

Nesting biology of an African allodapine bee *Braunsapis vitrea*: female biased sex allocation in the absence of worker-like behavioural castes

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The life cycle and social behaviour of an African allodapine bee, *Braunsapis vitrea*, was examined using nest contents data from thorn-nests in *Acacia karroo* in Limpopo Province, South Africa. The majority of *B. vitrea* colonies were subsocial, with 34% of nests collected containing more than one adult female. Although brood numbers increased with colony size, per capita brood numbers did not increase. Within multifemale colonies, reproductively active females had greater wing wear than their nestmates, which tended to be uninseminated, and there was no relationship between relative ovary and body size. This suggests that females who were not reproductively active were not workers in the nest, but were instead waiting to either disperse or inherit the nest. There is evidence that these younger females may help rear their mother's brood in the event of orphaning, and the number of brood in multifemale nests was greater than for single-female nests, suggesting that egg layers increase brood production when a potential alloparent is present. Sex allocation was strongly female biased, with a numerical population ratio of $r \approx 0.3$. This bias is interesting because our results suggest that it occurs in the absence of behavioural castes, where most colonies are occupied by single females, and where capita brood production does not increase with colony size. We argue that such bias might be selected by the benefits of having potentially alloparental 'insurance' daughters, who can help rear their younger siblings in the case of orphaning.

KEY WORDS: Allodapini, *Braunsapis*, life cycle, sex allocation, sociality.

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INTRODUCTION

The tribe Allodapini (family Apidae; subfamily Xylocopinae) provides exceptional opportunities for studying social evolution in bees. Social organisation varies widely among species and genera, giving rise to opportunities for comparative studies at multiple hierarchical levels (SCHWARZ et al. 1998). Allodapines are most abundant and diverse in southern Africa and Australia, with only two genera occurring outside these regions: the rare genus *Exoneuridia*, which occurs in the Middle East, and *Braunsapis*, which occurs throughout sub-Saharan Africa, Australia, and tropical and subtropical Asia. The most recent and detailed studies on allodapines have focussed on Australian genera (review in SCHWARZ et al. 1998), but MICHENER (1971) provided important data on African species that allow some broad life history and social patterns in many taxa from that region to be discerned.

Braunsapis is the most speciose allodapine bee genus and is also the most geographically widespread. Despite this, there have been relatively few detailed studies on species of this group, and only one species, *Braunsapis hewitti*, has been studied using multiple samples across different seasons (MAETA et al. 1984, 1985, 1992). MICHENER (1962, 1971) provided more limited data on African and Australian species, which allow some social traits to be discerned for a variety of species, but leave many aspects unresolved. There seems to be among-colony variation in sociality in many *Braunsapis* species, with many colonies being subsocial, while others contain multiple females with some indication of reproductive skew (MICHENER 1971). Workers in the genus appear to be common, possibly making up 50% or more of the total number of females in the population (MICHENER 1971). The diversity of *Braunsapis*, along with its wide geographical distribution, covering very diverse climates and ecosystems, suggests that it could provide important insights for comparative studies of social and life history evolution. Furthermore, recent studies based on DNA sequence data (SCHWARZ et al. 2004, FULLER et al. 2005) provided a well resolved and supported phylogeny for representative African, Asian and Australian *Braunsapis* species, setting the groundwork for phylogenetic comparative studies on social evolution. This genus therefore forms a potentially major source of information for studies into social evolution in insects.

In this report we examine the life history and sociality of a *Braunsapis* species from the north eastern region of South Africa. Our study is the first examination of an African *Braunsapis* species to use repeated sampling of a natural population throughout its life cycle.

MATERIALS AND METHODS

Study site

Allodapine nests were collected from Lesheba Wilderness (29°34'E; 22°59'S), a private game reserve located in the Soutpansberg mountain range, Limpopo Province, South Africa. The altitude is approximately 1400 m above sea level and the habitat is sour bushveld (Acocks 1988). Mean annual rainfall for the nearest major town (Makhado; approximately 35 km away) is 645.8 mm. The highest rainfall occurs from December to February (mean > 100 mm) and the lowest rainfall occurs from June to August (mean < 15 mm). Nest collections were made in September 1999, and in January, May, July, September, November and December 2000. In this area, *B. vitrea* nests were most commonly found in the swollen thorns of *Acacia karroo*, and only rarely in dead stems of other plant species. This study was based on thorn-nesting *B. vitrea* colonies only.

Field collection and processing of nests

Nests were collected from the study site in the early morning, late afternoon, or during cool, overcast conditions when all nest members were assumed to be present. Clippers were used to remove each thorn from its branch, and nest entrances were sealed with tape and the nests stored at < 8 °C until processing. Nests were split longitudinally using a knife and the contents fixed in Kahle's solution for later analysis and dissection.

