



Insect galls associated with *Copaifera sabulicola* J.A.S Costa & L.P Queiroz (Fabaceae): Characterization and new records

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ABSTRACT

Among the plant diversity of the Cerrado, the genus *Copaifera* is commonly associated with galling insects. Here, we expand the knowledge about insect galls on *Copaifera sabulicola* J.A.S Costa & L.P. Queiroz (Fabaceae) by characterizing their occurrence in a *stricto sensu* vegetation area of the Brazilian Cerrado, Western Bahia, Brazil throughout the year. We randomly sampled 60 branches (50 cm long) from individuals of *C. sabulicola*, from April 2015 to March 2016. We found 11 morphotypes for *C. sabulicola*, of which 5 are new records. Further, we identified six gall-inducing insects, all belonging to Cecidomyiidae family. Regarding other arthropods associated with *C. sabulicola*, we observed inquilines, successors, and parasitoids, belonging to the orders Collembola, Hemiptera, Hymenoptera and Acari. The parasitoid fauna included three families of the order Hymenoptera: Braconidae, Encyrtidae and Eulophidae.

Fabaceae is considered the third largest family of angiosperms and represents the group most commonly associated with a rich fauna of galling insects in the Neotropics (Maia and Monteiro, 1999; Fagundes et al., 2020; Santos-Silva and Araújo, 2020), particularly in the Cerrado (Gonçalves-Alvim and Fernandes, 2001; Lima and Calado, 2018; Vieira et al., 2018). Among its genera, *Copaifera* L. has been indicated as a super host of gall-inducing insects (Santos et al., 2018; Fagundes et al., 2020). However, the majority of studies dealing with the occurrence of galls on *Copaifera* are related to the species *Copaifera langsdorffii* Desf., (Fabaceae), which hosts a total of 23 gall morphotypes (Costa et al., 2010; Fagundes et al., 2020).

Information on galls in other *Copaifera* species are recent and limited to *Copaifera depilis* Dwyer, *Copaifera luetzelburgii* Harms, *Copaifera sabulicola* Costa & Queiroz (Santos et al., 2018), and *Copaifera oblongifolia* Mart. (Coutinho et al., 2019), which have a considerable number of galls, but less than *C. langsdorffii* (Costa et al., 2010; Fagundes et al., 2020). The lower gall richness in these four species may be associated with the scarcity of research assessments, which may

trigger underestimations regarding the number of species of inducers in each plant. *C. sabulicola* is a potential medicinal species known only from Brazil, occurring in the Cerrado of the central-west of Bahia and the extreme north of Minas Gerais. Here, we expand the knowledge about insect galls on *C. sabulicola* by characterizing their occurrence throughout the year in a Cerrado area.

This study was carried out in Serra da Bandeira (45° 02' 46" W and 12° 05' 23" S), located in the municipality of Barreiras (State of Bahia) within a *stricto sensu* vegetation area of the Brazilian Cerrado. The altitude of this area is approximately 800 meters. This phytogeographic domain has an Aw type climate according to Köppen classification (typical savanna), showing dry winter and average temperature varying between a minimum of 20.3° and a maximum of 31.5° C. The average annual precipitation is 1,500 ± 500 mm (Nascimento and Novais, 2020).

To avoid cutting away the same individual plant every month, we randomly sampled 60 branches (50 cm long) containing insect galls from different individuals of *C. sabulicola*, from April 2015 to March 2016. All branches were stored in properly identified plastic bags. In the laboratory, we inspected the branches to characterize the galls and obtain the associated fauna. Next, we photographed the galls and described

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them following the illustrated and annotated checklist for Brazilian gall morphotypes (Isaias et al., 2013). We inspected all plant organs.

To obtain gall-inducing insects and associated fauna, we dissected part of the material under a stereomicroscope. Then, we placed samples of each gall morphotype in plastic containers with moist paper towel and labeled them with tags. To identify the specimens of Hymenoptera, we used the keys of Goulet and Huber (1993). Later, we deposited all the sampled invertebrates on the UFOB Entomological Collection and a sample of fertile material from *C. sabulicola* on the BRBA UFOB Herbarium, under the number 6679. The host plant was identified by professor Jorge Antonio Silva Costa, who first described this species and later by members of the research group through comparisons in the herbarium.

