

Partial local mate competition and the sex ratio: A study on non-pollinating fig wasps

S. A. West^{1,2,*} and E. A. Herre³

¹*Department of Biology, Imperial College at Silwood Park, Ascot, Berkshire SL5 7PY, U.K.*

²*Current address: Institute of Cell, Animal and Population Biology, University of Edinburgh, Edinburgh EH9 3JT, U.K., e-mail: Stu.West@ed.ac.uk*

³*Smithsonian Tropical Research Institute, Apartado 2072, Balboa, Republic of Panama*

Key words: Fig wasps; parasitoid; sex ratio; Local Mate Competition; wing dimorphism.

Abstract

In many species, mating takes place in temporary patches where only a small number of females produce offspring. In this situation Local Mate Competition (LMC) theory predicts that the optimal sex ratio (defined as proportion males) should become increasingly female biased as the number of females contributing offspring to a patch decreases. However, in a large number of these species, some mating is also likely to occur away from the natal patch (termed partial LMC). In this case the degree of LMC is reduced, and theory predicts a relatively less female biased sex ratio. We tested these two predictions with field data from 17 species of New World non-pollinating fig wasps representing three genera. We present a model which suggests that the average number of females ovipositing in a fruit (i.e. patch) is positively correlated with the proportion of fruit of a given tree species in which that species of wasp occurs. Across species, the overall sex ratio was positively correlated with the proportion of fruit in which that species occurs. Furthermore, the males of some species are wingless, and in these species all mating must take place before females disperse from their natal fruit. In contrast, the males of other species are winged, and in these species mating may also take place away from the natal fruit. Species with winged males had less female biased sex ratios than species with wingless males that occurred in a similar proportion of fruit. Finally, the correlation between sex ratio and the proportion of fruit in which a species occurs was also observed within species when comparing between the fruit crops of different trees. This suggests that individual females facultatively adjust the sex ratio of their offspring in response to variable LMC.

* Author for correspondence

Introduction

The study of how sexually reproducing organisms allocate their resources between the two sexes (sex allocation) has provided one of the most productive areas in evolutionary biology (Charnov, 1982; Leigh et al., 1985). One particularly active avenue of research has centred on the evolution of female biased sex ratios in structured populations (where sex ratio is defined as the proportion of males). Hamilton (1967) demonstrated that when the offspring of one or a few mothers mate amongst themselves at their natal patch, before the daughters disperse, a female biased sex ratio is favoured. This has been termed Local Mate Competition (LMC). Species in the Hymenoptera, and in particular parasitoids, have played a central role in testing models of LMC (Werren, 1987; King, 1993; Godfray, 1994; Herre et al., 1997). However, future progress in this area is hindered severely by the lack of knowledge of parasitoid mating structures in nature (Nadel and Luck, 1992; Molbo and Parker, 1996). For example, considerable attention has been paid recently to situations in which some mating occurs away from the natal patch (termed partial LMC; reviewed by Hardy, 1994; Godfray and Cook, 1997). Theoretical models predict that in this case the degree of LMC is decreased and a relatively less female biased sex ratio is favoured for a given number of females ovipositing in a patch (Uyenoyama and Bengtsson, 1982; Frank, 1986; Nunney and Luck, 1988; Werren and Simbolotti, 1989; King and Skinner, 1991; Ikawa et al., 1993; Taylor, 1993; Greeff, 1995).

Two features of the non-pollinating fig wasps make these insects ideally suited to testing theories of partial LMC. Firstly, they have population structures that lead to LMC. Non-pollinating fig wasps are species of parasitoids or gall-formers that are able to develop only within fig fruit. Females of most species, and all those considered here, oviposit from the outside of the fig fruit. Fruit are suitable for oviposition for only a few days, and so in some species the number of females ovipositing in each fruit (i.e. patch) will be small. The wasps within a single fruit mature synchronously, and so males are able to mate with females before they leave their natal fruit. Hence, a small number of females lay their offspring in each fruit, and these offspring mate amongst themselves before the daughters disperse. Secondly, the extreme interspecific variation in male morphology allows us to distinguish between species in which either no or some mating away from the natal patch is likely to occur (Hamilton, 1979; Greeff, 1995; Cook et al., 1997; Herre et al., 1997). The males of some species are wingless, and in these species all mating must take place before females disperse from their natal fruit. In contrast, the males of other species are winged, and in these species mating may take place either before or after females disperse from their natal fruit (i.e. partial LMC).

Here we study 17 species of non-pollinating fig wasps representing three genera (*Idarnes* Walker, *Aepocerus* Mayr and *Physothorax* Mayr). Each of these wasp species develops within the fruit of one of 10 species of monoecious New World figs (*Ficus*, subgenus *Urostigma*, section *Americana*). We are unable to determine the precise number of females of each non-pollinating fig wasp species that oviposit in each fruit. However, we present a model showing that the proportion of a tree's fruit crop in which a wasp species occurs is likely to be positively correlated with

the average number of females of that species that oviposited in each fruit (Fig. 1a; see also Hamilton, 1979; Greeff, 1995; Read et al., 1995). Given this assumption, theoretical models predict that across species: (1). The average wasp sex ratio will be positively correlated with the proportion of a tree's fruit in which that species occurs (Fig. 1b), and (2). Wasp species with winged males will have a less female biased sex ratio than species with wingless males that occur in a similar proportion of fruit.

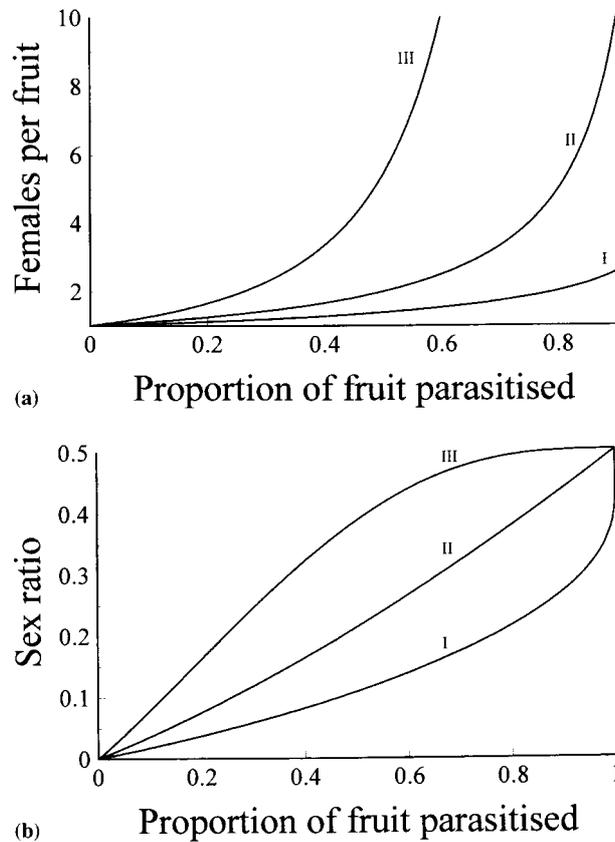


Fig. 1. The theoretical relationship between proportion of fruit in which a species appears and: (a) the average number of females of that species ovipositing in a fruit containing that species, and (b) the overall sex ratio of that species. The different lines represent situations when: (I) females are randomly distributed between the fruit, visiting each fruit with an equal probability (i.e. poisson distribution); (II) and (III) females aggregate at certain fruit (i.e. negative binomial distribution). The aggregation parameter k is equal to 1.0 in line (II) and 0.5 in line (III) (low values of k describe high degrees of aggregation).