Nest census

The following data were recorded for each nest: length, number of adult males and females, and the number of brood (male and female callows, male and female pupae, prepupae, larvae and eggs). Larvae were divided into three size categories, small (1st and 2nd instars), medium (3rd and early 4th instars), and large (older 4th instars) in order to obtain an indication of their developmental stage. Prepupae were classified separately to larvae. Newly emerged callow adults were identified by their lighter coloration (SCHWARZ 1986), and/or the presence of flattened body hairs, particularly on the ventral side of the thorax.

Dissection procedures

Adult females were dissected in 70% ethanol under a stereo-microscope fitted with an eyepiece micrometer. Wing length (the distance from the axillary sclerites to the apex of the marginal cell) was measured to obtain an indication of body size, and the number of wing nicks on each wing was counted to obtain an indication of wing wear (e.g. SCHWARZ 1986). Insemination status was determined by opacity of the spermatheca, either by examination *in situ* or as a temporary slide mount. The spermatheca of an inseminated female has an opaque appearance whereas an uninseminated female has a clear spermatheca (SCHWARZ 1986). However, in some cases insemination status could not be confirmed. The lengths of the three largest oocytes were measured and the values summed to provide a measure of ovary size.

Sex allocation

Numerical sex allocation was based on pupae and callows within nests (larvae could not be sexed). We estimated differential investment in males and females by measuring wet

and dry pupal weights. Wet pupal weights were measured by removing pupae from Kahle's solution, blotting for 5 min on filter paper and then weighing to ± 0.1 mg. Pupae were then dried at 50 °C for 3 days before weighing.

RESULTS

Nests comprised a short tunnel inside the thorn (mean nest length: 39.46 mm, range: 15-70 mm, $n = 177$). Although *Acacia* thorns are plentiful in the area, the exterior is relatively hard in comparison to the pith of broken stems usually hollowed out for allodapine nests. It is not clear if the bees can use all available thorns, or are limited to those thorns with a nick or small hole in the exterior where bees could burrow more easily. It is also possible that bees use abandoned ant nests, as ants also utilise the *Acacia karroo* thorns.

Life cycle patterns

Life cycle patterns were inferred by investigating the numbers of adults, eggs, larvae, prepupae, pupae and callows. Table 1 shows the number of adult females per nest for each collection period and Fig. 1 shows the mean number of brood per nest for the seven collection periods. All brood stages showed a significant difference in numbers between collection periods (Kruskal-Wallis non-parametric ANOVA, $P < 0.01$) with the exception of eggs ($H_{6, 205} = 10.86$, $P = 0.093$). There was a substantial difference between nest census data from September 1999 and September 2000 (early spring samples), with 1999 showing higher numbers of larvae ($H_{1, 90} = 7.37$, $P = 0.007$) and prepupae ($H_{1, 90} = 4.82$, $P = 0.028$). This suggests that brood abundance can exhibit yearly as well as seasonal variation. Eggs and all larval stages were present in at least some nests for all collection periods, though Fig. 1 indicates that egg production was greatest in spring and summer. Larvae also varied strongly with collection date. The largest numbers of prepupae and pupae occurred in summer, but again there appeared to be between-year variation. The overall number of callows was very low compared to the other stages, and no callows were found in September

Table 1.

The number of nests collected for each collection period as a function of the number of adult females per nest.

Collection date	1-female nests	2-female nests	3-female nests
September 1999	9	11	1
January 2000	45	17	0
May 2000	9	4	0
July 2000	7	13	3
September 2000	53	16	0
November 2000	5	2	0
December 2000	7	3	0

(1999 and 2000) and November (2000). The presence of callows at all other times, and their co-occurrence with eggs and larvae, suggests that opportunities for newly emerged females to help their younger siblings are frequent.

The maximum number of females per nest was three, but only four out of a total of 205 colonies of this size were found. To establish whether the prevalence of multifemale colonies varied with collection date, nests with 2 and 3 females per nest were combined and a Chi-square test was performed. Data from November 2000 were excluded because of the small sample size for this date ($n = 7$ nests). The analysis indicated significant variation in the proportion of multifemale nests over time ($\chi^2_5 = 22.78, P < 0.001$), with peaks in the spring 1999 and winter samples.

Brood numbers and multifemale nesting

We examined lack of brood as a function of multifemale status (single and multifemale), and then compared mean per capita brood production (PCBP) for one, two and three female nests for each collection period, excluding those nests without any brood at all. We excluded nests lacking brood because studies of Australian allodapine bees (SCHWARZ et al. 1998) have shown that a positive relationship between PCBP and increasing colony size can reflect total brood loss or failure to produce brood, rather than increased rearing efficiency.