We classified insect galls as constant, accessory and accidental following Silveira-Neto et al. (1976). Constancy (C) was estimated as follows $C = (p \times 100)/P$, where p = number of collections containing a given species and P = total collections. The species were considered constant when $C > 50\%$, accessory $C > 25 < 50$ and accidental $C < 25\%$.

From a total of 8,834 insect galls sampled, we identified 11 morphotypes for *C. sabulicola*, of which 5 are new records. (Table 1 and Table 2, Fig. 1 and Fig. 2). The three most abundant morphotypes for *C. sabulicola* throughout the year were 'A' (59.2%), 'C' (20.4%) and 'F' (7.9%) (Fig. 1A, Table 2). We observed that 9 morphotypes could be classified as constant and 2 accessory (Fig. 1B). Insect galls occurred primarily on the leaves (81.8%) and on the stems (18.2%). Although all plant organs were inspected, we did not record galls on flowers and fruits. The characterization of gall morphotypes are shown in Table 1 and Fig. 2.

Six gall-inducing insects were identified from the 11 morphotypes, all from the Cecidomyiidae family (Table 1). The associated fauna was represented by inquilines, successors, and parasitoids, belonging to the orders Collembola, Hemiptera, Hymenoptera, and Acari. The parasitoid fauna was formed by three families of Hymenoptera: Braconidae, Encyrtidae, and Eulophidae. The latter was the most common family, found in eight morphotypes (Table 1). Other arthropods associated with *C. sabulicola* galls were ants, springtails, mites, true bugs and,

Table 1
Characterization of gall morphotypes found on *Copaifera sabulicola* J.A.S Costa & L.P. Queiroz (Fabaceae) in Serra da Bandeira (Barreiras, Bahia, Brazil) from April 2015 to March 2016. Im= Immature; Ad= Adult; ND= Not determined.

Gall	Organ	Shape	Foliar surface	Color	Trichomes	Chambers	Inducer	Associated fauna	References	Figure
A	Leaf	Lenticular	Both	Green, Red, Brown	Absent	One	Cecidomyiidae	Eulophidae (Hymenoptera, Ad) Hymenoptera (Im) Entomobryidae (Collembola, Ad)	Barreiras, BA (Santos et al., 2018)	2A
B	Leaf	Globoid	Abaxial	Yellow	Present	Multiple	ND	Braconidae (Hymenoptera, Ad) Eulophidae (Hymenoptera, Ad) Encyrtidae (Hymenoptera, Ad) Hymenoptera (Im)	Caetit�, BA (Vieira et al., 2018)	2B
C	Stem	Lenticular	-	Brown	Absent	One	Cecidomyiidae	Eulophidae (Hymenoptera, Ad) Formicidae (Hymenoptera, Ad) Acari (Arachnida, Ad)	Barreiras, BA (Santos et al. 2018); Vieira et al. (2018)	2C
D	Leaf	Globoid	Both	Green	Absent	One	ND	Braconidae (Hymenoptera, Ad)	Barreiras, BA (Santos et al. 2018)	2D
E	Leaf	Cylindrical	Both	Green, Red	Absent	One	Cecidomyiidae	Encyrtidae (Hymenoptera, Ad) Eulophidae (Hymenoptera, Ad) Hymenoptera (Im)	New record	2E
F	Leaf	Lenticular	Both	Green	Absent	One	Cecidomyiidae	Eulophidae (Hymenoptera, Ad) Hemiptera (Im)	Barreiras, BA (Santos et al. 2018)	2F
G	Leaf	Fold	-	Green	Absent	One	Cecidomyiidae	Hymenoptera (Im)	New record	2G
H	Leaf	Globoid	Adaxial	Green	Absent	Multiple	Cecidomyiidae	Eulophidae (Hymenoptera, Ad) Encyrtidae (Hymenoptera, Ad) Braconidae (Hymenoptera, Ad) Hemiptera (Im)	New record	2H
I	Leaf	Globoid	Abaxial	Green	Absent	One	ND	Eulophidae (Hymenoptera, Ad)	New record	2I
J	Leaf	Lenticular	Abaxial	Brown	Absent	One	ND	ND	New record	2J
K	Stem	Globoid	-	Brown	Absent	Multiple	ND	Eulophidae (Hymenoptera, Ad) Formicidae (Hymenoptera, Ad) Hymenoptera (Im)	Barreiras, BA (Santos et al. 2018); Vieira et al. (2018)	2K

