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SPECIAL ISSUE – INSECT-PLANT INTERACTIONS: HOST SELECTION, HERBIVORY, AND PLANT RESISTANCE

Style length variation in male and female figs: development, inheritance, and control of pollinator oviposition

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Abstract

Fig trees (Ficus spp., Moraceae) depend on female fig wasps to transport their pollen and as a reward provide nutrition for pollinator fig wasp larvae. Most pollinators are host specific, but natural plant hybrids occur if atypical hosts are pollinated. Female fig wasps lay their eggs into ovaries of female flowers in receptive figs by inserting their ovipositors through the styles, and relative style and ovipositor lengths are believed to largely determine their ability to oviposit. In dioecious fig trees, flowers in receptive male figs have short styles and support larval development, whereas flowers in receptive female figs have long styles and only develop into seeds. Using the dioecious fig tree species Ficus montana Burm. f. and Ficus asperifolia Miq., we recorded the comparative ontogeny of style lengths to determine whether style growth in the two sexes is coordinated with the attraction of pollinators, and how maximum style lengths relate to the ovipositor lengths of their pollinators. F1 hybrids between these species produce viable seeds, but no pollinator offspring. We examined style lengths in F1s and backcrosses, relative to those of their parents, and whether coordination of style growth with pollinator attraction is disrupted. The ovipositor of Kradibia tentacularis (Grandi) (Hymenoptera: Agaonidae), the pollinator of F. montana, could reach some of the ovaries in female figs, but fails to lay eggs there. Styles in male F1 figs were intermediate in length between those of the two parents, but in female F1 figs styles grew longer than in either parent. Maximum style lengths in F1s were partially decoupled from receptivity, especially in female figs. Our results illustrate how timing of stigmal growth is coordinated with pollinator attraction, and that this coordination can be disrupted in hybrid individuals.

Introduction

Mutualisms are interactions between species where the species involved receive mutual benefits, but in many mutualisms at any one time some individuals may be gaining from the interaction whereas others are gaining no benefits, or are even being harmed (conditionality sensu Cushman & Whitham, 1989; context dependency sensu Hoeksema & Bruna, 2015). Partners in a mutualism routinely have conflicts of interest, and if reproductive advantage can be achieved at the expense of other individuals, then selection will favour the evolution of 'cheating' (Bronstein, 1994; Herre et al., 1999; Sachs & Simms, 2006; Sachs, 2015). Some plant species achieve pollination by deception, without ever providing a reward, and in some dioecious species only one sex provides rewards to the pollinator (Willson & Agren, 1989).The negative consequence of a failure by one sex to provide rewards to pollinators is particularly apparent for insects in nursery mutualisms. Here the expected reward is in the form of food for larval development (Dufaÿ & Anstett, 2004) and in the extreme example provided by females of dioecious fig trees (Ficus spp., Moraceae), 'cheating' female inflorescences reduce pollinator lifetime reproductive success to zero.

Fig trees are characterised by having tiny flowers distributed inside the cavity of their inflorescences (figs, also known as syconia) (Anstett, 2001). The several hundred species of fig trees are distributed in warmer countries worldwide (Berg & Corner, 2005), and all are pollinated by tiny fig wasps belonging to the family Agaonidae (Hymenoptera). Each pollinator species is usually associated with a single species of fig tree, but pollination of atypical hosts can result in hybrid plants, and hybridisation may have been a significant force in the evolution of the genus (Kusumi et al., 2012). Fig trees and fig wasps are mutually dependent, with fig wasp females laying their eggs only in the female flowers of receptive figs and the figs only being pollinated by these fig wasps. Around half of all fig species are monoecious and the rest are dioecious. In monoecious species the relationship is consistently mutualistic with both seeds and fig wasp offspring developing within the same figs. However, in dioecious fig trees, some individuals have figs that produce pollen and support development of the fig wasps that will carry it (male trees) and others produce only seeds (female trees) and exploit pollinators without providing any reward.