Twenty-three percent of single-female colonies (31 of 135 nests) but only 1.4% of multifemale nests (1 of 70 nests) lacked any brood at all. These two proportions are significantly different ($\chi^2_1 = 16.22, P < 0.001$). It is possible that lack of brood is related to nest age, whereby newly founded single-female nests have not yet com-

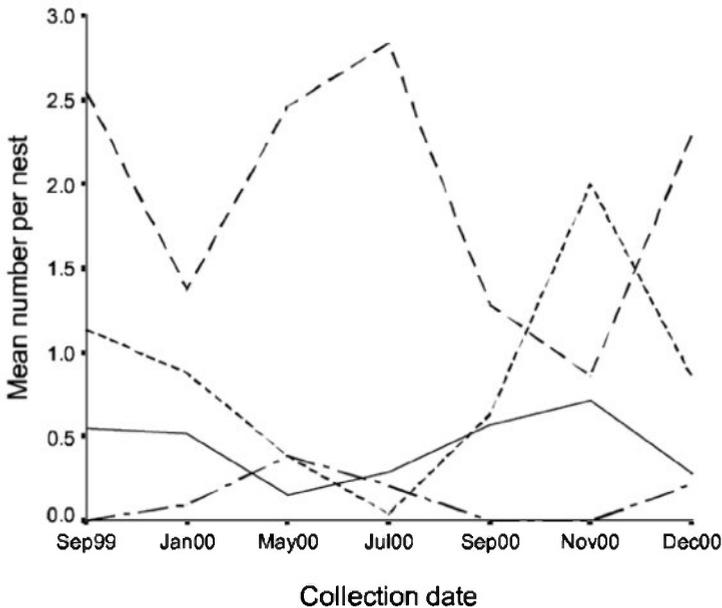


Fig. 1. — Mean number of brood stages for the seven collection periods. Brood stages are: eggs (solid line); larvae (dashed line); prepupae + pupae (dotted line); and callows (dot-dashed line).

menced egg laying. We would expect newly initiated nests to be shorter than older nests and we therefore compared nest lengths for single-female nests with and without brood for all collection periods using a 2-way ANOVA with lack of brood as a fixed factor and collection date as a random factor. No effect was found for either collection number ($F_{5, 115} = 1.687$, $P = 0.141$) or lack of brood ($F_{1, 115} = 0.427$, $P = 0.515$) on nest length.

We then examined total and per capita brood production as a function of collection date and the number of adult females per nest for those nests with at least some brood. Data could not be transformed to meet homoscedasticity requirements. Therefore, we compared total number of brood and PCBP between collection dates separately for single-, two-, and three-female nests, using a Kruskal-Wallis non-parametric ANOVA with a Bonferroni sequential adjustment (RICE 1989) for multiple tests. There were no collection date effects, so data were subsequently pooled across collections. A Kruskal-Wallis ANOVA on the pooled data indicated that the mean number of brood varied with the number of adult females ($\chi^2_2 = 41.484$, $P < 0.001$) and mean values summarised in Fig. 2 suggest that while two-female colonies have substantially more brood than single-female nests, there is little or no further increase for three-female colonies. A Kruskal-Wallis test on PCBP indicated no statistically significant effect of colony size ($\chi^2_2 = 1.608$, $P = 0.448$), suggesting that rearing efficiency does not alter with colony size. Because dissections (below) indicate that only one female is reproductively active at a time, and she is likely to be the mother of her adult nestmate, these data suggest that mothers increase their brood number when an adult daughter is present, but do not further increase brood numbers when additional daughters are available.

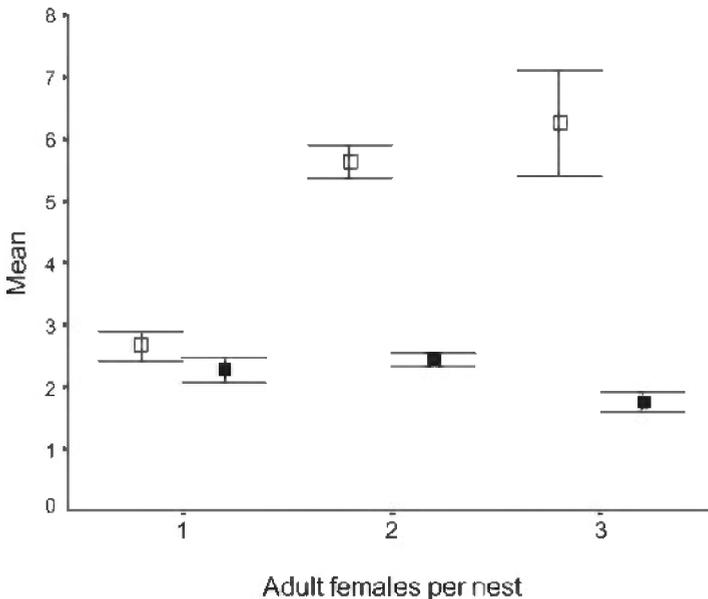


Fig. 2. — Mean number of brood (hollow squares) and mean per capita brood (filled squares) as a function of the number of females per nest. Error bars indicate \pm mean standard error.

