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## Superparasitism of larval hosts by the walnut fly, *Rhagoletis juglandis*, and its implications for female and offspring performance

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**Abstract** The oviposition-preference–offspring-performance hypothesis predicts that female insects should prefer to deposit clutches on or in hosts that maximize offspring performance. An important assumption behind this prediction is that female fitness is tightly correlated with the fitness of any one offspring. In this study, we evaluate offspring performance in the walnut fly, *Rhagoletis juglandis* Cresson (Diptera: Tephritidae), in relation to a previously described oviposition preference for previously exploited host fruit. In particular, we examined how superparasitism of walnut hosts influences offspring survival and weight at pupation under field conditions. We found that superparasitism was common and that increases in larval densities within fruit were associated with reduced larval survival and weight at pupation. In a laboratory experiment, female size was correlated with lifetime fecundity. In this system, oviposition preference is therefore negatively, not positively, correlated with offspring performance. We argue that patterns of female preference in this system reflect direct benefits to females

that are traded off against costs in terms of offspring fitness. Because female fitness is a product not only of offspring quality but also of the total number of offspring produced, female walnut flies may be optimizing their fitness by producing many less fecund offspring. Studies examining the preference-performance hypothesis should consider the reproductive conflicts between parents and offspring as potential factors that influence the congruence between parental preference and offspring performance.

**Keywords** Larval competition · Oviposition-preference–offspring-performance · Parent-offspring conflict · *Rhagoletis* · Reproductive trade-offs · Superparasitism

### Introduction

The oviposition-preference–offspring-performance hypothesis was proposed to explain patterns of host specificity in herbivorous insects (Jaenike 1978; Thompson 1988; Mayhew 1997). The hypothesis states that, in insects that utilize discrete host resources or environments, and in which progeny are limited in their ability to disperse to other hosts, females should be under strong selection to choose hosts that are optimal for larval development. As a result of such selection, a female insect's oviposition preference is expected to correspond to patterns of host suitability that optimize larval performance.

In support of the preference–performance hypothesis, a good correspondence between female preference and offspring performance has been found in some systems (Copp and Davenport 1978; Wiklund 1981; Williams 1983; Leather 1985; Craig et al. 1989; Rossi and Strong 1991; Price and Ohgushi 1995; Nylin and Janz 1996). However, in many other systems, the correspondence has been found to be poor or nonexistent (Messina 1982; Karban and Courtney 1987; Valladares and Lawton 1991; Fox 1993; Larsson and Ekbom 1995; reviewed in Mayhew 1997).

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A failure to find a strong correspondence between female preference patterns and offspring performance has sometimes been attributed to physiological constraints that prevent females from making optimal decisions. For instance, ovipositing females may simply be limited, at a sensory level, in their capacity to discriminate between juvenile-suitable and unsuitable hosts (reviewed in Courtney and Kibota 1990; Bernays 2001). Alternatively, a weak correlation between preference and performance may reflect an incomplete measure of performance. For example, hosts associated with high juvenile growth and survival under controlled laboratory conditions may be associated with high levels of juvenile predation or parasitism in the field (Denno et al. 1990; Ballabeni et al. 2001). Finally, the lack of a correspondence between female preference and offspring performance has also been attributed to the recentness of certain insect–host plant associations (Thompson 1988). Given enough time, it is proposed that these insects will evolve the capacity to make optimal host choices.

Less often acknowledged in the female-preference–offspring-performance literature is the possibility that oviposition patterns that maximize parental fitness are not the same as those that maximize the performance of any one offspring (reviewed in Mayhew 1997; Scheirs and De Bruyn 2002). Studies often fail to acknowledge that a female may potentially increase her fitness by increasing the number of offspring she produces over a lifetime (either by increasing her reproductive life span or rate of oviposition), even if this is at the expense of some decrease in offspring performance. Under conditions where juvenile-optimal hosts are less common than other hosts (Etges and Heed 1987; Mayhew 1997) or are associated with higher female mortality (Weisser et al. 1994), for example, a female may opt to search selectively for a juvenile-suboptimal host because it permits her to increase the number of eggs laid over her lifetime (Rausher 1980). Similarly, juvenile-optimal hosts might also be suboptimal female-feeding sites and as such females may choose to oviposit into juvenile-suboptimal hosts that are associated with increases in female performance (Schiers et al. 2000). So long as the gains that a female accrues in terms of numbers of offspring produced more than offset her losses in terms of per capita offspring performance, her decisions may be optimal for herself, yet in conflict with the interests of any one offspring.

