

Electronic Supplementary Material

MATERIALS AND METHODS

(a) *Selective exclusion experiments*

Fifty *Phyllanthus lepidocarpus* plants were collected from a wild population in Kyoto and transplanted individually to 500-ml pots. The plants were kept in a greenhouse for 1 week, and 15–39 female flowers that opened during this period were marked for each plant. We then applied one of the following four treatments: (i) the plants were bagged with fine netting to exclude all flower visitors ($N = 15$), (ii) bagged as above, but ants were allowed to freely forage through a slit (width, 5 mm) at the bottom of the bagging ($N = 15$), (iii) bagged and self-pollinated by hand ($N = 5$), or (iv) left unbagged as controls ($N = 15$). The experimental pots were placed in the Kyoto University campus, where ants were abundant, for 1 week, and the proportion of marked flowers that developed into fruits was calculated for each plant. During the experiment, we monitored the “ants-only” plants at various times of the day, between 800 and 2200 h, for a total of 12 h and confirmed that no insects other than the ant *Aphaenogaster famelica* were present on the plants.

(b) *Taxon sampling for the Epicephala phylogenetic analysis*

The genus *Epicephala* currently consists of > 40 described species with little known larval biology (de Prins & de Prins 2005) and most likely includes a large number of undescribed species as sampled in this study. However, many of the described species are known only from the type collections and lack sufficient descriptions of genitalia and wing venation, which provide explicit features for the grouping of genera in Gracillariidae (Vári 1961). Because we did not include any described species of *Epicephala* or thoroughly sample species from related genera, we were unable to test the monophyly of the genus or explicitly investigate the origin of seed feeding within the family. Nevertheless, our extensive sampling of *Epicephala* moths from a broad range of Phyllanthaceae hosts (table 1) provides a sound basis for investigating overall evolutionary pattern of pollinating behavior and host associations in the genus, and estimating their divergence times. Sampling of *Cuphodes diospyrosella* and *Stomphastis labyrinthica* is based on distinct morphological apomorphies in male and female

genitalia that they share with species of *Epicephala* (A. K., unpublished data).

Melanocercops ficuvorella was sampled as the farthest outgroup to allow estimation of branch length leading to the ingroup, which is required in the analyses of ancestral character states and divergence times.

(c) *Cophylogenetic analysis*

To test whether the plant and moth phylogenies are associated at random at higher taxonomic levels, we performed the ParaFit (Legendre *et al.* 2002) analysis based on patristic distances calculated along the ML phylogeny using PATRISTIC 1.0 (Fourment & Gibbs 2006). Because the phylogenetic results for *Epicephala* indicated that major moth clades have specific associations with particular host taxonomic groups, we reduced the following six clades to single terminal taxa: Malagasy *Phyllanthus* clade, *Glochidion* clade, *Phyllanthus* section *Gomphidium* clade, *Breynia* clade, *Phyllanthus* subgenus *Kirganelia* clade, and *Phyllanthus* section *Adenoglochidion* clade (see fig. S2 for clade names). Pruning was done by averaging patristic distances across all constituent species in each clade. Similarly, we extracted the species hosting *Epicephala* from the plant phylogeny and constructed a patristic distance matrix by reducing the above-mentioned taxonomic groups to single taxa.

(d) *Phyllanthaceae fossils*

A list of known Phyllanthaceae fossils is given in table S7. Of these, we selected the following four non-redundant fossils as minimum age constraints in the divergence time estimation of the Phyllanthaceae phylogeny: *Bischofia*-type pollen from Bartonian, Middle Eocene (37.2 mya); *Actephila*-type pollen from Late Eocene (33.9 mya); *Phyllanthus*-type pollen from Early Eocene (48.6 mya) (Gruas-Cavagnetto & Köhler 1992); and *Glochidion* leaf impressions from Middle Miocene (11.6 mya) (Prasad 1994; Antal & Prasad 1996). Ages of the fossils are given as the upper boundaries of epochs/stages to which they belong (geologic times based on Gradstein *et al.* 2004). The *Actephila*-type pollen had palynological features most closely related to *Actephila* but also showed some affinity to genera such as *Andrachne*, *Leptopus*, and *Meinekia* (Gruas-Cavagnetto & Köhler 1992), all of which are members of Poranthereae; thus, we used this fossil to provide minimum age for the Poranthereae–Wielandieae split (fig. S3). We did not use the *Reverchonia*-type pollen to constrain the age of *Reverchonia*,

