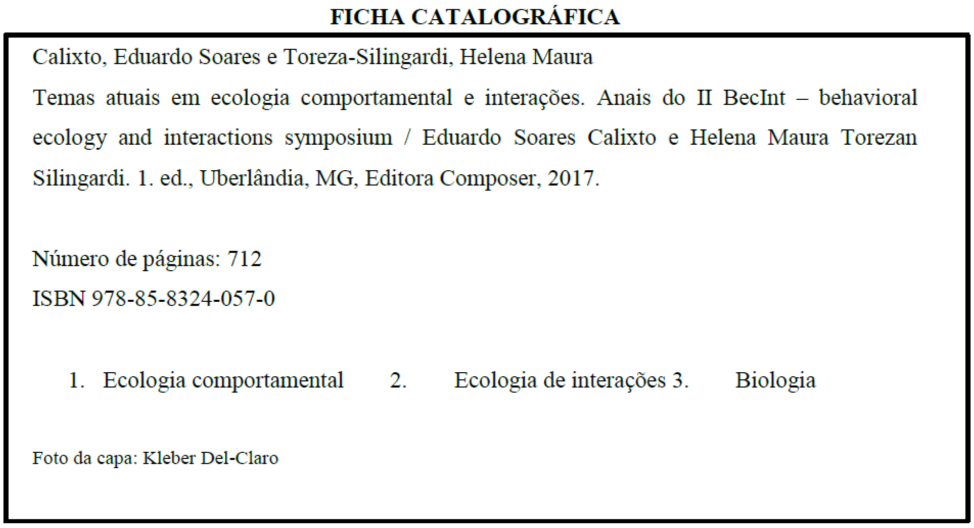


Figure 3. Male of *Melissodes nigroaenea* ingesting nectar in flower of *Cosmos sulphureus*.

APÊNDICE GERAL 3

Capítulo de livro publicado durante o doutorado



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1. Ecologia comportamental 2. Ecologia de interações 3. Biologia

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O óleo floral coletado pelos machos das abelhas *Tetrapedia* (Hymenoptera, Apidae): presente nupcial ou camuflagem química?

Floral oil collection by male of *Tetrapedia* bees (Hymenoptera, Apidae): nuptial gift or chemical camouflage?

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Resumo: As abelhas do gênero *Tetrapedia* coletam óleo floral como recompensa, usando estruturas especializadas nas pernas, mais pronunciadas nas fêmeas. Este recurso e potencialmente utilizado na alimentação larval e construção do ninho. Os machos nao atuam no aprovisionamento dos ninhos e, portanto a coleta de óleo nos machos sugere dois possíveis questionamentos: o óleo funciona como um presente nupcial para as fêmeas ou como uma camuflagem química para facilitar a cópula. Deste modo, este estudo teve como objetivo analisar o comportamento de pareamento de machos e fêmeas de duas espécies de *Tetrapedia* com fins de avaliar o possível papel deste recurso quando coletado pelos machos. Em março de 2017, no município de Cavalcante, Chapada dos Veadeiros (GO), machos e fêmeas de *Tetrapedia* foram coletados em flores de *Banisteriopsis argyrophylla* e observados quanto ao pareamento para cópula. Foi possível observar duas formas de transferencia do óleo entre machos e fêmeas. As fêmeas foram observadas coletando ativamente o óleo das escopas das pernas e dos tergos dos machos com as pernas anteriores. E os machos, durante o acoplamento sobre as fêmeas, realizaram o contato do basitarso posterior com a escopa das pernas das fêmeas. Desta forma sugerimos fortemente que há transferência do óleo floral dos machos para fêmea como presente nupcial. Observações adicionais são necessárias para avaliar com mais detalhes a sequência de transferência para cópula.

Palavras-chave: Acasalamento; abelhas solitárias; comportamento.

Abstract: Bees of the genus *Tetrapedia* collect floral oils as a reward using specialized structures in the legs, more pronounced in females. This resource is potentially used in larval feeding and nest construction. For not acting in the provision of the nest, the collection of oil in males suggests two possible questions: the oil is used as a nuptial gift or as a chemical camouflage to facilitate copulation. In this way, this study aimed to describe the pairing behavior of two species of *Tetrapedia* in order to analyze the possible effects of oil when collected by males. In March of 2017, in the municipality of Cavalcante, Chapada dos Veadeiros (GO), males and females of *Tetrapedia* were collected in flowers of *Banisteriopsis argyrophylla* and observed for pairing for copulation. It was possible to observe two forms of oil transfer between males and females. The females were observed actively collecting oil with the fore legs in the scopes and tergos of the males. And males, during the coupling with females contact the hind basitarsi with the hind basitarsi of females. We strongly suggest that there is transfer of oil from males to females as a nuptial gift. Further observations are necessary to evaluate the transference sequence for mating.

