

Trade-offs of host use between generalist and specialist *Helicoverpa* sibling species: adult oviposition and larval performance

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Received: 21 September 2010 / Accepted: 1 August 2011 / Published online: 24 August 2011
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Abstract Much attention has been paid to the question of the relative importance of female behaviour versus larval feeding capacities in determining the host range of herbivorous insects. Host-use trade-offs displayed by generalist and specialist sister species of the genus *Helicoverpa* were evaluated to examine the relationship between maternal choice and offspring performance. The prediction of optimal oviposition theory, that females will choose to lay eggs on plants on which their offspring perform best as larvae, was tested by measuring oviposition preference and larval performance of *Helicoverpa armigera* and *H. assulta* on tobacco, sunflower, and hot pepper. These two measures were more highly correlated in the specialist *H. assulta*. Both species exhibited the same oviposition preference ranking: tobacco > sunflower > hot pepper. *H. armigera* larvae preferred sunflower, followed by tobacco and hot pepper; while *H. assulta* larvae preferred tobacco to sunflower and hot pepper, consistent with their mothers' oviposition preference. Duration of the total period from egg to adult emergence for each species was significantly

shorter on the host plant preferred by the larvae. *H. assulta* had shorter larval duration and higher relative growth rate than *H. armigera* on tobacco and hot pepper, and vice versa for sunflower, indicating species differences in host utilization. Thus, while only the specialist *H. assulta* displayed the predicted optimal oviposition pattern, females of both species show the least preference for the plant on which their offspring perform worst. Selection for optimal oviposition may be stronger on the specialist, which has fewer choices and lower lifetime fecundity than the generalist.

Keywords *Helicoverpa armigera* · *Helicoverpa assulta* · Oviposition choice · Feeding preference · Larval performance · Host-use trade-offs

Introduction

Optimal oviposition theory is the dominant paradigm that attempts to explain host plant selection by ovipositing females (Thompson 1988; Courtney and Kibota 1990; Mayhew 2001; Poore and Steinberg 1999; Scheirs et al. 2000). This theory, also called the oviposition preference–offspring performance hypothesis, predicts that females will maximize chances of success for their offspring by choosing those host plants for oviposition on which their larvae perform best (Jaenike 1978). However, females do not always oviposit on plant species on which their offspring grow and survive, and on which their performance is maximal (Kitching and Zalucki 1983; Courtney and Kibota 1990; Nylin et al. 1996; Berdegué et al. 1998; Scheirs and De Bruyn 2002; Jallow and Zalucki 2003). Female fitness is not only a function of the performance of her offspring, it is also determined by adult performance (Reavey and Lawton 1991; Nylin and Janz 1996; Krebs and Davies

Communicated by Roland Brandl.

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1997), and several studies have shown the contribution of host selection driven by adult behavior in which location of nectar sources for adult feeding plays an important role (Firepong and Zalucki 1990; Cunningham et al. 1998, 2001; Forister et al. 2009; Liu et al. 2010). Therefore, there may be trade-offs between maternal choice and offspring performance in the choice of host plants.

Offspring performance ultimately depends on survival and fecundity beyond the larval stage, and so the nutritional quality of larval host plants is not necessarily the ultimate factor balancing maternal choice in these trade-offs. Larval growth rates may be affected by abiotic factors such as thermal constraints which may also influence the ecological suitability of maternal choices via phenotypic plasticity (Nylin and Gotthard 1998). If thermal constraints on voltinism success are relaxed, the broader host range afforded may provide opportunities to escape from host-specific pathogens, parasites, or predators (Lederhouse et al. 1992; Scriber 2002). The greater physiological efficiency of specialized feeders may interact with climatic variables, and resulting differences in the time available for larval growth can in turn produce geographic variation in host preferences and degree of specialization over evolutionary time (Scriber 2010). Thus, biotic and abiotic factors interact with the rank order of preferences and their specificity (how far down the rank order a female will go) to shape the selective pressures on female oviposition choice (Mercader and Scriber 2007).

Neural or information-processing constraints have also been hypothesized to constitute an important source of trade-offs in the evolution of insect diet breadth (Bernays 2001; Bernays and Funk 1999; Janz 2003; Egan and Funk 2006). The information-processing hypothesis states that specialist herbivores make host-associated decisions more effectively than generalists, providing a fitness advantage that helps to explain the evolutionary prevalence of host-specific insects. Several studies have found that generalists are more likely to make mistakes, and are slower in deciding to oviposit than specialists (Janz 2003; Vargas et al. 2005; Egan and Funk 2006); yet the empirical evidence to support this idea is still rather scarce. Gene families such as cytochromes P450 that are involved both in olfaction by parents and detoxification by larvae have been suggested to provide a genetic substrate for correlations or trade-offs in the preference–performance relationship (Berenbaum and Feeny 2008).