Background biology

Wasp species that are able to develop only within the fruit of fig trees are collectively termed fig wasps. These species include both mutualistic pollinators upon which the fig trees depend, and non-pollinators, which provide no apparent benefit to their host figs. The fig species that we have examined are grouped in the subgenus *Urostigma*, section *Americana*, and are each pollinated by a single species specific pollinating wasp species from the genus *Pegoscapus* (Wiebes, 1995; Herre et al., 1996). Individual trees of all fig species may produce one to three fruit crops per year (Windsor et al., 1989). The development and maturation of the fruit within the crop of a single tree is synchronous. When the fruit are receptive, mated pollen-bearing female pollinating wasps (foundresses) arrive at the tree, enter the fruit, pollinate the receptive female flowers, and lay eggs in the ovaries of some of these flowers (Frank, 1984; Herre, 1989, 1996; Herre and West, 1997). These foundress wasps subsequently die inside the fruit. Just before final ripening of the fruit takes place, the wingless males of the pollinating wasps chew their way out of the flowers in which they have developed. They then crawl around the interior of the fruit searching for flowers which contain female wasps. The males chew open these flowers and mate with the females. The females then emerge from their flowers and gather pollen, before leaving through a hole in the fruit wall chewed by the male wasps (Herre, 1989).

The non-pollinating wasps also appear to be generally restricted to a single host fig species (Boucek, 1988, 1993; Machado et al., 1996). However, a single fig species may have a large number of non-pollinating wasp species associated with it (Compton and Hawkins, 1992; Cook and Power, 1996). The most common non-pollinating wasps found emerging from the *Urostigma* figs in Panama are those in the genera *Idarnes*, *Aepocerus* and *Physothorax* (West et al., 1996). The *Idarnes* wasps can be split further into two morphologically distinct groups (Boucek, 1993; Machado et al., 1996). Those belonging to the *flavicollis* and *carne* species-groups have wingless males and females with very long ovipositors. We refer to this group as *Idarnes*. In contrast, the *Idarnes* belonging to the *incerta* species-group have winged males and females with relatively short ovipositors. We refer to this group as *Idarnes (incerta)*.

Unlike pollinating wasps, the females of the non-pollinating species considered in this paper do not enter the fruit in order to lay their eggs. Instead females oviposit into a fruit from the outside. Different species may oviposit at different stages of the fruits development, and use different resources for larval development (Bronstein, 1991; West and Herre, 1994; West et al., 1996). For example, the *Idarnes* wasps develop within female flowers, for which they appear to compete with the pollinating wasps. In contrast, the *Aepocerus* and the *Idarnes (incerta)* wasps develop within much larger galls which protrude into the centre of the fruit. The final group, the *Physothorax*, are parasitoids of the *Aepocerus* wasps. However, within any wasp species, oviposition, development and emergence are synchronised across the fruit crop of a single tree. In addition, the different species within each of the genera/species groups appear to use the same resources for larval development (West and Herre, 1994; West et al., 1996). The adult non-pollinating wasps emerge from their flowers

at approximately the same time as the pollinators. After mating within the fruit the females of all species, and any winged males, leave through the hole in the fruit wall chewed by the pollinating males. While the *Idarnes* species produce wingless males, as the pollinators do, the *Idarnes (incerta)*, *Aepocerus* and *Physothorax* species have winged males. Males of all the winged species that we have considered will mate readily with females inside their natal fruit. Furthermore, we have observed winged males of three species (*Idarnes (incerta)* sp. from *F. citrifolia*; *Aepocerus* sp. and *Physothorax* sp. from *F. dugandi*) chew open galls containing females of their own species within their natal fruit and then climb into those galls to mate with the females. Males will mate multiple females (West et al., 1998).

Estimating the degree of LMC

The basic model

The purpose of this section and the accompanying appendix is to argue that the proportion of a tree's fruit crop in which a wasp species occurs is likely to be positively correlated with the average number of females, of that species, that oviposit in each fruit (Fig. 1a). In order to demonstrate this, consider a number of females, of a certain species, who arrive at the fruit crop of a single tree and then move around laying a few offspring in each fruit they visit. We assume that females may distribute themselves amongst the fruit in one of three possible ways. First, females may be randomly distributed among the fruit, with each female visiting each fruit with an equal probability (poisson distribution). Second, females may aggregate at certain fruit (negative binomial distribution). Third, females may be underdispersed, and spread evenly among the fruit. Reasons why females might not be distributed at random will be considered in the discussion.

Nonetheless, a feature common to all these possible distributions is that increasing numbers of females visiting a tree will lead to an increase in both the average number of females ovipositing in a fruit, and the proportion of fruit in which females of that species oviposit. Consequently, the average number of females that oviposited in fruit that contains wasps of a certain species should be positively correlated with the proportion of fruit in that crop containing wasps of that species (Fig. 1a). This, therefore, leads to the prediction that the overall sex ratio of a species should be positively correlated with the proportion of a tree's fruit in which that species occurs (Fig. 1b; see Read et al., 1995) for a similar argument applied to parasitic protozoa).

