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SHORT COMMUNICATION

A behavioral guard caste in a primitively eusocial orchid bee, *Euglossa viridissima*, helps defend the nest against resin theft by conspecifics

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Abstract Defense castes are know from highly eusocial insects yet have rarely been described in social species with a small colony size. In nests of *Euglossa viridissima*, an orchid bee exhibiting primitively eusocial behavior, we recorded one subordinate female per nest to specialize in guarding in the presence of a dominant and a second subordinate who specialized in foraging. Guarding may have arisen as a response to cleptobiosis by conspecifics, as nests with a guard more successfully avoided intrusion and resin theft.

Keywords Apidae · Euglossini · Social evolution · Intraspecific parasitism

Insect nests are a rich source of resources for conspecifics, parasites and predators alike. Thus, it is not surprising that theft of nest materials, termed cleptobiosis (Breed et al. 2012), is common in various taxa, including solitary (e.g. *Trachusa byssina*) and eusocial species (Ribbands, 1954). Many species of the highly eusocial ants, termites, bees and wasps have evolved a well-defined morphological or behavioral guard caste with often bizarre adaptations to help defend the nest from potential intruders (Ayasse and Paxton 2002; Wilson 1971; Michener 1974). However, in primitively eusocial species, the existence of a distinct guard caste is not well defined. For instance, small colonies of social sweat bees often possess a guard at the nest entrance (e.g. Wilson 1971; Michener 1974), but it is unclear whether a single individual specializes in guarding.

Euglossa viridissima is a facultatively social orchid bee (Euglossini) that forms multi-female nests of genetically related individuals in a matrifilial association. All orchid bee species are mass-provisioners, rearing brood in resinous cells filled with pollen and nectar. A social nest usually starts with a single female that, after emergence of offspring, is dominant over its putative daughters (Cocom-Pech et al. 2008). May-Itzá et al. (2014) recently described a seasonal pattern in the sociality of *E. viridissima* and suggested that the development of social behavior in this species may reflect increased parasitism concomitant with a lack of resources during the dry season, such as nesting materials (resin) and brood cell provisions (pollen and nectar).

Our investigation was undertaken during the dry season in Merida, Mexico (at the Departamento de Apicultura, Universidad Autónoma de Yucatan, Xmatkuil, 87.37°W, 20.52°N) from 1 December 2013 to 15 January 2014. This is a season when resources, including plant resin used for brood cells, have been hypothesized to be in short supply and when nests often transition from solitary to social (May-Itzá et al. 2014). We conducted observations on four social nests, each composed of a dominant and two subordinate females. We did not take observations on nests with solitary females and others with two females. Each female could be identified by individual paint marks (Edding® 780) on the

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thorax ($n = 12$). Two independent observers monitored a nest continuously circa 6 h per day across 10 consecutive days. We used previous descriptions of intranest behavior (Cocom-Pech et al. 2008) to define female hierarchy. We also recorded the total amount of time each female was present inside the nest (TI) or foraging (TF).

We defined intraspecific robbing as a non-nestmate approaching a monitored nest, and we recorded whether it invaded the nest and if it was successful in removing resin or food. The effect of the presence of a guard on the success of an intruder in stealing nest material was tested with a generalized linear mixed model (GLMM), with the success of stealing resin as the response, the presence of a guarding subordinate as a fixed factor, and nest identity as a random factor. Incorporation of a repeated measures design accounted for multiple observations from the same nest.

Our observations confirmed that the dominant female, always the oldest bee (nests monitored the single female stage) and probably the female that had provisioned or usurped the nest, spent 90 % (SE ± 4 % per nest) of its time in its nest, mainly over the brood cells. Dominant females exhibited agonistic behavior toward subordinates, which were all females that had eclosed in the nest, when subordinates approached brood cells, except when a subordinate was building a new cell. Eating of a subordinate's egg by the dominant was observed three times, each time when the subordinate was absent from the nest. We could not determine if all the eggs laid by a subordinate were replaced, or if a subordinate replaced eggs of the dominant female.

Interestingly, in each nest, one of the subordinates acted as an entrance guard, which we term the subordinate guarding female (SGF) (Fig. 1, Supplementary material). The SGF was in all nests the most recently eclosed female (i.e. the youngest in a nest). We classified this female as a specialist as it spent most of its time guarding the entrance; it only rarely performed foraging activities (TI = 84 ± 2 %). Concomitantly, the activity of the other subordinate female was centered on building cells and foraging to collect resin or pollen (TI = 69 ± 5.4 %); we term it the subordinate foraging female (SFF). The dominant and SGF females both spent significantly more time inside the nest than the SFF ($p = 0.021$, ANOVA) but the dominant spent most of its time upon cells while the SGF spent most of its time at the nest entrance. The SFF foraged 2.3 ± 0.8 times per day, significantly more often than the SGF and the dominant ($p = 0.034$, ANOVA) (1.2 ± 0.3 and 1.1 ± 0.2, respectively). We did not observe the dominant bringing any resource into the nest, although the SGF was observed carrying resin (but not pollen) to the nest.