Here, we compare oviposition choice and offspring performance in the sister species *Helicoverpa armigera* and *H. assulta* (Lepidoptera: Noctuidae). These species occur sympatrically and are serious crop pests in China and other areas of eastern Asia (Mitter et al. 1993; Chen 1999). They have a similar appearance and feeding behavior, but their host plant ranges are quite different. *H. armigera* is a highly

polyphagous herbivore; its host plant range includes at least 60 crop species such as cotton, corn, wheat, soybean, tobacco and tomato and 67 wild plant species from about 30 plant families including Malvaceae, Solanaceae, Gramineae, and Leguminosae (Xu et al. 1958; Fitt 1989; Zalucki et al. 1986; Jallow et al. 2004). *H. assulta* is an oligophagous species, with a relatively narrow host plant range, and mainly specializes on Solanaceae such as tobacco, hot pepper, and several *Physalis* species (Wang and Li 1984; Fitt 1989; Mitter et al. 1993; Wu et al. 2006). Their phenology overlaps from mid-May to mid-October, during which period five generations occur. Since Wang and Dong (2001) found interspecific hybridization of these two sister species is feasible, it has been used as a model to explore the genetic basis of host use differences and sex pheromone evolution between a generalist and specialist (Wang et al. 2005; Zhao et al. 2006; Zong and Wang 2007).

Recently, some studies (Mayhew 2001; Scheirs et al. 2004; Johnson et al. 2006) have re-ignited the debate about the preference–performance hypothesis. However, whether closely related generalists and specialists also fit the preference–performance hypothesis is unclear. It has been hypothesized that in species with a broad diet range the match between female preference and offspring performance may generally not be as tight as in species with narrower host ranges. Here, we investigated the relationship between female oviposition preference, offspring feeding preference, and offspring performance in these two *Helicoverpa* species in order to address both the preference–performance hypothesis and the information-processing hypothesis. The first predicts a correlation between female oviposition preference and offspring performance, which however could be compromised between maternal and offspring interests. The second predicts that the correlation should be stronger in specialists than generalists. We examined the following questions. First, do *H. armigera* and *H. assulta* females share the same oviposition patterns among the tested host plants? Second, do offspring share the same pattern in host use among the tested plants? Third, is there a better correspondence between female choice and offspring performance in the specialist rather than the generalist? Fourth, what are the differences between *H. armigera* and *H. assulta* in measures of performance on different hosts, such as larval duration, pupal weight, and relative growth rate?

Materials and methods

Insect colony and plant sources

A laboratory population of *H. armigera* was established from larvae collected near Toowoomba in south-eastern

Table 1 Homogeneity of female *Helicoverpa* species oviposition preference over time

	Tobacco × sunflower Eggs laid on tobacco (%)	Sunflower × hot pepper Eggs laid on sunflower (%)	Tobacco × hot pepper Eggs laid on tobacco (%)
<i>H. armigera</i>			
1st day	70 ± 3.4 (91) a	93 ± 5.3 (20) a	91 ± 3.1 (42) a
2nd day	66 ± 3.8 (99) a	90 ± 5.3 (23) a	89 ± 5.8 (40) a
3rd day	73 ± 3.7 (87) a	95 ± 3.0 (23) a	98 ± 0.9 (32) a
4th day	73 ± 4.1 (71) a	99 ± 0.7 (19) a	99 ± 0.7 (24) a
<i>F</i>	0.702	0.792	2.041
<i>df</i>	3,344	3,81	3,117
<i>P</i>	0.551	0.502	0.101
<i>H. assulta</i>			
1st day	92 ± 3.9 (37) a	93 ± 3.0 (43) a	95 ± 3.2 (36) a
2nd day	97 ± 1.9 (32) a	79 ± 4.9 (45) a	97 ± 2.7 (37) a
3rd day	93 ± 4.3 (32) a	86 ± 4.7 (40) a	99 ± 0.1 (37) a
4th day	99 ± 0.9 (24) a	73 ± 8.5 (18) a	99 ± 0.8 (32) a
<i>F</i>	1.1	2.5	0.8
<i>df</i>	3,121	3,142	3,138
<i>P</i>	0.320	0.058	0.520

Data are shown as mean % eggs laid on the preferred plant ± SE (number of replicates). Means not significantly different from each other at $P < 0.05$ are indicated by the same letter. ANOVA was performed on arcsine-transformed percentages

Queensland, Australia, in 2004, kindly provided by Dave Murray, Queensland Department of Primary Industries. The population of *H. assulta* was established by collecting larvae in Hunan Province, China, in 2007. Larvae were reared on a pinto bean-based artificial diet (Joyner and Gould 1985) to minimize the possible influence of experience of host plants at this stage (Firempong and Zalucki 1991). The larvae were reared at 27°C under a photoperiod of L:D 14:10 h to prevent diapause. Newly hatched larvae were reared in groups in a Petri dish with diet until the 3rd instar, after which they were separated in individual 30-ml plastic cups (Solo Cup, Illinois, USA) to prevent cannibalism (Twine 1971) and were allowed to pupate in the cup. Pairs of eclosed adults were allowed to mate in 350-ml paper mating cups (Solo Cup), and provided with 10% honey solution. Eggs were collected and the larvae of the new generation were put on a fresh artificial diet in Petri dishes. Both *H. armigera* and *H. assulta* colonies had been maintained for more than 6 generations in the laboratory before the start of the experiments.