The constraint of finite brood size

The above model assumes that females can produce any offspring sex ratio and that all females produce broods of equal size. Relaxing these assumptions can cause a number of complications (Stubblefield and Seger, 1990; Read et al., 1992, 1995; Nagelkerke, 1994, 1996; Nagelkerke and Hardy, 1994; Shutler et al., 1995; Greeff,

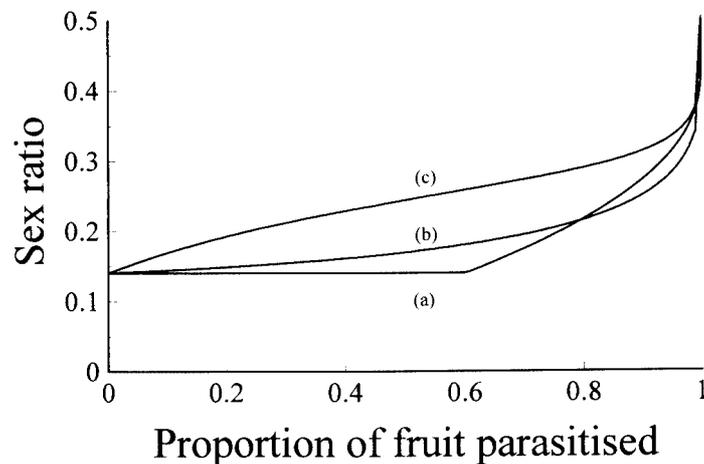


Fig. 2. The effect of the constraint imposed by small brood size on the theoretical relationship between the sex ratio of a certain species and the proportion of fruit in which that species appears. In all cases females are assumed not to be able to produce an offspring sex ratio lower than 0.14. The different lines represent situations when: (a) females adjust their sex ratio according to the average number of females that oviposit in a fruit, (b) and (c) females adjust their offspring sex ratio in response to the precise number of females that oviposit in each fruit. For (b) we assumed that the total number of offspring that developed in a fruit was independent of the number of females that oviposited in it. For (c) we assumed that the total number of offspring that developed in a fruit was linearly dependent upon the number of females that oviposited in it.

1997; West et al., 1997; West and Herre, 1998). For example, finite brood sizes limit the offspring sex ratios that a female can produce. One consequence of this is that the most female biased offspring sex ratio that a female can produce, whilst still producing sons, is $1/(\text{brood size})$. It is therefore possible to produce a more female biased sex ratio with a large brood than with a small brood (Griffiths and Godfray, 1988). Here, we consider the implications of this for the sex ratio pattern that we would expect to observe in nature.

The general conclusion is that the constraint of finite brood sizes leads to less female biased sex ratios being predicted for a given proportion of a tree's fruit in which a species occurs. In Figure 2 we show the possible consequences of incorporating this constraint into our model in three ways. The equations for deriving these relationships are given in the Appendix, and the predicted sex ratios represent the overall population sex ratio. In all cases we assumed that females are distributed amongst the fruit at random (i.e. poisson), and that there is a minimum possible sex ratio that a female can produce. This minimum sex ratio was set as 0.14, the predicted Y-intercept of the best fitting lines through the data in Figure 3, and corresponds to one male in a brood of seven offspring. Line (a) was calculated assuming that females adjust their sex ratio according to the average number of females that oviposit in a fruit, as in Figure 1b. For lines (b) and (c) we assumed that females adjusted their sex ratio precisely, as predicted by Hamilton's original

model, in response to the number of females that oviposited in each fruit. However, if a single female oviposited in a fruit we assumed that she produced a sex ratio of 0.14. To obtain line (b) we assumed that the total number of offspring that developed in a fruit was independent of the number of females that oviposited in it. This represents the case when a single female produces the maximum brood size that can develop in a fruit, and so there is extreme density dependence. For line (c) we assumed that the total number of offspring that developed in a fruit was linearly dependent upon the number of females that oviposited in the fruit. This represents the situation where oviposition sites are not limiting and larvae in a fruit do not compete for resources. Lines (b) and (c) therefore represent two extreme possible scenarios between which species could occur, and we might expect intermediate situations. For example, consider a species in the genus *Idarnes*, which competes with the pollinating wasps for female flowers (West and Herre, 1994; West et al., 1996). Although the total number of oviposition sites (female flowers) may not be limiting, increasing numbers of females ovipositing in a fruit could lead to females laying eggs in flowers in which other *Idarnes* females had already laid an egg, and/or being forced to lay their eggs in flowers that provide less resources for a developing larvae (e.g. those containing pollinator larvae). The difference in these two lines arises if fruit in which several foundresses oviposited produce more offspring, thus having a greater effect on the overall population sex ratio.

In the extreme case, it is possible that brood size may be a major determinant of sex ratio variation between species. As discussed above, it is possible to produce a more female biased sex ratio, and still produce both sexes, with a larger brood size. Consequently, under conditions of extreme LMC, negative correlations are predicted, and have been observed, between sex ratio and brood size (Griffiths and Godfray, 1988; Morgan and Cook, 1994; Nagelkerke and Hardy, 1994; Hardy and Cook, 1995; Nagelkerke, 1996; Herre et al., 1997; West et al., 1997; West and Herre, 1998). If the sex ratios of the species that we are considering here were determined primarily by the constraint of small brood size then we would expect to observe a negative correlation across species between sex ratio and brood size.

Methods

We sampled trees in the vicinity of Barro Colorado Island, Republic of Panama. Each of these trees was visited approximately twice a week between January and May 1994. This procedure enabled us to monitor fruit development on a large number of trees. Any trees bearing a fruit crop near maturation were visited more frequently, and a sample of 40–50 fruit was collected during the period of wasp emergence. These fruit were collected before any wasps had exited. Each fruit was then cut open and sealed between two matching petri dish halves, and all the wasps were allowed to emerge before being frozen. Later, the fruit was dissected for unemerged wasps, and the number and sex of each species of wasp that the fruit had contained were recorded. We then calculated, for each wasp species, the proportion of fruit in which they occurred (considering only fruit from their host fig species), their overall sex ratio,

and the average number of wasps developing in fruit which contained that species (defined as the combined brood size).

Statistical analysis

We first analysed our data treating species as independent data points. Proportional data such as sex ratios often have non-normally distributed error variance and unequal sample sizes (McCullagh and Nelder, 1989; Crawley, 1993). To avoid these problems we analysed the data with a general linear model analysis of deviance, assuming binomial errors in the GLIM statistical package (GLIM 3.77, Numerical Algorithms Group, Oxford, 1985). The number of males in a species was used as the response variable and the total number of males and females in that species as the binomial denominator. Initially, a full model was fitted to the data, including all explanatory variables and their interactions. All continuous explanatory variables were assessed for non-linearity by fitting quadratic terms. Terms were then removed from the full model by step-wise deletion (Crawley, 1993). Whether the removal of a term caused a significant increase in deviance was assessed with a χ^2 -test. In all cases the residual deviance was approximately equal to the residual degrees of freedom, and so the assumption of binomial errors appeared to be valid (Crawley, 1993). In addition to examining the pooled data set, we also examined the data for the wingless (i.e. *Idarnes*) and the winged species (i.e. *Idarnes (incerta)*, *Aepocerus* and *Physothorax*) separately. This was done to investigate if the same relationships occurred within groups.