Pollinators are attracted to their hosts by chemical signals released by receptive figs (Proffit et al., 2009). After a female has landed on a receptive fig, she needs to squeeze through its narrow ostiole in order to reach the female flowers that line its inner surface. Once inside she attempts to insert her ovipositor down the full length of the styles, and if she reaches an ovary she can lay an egg (Weiblen, 2002). Larvae complete their development inside the galled flowers. Males of the next generation of fig wasps emerge first, mate with the females that are still in their galls, then chew a hole through the fig wall to allow the females to depart in search of receptive figs, which are usually on other trees (Weiblen, 2002; Suleman et al., 2012). Before departing their natal figs the female fig wasps either become covered passively with pollen from dehiscent anthers, or actively collect pollen into thoracic pollen pockets (Kjellberg et al., 2001, 2014), which they later unload while ovipositing.

Many Ficus species are pollinated by a single fig wasp species (Ramirez, 1970; Bronstein, 1987), but increasing numbers of tree species are known to have two or more pollinators (Molbo et al., 2003; Compton et al., 2009; Lin et al., 2010). Host specificity is believed to be maintained by a combination of unique chemical cues (Grison-Pigé et al., 2001; Wang et al., 2013), the physical barrier generated by ostiole size and shape (van Noort & Compton, 1986), style lengths relative to the length of a pollinator's ovipositor (Nefdt & Compton, 1996; Liu et al., 2013), and the ability of the fig wasps to induce gall development (Ghana et al., 2015a). The relationship between style lengths in figs and the lengths of their pollinator's ovipositor are significant because pollinators insert their ovipositors down the length of the styles and they must have sufficient length to reach the ovules. The style lengths are typically distributed unimodally (Kathuria et al., 1995; Otero & Ackerman, 2002) and the ovipositors of a tree's pollinator are usually long enough to reach most or all of the ovaries in monoecious figs or male dioecious figs (Nefdt & Compton, 1996; Zhang et al., 2009). Variation in style lengths is linked to the fate of the ovules in monoecious figs, with longerstyled flowers tending to produce seeds, and flowers with shorter styles more likely to support fig wasp development (Compton et al., 1994; Kathuria et al., 1995; Nefdt & Compton, 1996; Jousselin et al., 2001; Yu et al., 2004; Dunn et al., 2008).

Dioecious fig trees have modified their relationship with pollinators by having seeds and pollinator offspring developing on different trees. In male figs the style lengths are short and ovipositors can easily access all the styles, whereas in female figs the styles of the flowers are typically longer than the ovipositor, so pollinators cannot reach the ovules to lay their eggs (Nefdt & Compton, 1996; Weiblen, 2004). The stigmas of female flowers in male and female figs also differ in shape, with the former facilitating oviposition and the latter adapted for pollen collection (Verkerke, 1987; Beck & Lord, 1988). Pollination of female dioecious figs is achieved by mutual mimicry between males and females, with receptive figs of the two sexes having a similar appearance and smell (Grafen & Godfray, 1991; Grison-Pigé et al., 2001). Pollinator females lose their wings when passing through the ostiole into a fig, so they have no chance of re-emerging to fly to other trees after they enter a female fig.

Once inside, they continue to behave as if they were in a male fig, and pollination is achieved (Raja et al., 2008a).

Ficus montana Burm. f. is a small dioecious fig tree from Southeast Asia, where its pollinator is Kradibia (= Liporrhopalum) tentacularis (Grandi) (Hymenoptera: Agaonidae). Ficus asperifolia Miq. is a closely related species that in East Africa is pollinated by Kradibia hilli Wiebes. Hybrid seeds generated from crosses between the two Ficus species germinate readily, grow rapidly, and mature about as rapidly as their parents (Ghana, 2012). Together with their parent species they provide an opportunity to investigate patterns of inheritance and growth of style lengths in figs, whether style lengths are solely responsible for preventing the reproduction of fig wasps inside female figs and whether style lengths in F1s can reduce their reproductive success. Dissections of flowers in female figs of F. montana that had been entered by K. tentacularis females have shown that they cannot reproduce in female figs of their host because females of this wasp species do not lay any eggs there (Ghana et al., 2015b). In contrast, when wasps enter F1 male figs they do lay eggs, but larvae fail to complete development (Ghana et al., 2015a,b).