Two members of the Solanaceae, tobacco (*Nicotiana tabacum* L., SR1) and hot pepper (*Capsicum annum* L.), and one of the Asteraceae, sunflower (*Helianthus annuus* L., Bambino Dwarf), were selected for the oviposition preference trials and subsequent larval preference and performance measurements. Tobacco and hot pepper are common natural host plants for *H. armigera* and *H. assulta* (Wang and Li 1984; Liu et al. 2004) with significant variation in suitability. Sunflower is a recorded host for *H. armigera* (Zalucki et al. 1994). Greenhouse growth conditions for tobacco were: day 23–25°C, night 19–23°C, humidity 45–55%; hot pepper and sunflower: day and night

21–23°C, humidity 50–60%. Whole plants and leaves of 1-month-old tobacco, sunflower, and hot pepper plants were used in the oviposition preference and offspring performance experiments.

Oviposition preference

Oviposition preference was evaluated in a dual choice test conducted in large cages (61 cm × 61 cm × 61 cm) in an environmental chamber at 27°C and L:D 14:10 h with three combinations: tobacco × sunflower, sunflower × hot pepper, tobacco × hot pepper. One-month-old tobacco, sunflower, and hot pepper plants of the same height (30 cm) were used to minimize effects of height on host preference (Firempong and Zalucki 1990). Tested insects were allowed to mate in single pairs in mating cups provided with 10% honey solution for two nights and were then transferred to large cages (one pair per cage) for the oviposition choice test. Each cage contained two plants and was provided with 10% honey solution. Twenty-four hours after adults were released in cages, plants were checked for eggs; afterwards, they were replaced by new plants of each combination and the position of the two plants was rotated every day to minimize position effects. The oviposition choice by each female was observed for four consecutive days. Oviposition preference of a female adult was expressed as the proportion of eggs laid on the choice plant of interest, of the total eggs laid over the four experimental days, including these eggs laid on the walls of cages, if any (generally less than 30% of the total). About 30 replicate cages (Table 1) were used for each treatment for both *H. armigera* and *H. assulta*.

Larval feeding preference

Neonates and 3rd instar larvae were tested for feeding preferences, as there is evidence that preference may differ for different instars (Zalucki et al. 2002; Perkins et al. 2008; Yang et al. 2008). Before females were released in cages, some individuals had already begun to lay fertile eggs on egg-mesh, and these were collected. Some were kept for neonate feeding choice tests, others were fed on artificial diet after larval hatching and subsequently used for feeding choice as 3rd instar larvae. Two-day-old eggs, ranging from 20 to 60, were put in the center of a 12-cm-square Petri dish and provided with 4 leaf discs with the diameter of 3 cm at each corner, 2 discs from each plant species. For each plant combination, 20 replicates were carried out. The Petri dishes were placed in a dark container at 27°C to avoid the effects of light on the behavior of the phototactic neonates. After 24 h, each leaf disc was checked to see how many neonates were resting or feeding there. These counts were highly correlated with the amount of feeding damage on the leaf discs. Leaf discs with no larvae usually had some small holes, indicating that some feeding occurred there before the larvae moved to another disc. Neonate feeding preferences were expressed as the proportion of neonates on the choice leaf disc of interest to all tested neonates.

The larvae kept on artificial diet were tested for feeding choice tests when they reached the 3rd instar and are considerably more mobile. In this experiment, ten 3rd instar larvae were placed in the center of the Petri dish and supplied with 4 leaf discs, 2 from each plant as described above. These Petri dishes were placed in a dark container at 27°C. Twenty-four hours later, larvae were checked to see how many were resting or feeding on each kind of leaf disc. Discs on which no larvae were resting usually had a small amount consumed (less than 10%), while discs with the majority of 3rd instar larvae were about 60% consumed. Feeding preferences were measured as the proportion of larvae on a given plant leaf disc in relation to all tested larvae. Each pairwise plant combination was tested in 20 replicates for each insect species and larval stage.