However, species may not be independent data points because they are phylogenetically related (Felsenstein, 1985; Burt, 1989). This leads to potential problems if a trait is evolutionarily conserved, or highly correlated with an unknown phylogenetically inert third variable (Ridley, 1989). Put simply, closely related species tend to share many characters through common descent rather than through independent evolution. We therefore also used the molecular phylogeny constructed by Machado et al. (1996) and the method of independent contrasts (Felsenstein, 1985; Grafen, 1989; Pagel, 1992) to examine the relationship between the overall sex ratio of a species and the proportion of fruit in which it appears. Independent contrasts are derived by calculating the difference in the response and the explanatory variables across pairs of species, or higher nodes that share a common ancestor. Hypothesis testing can then be carried out on these contrasts with a regression fixed through the origin or a sign test (Harvey and Pagel, 1991; Garland et al., 1992). We derived contrasts using the statistical package of Purvis and Rambaut (1995), which allows both the Grafen (1989) and the Pagel (1992) methods of assigning branch lengths to be used.

Results

The characteristics of each species are shown in Table 1. An estimate of the average number of developing offspring that were laid by individual females

(defined as the average brood size) can be made if we assume that ovipositing females were distributed at random amongst fruit. Given this assumption, the proportion of fruit in which each species occurs allows us to estimate the average number of females ovipositing in a fruit that contains that species (equation 1 of the appendix). Dividing the combined brood size by this number gives an estimate of the average brood size. Across species this estimate of the average brood size was significantly positively correlated with the combined brood size ($F_{(1,15)} = 275.47$, $P < 0.01$, $r^2 = 0.948$, $n = 17$).

Across species there was a significant positive relationship between the proportion of fruit in which a species occurs and its sex ratio ($\chi^2 = 113.50$, d.f. = 1, $P < 0.001$, $n = 17$; Fig. 3). Species with winged males had a significantly steeper relationship between sex ratio and proportion of fruit in which a species occurs than species with wingless males ($\chi^2 = 90.72$, d.f. = 1, $P < 0.001$; Fig. 3). The predicted intercept for the two lines was not significantly different ($\chi^2 = 0.35$, d.f. = 1, $P > 0.1$). In contrast, there was no significant correlation across species between sex ratio and either combined brood size ($\chi^2 = 0.04$, d.f. = 1, $P > 0.1$) or the estimated average brood size ($\chi^2 = 2.36$, d.f. = 1, $P > 0.1$). Together the proportion of a fruit in which a species occurs and whether or not a species is winged explained 94.5% of the variation in sex ratio across species. Models which also contained the combined brood size or the estimated average brood size explained an extra 0.02% and 1.1% of the variance respectively.

Considering the winged and wingless species separately there were significant positive correlations between the average sex ratio of a species and the proportion of fruit in which it occurred (wingless: $\chi^2 = 129.50$, d.f. = 1, $P < 0.001$, $n = 10$; winged: $\chi^2 = 34.55$, d.f. = 1, $P < 0.001$, $n = 7$). In contrast, there were no significant correlations across species between sex ratio and either combined brood size (wingless: $\chi^2 = 1.91$, d.f. = 1, $P > 0.1$; winged: $\chi^2 = 0.63$, d.f. = 1, $P > 0.1$).

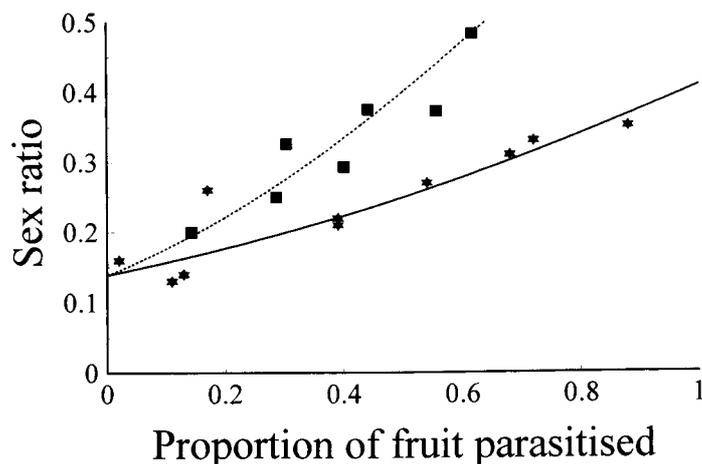


Fig. 3. The relationship between the sex ratio of a species and the proportion of fruit in which that species occurs (proportion of fruit parasitised). The square points represent species with winged males, while the star points represent species with wingless males. The dashed and solid curves represent the best fit lines for the species with winged and wingless males respectively.

Table 1. Characteristics of the non-pollinating fig wasp species studied. Species with a higher proportion of fruit in a crop parasitised were likely to have a higher number of foundresses contributing to each fruit. Species with winged males were able to mate with females away from their natal patch. *Idarnes* represents species from the *flavicollis* and *carne* species groups, while *Idarnes (i)* represents species from the *incerta* species group.

Fig species <i>Ficus</i>	Wasp genera	Males winged/ wingless	Number of trees sampled	Combined brood size	Proportion parasitised fruit	Sex ratio
<i>perforata</i>	<i>Idarnes</i>	wingless	2	15.0	0.02	0.16
<i>nr. trigonata</i>	<i>Idarnes</i>	wingless	2	5.9	0.11	0.13
<i>bullenci</i>	<i>Idarnes</i>	wingless	2	8.8	0.14	0.13
<i>columbrinae</i>	<i>Idarnes</i>	wingless	1	9.8	0.17	0.26
<i>dugandi</i>	<i>Idarnes</i>	wingless	2	22.5	0.39	0.22
<i>citrifolia</i>	<i>Idarnes</i>	wingless	5	20.0	0.39	0.21
<i>costaricana</i>	<i>Idarnes</i>	wingless	2	30.6	0.54	0.27
<i>obtusifolia</i>	<i>Idarnes</i>	wingless	3	53.6	0.68	0.31
<i>popenoei</i>	<i>Idarnes</i>	wingless	2	100.3	0.72	0.33
<i>trigonata</i>	<i>Idarnes</i>	wingless	4	126.4	0.88	0.35
<i>costaricana</i>	<i>Aepocerus</i>	winged	1	4.0	0.29	0.25
<i>obtusifolia</i>	<i>Aepocerus</i>	winged	1	10.9	0.30	0.33
<i>trigonata</i>	<i>Aepocerus</i>	winged	2	8.6	0.56	0.37
<i>dugandi</i>	<i>Aepocerus</i>	winged	2	6.7	0.62	0.48
<i>dugandi</i>	<i>Idarnes (i)</i>	winged	2	8.8	0.14	0.20
<i>citrifolia</i>	<i>Idarnes (i)</i>	winged	2	5.6	0.44	0.38
<i>dugandi</i>	<i>Physothorax</i>	winged	2	3.5	0.40	0.29

If the overall sex ratio, and proportion of sibmating are known for a species with winged males then equation (7) of Greeff (1995) allows an estimate of the amount of mating away from the natal fruit to be obtained. We can estimate the degree of sibmating by assuming that ovipositing females were distributed at random amongst fruit (Hamilton, 1979; Greeff, 1995; equation 1 of Appendix). Making this assumption, our estimates of the proportion of mating that occurs away from the natal patch range from 0.43 (*Idarnes (incerta)* sp. in *F. dugandi*) to 0.95 (*Aepocerus* sp. in *F. dugandi*).