The walnut fly, *Rhagoletis juglandis*, the focus of this study, is a specialist tephritid species that utilizes the husks of developing walnut fruit as a larval resource. Like many other tephritid flies, *R. juglandis* engages in host-marking behavior following the deposition of a clutch of eggs into a fruit. Paradoxically, while marking pheromones in *Rhagoletis* species that use other hosts cause arriving females to reject occupied hosts (reviewed in Prokopy 1981; Landolt and Averill 1999), *R. juglandis* and the other walnut-infesting *Rhagoletis* species commonly deposit clutches of eggs into previously exploited hosts (a behavior often referred to as superparasitism), often

depositing numerous clutches into existing oviposition cavities (Papaj 1994; Nufio et al. 2000). Despite possible costs associated with superparasitizing larval hosts, in terms of increased larval competition, walnut flies prefer to lay eggs in previously attacked hosts and actively superparasitize hosts early in the season when unattacked hosts are still available (Lalonde and Mangel 1994; Nufio et al. 2000).

If larvae are competing within host fruit, as they do in congeneric species (Averill and Prokopy 1987), *R. juglandis*' early-season preference for infested fruit would likely be negatively correlated with offspring performance. Alternatively, it is possible that larvae do not compete within fruit but gain some advantage from being deposited together, for example, an advantage due to a sharing of microbial symbionts thought to generate the rot on which larvae feed (Howard et al. 1985; Howard and Bush 1989). In that case, the correlation between preference and performance might be positive, at least over a range of larval densities. In this study, we quantify field patterns of host utilization by the walnut fly, *R. juglandis* (Tephritidae), and examine offspring performance in relation to the females' unusual preference for previously exploited fruit.

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## Materials and methods

### Natural history

*Rhagoletis juglandis* is a member of the walnut-infesting *R. suavis* group (Bush 1966). In southern Arizona, this species is found on the Arizona walnut, *Juglans major* Torr, which is common in montane canyons between 1,200 and 2,700 m. These flies are univoltine and females deposit clutches of ca. 16 eggs ( $\pm$ SE 1.5) (Nufio et al. 2000) after puncturing the fruit surface with their ovipositor. The larval stages feed on the husk of developing fruit, pupate in the soil beneath the natal tree, diapause as pupae through the winter and spring and emerge as adults in mid to late summer.

### Field patterns of superparasitism and offspring performance

In mid June, 1996, five *J. major* trees in lower Garden Canyon in the Huachuca Mountains in Cochise County, Arizona (31°28.69'N, 110°20.53'W; 1,500 m elevation) were selected for study. On each tree, 25–60 fruit that were accessible from ground level were chosen and tagged for the study. The fruit from a given tree used in this study constituted roughly 2–5% of the total fruit yield of that tree.

Walnut flies were first observed on a study tree on 2 July. After 8 July, when the first oviposition punctures on fruit were observed, study trees were censused every 2 days as follows. From 0900 to 1100 hours, tagged fruit within each tree were examined for signs of walnut fly oviposition punctures. The minimum and maximum diameters of each of the recently punctured fruit were recorded with digital callipers. These measurements were later used to estimate the volume of a given walnut, by assuming a walnut was spherical, taking the average of the axes measurements as an estimate of sphere diameter, and then computing fruit volume as  $4/3\pi r^3$ .

To measure the impact of host superparasitism on offspring fitness, we manipulated the period of time over which cohorts of infested fruit were exposed to additional oviposition events. At the end of a census day, newly-infested fruit were grouped according to size and location and then haphazardly assigned to one of three treatments. Fruit in the first treatment were bagged immediately with

thin wedding veil, and thus represented fruit that had been infested that day and/or the previous day and thus this cohort was considered to have been exposed to gravid females for 1–2 days. Fruit placed into the second and third treatments were bagged 2 and 4 days later and thus represented fruit that had been infested and exposed to gravid females for 3–4 and 5–6 days, respectively.