Table 2

Abundance of galls per morphotype found on *Copaifera sabulicola* J.A.S Costa & L.P. Queiroz (Fabaceae) in Serra da Bandeira (Barreiras, Bahia, Brazil) from April 2015 to March 2016.

Gall	Apr	May	June	July	Aug	Sept	Oct	Nov	Dec	Jan	Feb	Mar	Total
A	123	373	354	192	217	147	224	668	601	558	1023	748	5,228
B	31	38	46	60	66	108	12	18	42	14	11	9	455
C	123	199	157	176	192	133	93	188	170	124	104	140	1,799
D	7	0	3	2	5	7	0	3	0	1	1	7	36
E	2	31	46	57	45	29	21	61	31	41	24	48	436
F	107	3	28	55	38	59	11	77	49	21	129	117	694
G	0	6	0	2	0	6	0	0	0	0	0	12	26
H	0	6	17	10	13	0	4	9	9	4	2	2	76
I	0	0	3	1	0	0	7	4	13	2	0	1	31
J	0	0	0	0	2	0	7	3	12	9	2	0	35
K	0	0	0	0	0	0	0	7	6	2	3	0	18
Total													8,834

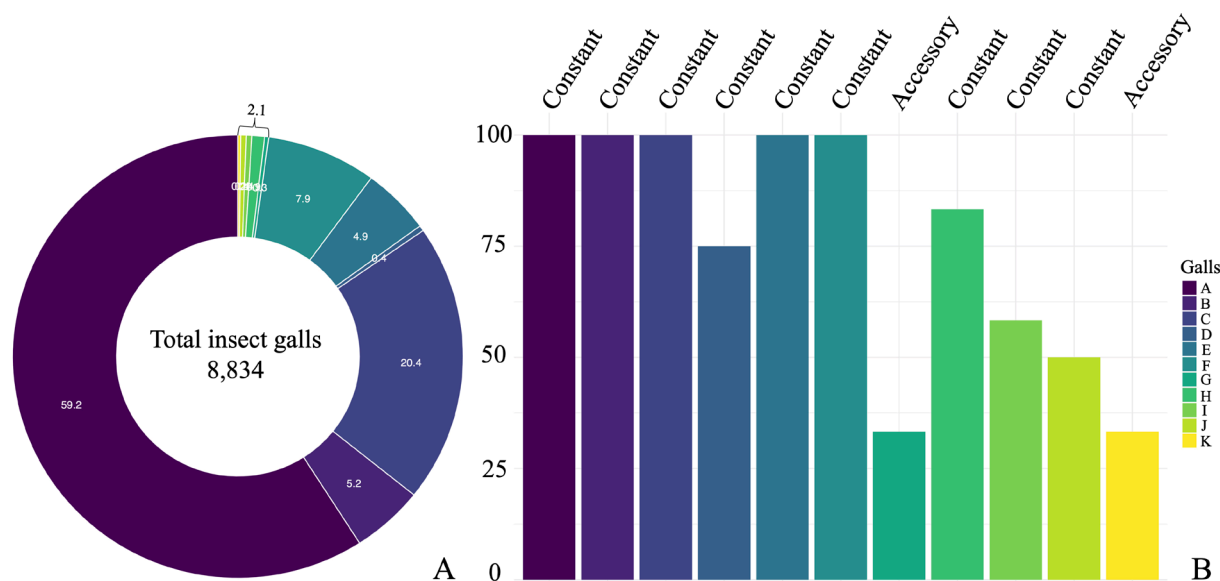


Figure 1 (A) Abundance and (B) frequency of gall morphotypes (%) found on *Copaifera sabulicola* J.A.S Costa & L.P. Queiroz (Fabaceae) in Serra da Bandeira (Barreiras, Bahia, Brazil) from April 2015 to March 2016.