Larval performance

The eggs of another 30 families for both *H. armigera* and *H. assulta*, different from those families tested for larval preference above, were collected for tests of larval performance. Eggs laid on the same day were divided into three groups and kept in separate containers, and provided with young leaves (about 1 week old) from one of the three host plants, on which they started to feed after hatching. All the larvae were reared at 27°C with a photoperiod of L:D 14:10 h. Neonates were transferred to individual test tubes (one larva per tube to prevent cannibalism) and

provided with tender leaves from one of these three host plants. Leaves were changed every other day until pupation. Larval duration, pupal weight, relative growth rate, and sex were recorded for each individual. Larval duration was defined as the period from the day of hatch to the prepupal state. Pupae were weighed 24 h after pupation and then placed in individual cups until adult emergence. Relative growth rate (RGR in milligrams of increase per milligram body weight per day), which quantifies mass gained per unit time, was calculated based on an exponential growth model (Kogan and Cope 1974; Nitao et al. 1991) as, $RGR = [\ln(W_p) - \ln(W_1)]/D$, where W_p is pupal weight, W_1 is initial larval weight, and D is larval duration up to the prepupal stage. During the development from neonates to pupae, some larvae ($n = 100$) were weighed at 3, 6, 9, 12, and 15 days post-hatching to compare the growth rate curves.

Data analysis

Percentage data of female oviposition on plants were tested for homogeneity across the 4 days by a one-way ANOVA using the PROC GLM procedure in SAS (SAS Institute 1990) to investigate the egg laying pattern across different days, since some variation has been found in *Helicoverpa* species (Jallow and Zalucki 1996). Percentage data of each female on the preferred plant was arcsine-transformed in order to meet the assumptions of ANOVA. Preference (female oviposition and larval feeding) was analyzed using the General Linear Model (GLM). Species (*H. armigera* and *H. assulta*), and host plant were tested as main effects. Data of the dual choice experiments (oviposition and feeding preference) were subsequently analyzed using paired-sample *t* tests for *H. armigera* and *H. assulta* separately to clarify the species-specific pattern revealed by the full model.

All measures of progeny performance (pupal weight and larval duration) were analyzed using a General Linear Model (GLM). Species, host plant, and sex were treated as main effects. Similarly, data were subsequently analyzed using GLM for both *H. armigera* and *H. assulta* separately to clarify species-specific patterns revealed by the full model. Host and sex were tested as main effects. Data were square root-transformed before analysis to meet the assumptions of ANOVA.

Results

Oviposition preference

Oviposition preferences were homogeneous across the four tested days for both *H. armigera* and *H. assulta* (Table 1).

Table 2 Analysis of variance of oviposition preference, neonate feeding preference, and 3rd instar larva feeding preference of the two insect species on three plant hosts, partitioning effects of species and host (GLM)

Source of variation	df	Oviposition preference		Neonates feeding preference		3rd larvae feeding preference	
		MS	F	MS	F	MS	F
Species	1	3,021.2	9.0*	4,472.5	32.1****	1,995.6	17.9****
Host	2	4,725.6	13.3****	14,565.8	104.7****	179.3	1.6
Species × host	2	4,707.3	13.3****	8,087.0	58.1****	11,927.7	107.4****
Error	325	355.2		139.1		111.0	

Proportion data were arcsine-transformed before analysis to meet the assumption of GLM

* $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, **** $P \leq 0.0001$

Oviposition was affected significantly by insect species and host plant (GLM; Table 2). Both *H. armigera* and *H. assulta* females had the same overall oviposition preference patterns: tobacco was preferred, followed by sunflower, then hot pepper (Table 3A). Preferences in the pairwise tests were highly consistent, with the great majority of females in a test preferring the same plant, except for the 28% of *H. armigera* which chose sunflower over tobacco (Fig. 1).

Larval feeding preference

Both *H. armigera* and *H. assulta* neonates showed strong feeding preference patterns among tobacco, sunflower and hot pepper, but the order was different (Table 3B). For neonates, the feeding preference was significantly affected by species, host plants, and species × host (Table 2). *H. armigera* neonates chose to feed most on sunflower, then tobacco, and least on hot pepper. However, *H. assulta* neonates preferred tobacco, then sunflower, and last hot pepper. Third instar larvae feeding preference was significantly affected by species and the species × host interaction, but the marginal effect of host was not significant, differing from neonates (Table 2). This is due to a switch in preference by *H. assulta* 3rd instar larvae. *H. armigera* shared the same pattern with its neonates: sunflower > tobacco > hot pepper. However, *H. assulta* 3rd instar larvae did not; they preferred tobacco and hot pepper over sunflower (Table 3C).

Larval performance

Larval duration from egg hatching to prepupal state was significantly affected by host, species, sex, and species × host (Table 4). When *H. armigera* and *H. assulta* were analyzed separately, only the host plants effect remained significant (Table 5). *H. armigera* and *H. assulta* showed different patterns among these three host plants (Table 6A). The shortest larval duration for *H. armigera* (about 14 days) occurred on sunflower, significantly less

than that those fed on tobacco (15 days) and hot pepper (20 days). However, *H. assulta* larvae developed fastest on tobacco (14 days), significantly faster than those fed on sunflower (15 days) and hot pepper (17 days). *H. assulta* developed faster than *H. armigera* on tobacco and hot pepper, but not sunflower (Table 6A).