Here we address the following questions: (1) Does the maximum length of styles in male and female figs coincide with receptivity? (2) Are growth patterns similar in male and female figs and between hybrids and their parents? (3) What are the relationships between style lengths in male and female figs and the lengths of the ovipositors of the fig wasps that pollinate F. montana and F. asperifolia? (4) Do style lengths alone explain the lack of oviposition in female figs? And (5) do hybrids display style length characteristics intermediate between those of their parents?

Materials and methods

Study species

This study was conducted in the glasshouses of the University of Leeds Experimental Gardens (Leeds, UK) using F. montana, F. asperifolia, their F1 hybrids, and backcrosses to F. montana. F. montana and its pollinator had been maintained at Leeds University since 1995 (Suleman, 2007). The seeds of F. montana and mature figs containing K. tentacularis were obtained from the Centre of International Forestry Research (CIFOR) plantation near Bogor, Java and Rakata (Krakatau Islands) in Indonesia (Moore, 2001). Female K. tentacularis pollinate figs actively (Compton et al., 2010), can reemerge from the first figs they enter, and are capable of utilizing up to four additional figs nearby (Suleman et al., 2013b), but after losing their wings on initial entry they cannot fly. Their offspring sex ratios are biased and they lay mostly male eggs when they first enter a fig (Moore et al., 2002; Raja et al., 2008b).

The F. asperifolia population at Leeds was established using seeds collected from Kibale Forest in Uganda in August 2004 (Ghana, 2012). Trees of this fig species produced few figs in winter. Kradibia hilli is the pollinator of F. asperifolia in East Africa, whereas in West Africa the plant is pollinated by another Kradibia species (Berg & Wiebes, 1992). There are large differences between the number of flowers in the figs from the two areas and they may eventually be recognised as distinct species (Verkerke, 1987; SG Compton, unpubl.).

Although they occur on different continents, F. montana and F. asperifolia are closely related within the F. montana species group in Ficus subgenus Sycidium, Section Sycidium (Berg & Corner, 2005). Hybrids were generated by crossing F. montana (paternal parent) and F. asperifolia (maternal parent) and backcrosses were between male F. montana and female F1s using K. tentacularis as the pollen vector. No live K. hilli were available to generate reciprocal crosses, and K. tentacularis cannot develop in F1 hybrid figs, which prevented breeding of F2s (Ghana et al., 2015b). Experimental crosses were achieved by placing K. tentacularis females from known male host plants at the ostiole of figs on female plants. The figs were bagged before and after wasp entry into individual figs to prevent entry by other fig wasps. F1s were generated in 2005 and backcrosses in 2006.

Style length in figs of different diameters

Fig development is generally characterised using the stages described by Galil & Eisikowitch (1968) and Valdeyron & Lloyd (1979). Phase B figs are receptive and attract adult female wasps (loaded with pollen) to enter them. Phase C female figs contain developing seeds, whereas phase C male figs contain wasp larvae developing in galled ovules. Male figs at D phase release the next generation of fig wasps, with female pollinators then flying away in search of receptive figs to enter. The subsequent E phase male figs wither and eventually abort. Female figs have an extended C phase, which ends when the figs become attractive to seed dispersers (E phase).

Ficus montana, F. asperifolia, F1s, and backcrosses were selected haphazardly from the plants available in the general glasshouse population, with six plants from each group and three plants from each sex. Figs from each plant were collected at a range of diameters from 2.5 to 7.5 mm (one fig representing each 0.5 mm increment, if available, from each plant).

Pollination of the figs once they were receptive was allowed to occur naturally. The figs were examined under a binocular microscope (after being split longitudinally) and the style lengths of all the female flowers from a quarter of each fig were measured under a compound microscope (40× magnification) using an ocular scale. The measurements followed Nefdt & Compton (1996), where style length was taken as the distance between the top of the stigma and the end of the style, where it is connected to the ovule.