Reproductive differentiation

The possibility of reproductive skew and caste differentiation within multi-female nests was investigated by comparing ovary size, insemination status, wing wear and body size among nestmates in multifemale colonies. Because many collections had only small numbers of multifemale colonies recovered, statistical analyses were restricted to four collections: September 1999 and January, May and September 2000. These samples covered very different parts of the seasonal life cycle (spring, summer and autumn), and since there were significant differences in brood demography between seasons, we began by investigating whether mean ovary size varied among samples. ANOVA showed that mean ovary sizes did not significantly vary among samples for either solitary nesting females ($F_{3, 30} = 1.090$, $P = 0.368$) or for females living in groups ($F_{3, 96} = 1.599$, $P = 0.195$). We also investigated whether some aspects of ovary sizes in multifemale nests varied among samples, but because the number of 3-female nests was small, we restricted these analyses to only 2-female nests. We tested whether maximum and minimum ovary sizes within a colony, and the difference between these, varied among samples, using ANOVA for the two latter variables and a Kruskal-Wallis test for maximum ovary size because this variable could not be transformed to produce homoscedasticity. These tests showed no significant among-sample differences for any of the variables ($P > 0.2$ for all comparisons). We therefore pooled data from the four samples for subsequent analyses.

Of the 49 2-female nests that were dissected, only one had both nestmates positively identified as being inseminated, 19 contained one inseminated and one uninseminated female, 23 contained one inseminated female and one female whose status was undetermined and the remaining 6 nests contained one uninseminated female and one with undetermined status ($n = 4$ nests) or both nestmates were undetermined ($n = 2$ nests). Since spermathecae that do not contain sperm are more difficult to find and classify in these very small bees, it seems likely that the large majority of 2-female nests in our sample contained one inseminated and one uninseminated female. We then compared ovary sizes for females whose status was inseminated, uninseminated and unknown using ANOVA. This showed a significant effect of insemination status ($F_{2, 95} = 24.8$, $P < 0.001$) and post hoc LSD and Scheffe's tests indicated that inseminated females had larger ovaries than both the uninseminated or undetermined females, but that these two latter groups did not differ from each other. Ovary sizes for females from multifemale nests who were inseminated, uninseminated and undetermined are summarised in Fig. 3.

We compared wing wear among the three insemination status females from 2-female nests using a Kruskal-Wallis test. This indicated a strong effect of insemination status ($\chi^2_2 = 31.925$, $P < 0.001$) where inseminated females had much more mean wing wear (mean = 8.7, $n = 44$) than uninseminated females (mean = 0.22, $n = 23$) or females whose status was unknown (mean = 2.4, $n = 31$). The distribution of wing wear for females from multifemale nests which were inseminated, uninseminated or whose status was unknown is summarised in Fig. 4, in which wing wear is categorised into 6 groups (0, 1-2, 3-4, 5-10, 11-20 and 21+ wing nicks). We then ranked nestmates according to ovary size and compared the incidence of wing wear. Of the 49 females which had the largest ovaries in their colony, 14 did not have any wear at all, whereas of the 49 lower ranked females, 43 had no wing wear. These results when combined strongly suggest that 2-female colonies nearly always comprise a single, older female which is or has been reproductive and has engaged

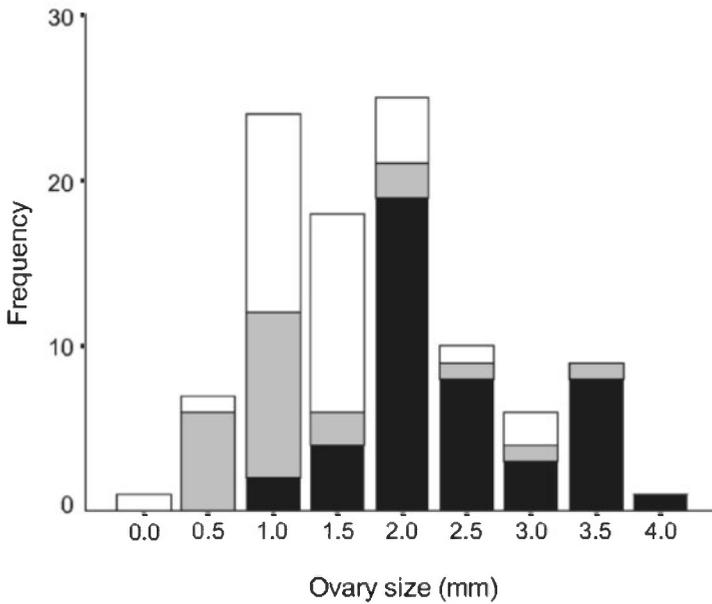


Fig. 3. — Histogram of ovary sizes for females from multifemale nests. Inseminated females are indicated by black bars, uninseminated females by grey bars, and females whose insemination status was unknown by white bars. Ovary sizes were measured as the summed length of the three largest oocytes and these were then grouped into size classes with class 0 being no measurable oocytes, and subsequent classes comprising eight 0.5 mm intervals up to 4 mm.