Assessing the precise number of ovipositions on a host was not possible in our study because females add clutches to existing oviposition cavities and it was not possible to distinguish individual clutches deposited within a single cavity. We instead used total number of eggs deposited within a host as an indication of the degree to which hosts were superparasitized. We estimated the number of eggs deposited in a fruit as follows. On each census date, all study fruit not previously bagged were examined, and new punctures on previously infested fruit were circled and dated. After a given oviposition site was at least 6–7 days old, it was removed by excavating a cylinder around the puncture, roughly 6 mm long and 8 mm wide. Because larvae move towards the seed and away from the area surrounding their oviposition sites shortly after hatching, only very rarely were larvae found or noticeably damaged while making this excavation. To keep the fruit from drying out and larvae from prematurely leaving the host fruit, the space previously occupied by the oviposition cylinder was covered with a piece of parafilm over which was placed a 15×20-mm strip of black electrical tape. Excavated oviposition cavities were placed in vials with alcohol and brought to the laboratory where they were dissected and the hatched and unhatched eggs within were counted. The number of hatched eggs was used as an estimate of the number of larvae initially present within a fruit.

After 10 days a given study-fruit was initially punctured on a tree, it was removed, brought to the laboratory and placed individually into an “incubator”. These incubators were stored in a growth chamber set at 30°C and 50% humidity. The incubators consisted of inverted 16-fluid ounce (473-ml) plastic cups into which infested fruit were placed on top of a 4-cm-long by 3.5-cm-wide section of PVC tubing that was inserted into a 3-cm-deep bed of mixed vermiculite and sand. After placing the fruit within the incubators, the incubators were covered by a thin wedding veil mesh that was held in place by a rubber band. The vermiculite/sand layer was periodically kept moist by adding water until day 15, when larvae began to emerge from the fruit. Fruit were placed on PVC tubing to keep them from becoming moldy and from absorbing water. The live and dead pupae associated with a fruit were counted, and live pupae were weighed.

#### Effects of female size on reproductive potential

We conducted the following laboratory experiment to examine the relationship between female size and lifetime fecundity. Adult flies used in each of two replicates of this experiment, which were conducted in separate years, were collected as larvae 1 year previously from fruit collected at the lower Garden Canyon site. Pupae were stored in darkness at 4°C until ca. 4–5 weeks prior to the experiment, when they were removed from cold storage and warmed to 28°C under a 14:10 h light:dark cycle. Approximately 24 h after emergence, a female and male were placed into a clear 16-fluid ounce plastic cup, fitted with a petri dish lid, in which they were provided with ad libitum water, sugar and a yeast hydrolysate and sugar mixture. If the male died before the female, he was replaced with a reproductively mature male that was 10–20 days post-emergence. Fly pairs placed into the rearing cups were stored in a room with a 14:10 h light:dark cycle and a day temperature of 28°C.

Starting on day 1, when females were first placed into the rearing cups, a ripe walnut fruit hung from the top of the cup. All fruit provided to females bore four punctures made with a no. 00 insect pin, placed equal distances apart on the fruit surface. While females actively oviposited into the artificial punctures provided, females in this experiment also commonly deposited clutches into female-made oviposition cavities.

Every 2 days, a new fruit replaced the older fruit. The old fruit was dissected and the eggs deposited within the host over the previous 2 days were counted. After an experimental female died, her size was estimated under a dissecting microscope by measuring the length of the discal medial cell. This wing measure was used as an estimate of female size because previous laboratory experiments showed that it was strongly correlated with other indicators of female size such as thorax and head width and femur length (A. Lachman, unpublished data) and because of the relative ease of using the wing measure as opposed to other indicators. Female size was regressed against the number of days that she lived, the number of days until she laid her first clutch, the size (number of eggs) of her first clutch, the total number of eggs she deposited over a lifetime and the average number of eggs deposited per day that she was alive.