because this fossil pollen does not precisely match those of the modern species (the monotypic *Reverchonia arenaria*; Sagun & van der Ham 2003) and exhibits some features associated with members of *Phyllanthus* (Gruas-Cavagnetto & Köhler 1992). Thus, we regard this fossil as *Phyllanthus* to be conservative, which consequently becomes redundant to the older *Phyllanthus*-type pollen. We also did not use the *Bridelia* wood fossil, because the assignment of Phyllanthaceae wood fossils to modern taxa is not straightforward (Prakash *et al.* 1986) and the age of the present fossil is too young to provide a meaningful lower bound on node ages. Because precise attribution of some of the Phyllanthaceae fossils may still require refinements (Gruas-Cavagnetto & Köhler 1992), the estimated divergence times may be revised when fossils with a more reliable taxonomic placement become available. Nevertheless, taxa belonging to Phyllanthaceae are well represented in the fossil record during the Eocene–Miocene times (table S7), providing evidence for the antiquity of the tribe as a whole.

(e) *Priors for Bayesian divergence time estimation*

The following prior distributions on parameters were used for the Bayesian divergence time estimation in Phyllanthaceae. The prior mean \pm SD for the age at root node was set to either 125 ± 20 myr (based on the earliest occurrence of eudicot pollen; Magallón *et al.* 1999) or 108 ± 20 myr (based on the estimated maximum age of the corresponding node in Davis *et al.* 2003). These values were divided by 100 so that they fall between 0.1 and 10, which is the recommended range of prior mean for the root age. The corresponding prior distributions of rate at root node were 0.88 ± 0.44 and 0.1 ± 0.05 , which were obtained by dividing the median of the distances between the ingroup root and the tips by the respective prior root age (standard deviations were set to half the values of the means). The prior for the Brownian motion parameter was 1.0 ± 1.0 for both analyses, which fall between 1 and 2 when multiplied by the prior root age, as recommended by the program manual.

The prior distributions on parameters for the *Epicephala* divergence time estimation were as follows. The root node age was set to 1.0 ± 0.0 , which is equivalent to fixing the root age with one unit time, the rate at ingroup root was 0.12 ± 0.06 , based on median distance between the ingroup root and the tips, and the Brownian motion parameter was 1.0 ± 1.0 , as recommended by the manual.

RESULTS

(a) Associations with *Epicephala* in *Phyllanthaceae*

The examination of flowers and fruits in *Phyllanthaceae* showed that 18 of the 26 species studied did not have associations with *Epicephala* moths (table S1), indicating that they are not involved in pollination of these species. In turn, *Epicephala* moths parasitized fruits in eight of the 26 species studied. Of these, four species (*P. reticulatus*, *P. sp.*, *P. marojejiensis*, and *P. humbertii*) belonging to two major *Phyllanthus* taxonomic groups (section *Anisonema* and unclassified Malagasy clade) were newly found to have the *Epicephala* pollination system on the grounds that there was a significant association between pollinated status and the presence of *Epicephala* eggs in female flowers (table S2), the styles were simple and not spread (fig. 2 in the printed paper), and/or female moths were observed pollinating flowers. *Epicephala* moths associated with *P. reticulatus* were observed pollinating female flowers in a manner typical of other actively pollinating species. We also collected a single female on a *P. marojejiensis* plant that bore > 100 pollen grains on her proboscis, suggesting that this species is also a pollinator.