Style lengths in receptive phase figs

Thirty-one pre-receptive (A phase) figs from three male plants and 33 figs from three female plants of F. montana were chosen haphazardly. The figs were bagged until they became potentially receptive and were then tested for receptivity by introducing wasps to the ostiole area. Any attempt by the wasps to enter the figs was considered to indicate that the figs were receptive. These figs were collected and fig diameter was measured to record the range of sizes of figs at receptivity and then dissected under a binocular microscope. All the flowers in the figs were removed and positioned horizontally on slides to measure the styles, as described above.

With F1s, backcrosses, and F. asperifolia, three plants from each sex were chosen haphazardly and five figs of varying sizes from each plant were scored for receptivity. Due to the absence of its own pollinator, potentially ‘receptive’ figs of F. asperifolia were tested by first rubbing the ostiole of receptive F. montana figs onto the ostiole of F. asperifolia figs before introducing K. tentacularis females to the area of the ostiole. Successful entry of the pollinators was used as an indicator of receptivity. The figs were then divided longitudinally into four pieces, with the flowers counted and styles measured as indicated above.

Ovipositors of K. tentacularis were measured by collecting females as they emerged from 25 mature (D phase) F. montana figs obtained from five trees (five figs from each plant). Up to 10 wasps from each fig were collected, slide mounted, and ovipositors were measured under a compound microscope using an ocular scale after displacement of the sheaths (Nefdt & Compton, 1996). In addition, the ovipositors of K. hilli, the pollinator of F. asperifolia, were also measured. These were reared from their host figs by SG Compton & C Nuttman in Kibale Forest, Uganda.

Data analysis

All statistical analyses were performed in R (v.2.12.2). We used Wilcoxon’s rank sum tests to compare ovipositor lengths of the pollinators of F. montana and F. asperifolia (K.

tentacularis and K. hilli, respectively). Nested ANOVA (figs within plants) was used to compare differences in style lengths of female flowers at receptivity, with trees nested within groups (parental species and offspring generations). F tests (R procedure ‘variance’) were used to compare the equality of variances in style lengths between sexes.

Results

Style lengths during fig development

In figs of both sexes, and throughout their development, variance in the lengths of the styles was low (Figure 1). The mean style lengths of female flowers in female figs of F. montana were higher than in male figs at all stages of fig growth from early phase A to phase C, when seeds and the next generation of fig wasps are developing (Figure 1A). In male figs the maximum mean style length was 0.38 mm, at a time when fig diameter was 5.0 mm, towards the end of the receptive phase (when diameters were 4.0–5.5 mm). In female figs the mean maximum style length was 1.15 mm, recorded at a fig diameter of 6.5 mm, which was just after the end of the receptive phase (diameters 4.0–6.0 mm). In general, the styles in male and female figs of F. montana started to shrink when receptivity ended, at the time when the fig wasp offspring and seeds would have started developing.

Ficus asperifolia figs were available only from early phase A to late phase B, because these figs were rarely entered by K. tentacularis females. Style lengths in female figs of F. asperifolia were again longer than in male figs throughout (Figure 1B). The maximum mean style length in male figs was 0.43 mm, reached at a fig diameter of 7.5 mm, which coincided with the end of the putative receptive phase (the range at which pollinators would attempt entry was 5.5–7.5 mm). In female figs the maximum mean style length was 1.05 mm, at a fig diameter of 6.5 mm, which again coincided with the putative receptive phase (range 6.0–7.5 mm).

In very young figs on F1 plants, style lengths in female figs were marginally shorter than in male figs of the same diameter, but they had become much longer by the receptive phase, as in both parents (Figure 1C). The maximum mean style length in male figs was 0.39 mm, at a diameter of 6.5 mm, which was after the receptive phase (diameters 4.0–5.5 mm). In female F1 figs the maximum mean style length was 1.18 mm, at a diameter of 7.0 mm, which again was well after the end of the receptive phase (range 4.0–5.5 mm).