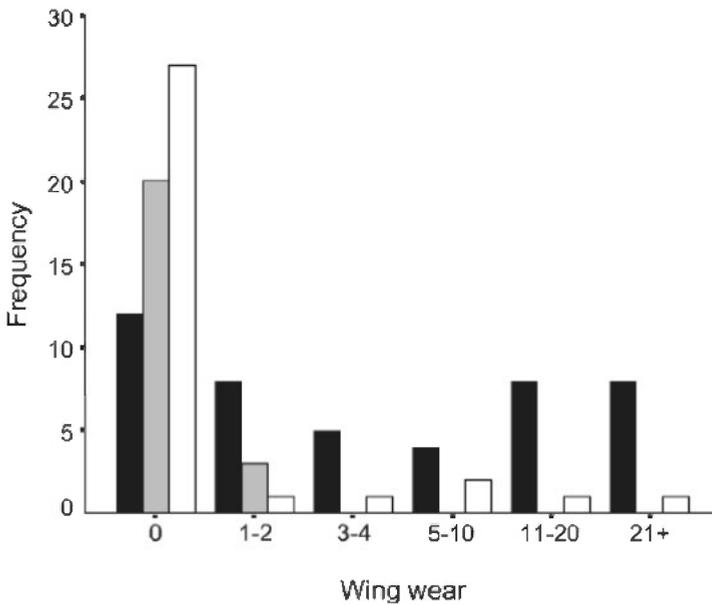


Fig. 4. — Wing wear for females from multifemale nests who were inseminated (black bars), uninseminated (grey bars) or whose insemination status was unknown (white bars). Wing wear was categorised into six classes: 0, 1-2, 3-4, 5-10, 11-20 and 21+ wing nicks.

in foraging, and a younger, unseminated female which is not reproductive and has undertaken little or no extra-nidal activity.

Lastly, for 2-female nests, we compared body size between inseminated females and females which were unseminated or whose insemination status was unknown. We combined these two latter groups since the previous analyses suggested that these latter females were very likely to be unseminated. A t-test indicated that the two groups differed significantly ($t_{87.7} = 2.679$, $P = 0.009$) and inseminated females had a larger body size, as determined by wing length (mean = 3.442 mm, $n = 44$) than the other group (mean = 3.364 mm, $n = 54$). We then visually compared the distribution of wing lengths for these groups (Fig. 5) and concluded that the predominant difference between the two groups was the relative lack of particularly small body size in inseminated females, rather than a difference in mode or maximum. When the 10 smallest unseminated/undetermined females (2.9 to 3.1 mm) were removed, there was no longer a significant difference between the two insemination status groups ($t_{86} = 0.573$, $P = 0.568$). In addition, when body size in dissected single-female nests was examined ($n = 34$), there was only one nest containing a small female (wing length 2.66 mm, insemination status unknown) and she was likely to be the sibling, rather than mother, of the brood in the nest (see below). All other females from single-female nests were within the range for inseminated females from 2-female nests shown in Fig. 5.

The data summarised above suggest that some newly emerged females could provide alloparental help to their younger, immature siblings, and one might expect this to be most evident if the original mother dies. We therefore examined single-female nests to see if there was evidence for alloparental care in the absence of a

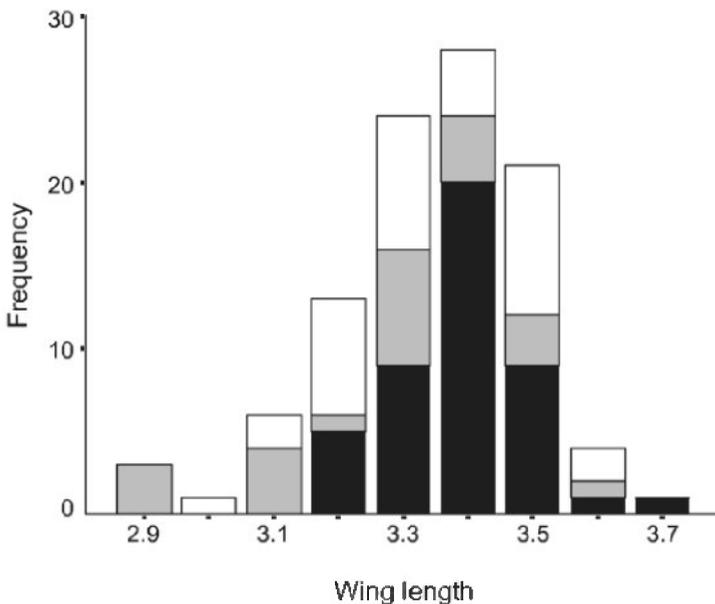


Fig. 5. — Histogram of wing lengths for females from 2-female nests who were inseminated (black bars), unseminated (grey bars) or whose insemination status was unknown (white bars). Wing lengths are grouped into intervals of 0.1 mm.