The remaining four species that had associations with *Epicephala* (*F. suffruticosa*, *P. ussuriensis*, *P. lepidocarpus*, and *P. amarus*) were visited by abundant diurnal insects (table S3). *Flueggea suffruticosa* attracted a wide variety of insects that foraged for nectar and pollen, which are likely pollinators of this plant. The other three species, which are all herbaceous, were visited predominantly by ants, and pollination experiments in *P. lepidocarpus* indicated that ants are effective pollinators (table S8). Observation of *Epicephala* moth behaviour suggested that the moths do not pollinate these flowers. In *F. suffruticosa*, female moths laid eggs in flower buds, flowers, or developing fruits and did not carry pollen grains on the proboscis ($N = 32$). In *P. lepidocarpus* and *P. amarus*, *Epicephala* females laid eggs only in young fruits and carried no pollen grains ($N = 15$ and 9, respectively). The absence of *Epicephala* moths in some of the studied populations of these plants further indicates that they are not dependent on *Epicephala* for pollination. Based on these observations, we conclude that *Epicephala* moths associated with the above four species are truly parasitic, non-pollinating seed parasites.

(b) *Divergence time estimation in Epicephala*

Calibration of the *Epicephala* tree based on the COI molecular clock (1.5% myr⁻¹) suggested that the crown group *Epicephala* most likely diverged 20–35 mya (fig. 4 in the printed paper). The estimated constant rate of COI sequence divergence in arthropods ranges between 1.3% and 2.3% myr⁻¹ (Brower 1994; Quek *et al.* 2004). However, the use of higher rates would only result in younger estimates for the *Epicephala* root node; thus, our method is conservative with respect to providing young ages. The outlying old age estimates (fig. 4) were contributed by the shallowest nodes on the *Epicephala* chronogram, which are most prone to stochastic error in divergence time estimations (Ho *et al.* 2005; Sota & Hayashi 2007). On the other hand, ages provided by deeper nodes clustered within a narrow time frame between 20 and 30 mya (fig. 4). When the moth tree is calibrated with 25 mya at the *Epicephala* crown group (fig. 3), roughly concordant ages were obtained for the associated plant and moth clades, indicating the robustness of our age estimates (e.g., *Phyllanthus* section *Gomphidium* and their pollinators, 7.75–15.16 and 7.36–10.47 mya, respectively; section *Adenoglochidion* and pollinators, 2.56–7.98 and 5.38–9.24 mya, respectively; *Breynia* and pollinators, 5.01–13.26 and 3.32–5.34 mya, respectively; *Glochidion* and pollinators, 1.46–4.69 mya and 5.26–8.08 mya, respectively).

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Figure legends

Figure S1. Bayesian majority consensus cladogram of Phyllanthaceae based on 4,059-bp aligned sequences of combined plastid *matK*, *ndhF*, *atpB*, and nuclear *PHYC* genes. Numbers indicate maximum parsimony and likelihood bootstrap values, and Bayesian posterior probability (from top to bottom; shown only when > 50). Asterisks indicate maximal nodal support (100 for all three measures).

Figure S2. Bayesian majority consensus cladogram of *Epicephala* based on 2,933-bp aligned sequences of combined mitochondrial *COI* and nuclear *ArgK*, *EF-1 α* , *Wg*, and *18S rDNA* genes. Numbers indicate parsimony and likelihood bootstrap values, and Bayesian posterior probability (from top to bottom; shown only when > 50). Asterisks indicate maximal nodal support (100 for all three measures). *Epicephala* species are currently all undescribed; thus, host associations are given as taxon labels. Associated host taxonomic groups are also given.

Figure S3. Penalized likelihood chronogram of Phyllanthaceae based on 4,059-bp aligned sequences of combined plastid *matK*, *ndhF*, *atpB*, and nuclear *PHYC* genes. Maximum age constraint (*a*) and minimum age fossil constraints (*b–e*) are indicated. 95% confidence intervals are shown with shaded bars.

Figure S4. Penalized likelihood chronogram of *Epicephala* based on 2,933-bp aligned sequences of combined mitochondrial *COI* and nuclear *ArgK*, *EF-1 α* , *Wg*, and *18S rDNA* genes. Branches are scaled to one arbitrary time unit fixed at the root node. 95% confidence intervals are shown with shaded bars.