On backcross plants, the mean style lengths in female figs were longer throughout (Figure 1D). In male figs the maximum mean style length was 0.5 mm at a diameter of 6.0

mm, during the receptive phase (4.5–6.5 mm). Maximum mean style length in female figs reached 1.0 mm at a fig diameter of 6.5 mm, towards the end of the receptive phase (5.0–6.5 mm).

In summary, fig diameters during the receptive periods of male and female F. montana, F. asperifolia, their F1s, and backcrosses were generally the same, or at least largely overlapped. The styles in female figs were generally slightly longer even in very young figs, and then elongated much more quickly. Style lengths in both sexes were generally at their longest during the receptive period, before starting to shrink after receptivity, but there was an apparent disconnect in development in F1s, where in both sexes styles continued to elongate after the receptive period.

Style lengths of receptive figs in relation to ovipositor lengths

There was a consistent unimodal distribution in style lengths in both male and female figs and the ovipositor lengths of the pollinators. On average, styles in receptive-phase male figs were consistently much shorter than in female figs. The ovipositors of K. tentacularis females ranged from 0.64 to 0.92 mm, and they were much longer than the styles of any flowers in male figs of F. montana (range 0.25–0.46 mm; mean ± SE = 0.33 ± 0.0007 mm, n = 2 306; Figure 2A). The styles in female figs of F. montana were much longer than in male figs (range 0.61–1.41 mm; mean ± SE = 0.98 ± 0.002 mm, n = 3 342; Figure 2A) and the variance in style lengths of male and female figs was also significantly different ($F_{2305,3341}$ = [MP1]0.06, P<0.001), with more variation amongst styles in female figs[SC2]. Although styles in female figs were mostly longer than the K. tentacularis ovipositors, up to 33% of the female flowers in these figs were potentially reached by ovipositors of F. montana females (based on the longest recorded ovipositor) and 8.9% of the styles were shorter than the mean length of the fig wasps' ovipositors.

Styles of female flowers in receptive male figs of F. asperifolia (not a natural host for K. tentacularis) were again all shorter than the fig wasp's ovipositor (style lengths 0.25–0.62 mm; mean ± SE = 0.47 ± 0.001 mm, n = 914) and much shorter than the styles in female figs (0.74–1.36 mm; mean ± SE = 1.03 ± 0.002 mm, n = 1 157; Figure 2B). As in figs of F. montana, the variance in style lengths was also greater in female figs ($F_{913,1155}$ = 4.76, P<0.001). Up to 13.7% of the ovules of female flowers could be reached by the ovipositor of K. tentacularis (based on the longest recorded ovipositor), but only 0.4% were accessible for a female with an ovipositor of average length.

Kradibia hilli is the routine pollinator of F. asperifolia in East Africa. Its ovipositor (mean ± SE = 0.80 ± 0.005 mm; n = 44) is significantly longer than that of K. tentacularis (mean ± SE = 0.76 ± 0.003 mm; n = 214; Wilcoxon test: W = 1 838, $P<0.001$) and potentially reaches all the ovules in male figs of F. asperifolia (Figure 2C). It could also reach 1.5% of the ovules in female figs (based on its mean ovipositor length).

All ovules in male F1 hybrids were accessible to K. tentacularis (style length range 0.32–0.57 mm; mean ± SE = 0.45 ± 0.001 mm, n = 862; Figure 3A). Style lengths of flowers in female figs were again much longer (range 0.87–1.36 mm; mean ± SE = 1.11 ± 0.003 mm, n = 795; Figure 3A) and the variance between style lengths of female flowers in male figs and female figs was again significantly different ($F_{794,861} = 3.87$, $P<0.001$). Around 1.9% of female flowers in female figs of F1 plants could be reached with the longest recorded ovipositor of K. tentacularis, but all the female flowers in female figs were longer than the mean ovipositor length.

Styles of female flowers in male figs of backcrosses were again all shorter than the ovipositor of K. tentacularis (range 0.3–0.47 mm; mean ± SE = 0.38 ± 0.001 mm, n = 651; Figure 3B). The much longer styles in female figs (0.67–1.31 mm; mean ± SE = 0.99 ± 0.006 mm, n = 558; Figure 3B) again displayed a larger variance in length than in male figs ($F_{650,557} = 25.76$, $P<0.001$). Around 36% of the ovaries of female flowers in female figs could be reached with the longest recorded ovipositor of K. tentacularis and 8.2% of the female flowers were shorter than its mean ovipositor length.