possible mother. Of the 34 females from single-female nests that were dissected, we found five nests where the female had small or moderate sized ovaries (ovary size 1.03-1.8 mm), few wing nicks (0-2) and was either uninseminated or had unknown insemination status. Two of these nests contained an egg and/or larva, and three nests included older brood (female pupae/callows). These data (particularly the nests with older brood) suggest that not all the brood in these nests were laid by the female in the nest, and were likely her orphaned siblings.

Sex allocation

Before analysing numerical sex ratios, we compared wet and dry pupal weights for a subsample of males and females. Because the total number of pupae was small, we pooled data across all sample dates. Mean wet weights were higher for females (mean = 9.220 mg, SE = 0.348, n = 30) than for males (mean = 8.027 mg, SE = 0.440, n = 15) but this difference was only marginally significant ($t_{43} = 2.046$, $P = 0.047$). Mean dry weights were also higher for females (mean = 2.153 mg, SE = 0.117, n = 30) than for males (mean = 1.807 mg, SE = 0.122, n = 15) and this difference was marginally non-significant ($t_{43} = 1.848$, $P = 0.071$). In order to take possible nest effects into account we also compared wet and dry weights using paired t-tests, where mean male and female weights were calculated for all nests where at least one pupa of each sex was present. In the 9 nests meeting this criterion, there was a significant difference for wet weights ($t_8 = 3.246$, $P = 0.012$) and a marginally non-significant difference for dry weights ($t_8 = 2.258$, $P = 0.054$), with the mean difference being 1.589 mg for wet weights and 0.378 mg for dry weights, females being heavier. These results suggest that numerical ratios will tend to over-estimate relative investment in males, but only slightly.

Nest census data on male and female pupae and callows were used to investigate sex allocation. Because sex ratios for each colony were nearly always based on small numbers of pupae and callows, we used non-parametric analyses. Sex ratio data were obtained for all collection periods (Table 2), and the total number of nests with pupae and callows was n = 82. Median colony sex ratios did not vary among collection dates when all nests were combined (Kruskal Wallis test, $\chi^2_6 = 6.603$, $P = 0.359$) or when sex ratios were calculated separately for single-female colonies ($\chi^2_5 = 7.842$, $P = 0.165$) or multifemale colonies ($\chi^2_6 = 5.274$, $P = 0.509$). Data were therefore pooled across all samples for subsequent analyses. Mean sex ratio per nest was female biased ($r = 0.238$) when all nests were combined and also when calculated separately for single-female nests ($r = 0.154$) and multifemale nests ($r = 0.314$). The two latter values differed significantly (Mann-Whitney U test, $Z = -2.032$, $P = 0.042$) suggesting that single-female nests tend to produce a greater proportion of daughters. We then examined whether sex ratio varied with brood size, a pattern that has been found to occur in several *Exoneura* species (SCHWARZ 1988, BULL & SCHWARZ 2001). Because of small sample sizes, total brood size was grouped into five categories (1-2, 3-4, 5-6, 7-8 and 9-10 brood) and mean sex ratios for each brood size category are shown for both single-female and multifemale colonies (Fig. 6). Neither Kruskal-Wallis tests (using the above brood size categories as the factor) nor non-parametric correlations between total brood size and sex ratio per nest indicated significant relationships for either single-female or multifemale colonies (single-female nests: $\chi^2_5 = 7.842$, $P = 0.165$, Spearman R = 0.194, $P = 0.237$; multifemale nests: $\chi^2_6 = 5.274$, $P = 0.509$, Spearman R = 0.227, $P = 0.142$). Lastly,

we used one-sample t-tests to compare mean sex ratio against $r = 0.5$. These indicated that sex ratios were significantly lower than 0.5 for both single-female ($t_{38} = 6.992, P < 0.001$) and multifemale colonies ($t_{42} = 3.059, P = 0.004$).

We also estimated population wide numerical sex allocation by summing male and female pupae across all collections and this also suggested strong female bias (total of 89 females: 38 males, $r = 0.30$), close to the estimate based on per nest sex ratios.

Table 2.
The number of female and male pupae and callows for each collection period.