Pupal weight was significantly affected by host plant, and host × species (Tables 4 and 5). Both *H. armigera* and *H. assulta* produced heavier pupae on tobacco and sunflower than hot pepper, and further comparison showed there were no significant differences between these two species on each test host plant (Table 6B).

The relative growth rate (RGR) was significantly correlated with host, species, and host × species (Tables 4 and 5), indicating differences in rates of weight increase over the larval period between the generalist and specialist. *H. armigera* had its highest RGR feeding on sunflower whereas *H. assulta* had the highest RGR on tobacco, and both species grew slowest on hot pepper (Table 6C). Moreover, *H. assulta* had a higher RGR than *H. armigera* on tobacco and hot pepper, but not sunflower (*H. assulta*'s faster weight gain on this plant was offset by the additional 2 days taken in the larval period before pupation). These trends are shown in more detail by the growth curves (Fig. 2), which showed that *H. armigera* larvae increased their body mass fastest on sunflower and *H. assulta* larvae grew fastest on tobacco, and *H. assulta* grew faster than *H. armigera* on all three plants (Fig. 2).

Discussion

In all pairwise choice situations, both species exhibited the same strong and consistent oviposition preferences, with tobacco ranked highest and pepper lowest. *H. assulta* consistently preferred sunflower to pepper, even though pepper is commonly utilized in the field and sunflower is not recorded as a host for this species. The moths were experimentally confined to plants that were not in flower, and it is known that the presence of flowers can affect the

Table 3 *Helicoverpa armigera* and *H. assulta* adult oviposition preference and neonate and third-instar feeding preference in dual choice tests with three hostplants

	Rank of plant	Tobacco × sunflower	Sunflower × hot pepper	Tobacco × hot pep
A: Adult oviposition preference				
<i>H. armigera</i>				
Tobacco	1	66 ± 3.0		92 ± 3.0
Sunflower	2	28 ± 2.8	91 ± 4.0	
Hot pepper	3		7 ± 3.6	5 ± 2.3
<i>t</i>		−6.094*	10.562*	15.691*
<i>H. assulta</i>				
Tobacco	1	93 ± 2.5		98 ± 1.6
Sunflower	2	7 ± 2.5	86 ± 3.2	
Hot pepper	3		12 ± 2.9	1 ± 0.2
<i>t</i>		−15.308*	11.441*	53.045*
B: Neonates feeding preference				
<i>H. armigera</i>				
Tobacco	2	26 ± 1.4		83 ± 1.9
Sunflower	1	74 ± 1.4	91 ± 1.5	
Hot pepper	3		6 ± 0.9	14 ± 1.5
<i>t</i>		14.715*	18.848*	12.646*
<i>H. assulta</i>				
Tobacco	1	72 ± 1.8		76 ± 4.8
Sunflower	2	27 ± 1.8	83 ± 2.6	
Hot pepper	3		17 ± 2.6	24 ± 4.8
<i>t</i>		−10.76*	9.462*	5.277*
C: 3rd instar larvae feeding preference				
<i>H. armigera</i>				
Tobacco	2	29 ± 1.8		80 ± 4.0
Sunflower	1	62 ± 2.1	82 ± 3.4	
Hot pepper	3		11 ± 2.4	13 ± 2.9
<i>t</i>		9.411*	9.243*	8.050*
<i>H. assulta</i>				
Tobacco	1	83 ± 1.9		45.0 ± 3.2
Sunflower	3	15 ± 1.9	29 ± 3.3	
Hot pepper	2		64 ± 2.8	36 ± 4.7
<i>t</i>		9.966*	−6.303*	1.625

Data are shown as mean (percentage of plant chosen) ± SE. Percentage data was arcsine transformed before analysis by the paired-samples *t* test

* Means significantly different at $P < 0.05$

tendency of *Helicoverpa* spp. to oviposit (Firempong and Zalucki 1990; Liu et al. 2010). Preference for flowering plants that females can use as a nectar source as well as an oviposition site has been interpreted as evidence of a trade-off between maternal and neonatal nutritional interests (Nylin and Janz 1996; Krebs and Davies 1997), but in our experiment the opportunity for this trade-off did not exist, as adults were provided a source of 10% honey throughout. The absence of flowers made the preference–performance hypothesis easier to validate. If flowering had increased the attractiveness of pepper to *H. assulta* for oviposition, as might be suspected by its well-documented host status in the field, the poor larval performance on pepper would have contributed evidence against the preference–performance hypothesis. In the absence of flowers on any of the

plants offered as a choice, pepper leaves were generally least preferred for oviposition, least preferred by larvae, and least nutritionally suitable for larval growth; thus, the low maternal ranking of this resource appears to be an accurate assessment of its relative insuitability for offspring, for both species.