Table S1. Presence/absence of *Epicephala* larvae in fruits and eggs in flowers

Species	Locality	Fruits		Fruits infested (%)	Flowers		
		No. of fruits (plants)	Ovules per fruit [†]		No. of flowers (plants)	No. of flowers pollinated	Pollinated with eggs (%)
<i>Margaritaria</i>							
<i>M. discoidea</i>	Bossou	28 (1)	6	0	n. a.	n. a.	n. a.
<i>Flueggea</i>							
<i>F. suffruticosa</i>	Hyogo	461 (4)	6	51	379 (2)	379	13.2
	Hiroshima	n. a.	n. a.	n. a.	393 (8)	365	6.3
	Amami Is.	115 (2)	6	0	n. a.	n. a.	n. a.
<i>F. virosa</i>	Vieng Xai	243 (3)	6	0	n. a.	n. a.	n. a.
<i>Phyllanthus</i>							
<i>P. (Is.) ussuriensis</i>	Tokyo	29 (12)	6	44.8	n. a.	n. a.	n. a.
	Kyoto	148 (6)	6	0	275 (6)	217	0
<i>P. (Is.) virgatus</i>	Vientiane	233 (31)	6	0	n. a.	n. a.	n. a.
<i>P. (Er.) liukuensis</i>	Okinawa Is.	139 (16)	6	0	82 (11)	53	0
<i>P. (Er.) pulcheroides</i>	Mahaxai	53 (6)	6	0	38 (4)	17	0
<i>P. (Ki.) flexuosus</i>	Kyoto	193 (4)	6	0	81 (4)	72	0
	Hyogo	51 (1)	6	0	n. a.	n. a.	n. a.
	Miyazaki	79 (2)	6	0	n. a.	n. a.	n. a.
<i>P. (Ki.) oligospermus</i>	Yonaguni Is.	123 (5)	6	0	182 (3)	167	0
<i>P. (Ki.) reticulatus</i>	Henchun	61 (3)	16.4 (12–20)	78.7	251 (3)	120	89.2
<i>P. (Ki.) sp.</i>	Lakxao	n. a.	n. a.	n. a.	170 (2)	54	94.4
<i>P. (Ki.) tenellus</i>	Okinawa Is.	322 (7)	6	0	190 (10)	169	0
<i>P. (Ph.) lepidocarpus</i>	Kyoto	100 (5)	6	0	106 (5)	104	0
	Miyako Is.	184 (10)	6	17.4	129 (10)	114	0
	Ishigaki Is.	207 (10)	6	49.8	176 (10)	162	0
<i>P. (Ph.) amarus</i>	Ishigaki Is.	212 (6)	6	0	171 (6)	148	0
<i>P. (Ph.) debilis</i>	Ishigaki Is.	76 (7)	6	0	68 (7)	62	0
<i>P. (Ci.) acidus</i>	Vientiane	200 (2)	6	0	n. a.	n. a.	n. a.
<i>P. (Em.) emblica</i>	Ban Chomesy	111 (2)	6	0	n. a.	n. a.	n. a.
<i>P. (Pd.) roseus</i>	Phialat	53 (3)	6	0	158 (3)	120	0
<i>P. marojejiensis</i>	Mt. Marojeji	49 (6)	6.2 (6–8)	77.8	116 (11)	59	56
<i>P. humberitii</i>	Mt. Marojeji	27 (5)	6	88.9	n. a.	n. a.	n. a.
<i>Reverchonia</i>							
<i>R. arenaria</i>	New Mexico	266 (18)	6	0	n. a.	n. a.	n. a.
<i>Breynia</i>							
<i>B. retusa</i>	Vientiane	138 (4)	6	0	226 (4)	112	0
<i>Sauropus</i>							
<i>S. quadrangularis</i>	Vientiane	43 (11)	6	0	128 (13)	73	0
<i>S. brevipes</i>	Vientiane	19 (8)	6	0	88 (11)	37	0
<i>S. androgynus</i>	Thakhaek	78 (6)	6	0	70 (6)	34	0
<i>S. granulosus</i>	Vientiane	n. a.	n. a.	n. a.	25 (5)	15	0

[†]Mean and range in parentheses for species with variable ovule number.