Month	Females:Males
September 1999	7:6
January 2000	34:15
May 2000	8:2
July 2000	6:0
September 2000	20:7
November 2000	6:3
December 2000	8:5

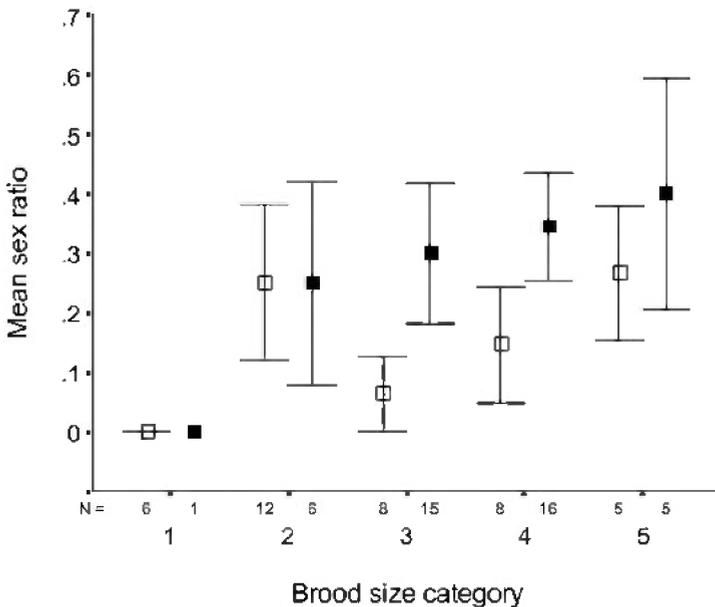


Fig. 6. — Mean sex ratio (\pm mean standard error) as a function of brood size for single-female (hollow squares) and multifemale nests (filled squares). Brood sizes were categorised as 1-2, 3-4, 5-6, 7-8 and 9-10 total brood. Sample sizes are given for each category underneath the abscissa.

DISCUSSION

Life cycle

Our results show that egg production and brood maturation occurs throughout the year, but that brood sizes vary with season and also across years. Continuity of egg production and brood maturation may be due to the relatively mild winters in the study site, which lies just within the tropic of Capricorn. The mean number of eggs per nest never exceeded one and, along with mean values for other immature stages, indicates that brood production is staggered, without the strong seasonal pulses leading to egg stockpiling in allodapines from more southern latitudes. Brood development of *B. vitrea* contrasts with certain other allodapines from southern South Africa (MICHENER 1971) and temperate Australia (SCHWARZ et al. 1997, 1998) where species have very distinct seasonal patterns of egg production and brood maturation.

In terms of colony phenology, *B. vitrea* is similar to many other African *Braunsapis* species, such as *B. facialis* and *B. bouyssoui* collected from south and east Africa (MICHENER 1971). Although MICHENER's (1971) studies of *Braunsapis* did not use repeated sampling, there was good evidence for continuous egg production and asynchronous brood development. The colony phenology of *B. vitrea* is also similar to the subtropical Asian *Braunsapis* species *B. hewitti* (MAETA et al. 1985) and the tropical species *Macrogalea zanzibarica* (TIERNEY et al. 2002). Year-round brood development, with a very small number of eggs at any one time and near continuous maturation of brood, is important for understanding social behaviour in *B. vitrea* because it creates extended opportunities for alloparental care.

Sociality

Although about 1/3 of the nests in our sample contained two or three females, we found no evidence for the existence of queen-like or worker-like roles. Instead, nearly all 2-female colonies comprised a mated female with worn wings and evidence of ovarian enlargement, along with a usually unmated female with little or no wing wear and with smaller ovaries, which suggests that they are younger and perhaps recently emerged females. Although the mean body size of these latter females was smaller than that of mated females, this was due to a skewed distribution of body size, with a longer tail of smaller sized females. It seems likely that smaller sized females may show higher levels of mortality and be less likely to survive to either found a new nest or inherit their natal nest. This is supported by the fact that only one small-sized female was found in single-female nests.

Lack of brood was much more common in single-female nests than in multi-female nests, and this was not associated with nest length. This suggests that nests lacking brood are not newly founded nests, but rather that the prevention of brood loss could be a benefit to multifemale nesting. Evidence for increased protection from predators as a result of group living has been shown for a number of Australian species such as *Exoneura robusta* and *E. nigrescens* (SCHWARZ 1994, BULL & SCHWARZ 1996). In *B. vitrea*, however, nest length is restricted to the length of the thorn, and it is possible that older nests are not able to increase nest length proportionally with increased age as they could do in stem nests.