The phylogeny given by Machado et al. (1996) allowed the calculation of nine independent contrasts. For both the Grafen (1989) and Pagel (1992) methods of assigning branch lengths there was a significant positive relationship between the sex ratio contrasts and the proportion of fruit parasitized contrasts. This was the case whether analysis was carried out with a regression fixed through the origin (Grafen: $F_{(1,8)} = 7.63$, $P < 0.05$, $n = 9$, Fig. 4; Pagel: $F_{(1,8)} = 7.42$, $P < 0.05$, $n = 9$) or a sign test (Grafen and Pagel: $P = 0.02$).

Discussion

Across 17 species of non-pollinating fig wasps the average sex ratio of a species was positively correlated with the proportion of fruit in which that species occurred

(Fig. 3). This same relationship was found within each genus/species group. In addition, species with winged males had less female biased sex ratios than species with wingless males that occurred in a similar proportion of fruit (Fig. 3). Given our assumptions, these results support the theoretical predictions that the average sex ratio should be positively correlated with the number of females which oviposit in a patch, and that increased mating away from the natal patch leads to relatively less female biased sex ratios due to lowered LMC.

These results were supported by a formal comparative analysis using independent contrasts. The difference in the sex ratio of two species/taxa was positively correlated with the difference in the average proportion of fruit in which each of those two species/taxa occurred. In addition, the position of two outlying data points (marked *a* and *b* on Fig. 4) supports the suggestion that species with winged males had less female biased sex ratios than species with wingless males. Point *a* was the only comparison in which the taxon which occurs in the highest proportion of fruit had a more female biased sex ratio. This point was a comparison of the *Idarnes* and *Idarnes (incerta)* with the *Aepocerus*. In point *b* there was little difference in the average sex ratio of the taxa despite a considerable difference in the proportion of fruit in which they occur. This point was a comparison of the *Idarnes* with the *Idarnes (incerta)*. In both these cases (*a* and *b*) the taxa consisting entirely of winged species had a less female biased sex ratio than we would have expected on the basis of the other comparisons. More generally, within fig wasps, wingless males have evolved on at least seven separate occasions (Cook et al., 1997), and so future work could examine this question in more detail.

Hamilton (1979) has previously shown that non-pollinating fig wasp species with winged males tend to have less female biased sex ratios than those species with wingless males. However, the winged males of the species studied by Hamilton were

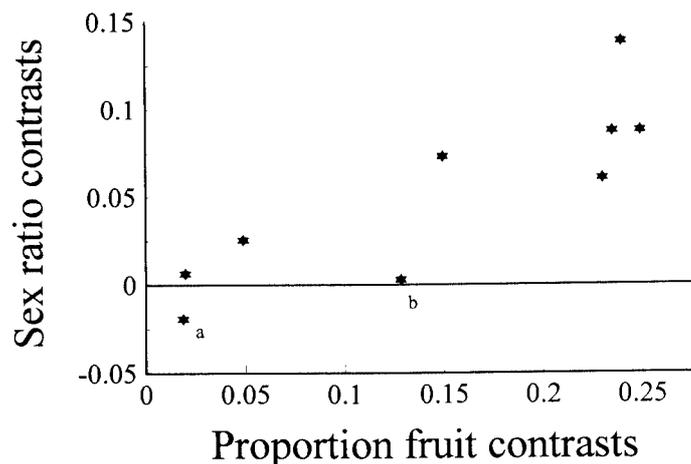


Fig. 4. The relationship between the sex ratio contrasts and the proportion of fruit in which a species occurs contrasts. Point *a* represents a comparison of the *Idarnes* and *Idarnes (incerta)* with the *Aepocerus*. Point *b* represents a comparison of the *Idarnes* with the *Idarnes (incerta)*.

not observed to mate within their own fig. The distinction between species with winged and wingless males was therefore one between local mating and outbreeding species. In contrast, the winged males of the species studied here actively searched within their natal fruit for females with whom they mated before leaving to search for females from other fruit (see background biology). The distinction between our species with winged and wingless males was therefore one between local mating partially local mating species.

The variation in the average sex ratios of the different species was in qualitative agreement with the predictions made by our model (Fig. 1). However, the observed sex ratios of the species with wingless males were all less female biased than we would have expected if ovipositing females were randomly distributed amongst the fruit of trees. A possible explanation for this is that ovipositing females were not distributed at random. If females aggregated at certain fruit then the average number of females ovipositing in a fruit would be higher for a given proportion of fruit parasitised (Fig. 1a) and we would expect less female biased sex ratios (Fig. 1b). Females would be expected to aggregate if fruit differed in their quality as oviposition sites, and if females could detect this variation, or if different fruit were at the correct stage for oviposition at different times. An alternative, and non-exclusive explanation is suggested by the extension of our model to incorporate the constraint imposed by finite brood size (Fig. 2). Nonetheless, a consequence of both of these possibilities is that our estimates of the amount of mating away from the natal patch, in species with winged males, are likely to be overestimates. The extent of this possible overestimation is shown if we use the same assumptions (Greeff, 1995) to calculate the predicted amount of mating away from the natal patch in species with wingless males. Instead of the predicted 0, we obtain values ranging from 0.28 (*Idarnes* sp. from *F. bullenei*) to 0.49 (*Idarnes* sp. from *F. popenoei*).

The average sex ratios of the species may represent different fixed strategies, or different manifestations of facultative strategies. Population densities of most of these species fluctuate enormously in both space and time (Herre, 1989; West et al., 1996), and so there is likely to have been selection for facultative strategies (Herre, 1987; Herre et al., 1997). Indeed, given such fluctuations if individuals use only fixed strategies we would expect considerably more scatter in our data (Fig. 3). Further suggestive evidence for facultative strategies is found if we examine the sex ratios produced in the fruit crops of trees, for the species in which more than two crops were sampled (The *Idarnes* species from *F. citrifolia*, *F. obtusifolia* and *F. trigonata*). In all three species there was a perfect rank order correlation between the sex ratio of a sample and the proportion of fruit in which they occurred (Fig. 5). This is the same pattern that we observed between species, and suggests that females are facultatively producing less biased offspring sex ratios in crops where local densities of females are higher, and higher average numbers of females are ovipositing in each fruit.