In *H. armigera*, larval food preference was the same for neonates and third instars, but only partly corresponded to female oviposition choice. Although mothers and offspring preferred pepper least, neonates and 3rd instars of *H. armigera* consistently preferred sunflower, despite it being the second choice of most *H. armigera* females. The correlation between maternal and offspring preference was stronger in *H. assulta*, but was complicated by a change in ranking with larval age. Neonates and third instars of *H. assulta* preferred

Fig. 1 Oviposition preference variance between *H. armigera* and *H. assulta*. *I* tobacco versus sunflower, *II* tobacco versus hot pepper, and *III* sunflower versus hot pepper

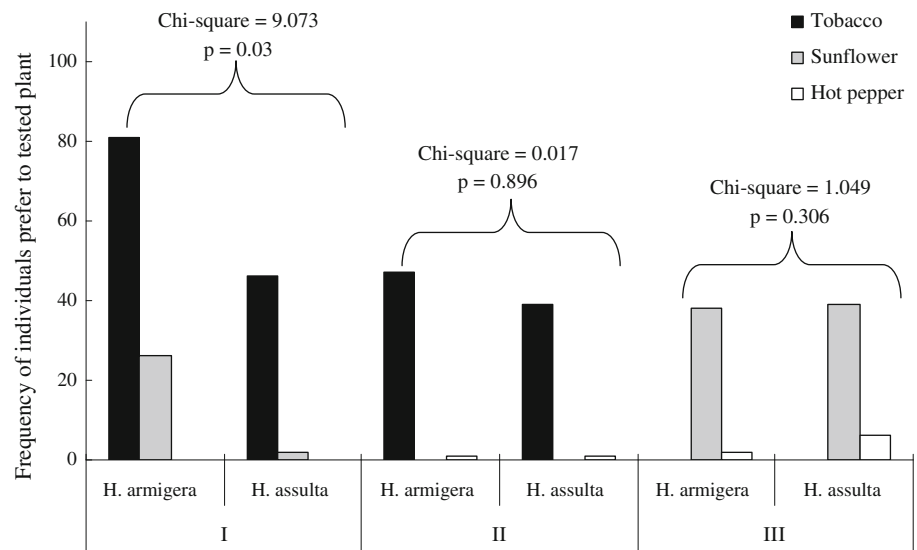


Table 4 Analysis of variance of measures of offspring performance of *Helicoverpa* spp., partitioning effects of species, host and sex (GLM)

Source of variation	df	Larval duration		Pupal weight ^a		Relative growth rate ^b	
		MS	F	MS	F	MS	F
Host	2	5.8	118.3****	67.7	51.2****	0.2	120.5****
Species	1	1.3	25.7****	1.6	1.2	0.02	19.5****
Sex	1	0.2	4.1*	0.03	0.02	0.004	3.1
Host × species	2	2.7	54.4****	6.9	5.2**	0.05	40.3****
Host × sex	2	0.01	0.3	2.4	1.8	<0.0001	0.3
Species × sex	1	0.1	2.7	0.05	0.03	0.002	1.4
Host × species × sex	2	0.04	0.9	1.8	1.3	<0.0001	0.2
Error	717	0.05		1.321		0.001	

Larval duration and pupal weight data were square root-transformed before analysis to meet the assumptions of GLM

* $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, **** $P \leq 0.0001$

^a Error df = 688

^b Error df = 680

tobacco, the same plant as their mother. The host plant least preferred changed from pepper for neonates (as for their mothers) to sunflower by the third instar. These older larvae are considerably more mobile and a switch in preference could conceivably result in inter-plant movement; however, movement between these two plant species would depend on their proximity in the field, which is unlikely. The type of shift most often seen in the field is from leaves upwards to fruits and flowers of the same plant, and larvae were not given this opportunity in our study.

For *H. armigera*, different measures of offspring performance were somewhat discordant. Pepper was inferior by all three measures, especially in larval duration and pupal weight. Larval duration was significantly shorter and relative growth rate significantly higher on sunflower than tobacco, corresponding to larval preference for sunflower but not maternal preference for tobacco. However, the

longer larval duration on tobacco evidently allowed for greater total food consumption, which compensated for the lower growth rate by resulting in pupae that tended to be slightly (but not significantly) larger on tobacco. The expected net effect on fitness in the field is difficult to assess; longer larval duration would likely increase mortality due to increased exposure to predation and parasitism, but larger pupal size would likely increase lifetime fecundity.