Table S2. Associations between pollination and presence/absence of *Epicephala* eggs in female flowers.

Species		No eggs	With eggs	Significance [†]
Locality (No. of plants)				
<i>Flueggea suffruticosa</i>				
Hyogo (2)	Pollinated	329	50	$P = 1$
	Un-pollinated	0	0	
Hiroshima (8)	Pollinated	342	23	$P = 0.1735$
	Un-pollinated	28	0	
<i>Phyllanthus reticulatus</i>				
Henchun (3)	Pollinated	13	107	$P < 0.0001$
	Un-pollinated	131	0	
<i>Phyllanthus</i> sp.				
Laksao (2)	Pollinated	3	51	$P < 0.0001$
	Un-pollinated	116	0	
<i>Phyllanthus marojejiensis</i>				
Mt. Marojeji (11)	Pollinated	3	56	$P < 0.0001$
	Un-pollinated	57	0	

[†]Significance of association between pollination and egg presence assessed by Fisher's exact test.

Table S3. Flower visitors of Phyllanthaeae plants

Genus Species	Study site	Hours observed		Insect species	Family: order [†]	Visitation rate [§]	Pollen attachment [‡]
		Day	Night				
<i>Flueggea</i>							
<i>F. suffruticosa</i>	Hyogo	0.2	11.0	<i>Adoretus tenuimaculatus</i>	Sca: Col	5.0	++
				<i>Eucetonia roelofsi</i>	Sca: Col	5.0	+++
				<i>Betasyrphus serarius</i>	Syr: Dip	5.0	+++
				<i>Phytomya zonata</i>	Syr: Dip	5.0	+++
				<i>Sphaerophoria philanthus</i>	Syr: Dip	25.0	+++
				<i>Odontomyia garatas</i>	Str: Dip	5.0	+++
				<i>Stomorhina obsoleta</i>	Cal: Dip	130.0	+++
				<i>Phasia crassipennis</i>	Pha: Dip	5.0	+++
				<i>Lasioglossum</i> sp. 1	Hal: Hym	10.0	+++
				<i>Lasioglossum</i> sp. 2	Hal: Hym	45.0	+++
				<i>Lasioglossum</i> sp. 3	Hal: Hym	70.0	+++
				<i>Apis cerana</i>	Api: Hym	65.0	+++
				<i>Epicephala</i> sp. 1	Gra: Lep	3.5	
				<i>F. suffruticosa</i>	Amami Is.	0.3	<i>Stomorhina obsoleta</i>
<i>Ophyra</i> sp.	Ant: Dip	3.3	+				
<i>Homoneura</i> sp.	Lau: Dip	3.3	+++				
<i>Polistes jokahamae</i>	Ves: Hym	6.7	+++				
<i>Lasioglossum</i> sp. 4	Hal: Hym	16.7	+++				
<i>F. virosa</i>	Fangliao	0.3	<i>Eristalinus quinquestriatus</i>	Syr: Dip	6.7	+++	
			<i>Ischiodon scutellaris</i>	Syr: Dip	6.7	++	
			<i>Paragus</i> sp.	Syr: Dip	10.0	++	
			<i>Stomorhina obsoleta</i>	Cal: Dip	50.0	+++	
			<i>Chrysomya</i> sp. 1	Cal: Dip	3.3	+++	
			<i>Chrysomya</i> sp. 2	Cal: Dip	10.0	+++	
			<i>Polleniopsis</i> sp.	Cal: Dip	6.7	+++	
			<i>Parasarcophaga</i> sp.	Sar: Dip	10.0	+++	
			<i>Siphona</i> sp.	Tac: Dip	20.0	+++	
			<i>Braunsapis</i> sp.	Api: Hym	3.3	+++	
			<i>Apis cerana</i>	Api: Hym	20.0	+++	
			<i>Apis mellifera</i>	Api: Hym	16.7	+++	
			<i>Amata</i> sp.	Arc: Lep	3.3	+++	
			<i>Princeps demoleus</i>	Pap: Lep	3.3	+++	
			<i>Eurema hecabe</i>	Pie: Lep	3.3	+	
			<i>Precis lemonias</i>	Nym: Lep	3.3	+	
			<i>Precis iphita</i>	Nym: Lep	3.3	+++	