#### Statistical analysis

In order to identify factors that influence offspring survival (from egg deposited to successful pupation), we conducted a multiple regression analysis with percent of total survival as the dependent variable and available fruit volume per egg, number of eggs deposited into a fruit and fruit cohort (number of days fruit were exposed to further oviposition) as the independent variables. We conducted an analogous analysis for percent of larval survival (from egg hatch to successful pupation) but in this analysis we included the available fruit volume per larva, the number of eggs deposited within a fruit, and fruit cohort age.

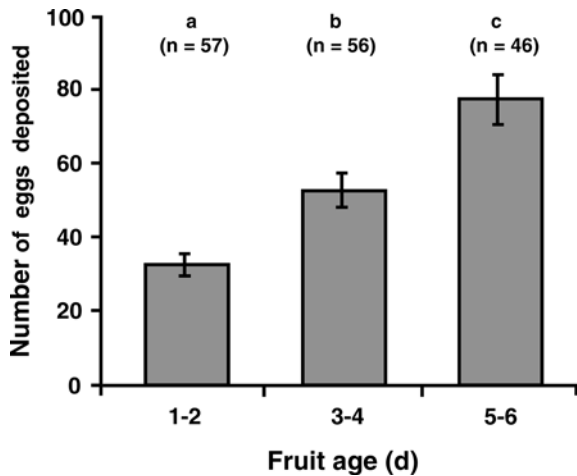
Percent total survival and percent larval survival data were normally distributed, but we log-transformed fruit volume per egg and per larva data to meet normality assumptions. Data for percent egg hatch were bimodally distributed, with one mode at 100%. For this latter variable, we generated a new nominal variable that took a value of 1 when the percent egg hatch was 100% and 0 when it was not. We then conducted a logistic regression analyses on the nominal variable to determine how the number of eggs deposited into a fruit affected percent egg hatch.

In order to identify factors that influenced pupal weight, we conducted a multiple regression analysis with pupal weight as the dependent variable and number of larvae that hatched within a fruit, fruit volume per larva, and fruit cohort as the independent variables. Because pupal weights were not always normally distributed on a per fruit basis, we felt that the median pupal weight of individuals from a given fruit was an appropriate measure of the pupal weights associated with a given fruit. The median pupal weight recorded for each fruit was found to be normally distributed among fruit and as such parametric statistics were used. To determine the factors that influence pupal weight, we conducted a multiple regression analysis with median pupal weight as the dependent variable and available fruit volume per larvae, number of eggs deposited into a fruit and fruit cohort as the independent variables. In all multiple regression analyses we tested for interactions between variables but no interactions were found. All statistical analyses were conducted using JMP-IN statistical software (SAS 2000).

## Results

### Field patterns of superparasitism

That egg density is a reasonable measure of the degree to which hosts were superparasitized is suggested by the pattern of egg density for fruit exposed for varying lengths of time since first infestations ( $F_{2, 153}=14.93$ ,  $P<0.0001$ ; Fig. 1). The number of eggs in a fruit clearly increased as the duration over which fruit were exposed to further



**Fig. 1** Mean ( $\pm$ SE) number of eggs deposited into a fruit as a function of fruit cohort age [the number of days (*d*) a fruit was exposed to oviposition]. Bars sharing the same letter are not significantly different (Tukey's honestly significant difference,  $P < 0.05$ )

superparasitism increased; on average, roughly 20 new eggs were placed into fruit every 2 days.

The number of eggs deposited within a fruit was positively correlated with fruit volume for each cohort ( $r = 0.45$ ,  $n = 57$ ,  $P = 0.0004$ ;  $r = 0.61$ ,  $n = 56$ ,  $P < 0.0001$ ;  $r = 0.57$ ,  $n = 46$ ,  $P < 0.0001$ ; 1–2, 3–4 and 5–6 days cohorts, respectively). Because more eggs were placed into larger fruit, available fruit volume per egg was not significantly different for offspring placed into small vs. large fruit ( $r = -0.005$ ,  $n = 156$ ,  $P = 0.95$ ).