For *H. assulta*, all measures of offspring performance gave the same ranking, and tobacco enabled the best performance, corresponding to both maternal and larval choice. As for *H. armigera*, pepper was inferior in all three measures, most notably in larval duration and pupal weight. However, *H. assulta* larvae performed markedly better on pepper than did *H. armigera*. Development to the pupal stage was possible on all three plants, yet

Table 5 Analysis of variance of measures of offspring performance of *Helicoverpa* spp., partitioning effects of host and sex (GLM)

Source of variation	df	Larval duration		Pupal weight		Relative growth rate	
		MS	F	MS	F	MS	F
(A) <i>H. armigera</i>							
Host	2	2.9	51.1****	29.8	25.8****	31.5	57.3****
Sex	1	0.2	3.5	0.04	0.04	<0.0001	0.2
Host × sex	2	0.03	0.5	2.8	2.4	0.003	2.2
Error	171	0.06		1.1 ^a		0.001 ^b	
(B) <i>H. assulta</i>							
Host	2	8.3	179.8****	89.5	65.1****	0.2	183.0****
Sex	1	0.01	0.2	0.005	0.003	0.001	0.5
Host × sex	2	0.03	0.6	0.08	0.06	<0.0001	0.2
Error	546	0.05		1.4 ^c		0.001 ^d	

Larval duration and pupal weight data were square root transformed before analysis to meet the assumptions of GLM

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$, **** $P < 0.0001$

^a Error df = 165

^b Error df = 161

^c Error df = 523

^d Error df = 516

Table 6 *H. armigera* and *H. assulta* offspring performance (larval duration and pupal weight) on three hostplants

Performance	Tobacco	Sunflower	Hot pepper	df	F	p
(A) Larval duration						
<i>H. armigera</i>	15.0 ± 0.19 (104) b	13.9 ± 0.21 (62) a	19.5 ± 0.56 (16) c	2,179	56.8	<0.0001
<i>H. assulta</i>	13.5 ± 0.11 (257) a	15.4 ± 0.11 (283) b	16.9 ± 0.15 (172) c	2,709	184.1	<0.0001
df	1,359	1,343	1,186			
F	48.6	33.5	27.5			
P	<0.0001	<0.0001	<0.0001			
(B) Pupal weight						
<i>H. armigera</i>	230.4 ± 3.29 (105) b	220.2 ± 4.22 (54) b	173.2 ± 6.28 (16) a	2,172	21.9	<0.0001
<i>H. assulta</i>	223.2 ± 2.61 (203) b	229.0 ± 2.13 (218) b	185.1 ± 3.08 (103) a	2,526	64.9	<0.0001
df	1,306	1,270	1,122			
F	2.7	3.4	2.0			
P	0.101	0.065	0.157			
(C) Relative growth rate (mg/mg/day)						
<i>H. armigera</i>	0.368 ± 0.005 (103) b	0.392 ± 0.005 (54) c	0.266 ± 0.008 (15) a	2,169	44.9	<0.0001
<i>H. assulta</i>	0.411 ± 0.003 (199) c	0.361 ± 0.003 (215) b	0.314 ± 0.003 (108) a	2,519	193.44	<0.0001
df	1,300	1,267	1,121			
F	58.3	23.0	27.5			
P	<0.0001	<0.0001	<0.0001			

Data are shown as mean ± SE (sample size). Row means followed by different letters are significantly different at $P \leq 0.05$ (Duncan's test). Larval duration and pupal weight data were square root-transformed before ANOVA; untransformed data are tabulated

performance would likely have been better if fruit were available for consumption. Our study design did not allow us to test how the availability of fruit might have changed preference rankings and larval performance; this is an important topic for future study.

The overall pattern of oviposition choice and larval performance is more consistent with the preference–

performance hypothesis for *H. assulta* than *H. armigera*. As previously mentioned, the absence of flowers in this experiment has removed one of the factors that may break this correlation when maternal nutritional interests compete with assessment of host quality for offspring. Such a trade-off may be more important for *H. armigera* in the field if the wider variety of potential oviposition sites used by this

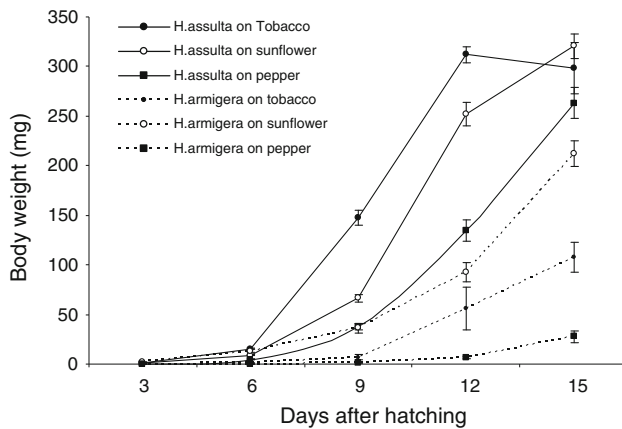


Fig. 2 Growth of *H. armigera* and *H. assulta* larvae feeding on sunflower, tobacco and hot pepper. Bars standard errors

host plant generalist provides more opportunities for encountering different species in flower. Yet, even when choices were restricted to non-flowering plants, *H. assulta* showed a closer overall correspondence between maternal choice and offspring performance. For *H. armigera*, the females' preferred plant, tobacco, came in second for larval preference and for 2 of the 3 measures of larval performance. The more accurate assessment by females of the more specialized of the two species thus provides some support for the information-processing hypothesis. Selection for optimal oviposition choice may be stronger on the specialist, which has fewer choices and lower lifetime fecundity than the generalist.