mainly, hymenopterans. Neither inducers or associated fauna were found on the 'J' morphotype.

Finding five new morphotypes for *C. sabulicola* indicates that long-term studies allow us to better characterize gall-inducing insect fauna. At least, eighteen gall morphotypes have already been recorded for *C. sabulicola*, confirming that this species is a "super host" for galling insects (Santos et al., 2018; Vieira et al. 2018). Although the number of galls identified on *C. sabulicola* is notable, *C. langsdorffii* can still be considered as the species with the greatest richness of galling insects, with 23 morphotypes (Costa et al., 2010). Other species of the genus can also be considered "super hosts", such as *Copaifera oblongifolia* (Fagundes et al., 2019) with 15 morphotypes, and *C. luetzelburgii* and *C. depilis*, which have seven gall morphotypes each (Santos et al. 2018).

Regarding the inducing insects, we identified larvae and pupae of cecidomyiids (Cecidomyiidae, Diptera) in six morphotypes. Cecidomyiids are considered the most common gall-inducing insects in the neotropical region (Maia and de Araújo, 2009; Gagné and Jaschhof, 2017) and several assessments confirm the great diversity of these insects in Brazilian ecosystems (Maia and Fernandes, 2004; Maia, 2013). Information on the fauna associated with *Copaifera* is still rare and the gall-inducing

species have not yet been described (Santos et al., 2018; Vieira et al., 2018). Although a large number of galls have been characterized on *C. sabulicola*, there is a difficulty in obtaining the galling inducers, since most of the collected material is immature, which does not allow further identification.

Almeida et al. (2006) observed that the Eulophidae family stands out as parasitoid of gall-inducing insects on *C. langsdorffii* in the Cerrado (Minas Gerais state). In several other studies, hymenopterans belonging to the Eulophidae family have been associated with galls, being described as parasitoids of Cecidomyiids (Fernandes et al., 1988; Maia and Monteiro, 1999; Almeida et al., 2006; Bregonci et al., 2010; Lima and Calado, 2018; Santos et al., 2018). Insects of the family Encyrtidae and Braconidae have also been identified as parasitoids associated with insect galls in previous studies in Cerrado (Bahia, Brazil) and restinga vegetation in the Atlantic forest (Rio de Janeiro, Brazil) (Maia and de Araújo, 2009; Santos et al. 2018). We observed ants (Formicidae) and mites (Acari) within empty galls, considering them as successors. We then noticed the presence of true bugs (Order Hemiptera) in some galls, however acting as inquiline, since they were found together with immature Cecidomyiidae. The occurrence of true