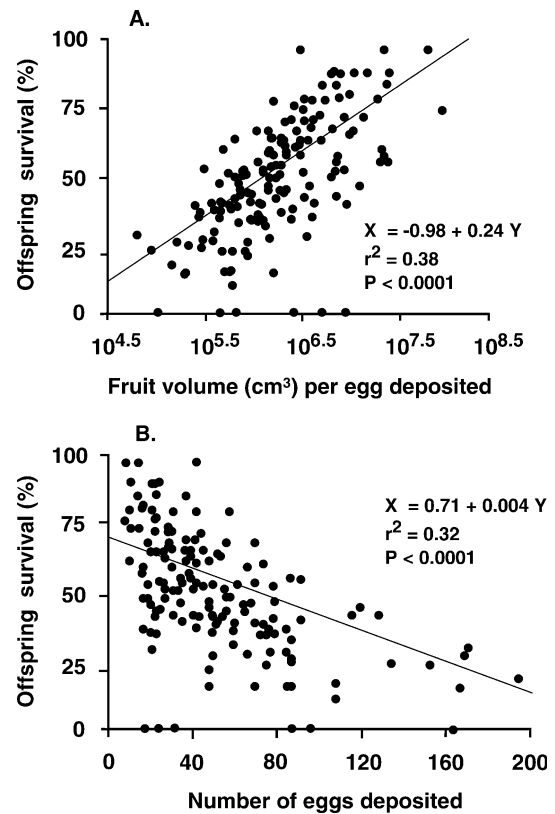
#### Superparasitism and offspring survival

On average,  $88 \pm 0.01\%$  ( $\pm$ SE) eggs deposited into a fruit hatched and of these hatched eggs,  $59 \pm 0.02\%$  of offspring successfully pupated (number of fruit = 156). A multiple regression analysis indicated that both the available fruit volume per deposited egg and the number of eggs deposited into a fruit were significant predictors of the percentage of eggs surviving to pupation (percent of total survival), while fruit cohort did not significantly improve the model (Table 1). Percent of total survival increased with increases in fruit volume per egg (Fig. 2a) and decreased with the number of eggs deposited into a fruit (Fig. 2b). The number of eggs deposited into a fruit may have remained in the model above, due to the effect of egg density on egg hatch. Egg hatch was, for example, found to be negatively related to increases in the number of eggs deposited into a fruit ( $\chi^2 = 15.58$ ,  $df = 1$ ,  $P < 0.0001$ ).

Of the eggs that hatched, percent of larval survival to successful pupation was best explained by the available fruit volume per larva while the number of eggs that hatched and fruit cohort did not significantly improve the model (Table 1). Offspring survival to successful pupation was positively correlated with fruit volume per larva (percent of larval survival =  $-0.68 + 0.20 \log$  fruit volume

**Table 1** Multiple regression results on factor affecting percent of total survival (from eggs deposited to successful pupation), percent of larval survival (from egg hatch to successful pupation) and percentage of pupal weight

Source	<i>df</i>	MS	<i>F</i>	<i>P</i>
<b>Total percent survival</b>				
Log fruit volume per egg	1	0.5135	17.11	<0.0001
Number of eggs deposited	1	0.1486	4.95	0.003
Fruit cohort age	2	0.01	0.13	0.88
<b>Percent of larval survival from hatching to successful pupation</b>				
Log fruit volume per larva	1	0.3128	9.54	0.0024
Number of eggs deposited	1	0.0654	1.99	0.38
Fruit cohort age	2	0.0631	0.96	0.16
<b>Median pupal weight</b>				
Log volume per larva	1	6634.91	40.21	<0.0001
Number of eggs hatched	1	694.01	4.21	0.003
Fruit cohort age	2	0.01	0.13	0.88

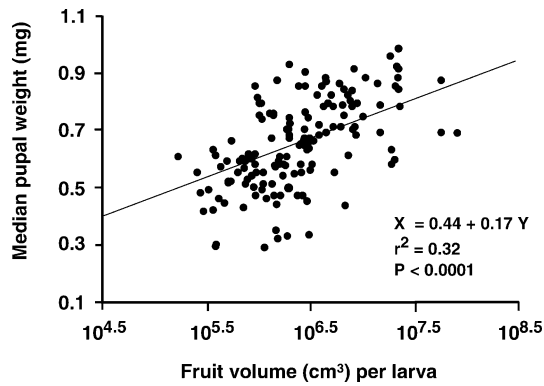


**Fig. 2** Percent of total survival of offspring as a consequence of (a) available fruit volume per egg deposited within a fruit, and (b) the number of eggs deposited within a fruit

per larva). Offspring survival did not peak at intermediate egg or larval densities.