Tobacco and hot pepper are common host plants for both insect species (Wang and Li 1984; Liu et al. 2004), and sunflower is a recorded host for *H. armigera* but not *H. assulta*. In China, sunflower and pepper are generally planted in the north and south, respectively, so a single ovipositing population of *H. assulta* is rarely if ever exposed to the choice between the two in the field. Yet, the oviposition acceptance and larval acceptance and performance documented here for sunflower suggests that it could be considered a potential host plant for *H. assulta* according to the criteria set by Kitching and Zalucki (1983). As long as larvae have the capability to grow on a wider variety of plants than the existing host range, adult behavior will serve as the limiting factor in host selection (Firempong and Zalucki 1990; Cunningham et al. 1998, 2001; Forister et al. 2009), and a change in adult behavior may drive an expansion of the host range.

Host-range differences in heliothines have been associated with specificity of crypsis, fecundity, and dispersal tendency (Matthews 1991). One factor affecting the oviposition preference–larval performance differences between these two *Helicoverpa* species might be egg load and overall lifetime fecundity. As reported, one of their main differences is higher fecundity for the generalist and

relatively low fecundity for the specialist (Mitter et al. 1993). Jaenike (1978) and Mangel (1987) suggested that oviposition strategies of herbivorous insects vary depending on whether a female is more limited by the time available for oviposition or the number of eggs she can lay. Jallow and Zalucki (1998) carried out a study of effects of egg load of *H. armigera* on host selection. They confirmed that the physiological state of a female moth greatly influenced her host specificity and propensity to oviposit, such that female moths were less discriminating against the low-ranked host when egg load increased. Furthermore, a study by Doak et al. (2006) on a time-limited butterfly suggested that low egg load may contribute to selection for strong host plant discrimination. *H. armigera* has a markedly higher maximum fecundity, with up to 2,300 eggs laid during the female adult's lifetime, than *H. assulta* with up to 730 (our unpublished data). Furthermore, females lay most of these eggs during the first 3–7 days, when the higher egg load may reduce the selectivity. Thus, the specialist may be under stronger selection for oviposition on the more restricted range of hosts on which the offspring can perform well, accounting for a closer correspondence to the preference–performance hypothesis and agreeing with the predictions of the information-processing hypothesis.

When larval performance on the two hosts commonly used by both species is compared, *H. assulta* grew faster and developed more quickly on tobacco and pepper than did *H. armigera*. This may reflect stronger selection for traits enabling rapid growth on the Solanaceous hosts in the specialist *H. assulta*, whereas the generalist *H. armigera* is relatively less efficient on these, due to possibly conflicting selective pressures for larval performance on the much wider taxonomic range of hosts that it utilizes. Conversely, *H. armigera* was able to grow faster on sunflower than *H. assulta*, which does not experience selection for performance on sunflower in the field. If *H. assulta* widens its host range to include sunflower, which it evidently has the potential to do given the oviposition and larval acceptance we have found, then selection for improved larval performance on sunflower might occur, or could be countered by any trade-offs due to decreased performance on its Solanaceous hosts.

When choosing between tobacco and sunflower, 28% of *H. armigera* females preferred sunflower; this is the only oviposition choice situation in either species where the minority preference exceeded 5%. Thus, these females, although in the minority, were choosing more appropriately according to the preference–performance hypothesis than the majority, given the overall better offspring performance on sunflower. If intraspecific variation in oviposition preference has a genetic component, we would expect more variation in the generalist species than the specialist, yet

such variation was not evident in the other two-choice situations. We did not assess the performance of offspring from the two types of choosing mothers, so we have no evidence for or against any intraspecific genetic correlation between maternal choice and offspring performance. But we are currently exploring the genetic basis of the inter-specific differences in larval performance, by inter- and intra-specific backcrosses among these two species and QTL analysis of the growth rates on tobacco, sunflower, and pepper.

Acknowledgments This research was supported by the Max-Planck-Gesellschaft (MPG). Z. Liu was supported by a postdoctoral research fellowship jointly funded by the Max Planck Institute for Chemical Ecology and the MPG initiative for co-operation with the Chinese Academy of Sciences. We thank Andreas Weber for culturing plants, Emily Wheeler for editorial assistance, and two anonymous reviewers for useful suggestions.

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