Styles of female flowers in male figs of F. montana, F. asperifolia, and its hybrids were generally at least twice as long as in the corresponding male figs (Figure 4). In male figs of F1s, style lengths were intermediate between those of the parents and so were longer than those of F. montana, but shorter than those of F. asperifolia (the maternal parent). Styles in the backcrosses were again longer than those of F. montana but were shorter than those of the F1s (the maternal parent). There was a significant overall difference in style lengths among male figs of the parental and other generations (nested ANOVA: $F_{3,4728} = 457.86$, $P<0.001$). Using pairwise t tests for differences between groups, significant differences were found among all four groups (all $P<0.001$).

In female figs, the styles were longer than in both parents (Figure 4), whereas backcrosses were intermediate between F. montana (paternal parents) and the F1s (maternal parents). Differences between groups in style lengths in female figs were again highly significant (nested ANOVA: $F_{3,5846} = 218.73$, $P<0.001$). Pairwise t tests detected significant differences among all four groups (all $P<0.002$).

Styles in female figs were consistently more variable in length than in male figs

(Figure 3), but coefficients of variation were almost similar between the sexes (Table 1). This suggests that the higher variance recorded in female figs can largely be attributed to their longer length.

Discussion

The styles in male and female figs of F. asperifolia and F. montana reached a maximum length at about the time that the figs were receptive, and then began to shrink. Growth of the styles before receptivity was much more rapid in female figs, resulting in them having much longer maximum lengths, but the ovipositors of K. tentacularis females were sufficiently long to reach some of their ovules in female figs of both the normal and atypical host figs. In contrast to the parental species, styles in F1 hybrids continued to grow after the receptive period, especially in female figs. Mean style lengths during receptivity were intermediate between the parents in male F1s, but longer than either parent in female F1s. Backcrosses to F. montana were consistently intermediate between F1s and F. montana.

Female figs of F. montana and F. asperifolia are receptive to pollinators at the same diameter as male figs, as is to be expected given their need for mutual mimicry (Grafen & Godfray, 1991; Moore et al., 2003). Pollinators that enter female figs will always fail to produce any offspring and this should generate strong selection to avoid these figs. Although the emission of similar (though not always identical) volatile blends from receptive figs is a key element of this deception (Grison-Pigé et al., 2001), pollinators searching for oviposition sites could also respond to visual cues if receptive figs were of different sizes.

Styles at receptivity in male figs were shorter than in female figs, as has been described for all other dioecious fig tree species (Weiblen, 2002; Shi et al., 2006; Ma et al., 2009). At the time of receptivity, styles in female figs are also much more variable in length than those in male figs. This is likely to be advantageous in terms of pollen capture, whereas the less variable and shorter styles in male figs facilitate oviposition by the pollinators. Development of the longer styles in female figs starts when they are very small, and accelerates up until receptivity, by which point the styles stop growing and then start to shrink. In F. montana, F. asperifolia, and backcrosses to F. montana the period of receptivity corresponds with the cessation of growth by the styles in both male and female figs. This was not the case with F1 figs, where receptivity ended at a time when the styles were still continuing to grow. Krabidia tentacularis cannot reproduce in male figs of F. asperifolia, F1s, and most backcrosses, a failure that seems linked to an inability to induce gall development, because eggs are laid in all three groups of male figs (Ghana et al., 2015a). The slight disconnect between style growth and receptivity in male F1s could possibly be linked to a failure to respond to gall induction, if responses to galling can only be initiated in flowers where the styles have reached their maximum length, although at present there is no evidence to support this claim. Female F1 figs (and backcrosses) produce viable seed without any apparent costs resulting from the lack of synchrony between maximum style length and receptivity (Ghana, 2012).