#### Superparasitism and pupal weight

In general, larvae emerging from infested fruit that were only exposed to female reuse for 1–2 days weighed significantly more than individuals from fruit that were



**Fig. 3** Relationship between median pupal weight of offspring as a function of the available fruit volume per larva

exposed to further reuse for 3–4 and 5–6 days ( $F_{2,174}=11.63$ ,  $P<0.0001$ ). There was no difference detected between the pupal weights of fruit exposed to female reuse for 3–4 or 5–6 days ( $t=11.63$ ,  $n=115$ ,  $P=0.23$ ). A multiple regression analysis showed that the median weight of viable pupae was best explained by fruit volume per larva and the number of eggs that hatched within a fruit (Table 1). Median pupal weight increased with increases in available fruit volume per larva (Fig. 3) but decreased with increases in the number of eggs hatching within a fruit.

#### Effects of offspring size on adult fitness characteristics

Although females used in the two replicates of our adult fitness study did not differ significantly in size (average discal medial cell length  $1.56\text{ mm}\pm 0.02$ ;  $F_{1,88}=0.43$ ,  $P=0.51$ ) or average lifespan ( $25.8\text{ days}\pm 1.40$ ;  $F_{1,83}=0.04$ ,  $P=0.84$ ), they nevertheless differed significantly in number of eggs they produced ( $126\pm 13.47$  and  $64.20\pm 11$  for the 1997 and 1999 samples, respectively;  $F_{1,83}=12.63$ ,  $P=0.0006$ ). In both the 1997 and 1999 replicates, there was a significant relationship between our index of body size and the size of the first deposited clutch, with larger females laying relatively larger first clutches (Table 2). Furthermore, in both replicates larger females also deposited significantly more eggs than did smaller ones. Larger females appeared to lay more eggs during the course of these experiments not because they lived longer than smaller females, but because they deposited significantly more eggs per day (Table 2).

## Discussion

Our study found that the walnut fly *R. juglandis* commonly superparasitizes hosts in the field and that, measured in terms of offspring survival and weight at pupation, superparasitism reduces offspring performance. The negative effects of superparasitism on offspring weight were further shown in laboratory assays to translate into a reduction in expected female lifetime fecundity.

**Table 2** Independent linear regression analyses summarizing factors correlated with female size for both the 1997 and 1999 replicates, respectively. All significant factors were positively correlated with female size (medial discal cell length)

	1997 Replicate			1999 Replicate		
	<i>n</i>	<i>r</i> <sup>2</sup>	<i>F</i>	<i>n</i>	<i>r</i> <sup>2</sup>	<i>F</i>
Days till first clutch deposited	33	0.00	0.09	46	0.30	19.01***
Size of first clutch	33	0.31	13.78**	46	0.26	16.76**
Total eggs deposited	34	0.17	6.55*	51	0.29	19.70***
Lifespan	34	0.03	1.12	51	0.00	0.01
Number of eggs produced per day	34	0.22	8.91**	51	0.30	19.87***

\* $P<0.05$ , \*\* $P<0.001$ , \*\*\* $P<0.0001$

Thus it appears that whereas the preference–performance hypothesis would predict that female walnut flies should avoid superparasitizing larval hosts so as to maximize both larval and female fitness, female walnut flies instead prefer to deposit eggs in hosts associated with low offspring performance. If females behave in a manner that improves their own fitness, our results imply that, with respect to superparasitism, female fitness and per capita larval performance are negatively, not positively, correlated.