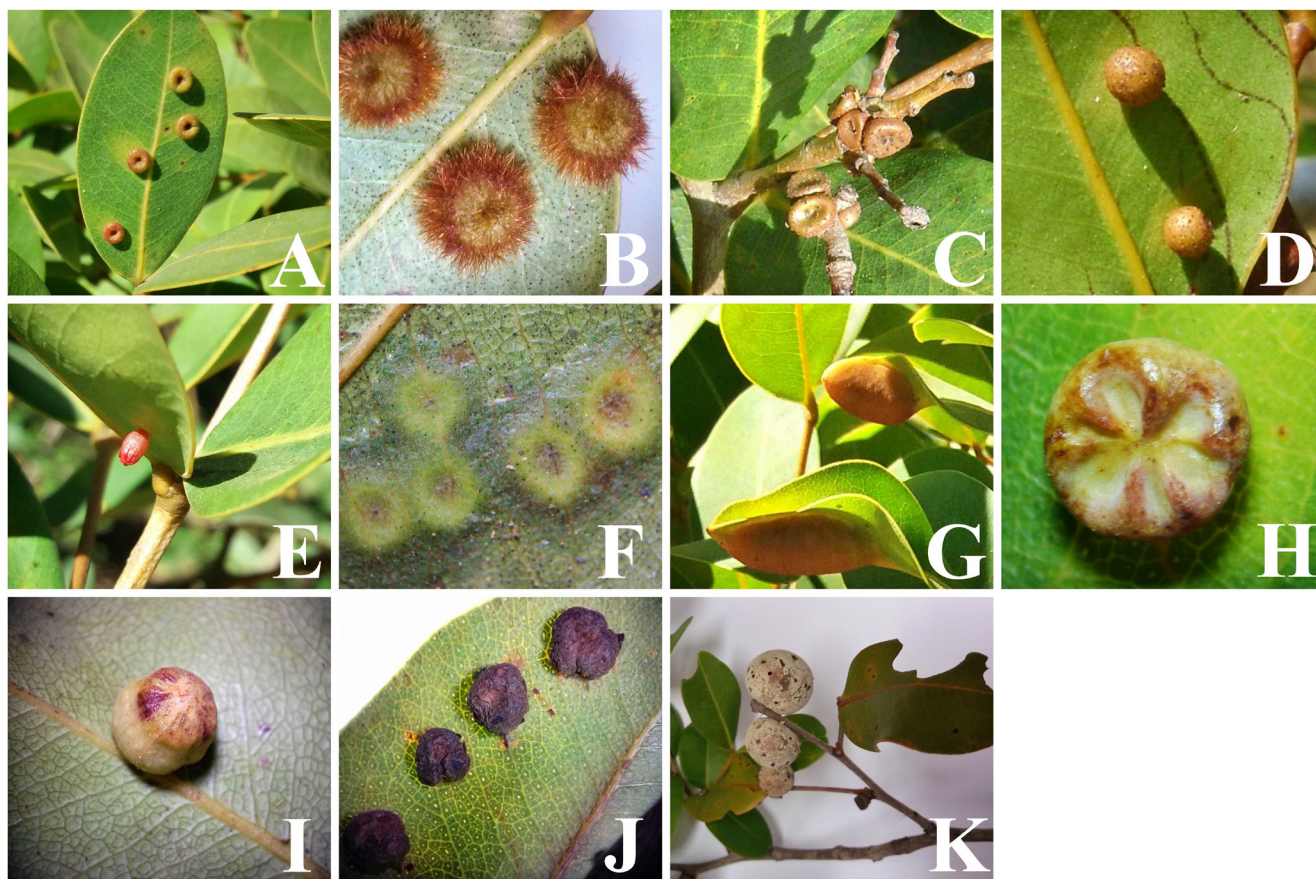


Figure 2 Gall morphotypes found on *Copaifera sabulicola* J.A.S Costa & L.P. Queiroz (Fabaceae) in Serra da Bandeira (Barreiras, Bahia, Brazil) from April 2015 to March 2016.

bugs as inquilines on galls of Cecidomyiidae has already been recorded by Maia and Fernandes (2004).

Here, we characterized insect galls as well as associated fauna on *C. sabulicola* throughout a year. We recorded five new insect galls for the species, indicating it as a “super host”, similarly to other species in the genus. Our results are fundamental for the conservation of this group, since they occur in a highly threatened ecosystem that routinely experiences threats to its survival. We highly recommend further scientific research on this insect-plant interaction in Western Bahia, particularly those to describe these new species, which are still unknown to science.

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Conflicts of interest

The authors declare no conflicts of interest.

Author contribution statement

EKKS, ABO, DC conceived the study; EKKS, ABO, DC, VPL analyzed the data and E.K.K.S. led the writing with contributions from all the authors. All authors approved the final version of this paper.

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