Keywords: Mating; solitary bees; behavior.

1. Introdução

O acasalamento representa uma importante etapa da evolução das espécies, envolvendo uma elevada diversidade de comportamentos entre as abelhas. Acredita-se que as interações entre machos e fêmeas de abelhas evoluíram de forma a tornar as fêmeas mais acessíveis às investidas dos machos (MICHENER, 2007). Todavia, o conflito sexual entre machos e fêmeas pode ser decorrente da diferença de interesses ao longo do processo evolutivo (PARKER, 1979). Como a maioria das abelhas solitárias fêmeas acasalam logo após emergirem, os machos necessitam desenvolver estratégias que aumentem as possibilidades de sucesso reprodutivo (EICKWORT & GINSBERG, 1980).

Diferenças entre os sexos nas abelhas também podem ser observadas em outras situações como, por exemplo, no cuidado parental, forrageamento e nidificação (MICHENER, 2007). As abelhas do gênero *Tetrapedia* (Apidae, Tetrapediini), são especializadas na coleta de óleos florais, principalmente de plantas das famílias Iridaceae, Krameriaceae, Malpighiaceae e Plantaginaceae. O óleo, que pode ser utilizado na alimentação larval ou revestimento das células, é coletado com o auxílio de estruturas especializadas nas pernas ou esternos, geralmente mais acentuadas nas fêmeas (NEFF & SIMPSON, 1981; ROIG- ALSINA, 1997). Contudo, pernas modificadas para coleta de óleos florais também podem ser observadas em machos de *Tetrapedia*. Todavia, estudos relatam que após emergirem, os machos de *Tetrapedia* deixam o ninho, não apresentando qualquer tipo de cuidado parental ou comportamento de nidificação, como é observado nas fêmeas (ALVES-DOS-SANTOS *et al.*, 2002; CAMILLO, 2005).

O gênero *Tetrapedia* é composto por 28 espécies de abelhas solitárias, de tamanho médio, pouco pilosas, restritas às regiões tropicais das Américas, cujas fêmeas nidificam em cavidades pré-existentes (ALVES-DOS-SANTOS *et al.*, 2002; SILVEIRA *et al*., 2002; MOURE, 2007). Nos machos do gênero *Tetrapedia* as pernas anteriores apresentam o aparato coletor com formato e tamanho semelhantes aos encontradas em fêmeas, que são utilizados para coleta de óleo floral, enquanto que o metassoma apresenta uma escopa tergal que é utilizada para armazenar este óleo (CAPPELLARI *et al.*, 2012). Entretanto, a funcionalidade das adaptações morfológicas para coleta e transporte do óleo dos machos de *Tetrapedia* foi pouco estuda, e ainda não foi esclarecida.

Segundo Neff & Simpson (1981), o óleo coletado pelos machos poderia ser utilizado na alimentação ou no acasalamento. De acordo com Alcock (2011) os machos de algumas espécies buscam atrair as fêmeas através de benefícios materiais, dentre eles, presentes nupciais ou cuidado parental. Corroborando esta hipótese, as estruturas presentes nas pernas anteriores, posteriores e nos tergos dos machos de *Tetrapedia*, podem estar associadas a coleta e transporte do óleo, utilizado de forma estratégica na busca por parceiras reprodutivas, tornando-os mais atrativos e receptivos a elas (CAPPELLARI *et al.*, 2012),

O óleo coletado pelos machos de *Tetrapedia*, possivelmente, é utilizado como presente nupcial, uma vez que em muitos casos as fêmeas recebem recursos durante a cópula (THORNHILL & ALCOCK, 1983). Seja para alimentação, nidificação ou sobrevivência da prole (BOGGS, 1995). Adaptações morfológicas observadas em machos, geralmente, estão mais relacionadas ao acasalamento do que aquelas observadas em fêmeas (BATERMAN, 1948). Assim sendo, machos que fazem uso de presentes nupciais teriam maior chance de obter sucesso no processo de seleção sexual (BUZATTO *et al.*, 2014).

Vários trabalhos apontam para algumas adaptações dos machos que estão relacionadas ao comportamento de cópula em abelhas solitárias (BATRA, 1978; SIHAG, 1986; WITTMANN

& BLOCHTEIN, 1995). Dessa forma, algumas abelhas fazem uso das pernas para reter a fêmea no momento da cópula, como observado por Sihag (1986) ao estudar o acasalamento e comportamento de cópula em quatro espécies de abelhas do gênero *Megachile* Latreille, 1802.