#### Preference for previously exploited hosts

An important point to emphasize with respect to the preference–performance hypothesis in this system is that females are not simply superparasitizing hosts because they cannot discriminate between hosts that have been previously exploited vs. those that have not been previously exploited. In previous experiments, gravid females were not only found to selectively visit fruit containing artificial punctures, these females also appeared to be depositing clutches directly into the punctures (Papaj 1994; Lalonde and Mangel 1994). In this study, superparasitism of walnut hosts by *R. juglandis* was found to be common; females placed nearly 20 new eggs into a given fruit every 2 days. Another important point to make is that *R. juglandis* females, like other fruit flies (Landolt and Averill 1999), also deposit a marking pheromone on the fruit surface following clutch deposition. In *R. juglandis*, this mark has been found to deter superparasitism, with the degree of deterrence being directly proportional to the amount of time that previous females marked the fruit (Nufio and Papaj 2004). In this and a previous study (Nufio et al. 2000), females as a whole also appear to be allocating more eggs to larger fruit. Hence, despite superparasitizing fruit to a significant degree, females nevertheless behave as though doing so is associated with some costs and as if they were rejecting hosts associated with high costs. These costs, as determined in our study, are expressed in terms of a decrease in per capita offspring survival and fitness.

## Offspring performance as a consequence of superparasitism

### *Superparasitism and offspring survival*

When we examined the components of offspring survival affected by superparasitism, we found that total survival declined with egg density and increased with available fruit volume per egg (Table 1). To our surprise, percent egg hatch in a fruit declined with egg density. The mechanism underlying this latter result is unclear. Possibly, newly hatched larvae spoil the cavity environment for eggs yet to hatch. It is also possible that females damage previously laid eggs while adding eggs to an existing cavity. Alternatively, the pattern in egg density may be confounded with changes that occur in the fruit over the time it takes eggs to accumulate. Such changes could be induced by egg deposition or be independent of it.

Increases in the number of eggs in a fruit were also associated with reduced survival after hatching (Table 1). This pattern presumably reflects competition for resources among offspring, especially since the amount of available resources per offspring is a good predictor of larval survival as well as weight at pupation (Figs. 2a, 3). While temporal spacing of clutches may be an important factor determining offspring survival, in our current field study we did not find that the amount of time fruit were available to females for further oviposition (fruit cohort) explained offspring survival over and above that which was explained by the available fruit volume per egg or larva and the number of eggs or larvae placed within a fruit.

### *Superparasitism and pupal weight*

Pupal weight was negatively related to the number of conspecific larvae within a host but was positively related to the amount of fruit volume available per larva (Table 1). As each of the above variables explained variation in pupal weight when controlling for the other variable, this suggests that both variables independently play a role in determining offspring weight. The number of larvae hatching within a fruit, for example, may negatively impact larval development independent of the manner in which increases in the number of conspecifics reduce the amount of resources available per larva. Perhaps the number of larvae within a fruit is a good indicator of factors such as the amount of waste products that might build up within a fruit, the number of antagonistic interactions larvae experience, or the rate at which a host resource is expected to degrade.

While laboratory experiments did show that the temporal asymmetry between when clutches were laid dramatically impacted the survival and weight of clutches laid later (C. R. Nufio and D. R. Papaj, in preparation), it appears that, as measured, under field conditions, the temporal asymmetry between when clutches were laid is not as important as the amount of available resources per

individual and the number of individuals placed within a fruit. Perhaps, although females placed more clutches within fruit as time passed, the allocation of more eggs to larger fruit may have minimized the effects of temporally staggering clutches within a host. The degree to which these results are influenced by female rejection patterns is unknown. Females may, for example, utilize marking pheromone to gauge larval infestation levels relative to fruit size and/or females may be more likely to reject hosts that appear to be degraded; these mechanisms may minimize the costs associated with staggering clutches.

### Pupal weight and its impact on offspring reproductive potential

Median pupal weight increased with fruit volume per larva. In turn, in both replicates of our laboratory experiments, we found that smaller females laid fewer eggs over their lifetime. Smaller females produced fewer eggs not because their life spans were shorter, but because larger females appeared to produce more eggs per day (Table 2). Similar reproductive advantages of female size have been noted in other fruit flies, such as *R. pomonella* Walsh (Averill and Prokopy 1987), as well as in other systems (Credland et al. 1986; Wickman and Karlsson 1989; Bonduriansky and Brooks 1999; Mills and Kuhlmann 2000). While not measured in our study, male tephritids may also experience costs associated with being small (Burk and Webb 1983; Taylor and Yuval 1999).