Unlike studies of *Exoneura* and *Brevineura* species (SCHWARZ et al. 1998; TIERNEY et al. 1997, 2000), per capita brood production did not increase from one- to two-female nests. However, we did find that total brood numbers were larger in multifemale than single-female nests. There is evidence that only one female in multifemale nests had laid eggs, and she was likely to be the mother of her adult nestmate(s). It is possible that the slow, continual production of brood results in more brood being present in the nest by the time that an adult daughter emerges. However, another possible explanation of this finding is that mothers increase brood sizes if an adult daughter is present. Such a pattern would be expected if daughters are potential alloparents, allowing their mothers to produce extra brood in anticipation that they will be cared for in the case of orphaning. Sib rearing following maternal death has been reported for experimentally orphaned colonies of *Exoneura nigrescens* (BULL & SCHWARZ 1997), and has been inferred for other allodapine species (SCHWARZ et al. 1998, TIERNEY et al. 2000, JOYCE & SCHWARZ in press) and is also likely for an arid-zone allodapine, *Exoneurella eremophila* (HOGENDOORN et al. 2001). Indeed, such sib rearing is expected to reap substantial indirect fitness gains for sibling alloparents via head start advantages (QUELLER 1989) and assured fitness returns (GADAGKAR 1990). However, in *B. vitrea* such alloparental care does not coincide with evidence for reproductive skew. Instead, it seems likely that after orphaning, adult daughters could reap indirect fitness benefits by completing the rearing of their immature siblings, but still go on to become reproductive themselves. Furthermore, such alloparental care may have direct fitness benefits as well, if some of the resulting adult siblings help rear their sister's brood in the case of her death.

The lack of queen-like or worker-like roles in *B. vitrea* contrasts with findings by MICHENER (1971) who reported that in most or perhaps all species of African *Braunsapis* that he examined, 30-45% of nests containing brood had one or more workers in the active season. Our findings also contrast with the Asian species *B. hewitti*, where a laboratory study showed that some daughters dispersed, while others remained as workers in their natal nests (MAETA et al. 1985). This contrast with *B. vitrea* could be due to the very short nest lengths imposed on this species by the size of *Acacia* thorns, which does not arise in species that nest in stems, where nest lengths and colony sizes can become much larger. If brood sizes are constrained in *B. vitrea*, then there may not be sufficient foraging or rearing work available for the evolution of a worker-like caste.

Sex allocation

As for most other allodapines (MICHENER 1971; TIERNEY et al. 1997, 2002; SCHWARZ et al. 1998), we found female biased sex ratios in *B. vitrea* and the degree of bias was quite substantial, with a numerical population ratio of about $r \sim 0.30$. Relative pupal weights suggest that this value will slightly underestimate the extent of female allocation bias. Female biased sex allocation in many Hymenoptera has been attributed to queen-worker conflict (CROZIER & PAMILO 1996), where workers would prefer to rear sisters than brothers because of their greater relatedness to the former. If workers have complete control over sex ratios, then for a colony comprising a single-mated queen and her daughter workers, we might expect (TRIVERS & HARE 1976) a brood sex ratio of 1:3 (male:female) and, for colonies where workers had less than complete control, a less female biased ratio. However, in our case we found that the most extreme female bias occurred in single-female colonies,

where there is no possibility for workers controlling sex ratios, whereas less-biased ratios occurred in colonies with one or more potential workers. This means that queen-worker conflict seems unlikely to explain the pattern of bias in *B. vitrea*. On the other hand, female biased allocation in allodapines has been attributed to several factors other than queen-worker conflict. Local resource enhancement (LRE, also termed local fitness enhancement, LFE, SCHWARZ 1994) has been suggested as a mechanism for female biased ratios in *Exoneura robusta* (SCHWARZ 1988) and *E. angophorae* (CRONIN & SCHWARZ 1997), and results from positive non-linear fitness interactions among sisters. This situation is unlikely to apply to *B. vitrea*, where there is no evidence for protracted cooperation among nestmates and where PCBP does not increase with colony size.

BULL & SCHWARZ (2001) argued that female biased allocation in *E. nigrescens* was due to brood insurance via protogyny (BIP). In this model, older adult daughters help rear their younger immature siblings in the event of orphaning, and investment in these 'insurance' daughters is discounted when reckoning maternal investment in sons and daughters overall, leading to an apparent female bias. We suggest that such a model may also help explain the female bias in *B. vitrea*. Although BULL & SCHWARZ applied the BIP model to a temperate univoltine bee with seasonal brood production, TIERNEY et al. (2002) suggested that it might also apply to a tropical allodapine, *Macrogalea zanzibarica*. We found evidence that in some of our *B. vitrea* single-female nests, the adult female was unlikely to be the mother of all the brood that were present, suggesting alloparental care following orphaning. Observations of *B. vitrea* colonies transferred to artificial nests indicated that, in at least one instance, a callow daughter appeared to provide alloparental care after the disappearance of the older females, providing more direct evidence for older daughters rearing their younger, orphaned siblings (AENMEY 2002). In *B. vitrea*, it may be to a female's advantage to maximise the probability that an adult daughter is present in the nest at any time, and given the small brood sizes and developmental spread of immatures within nests in this species, this may require a female biased sex ratio. In such a case, the presence of an alloparental daughter would enhance survival of her younger siblings, and female biased allocation would be selected for via positive inter and intra-sexual sibling fitness effects (TAYLOR 1981).

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