O óleo coletado pelos machos de *Tetrapedia* com o auxílio das pernas anteriores, é armazenado na escopa tergal e nas pernas posteriores (CAPPELLARI *et al.*, 2012). Adaptações morfológicas semelhantes são observadas nas abelhas das orquídeas (Euglossini), em que compostos aromáticos, coletados principalmente nas flores de Orchidaceae são armazenados nas pernas anteriores dos machos, e posteriormente, transferidos para as pernas posteriores (BEMBÉ, 2004). Dessa forma, a transferência do óleo para a escopa tergal e escopa das pernas posteriores, pode ser uma estratégia para facilitar o contato com os sistemas sensoriais das fêmeas, seja através de sinais visuais, mecânicos ou olfativos (CAPPELLARI *et al.*, 2012). Embora não exista uma total compreensão do papel do óleo coletado pelos machos de *Tetrapedia*, hipotetiza-se que possam estar associadas a um provável presente nupcial para as fêmeas durante a cópula ou como camuflagem química também para aproximação e facilitação da copula (CAPPELLARI *et al.*, 2012).

2. Objetivos

Analisar a função do óleo nos machos das abelhas coletoras de óleo, *Tetrapedia*, através da caracterização do comportamento de cópula.

3. Metodologia

O estudo foi realizado no município de Cavalcante, na região da Chapada dos Veadeiros (GO), em uma área de floresta de galeria, entre os dias 10 e 12 de março de 2017, totalizando 20 horas amostrais.

Para a observação do comportamento de cópula, machos (n=4) e fêmeas (n=18) de *Tetrapedia* foram coletados em flores de *Banisteriopsis argyrophylla* (A. Juss.) B. Gates, uma liana da família Malpighiaceae, bastante florida no período do estudo. Os indivíduos de ambos os sexos foram identificados ainda em campo e pareados para observação de forma independente quanto ao comportamento de cópula.

Observações focais e filmagens foram registradas no intuito de avaliar a duração das cópulas e o tempo dispendido pelas fêmeas ao resistirem às investidas dos machos, bem como os demais comportamentos exibidos por machos e fêmeas de *Tetrapedia*. Os espécimes coletados foram montados, identificados e depositados na coleção entomológica da Universidade de Brasília, no Departamento de Zoologia, Instituto de Ciências Biológicas.

4. Principais resultados

Foram feitos experimentos com três casais de *Tetrapedia* aff. *peckoltii* e um de *Tetrapedia* sp., onde foram observados pareamentos e possíveis comportamentos de pré-cópula. Em todos os experimentos, ocorreu interação entre machos e fêmeas, quase sempre de forma intensa nos primeiros minutos, com perceptiva diminuição das atividades ao longo do tempo de observação. Os machos e fêmeas foram coletados em sua maioria coletando óleo sobre as flores de *B. argyrophylla*. Dois padrões básicos de comportamento foram observados onde machos e fêmeas inverteram a posição um sobre o outro.

Inicialmente, foi observado que os machos de *Tetrapedia* tentaram segurar as fêmeas pelos flancos dos tergos metassomais com o auxílio das pernas anteriores e médias. Com as pernas posteriores, o macho esfregou a margem anterior do basitarso posterior contra a escopa da fêmea, possivelmente na tentativa de retirar o óleo. O que poderia explicar a presença de cerdas ramificadas nessa região do basitarso posterior dos machos de *Tetrapedia*, possivelmente relacionadas a transferência do óleo para as fêmeas durante a cópula. Esse comportamento de tentar segurar a fêmea foi observado no maior número de interações, com o macho realizando a contração do metassoma em alguns momentos.

O segundo padrão básico de comportamento foi observado quando o macho, em vários momentos da interação manteve-se em repouso por alguns segundos e levantando as pernas posteriores. Em seguida, a fêmea posicionava-se atrás do macho, esfregando o aparato coletor das pernas anteriores contra as escopas tergais e das pernas posteriores, claramente na tentativa de coleta de óleo. Em alguns momentos, foi observado que após o macho levantar as pernas posteriores, a fêmea posicionou-se a frente do macho, que tentou segura-la, bem como relatado no primeiro padrão de comportamento observado.