Styles in male figs of all groups were much shorter than the ovipositor of K. tentacularis, and an ability to reach the ovules has been confirmed by the presence of eggs laid in F. asperifolia, F1, and backcross figs (Ghana et al., 2012). The longer ovipositor of F. asperifolia's pollinator K. hilli, compared with K. tentacularis, is likely to reflect the slightly longer mean style length in its male figs (Nefdt & Compton, 1996). Kradibia hilli potentially reach all the flowers in male figs of F. montana and would also be able to reach more ovules inside female figs of F. montana than K. tentacularis, if the opportunity ever arose.

The female flowers in male figs of F1s had styles that were intermediate in length between those of its parents, but closer to those of F. asperifolia. Whether this was linked to F. asperifolia being the maternal parent is unclear, but the overall volatile profile of F1s during receptivity is also more like that of F. asperifolia than that of F. montana (C Soler, pers. comm.). Phenotypic characters in hybrids are not always intermediate between those of their parents (Arnold & Hodges, 1995; Rosas et al., 2010) and styles in female F1 figs are an example of this. They attained longer mean lengths than those seen in either parent, apparently as a result of style lengths continuing to grow post-receptivity. Style lengths in both sexes of backcrosses were intermediate between their parents (F. montana and F1s).

The ovipositors of the largest K. tentacularis individuals appear capable of reaching up to one third of the ovaries in female figs of F. montana and backcrosses to F. montana, and fewer ovaries in female figs of F. asperifolia and F1s. Despite this, no eggs were detected in female figs of F. montana (Ghana et al., 2012). Success in laying eggs in female figs may be influenced by stigma shape and the lack of a clearly defined synstigma (the platform formed from the tops of the stigmas, on which pollinators stand during oviposition), which are likely to make it harder for the fig wasps to insert the full length of their ovipositors to reach the ovules (Galil, 1973; Verkerke, 1986; Jousselin & Kjellberg, 2001). These factors, in combination with style length per se, appear to prevent oviposition by the pollinators when they enter female figs. The shape and presentation of the stigmas of fig flowers varies considerably between taxonomic groups (and sexes) within Ficus (Verkerke 1989; Berg & Corner, 2005). The significance of this variability may have been underestimated.

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Figure captions

Figure 1 Changes in the style lengths of female flowers in male (diamond-shaped symbols) and female (squares) figs of (A) Ficus montana, (B) F. asperifolia, (C) F1s, and (D) backcrosses to F. montana. The horizontal lines indicate the receptive phase when Kradibia tentacularis females attempted entry into the figs. Standard errors were too small to be visible in the figure.

Figure 2 Distribution of ovipositor lengths (hatched bars) of (A, B) Ficus montana’s pollinator (Kradibia tentacularis) and style lengths (at receptive phase) in male (white bars) and female (black bars) figs of (A) F. montana and (B) F. asperifolia, and of (C) F. asperifolia’s pollinator (K. hilli) and style lengths in male and female figs of F. asperifolia.

Figure 3 Distribution of ovipositor lengths (hatched bars) of Ficus montana’s pollinator (Kradibia tentacularis) and style lengths (at receptive phase) in male (white bars) and female (black bars) (A) F1 hybrid figs and (B) backcrosses to F. montana.

Figure 4 Mean (± SE) style lengths in receptive male (white bars) and female (grey bars) figs of Ficus montana, F. asperifolia, F1 hybrids, and backcrosses to F. montana.

Table 1 Style length (mm) in receptive male and female figs of Ficus montana, F. asperifolia, F1 hybrids, and backcrosses to F. montana

Ficus	Sex	No. figs	Style length (mm)		
			Mean	Variance	CV
F. montana	Female	33	0.977	0.020	0.146
	Male	31	0.334	0.001	0.104
F. asperifolia	Female	15	1.030	0.009	0.090
	Male	15	0.469	0.002	0.090
F1	Female	15	1.109	0.008	0.079
	Male	15	0.455	0.002	0.097
Backcross	Female	15	0.994	0.022	0.148
	Male	15	0.376	0.001	0.077

CV = Coefficient of variation.