### What benefits do females receive from superparasitizing hosts?

As we have argued previously, superparasitism of walnut hosts by *R. juglandis* may be influenced by three factors (Nufio et al. 2000). First, superparasitism may be influenced by the benefits that females gain not by simply superparasitizing a host fruit but more specifically by reusing oviposition punctures created by previous females. By reusing oviposition punctures, females may save time (Papaj 1993, 1994; Papaj and Alonso-Pimentel 1997), decrease the wear to their ovipositors (Papaj 1993), or gain access to fruit that are relatively impenetrable (Lalonde and Mangel 1994). These benefits have been proposed to increase the number of clutches that a female can lay over a lifetime.

Another reason that females may reuse fruit, independent of use of punctures, may be related to the host fruit's size. Most *Rhagoletis* species utilize relatively small hosts [e.g., hawthorn berries, cherries, blueberries and dogwood berries (Bush 1966)] that appear to offer fewer resources for developing offspring than do walnut fruit. With respect to the preference–performance hypothesis, in this system, the cost to larvae forced to compete with conspecifics, while meaningful, is thus not as severe as it could be in smaller hosts and this may in part explain why females commonly superparasitize these hosts.

The third factor that may influence superparasitism by walnut flies is the short temporal and limited numerical availability of larval hosts in the field. Since nearly all walnut hosts within an area will be synchronously utilized within 2–2.5 weeks, there will be a limit to the total amount of larval resources available to a population of walnut flies (Nufio et al. 2000). On an individual level, this may mean that females are time limited and must maximize the number of clutches deposited within the limited window of larval resource availability. One way to maximize the number of clutches deposited within the allotted time may be to superparasitize hosts as they ripen and become accessible to females.

#### Preference–performance and parent–offspring conflicts

Given an ability to discriminate and ample evolutionary time to adjust to novel hosts within their range, it is reasonable to expect that mothers would do best if they preferred only hosts that maximize their per capita offspring fitness as this should surely maximize their own fitness. However, models of parent–offspring conflict, progeny size–number trade-offs and even optimal foraging predict that, under a variety of conditions, females should devalue per capita offspring fitness if such behavior increases their own reproductive success (Forbes 1991; Lloyd 1987; Einum and Fleming 2000). In insects, such models are used primarily to understand the egg laying decisions made by parasitoids and seed predators (Charnov and Skinner 1985; Smith and Lessells 1985; Godfray 1987) and are generally not acknowledged in empirical studies of insect–plant systems in which the preference–performance hypothesis has been addressed (but see Rausher 1980; Larsson and Ekblom 1995; Nylin et al. 1996; Scheirs and De Bruyn 2002). In the superparasitism literature, for example, researchers have concluded that under conditions where the survival of the second progeny or clutch is  $>0$ , parasitism of an already parasitized host may be functional if unparasitized hosts are scarce, if search or handling time is high, if females are time limited or if multiple females are exploiting a patch simultaneously (van Alphen and Visser 1990; Speirs et al. 1991, van Alphen et al. 1992). Under such conditions, females benefit from superparasitizing hosts because their net returns are greater than if they rejected the host that was, from the larval perspective, inferior. The reproductive conflicts between offspring and parents explored in the field of superparasitism have not often been explored in regard to the preference–performance hypothesis primarily because researchers couple the outcomes of female decisions as they affect per capita offspring performance and not the different components of female performance (reviewed in Mayhew 1997).

The walnut fly *R. juglandis* appears to illustrate the reproductive conflicts that can arise when egg-laying strategies that maximize a female's reproductive success may not be the same strategies that maximize an

offspring's performance. We believe that a lack of a correlation between female preference and offspring performance should not always be interpreted as less than optimal decision making on the part of the female or sub-optimal conditions that limit offspring (and thus female) fitness, but alternatively, that such decision making can be optimal from the perspective of its impacts on female fitness. In other words, conditions may exist where even if a female is given a choice between a host where offspring perform best and a host where offspring do less well, females may, in the long run, actually do best choosing the latter host. We hope that investigators examine how alternative explanations, such as those provided by parent–offspring or optimal foraging models (Scheirs and De Bruyn 2002), may impact their interpretations and expectations regarding the evolution of female preference for oviposition sites and larval performance.

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