Future work on non-pollinating fig wasps could examine experimentally whether females facultatively adjust the sex ratio of their offspring, and if so, the ways in which they judge the number of females ovipositing in a patch. A possible experiment would be to determine how a female adjusts the sex ratio of her

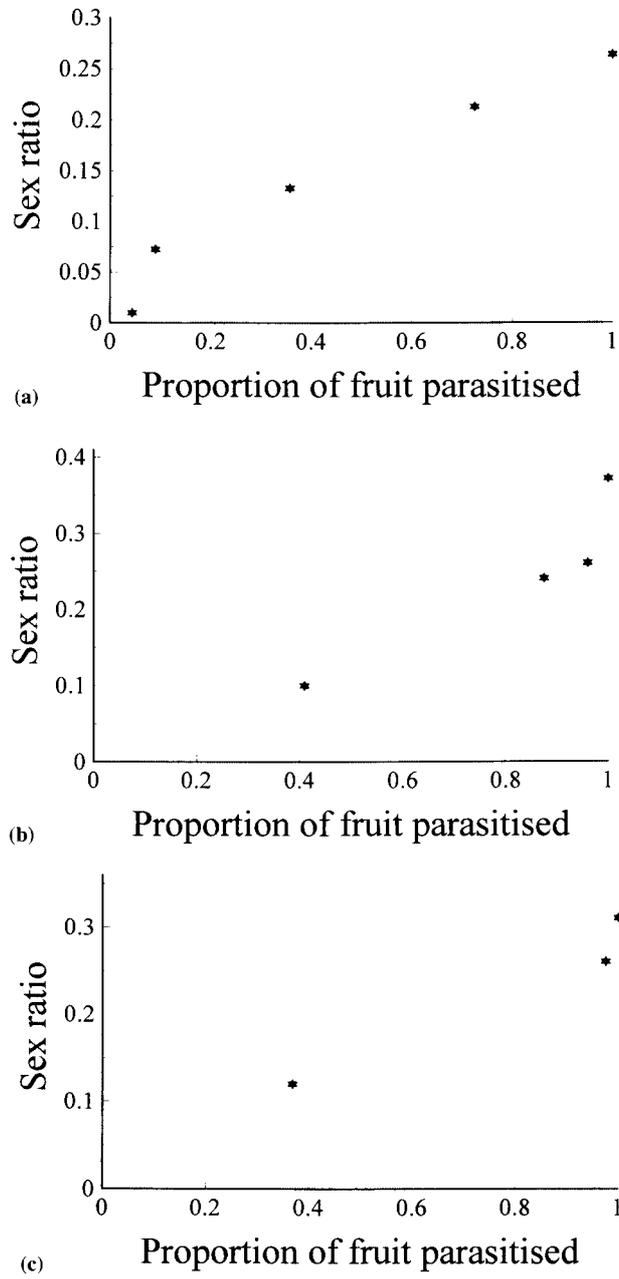


Fig. 5. The relationship across crops between the sex ratio of a single species and the proportion of fruit in which that species occurred. (a) *Idarnes* sp. from *F. citrifolia*. (b) *Idarnes* sp. from *F. trigonata*. (c) *Idarnes* sp. from *F. obtusifolia*.

offspring in response to confinement with various numbers of females before and/or during oviposition in a fruit. Alternatively molecular techniques, such as measuring allozyme polymorphism (see Molbo and Parker, 1996), could be used to examine how females adjust their offspring sex ratio in response to population structure in the field. We believe that understanding how females assess foundress number in natural conditions may help explain why the results of laboratory experiments with parasitoids often have not precisely matched the predictions of theory (e.g. Waage and Lane, 1984; Orzack et al., 1991; King and Skinner, 1991; see also Godfray, 1994). For example, in many species where females oviposit sequentially at a patch, an individual female may be unable to determine precisely the foundress number. In this case females may judge foundress number at least partially by their average contact with other females over time (e.g. Strand, 1988), and so the circumstances in which a female is kept prior to an experiment will effect the sex ratio that she produces in that experiment.

Acknowledgements

We thank Austin Burt, Steve Compton, James Cook, Kate Flanagan, Charles Godfray, Egbert Leigh, Carlos Machado, Dan Racey, Jack Werren, and in particular Jaco Greeff for useful discussion and comments on the manuscript. Simon van Noort and Jean-Yves Rasplus kindly helped identify the wasp species. Carlos Machado kindly provided the molecular phylogeny before its publication. Funding was provided by NERC, STR1 and the BSSRC.

References

- Boucek, Z. 1988. Australian Chalcidoidea (Hymenoptera). A biosystematic revision of genera of fourteen families, with a reclassification of species. CAB International, U.K.
- Boucek, Z. 1993. The genera of chalcidoid wasps from *Ficus* fruit in the new world. *J. Nat. Hist.* 27: 173–217.
- Bronstein, J. L. 1991. The nonpollinating wasp fauna of *Ficus pertusa*: exploitation of a mutualism? *Oikos* 61: 175–186.
- Burt, A. 1989. Comparative methods using phylogenetically independent contrasts. *Oxf. Surv. Evol. Biol.* 6: 33–53.
- Charnov, E. L. 1982. The theory of sex allocation. Princeton University Press, U.S.A.
- Compton, S. G. and B. A. Hawkins. 1992. Determinants of species richness in southern African fig wasp assemblages. *Oecologia* 91: 68–74.
- Cook, J. M. and S. A. Power. 1996. Effects of within-tree flowering asynchrony on the dynamics of seed and wasp production in an Australian fig species. *J. Biogeogr.* 23: 487–494.
- Cook, J. M., S. G. Compton, E. A. Herre and S. A. West. 1997. Alternative mating tactics and extreme male dimorphism in fig wasps. *Proc. R. Soc. Lond. B* 264: 747–754.
- Crawley, M. J. 1993. GLIM for Ecologists. Blackwell Scientific Publications, Oxford, U.K.
- Felsenstein, J. 1985. Phylogenies and the comparative method. *Am. Nat.* 125: 1–15.
- Frank, S. A. 1984. The behavior and morphology of the fig wasps *Pegoscapus ascutus* and *P. jimenezii*: descriptions and suggested behaviors for phylogenetic studies. *Psyche* 91: 289–307.
- Frank, S. A. 1985. Hierarchical selection theory and sex ratios. II. On applying the theory, and a test with fig wasps. *Evolution* 39: 949–964.
- Frank, S. A. 1986. Hierarchical selection theory and sex ratios. I. General solutions for structured populations. *Theor. Pop. Biol.* 29: 312–342.