5. Conclusão

O presente estudo sugere que o óleo coletado por machos de *Tetrapedia* é utilizado como presente nupcial durante o comportamento de cópula. A transferência do óleo pode ocorrer através de dois padrões de comportamento observados durante as interações macho-fêmea e podem estar diretamente relacionados, sendo etapas do comportamento pré-copulatório. Os resultados obtidos no estudo, ainda que pontuais, sugerem fortemente que o óleo é um presente nupcial fornecido pelo macho, sendo necessários observações adicionais para entender com mais detalhes o comportamento de cópula de *Tetrapedia* e suas variações.

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REFERÊNCIAS

ALCOCK, John. **Comportamento animal: uma abordagem evolutiva**. Artmed, Porto Alegre, 606 p. 2011.

ALVES-DOS- SANTOS, Isabel; MELO, Gabriel Augusto Rodrigues; ROZEN, Jerome George. Biology and Imature Stages of the Bee Tribe Tetrapediini (Hymenoptera: Apidae). **Am Mus Novit**, v. 3377, p. 1-45. 2002.

BATERMAN, A. J. Intra-sexual selection in *Drosophila*. **Heredity**, v.2, p. 349-368. 1948.

BATRA, Suzanne W. T. Aggression, territoriality, mating and nest aggregation of some solitary bees (Hymenoptera, Halictidae, Megachilidae, Colletidae, Anthophoridae). **J Kansas Entomol Soc**, v. 51, p. 547-559. 1978.

BEMBÉ, Benjamin. Functional morphology in male euglossine bees and their ability to spray fragrances (Hymenoptera, Apidae, Euglossini). **Apidologie**, v. 35(3), p. 283-291. 2004.

BOGGS, Carol L. Male nuptial gifts: Phenotypic consequences and evolutionary implications. In: LEATHER, Simon R.; HARDIE, Jim (Org.). **Insect reproduction**. New York, p.255. 1995.

BUZATTO, Bruno Alves; TOMKINS, Joseph L.; SIMMONS, Leigh W.; MACHADO, Glauco. Correlated evolution of sexual dimorphism and male dimorphism in a clade of neotropical harvestmen. **Evolution**, v.68, p.1671-1686. 2014.

CAMILLO, Evandro. Nesting biology of four *Tetrapedia* species in trap-nests (Hymenoptera, Apidae, Tetrapedini). **Revta Biol. Tropical**, v.53, p.175-186. 2005.

CAPPELLARI, Simone Caroline; MELO, Gabriel Augusto Rodrigues; AGUIAR, Antonio Jose Camillo; NEFF, John. L. Floral oil collection by male *Tetrapedia* bees (Hymenoptera: Apidae: Tetrapediini). **Apidologie**, v. 43, p.39-50. 2012.

EICKWORT, George C.; GINSBERG, Howard S. Foraging and mating behavior in Apoidea. **Annu. Rev. Entomol.**, v,25, p.421-446. 1980.

MICHENER, Charles Duncan. The bees of the world. Foraging and mating behavior in Apoidea. **Annu. Rev. Entomol.**, v.2, p.992. 2007.MOURE, Jesus Santiago. Tetrapediini. In: MOURE, Jesus Santiago; URBAN, Danuncian; MELO, Gabriel Augusto Rodrigues de (Org.). **Catalogue of Bees (Hymenoptera, Apoidea) in the Neotropical Region**. Sociedade Brasileira de Entomologia, Curitiba, p.632-637. 2007.

NEFF, J. L.; SIMPSON, B. B. Oil-collecting structures in the Anthophoridae: Morphology, function and use in systematics. **J Kans Entomol Soc**, 54: 95-123, 1981.

PARKER, Geoff A. Sexual selection and sexual conflict. In: BLUM, M. S. & BLUM, N. A. (Org.). **Sexual selection and reproductive competition in insects**. York: Academic Press, p.123-166. 1979.

ROIG-ALSINA, Arturo. A generic study of the bees of the tribe Tapinotaspidini, with notes on the evolution of their oil-collecting structures. **Mitt Muench Entomol Ges**, v.87, p.3-21. 1997.

SIHAG, Ram C. Reproduction of alfalfa pollinating subtropical megachilid bees. II. Reproductive isolating mechanisms. **Zool Anz**, v.216, p.204-217. 1986.

SILVEIRA, Fernando A.; MELO, Gabriel Augusto Rodrigues; ALMEIDA, Eduardo A. B. **Abelhas brasileiras: sistemática e identificação**. Belo Horizonte, p.253. 2002.

THORNHILL, Randy; ALCOCK, John. **The evolution of insect mating systems**. Harvard University Press, London, p.548. 1983.

WITTMANN, Dieter; BLOCHTEIN, Betina. Why males of leafcutter bees hold the females' antennae with their front legs during mating. **Apidologie**, v.26, p.181-195. 1995.

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