We registered a total of 28 events of non-nestmate, unmarked females (intruders) approaching the four monitored nests. A total of 47 % (8/17) of intruders succeeded in entering a nest when a SGF was present at the nest entrance, but only 11 % (2/17) succeeded in removing materials from that nest. In all cases the stolen material was resin; we never observed the removal of food. When a SGF was absent, we observed nine intruder approaches, and all (100 %) of them succeed in entering the nest; in seven of these nine occasions, they succeeded in robbing resin, significantly more often than when a SGF was present at the nest entrance (GLMM $z = -2.971$; $p = 0.003$, Fig. 2 Supplementary material). We observed two additional nests in which adult females were lost and, in both, non-nestmates occupied the nest and commenced cell construction. So it seems that intruders either look for resin to provision their nest or a potential site to initiate a new nest.

Intruder females usually hovered in front of a nest entrance, inducing agonistic behavior by the SGF, such as opening of the mandibles and jabbing the forelegs toward the intruder. If an intruder entered a nest, the dominant exhibited agitated behavior, e.g. beating wings, walking toward the SGF and antennating it, resulting in aggression (attacks, bites) by the SGF directed toward the intruder, and forcing the latter to leave the nest. The dominant was not observed defending the nest in the presence of an SGF, although she defended it when alone in the nest.

We describe here for the first time that behavioral specialization occurs among subordinate females of *E. viridissima* in social nests and that one of them can specifically engage in guarding. This behavior is unusual because *Euglossa* nests have hitherto been described as only defended by the dominant (Cocom-Pech et al. 2008; Augusto and Garófalo 2009, 2010; Andrade e Silva and Nascimento 2012). Based in our observations, guarding is another behavioral specialization, the other two being dominant female (lays most eggs and consumes and replaces those of subordinates), and subordinate forager (forages for resources and also lays eggs). We suggest that guarding behavior arose as a response to cleptobiosis by conspecifics, as nests with guards more successfully avoided intrusion and stealing of resin.

Two facts seem to support the idea that resin was a limiting resource. First, plasticine, a non-toxic synthetic material, was collected by foragers to build nests during our period of observations of putative resin shortage in the environment (Fig. 1, Supplementary material). Second, resin was the only item stolen from nests. Selection may favor a guarding caste in social euglossine nests, since loss of building materials must in the long run adversely influence reproduction (Michener 1974). The effect of the availability of resources on nest intrusion by conspecifics has already been described in other bee species as for *Xylocopa sulcatipes*, *Apis mellifera* and *Bombus terrestris* (Stark 1992; Downs and Ratnieks 2000; Carvell et al. 2008, respectively); for all three species, guarding bees are less tolerant toward non-nestmates when resources are scarce. For *E.*

viridissima, allocation of a potential forager at the nest entrance to protect resin from nest intruders when it is in short supply in the environment may be more advantageous than living solitarily.

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References

Andrade e Silva ACR, Nascimento FS (2012) Multifemale nests and social behavior in *Euglossa melanotricha* (Hymenoptera, Apidae, Euglossini). J Hymenoptera Res 26:1–16

Augusto SC, Garófalo CA (2009) Bionomics and sociological aspects of *Euglossa fimbriata* (Apidae, Euglossini). Genet Mol Res 8:525–538

Augusto SC, Garófalo CA (2010) Task allocation and interactions among females in *Euglossa carolina* nests (Hymenoptera, Apidae, Euglossini). Apidologie 42:162–173

Ayasse M, Paxton RJ (2002) Brood protection in social insects. In: Hilker M, Meiners T (eds) The chemoecology of insect eggs and egg deposition. Blackwell Science, Oxford, pp 117–148

Breed MD, Cook C, Krasnec MO (2012) Cleptobiosis in social insects. Psyche. doi:10.1155/2012/484765

Carvell C, Rothery P, Pywell RF, Heard MS (2008) Effects of resource availability and social parasite invasion on field colonies of *Bombus terrestris*. Ecol Entomol 33:321–327

Cocom-Pech ME, May-Itzá WJ, Medina Medina LA, Quezada-Euán JJG (2008) Sociality in *Euglossa (Euglossa) viridissima* Friese (Hymenoptera, Apidae, Euglossini). Insectes Soc 55:428–433

Downs SG, Ratnieks FLW (2000) Adaptive shifts in honey bee (*Apis mellifera* L.) guarding behavior support predictions of the acceptance threshold model. Behav Ecol 9:326–333

May-Itzá WJ, Medina Medina LA, Medina S, Paxton RJ, Quezada-Euán JJG (2014) Seasonal nest characteristics of a facultatively social orchid bee, *Euglossa viridissima*, in the Yucatan Peninsula. Mexico Insectes Soc 61:183–190

Michener CD (1974) The social behavior of the bees: a comparative study. Harvard University Press, Cambridge, p 418

Ribbands CR (1954) The defence of the honeybee community. Proc R Soc Lond B 142:514–524

Stark RE (1992) Cooperative nesting in the multivoltine large carpenter bee *Xylocopa sulcatipes* Maa (Apidae: Anthophoridae): do helpers gain or lose to solitary females? Ethology 91:301–310

Wilson EO (1971) The insect societies. Harvard University Press, Cambridge, p 562