- Garland, T., Jr., P. H. Harvey and A. R. Ives. 1992. Procedures for the analysis of comparative data using phylogenetically independent contrasts. *Systematic Biology* 41: 18–32.
- Godfray, H. C. J. 1994. *Parasitoids: Behavioural and evolutionary ecology*. Princeton University Press, U.S.A.
- Godfray, H. C. J. and J. M. Cook. 1997. Mating systems of parasitoid wasps. *In* J. Choe and B. Crespi (Eds.), *Social Competition and Cooperation in Insects and Arachnids. Volume I: The Evolution of Mating Systems*. Princeton University Press, U.S.A.
- Grafen, A. 1989. The phylogenetic regression. *Phil. Trans. Roy. Soc. B* 326: 119–156.
- Griffiths, N. T. and H. C. J. Godfray. 1988. Local mate competition, sex ratio and clutch size in bethylid wasps. *Behav. Ecol. Sociobiol.* 22: 211–217.
- Greeff, J. M. 1995. Offspring allocation in structured populations with dimorphic males. *Evol. Ecol.* 9: 550–558.
- Greeff, J. M. 1997. Offspring allocation in externally ovipositing fig wasps with varying clutch size and sex ratio. *Behav. Ecol.* 8: 500–505.
- Hamilton, W. D. 1967. Extraordinary sex ratios. *Science* 156: 477–488.
- Hamilton, W. D. 1979. Wingless and fighting males in fig wasps and other insects, pp 167–220. *In* M. S. Blum and N. A. Blum (Eds.), *Sexual Selection and Reproductive Competition in Insects*. Academic Press, London, U.K.
- Hardy, I. C. W. 1994. Sex ratio and mating structure in the parasitoid Hymenoptera. *Oikos* 69: 3–20.
- Hardy, I. C. W. and J. M. Cook. 1995. Brood sex ratio variance, developmental mortality and virginity in a gregarious parasitoid wasp. *Oecologia* 103: 162–169.
- Harvey, P. H. and M. Pagel. 1991. *The Comparative Method in Evolutionary Biology*. Oxford University Press, Oxford, U.K.
- Herre, E. A. 1985. Sex ratio adjustment in fig wasps. *Science* 228: 896–898.
- Herre, E. A. 1987. Optimality, plasticity, and selective regime in fig wasp sex ratios. *Nature* 329: 627–629.
- Herre, E. A. 1989. Coevolution of reproductive characteristics in twelve species of new world figs and their pollinator wasps. *Experientia* 45: 637–647.
- Herre, E. A. 1996. Implications of studies on a community of Panamanian figs: emerging patterns and future directions. *J. Biogeogr.* 23: 593–607.
- Herre, E. A. and S. A. West. 1997. Conflict of interest in a mutualism: documenting the elusive fig wasp/seed trade-off. *Proc. Roy. Soc. Lond. Ser. B* 264: 1501–1507.
- Herre, E. A., C. A. Machado, E. Bermingham, J. D. Nason, D. M. Windsor, S. S. MacCafferty, W. Van Houten and K. Bachmann. 1996. Molecular phylogenies of figs and their pollinator wasps. *J. Biogeogr.* 23: 521–530.
- Herre, E. A., S. A. West, J. M. Cook, S. G. Compton and F. Kjellberg. 1997. Fig wasp mating systems: pollinators and parasites, sex ratio adjustment and male polymorphism, population structure and its consequences, pp 226–239. *In* J. Choe and B. Crespi (Eds.), *Social Competition and Cooperation in Insects and Arachnids. Volume I: The Evolution of Mating Systems*. Princeton University Press, U.S.A.
- Ikawa, T., M. Shimada, H. Matsuda and H. Okabe. 1993. Sex allocation of parasitic wasps: local mate competition, dispersal before mating and host quality variation. *J. evol. biol.* 6: 79–94.
- King, B. H. 1993. Sex ratio manipulation by parasitoid wasps, pp 418–441. *In* D. L. Wrensch and M. A. Ebbert (Eds.), *Evolution and Diversity of sex ratio in insects and mites*. Chapman and Hall, New York, U.S.A.
- King, B. H. and S. W. Skinner. 1991. Sex ratio in a new species of *Nasonia* with fully winged males. *Evolution* 45: 225–228.
- Leigh, E. G., E. A. Herre and E. A. Fischer. 1985. Sex allocation in animals. *Experientia* 41: 1265–1276.
- Machado, C. A., E. A. Herre and E. Bermingham. 1996. Molecular phylogenies of fig pollinating and non-pollinating wasps and the implications for the origin and evolution of the fig-fig wasp mutualism. *J. Biogeogr.* 23: 531–542.
- McCullagh, P. and J. A. Nelder. 1983. *Generalized Linear Models*. Chapman and Hall, London, U.K.

- Molbo, D. and E. D. Parker. 1996. Mating structure and sex ratio variation in a natural population of *Nasonia vitripennis*. *Proc. R. Soc. Lond. B* 263: 1703–1709.
- Morgan, D. J. W. and J. M. Cook. 1994. Extremely precise sex ratios in small clutches of a bethylid wasp. *Oikos* 71: 423–430.
- Nadel, H. and R. F. Luck. 1992. Dispersal and mating structure of a parasitoid with a female-biased sex ratio: implications for theory. *Evol. Ecol.* 6: 270–278.
- Nagelkerke, C. J. 1994. Simultaneous optimization of egg distribution and sex allocation in a patch-structured population. *Am. Nat.* 144: 262–284.
- Nagelkerke, C. J. 1996. Discrete Clutch Sizes, Local Mate Competition, and the Evolution of Precise Sex Allocation. *Theo. Popul. Bio.* 49: 314–343.
- Nagelkerke, C. J. and I. C. W. Hardy. 1994. The influence of developmental mortality on optimal sex allocation under local mate competition. *Behav. Ecol.* 5: 401–411.
- Nunney, L. and R. F. Luck. 1988. Factors influencing the optimal sex ratio in structured populations. *Theor. Popul. Biol.* 33: 1–30.
- Orzack, S. H., E. D. Parker and J. Gladstone. 1991. The comparative biology of genetic variation for conditional sex ratio behaviour in a parasitic wasp, *Nasonia vitripennis*. *Genetics* 127: 583–599.
- Pagel, M. D. 1992. A method for the analysis of comparative data. *J. Theo. Biol.* 156: 431–442.
- Purvis, A. and A. Rambaut. 1995. Comparative analysis by independent contrasts (CAIC): an Apple Macintosh application for analysing comparative data. *CABIOS* 11: 247–251.
- Read, A. F., A. Narara, S. Nee, A. E. Keymer and K. P. Day. 1992. Gametocyte sex ratios as indirect measures of outcrossing rates in malaria. *Parasit. Today* 104: 387–395.
- Read, A. F., M. Anwar, D. Shutler and S. Nee. 1995. Sex allocation and population structure in malaria and related protozoa. *Proc. R. Soc. Lond. B* 260: 359–363.
- Ridley, M. 1988. Why not to use species in comparative tests. *J. Theor. Biol.* 136: 361–364.
- Shutler, D., G. F. Bennett and A. Mullie. 1995. Sex proportions of *Haemoproteus* blood parasites and local mate competition. *Proc. natn. Acad. Sci. U.S.A.* 92: 6748–6752.
- Taylor, P. D. 1993. Female biased sex ratios under local mate competition: an experimental confirmation. *Evol. Ecol.* 7: 306–308.
- Uyenoyama, M. K. and B. O. Bengtsson. 1982. Towards a genetic theory for the evolution of the sex ratio III. Parental and sibling control of brood investment ratio under partial sib-mating. *Theory. Popul. Biol.* 22: 43–68.
- Waage, J. K. and J. A. Lane. 1984. The reproductive strategy of a parasitic wasp. II. Sex allocation and Local Mate Competition in *Trichogramma Evanesces*. *J. Anim. Ecol.* 53: 417–426.
- Werren, J. H. 1987. Labile sex ratios in wasps and bees. *Bioscience* 37: 498–506.
- Werren, J. H. and G. Simbolotti. 1989. Combined effects of host quality and local mate competition on sex allocation in *Lariophagus distinguendus*. *Evol. Ecol.* 3: 203–213.
- West, S. A. and E. A. Herre. 1994. The ecology of the New World fig parasitizing wasps Idarnes and implications for the evolution of the fig-pollinator mutualism. *Proc. Roy. Soc. Lond. B* 258: 67–72.
- West, S. A. and E. A. Herre. 1998. Stabilizing selection and variance in fig wasp sex ratios. *Evolution* 52: 475–485.
- West, S. A., E. A. Herre, D. W. Windsor and P. R. S. Green. 1996. The ecology and evolution of the New World non-pollinating fig wasp communities. *J. Biogeogr.* 23: 447–458.
- West, S. A., E. A. Herre, S. G. Compton, H. C. J. Godfray and J. M. Cook. 1997. A comparative study of virginity in fig wasps. *Anim. Behav.* 54: 437–450.
- West, S. A., S. G. Compton, S. L. Vincent, E. A. Herre and J. M. Cook. 1998. Virginity in haplodiploid populations: a comparison of estimation methods. *Ecol. Ent.* 23: 207–210.
- Wiebes, J. T. 1995. Agaonidae (Hymenoptera Chalcidoidea) and *Ficus* (Moraceae): fig wasps and their figs, xv (Meso-American Pegoscapus). *Proc. Kon. Ned. Akad. v. Wetensch* 98: 647–653.

Windsor, D. M., D. W. Morrison, M. A. Estribi and B. de Leon. 1989. Phenology of fruit and leaf production by 'strangler' figs on Barro Colorado Island, Panama. *Experientia* 45: 647–653.

Received 9 June 1997;
revised 24 July 1997;
accepted 16 September 1997.

Appendix

Suppose that ovipositing females are randomly distributed between fruit, with each female visiting each fruit with an equal probability (i.e. poisson distribution). In this case the average number of females ovipositing in a fruit that contains that species (defined as \bar{n}) is

$$\bar{n} = \frac{-\ln(1 - P)}{P}, \quad (1)$$

where P is the proportion of fruit in which that species occurs (Line I of Fig. 1a). The numerator represents the average number of females ovipositing in all the fruit. This includes those in which no females oviposited and so must be divided by P to obtain \bar{n} .

If n females oviposit in a fruit, then assuming haplodiploid genetics, equal brood sizes, and that all mating takes place within the natal fruit, the optimal offspring sex ratio (defined as r) is

$$r = \frac{(n - 1)(2 - s)}{n(4 - s)}, \quad (2)$$

where s is the proportion of sibmating in the population (Frank, 1985; Herre, 1985; Werren, 1987). If all females produce the same sex ratio then the optimal sex ratio to produce is that associated with the average number of females ovipositing in a fruit (\bar{n} ; Nunney and Luck, 1988). Given this further assumption, and by substituting equation 1 into equation 2, we are able to predict the relationship between the sex ratio of a species and the proportion of fruit in which it occurs (Line 1 of Fig. 1b).

Let us now assume that females aggregate at certain fruit (i.e. negative binomial distribution). In this case the average number of females ovipositing in a fruit that contains that species (\bar{n}) is

$$\bar{n} = \frac{k((1 - P)^{-1/k} - 1)}{P}, \quad (3)$$

where k is the aggregation parameter (Lines II and III of Fig. 1a). Low values of k describe high degrees of aggregation. Substituting equation 3 into equation 2 we are able to predict the relationship between the sex ratio of a species and the proportion of fruit in which it occurs (Lines II and III of Fig. 1b; see also Read et al., 1995).

We now examine the consequences of partially relaxing the assumption that females can produce any offspring sex ratio, in three different ways (see also Nagelkerke, 1996;

West et al., 1997). For all cases we assume that females are randomly distributed between the fruit, visiting each fruit with an equal probability. We also assume that there is a minimum possible sex ratio that a female can produce, as a result of the constraint imposed by small brood size. The minimum sex ratio is set as 0.14, the predicted intercept of the best fit lines through the data in Figure 3, and corresponds to one male in a brood containing seven individuals.

Firstly, we assume that all females produce the same sex ratio in each fruit, as above. However, if the predicted optimal sex ratio (from equation 2) is below 0.14 then a value of 0.14 is assumed (Line a of Fig. 2). For the second and third case we assume that females facultatively adjust their sex ratio according to the number of females that oviposit in each individual fruit (n). In n females oviposit in a fruit and $n > 1$ then they produce the sex ratio predicted in equation 2. However, if only one female oviposits in a patch then she produces a sex ratio of 0.14 and not 0. The constraint of small brood size is therefore assumed to only affect the sex ratio produced when one female oviposits in a fruit ($n = 1$).

In the second case we assume that the total number of offspring that develop in a fruit is independent of the number of females that oviposit in it. Given this assumption we would expect to observe an overall sex ratio (defined as SR) of

$$SR = \frac{0.14P_0 + \left(\sum_{x=2}^{\infty} P_x r(x) \right)}{1 - P_0}, \quad (4)$$

where P_x is the proportion of fruit in which x females oviposit. The expected proportion of fruit in which x females oviposit is

$$P_x = \frac{e^{\ln(1-p)} (-\ln(1-p))^x}{x!} \quad (5)$$

(line b of Fig. 2).

The only difference in the third case is that we assume that the number of offspring developing in a fruit is linearly dependent upon the number of females that oviposit in the fruit. For example, twice as many offspring will develop in a fruit in which two females oviposit than in a fruit in which one female oviposits. Given this assumption we would expect to observe an overall sex ratio (SR) of

$$SR = \frac{0.14P_0 + \left(\sum_{x=2}^{\infty} P_x x r(x) \right)}{\sum_{x=1}^{\infty} P_x x} \quad (6)$$

(Line c of Figure 2).