				<i>Hypolimnas misippus</i>	Nym: Lep	3.3		+
<i>Phyllanthus</i>								
<i>P. (Is.) ussuriensis</i>	Kyoto	1.0		<i>Tetramorium</i> sp. 1	For: Hym	5.0		+
				<i>Tetramorium</i> sp. 2	For: Hym	2.0		
<i>P. (Is.) virgatus</i>	Vientiane	0.3		<i>Paragus</i> sp.	Syr: Dip	6.7		++
				<i>Camponotus</i> sp.	For: Hym	13.3		+
				<i>Solenopsis</i> sp.	For: Hym	10.0		+
<i>P. (Ki.) flexuosus</i>	Kyoto	8.0		<i>Pidonia puziloi</i>	Cer: Col	0.5		+++
				<i>Pidonia simillima</i>	Cer: Col	0.3		+++
				<i>Ezonacerda</i> sp.	Oed: Col	0.1		+++
				sp.	Cec: Dip	2.4		++
				<i>Rhamphomyia formidabilis</i>	Emp: Dip	0.3		++
				<i>Episyrphus balteatus</i>	Syr: Dip	0.1		+++
				<i>Sphegina</i> sp.	Syr: Dip	0.1		+++
				<i>Graptomyza itoi</i>	Syr: Dip	0.1		++
				<i>Phorticella</i> sp.	Dro: Dip	0.1		++
				<i>Spilichneumon nonagriæ</i>	Ich: Hym	0.1		+
<i>P. (Ki.) oligospermus</i>	Yonaguni Is.	3.0		<i>Episyrphus</i> sp.	Syr: Dip	0.7		++
				<i>Chrysomya</i> sp. 3	Cal: Dip	0.3		+++
				<i>Rhinia zanthogaster</i>	Cal: Dip	0.3		+
				<i>Lasioglossum</i> sp. 5	Hal: Hym	0.3		+++
<i>P. (Ki.) reticulatus</i>	Henchun	5.0	7.0	<i>Epicephala</i> sp. 2	Gra: Lep	2.9		+++
<i>P. (Ki.) tenellus</i>	Okinawa Is.	0.5		—				
<i>P. (Ph.) lepidocarpus</i>	Kyoto	0.5		<i>Formica japonica</i>	For: Hym	22.0		+++
				<i>Formica</i> sp.	For: Hym	24.0		++
	Miyako Is.	0.3	2.0	<i>Tetramorium</i> sp. 3	For: Hym	24.0		++
				<i>Anoplolepis gracilipes</i>	For: Hym	3.3		+
				<i>Epicephala</i> sp. 3 (on young fruit)	Gra: Lep	6.5		
	Ishigaki Is.	0.3		<i>Technomyrmex</i> sp.	For: Hym	10.0		++
				<i>Tapinoma</i> sp.	For: Hym	10.0		++
				<i>Tetramorium</i> sp. 4	For: Hym	26.7		++
<i>P. (Ph.) amarus</i>	Ishigaki Is.	0.3		<i>Monomorium</i> sp. 1	For: Hym	20.0		++
	Thakhaek		1.5	<i>Epicephala</i> sp. 4 (on young fruit)	Gra: Lep	4.0		
<i>P. (Ph.) debilis</i>	Ishigaki Is.	0.3		<i>Monomorium</i> sp. 2	For: Hym	16.7		++
<i>P. (Ph.) liukuensis</i>	Okinawa Is.	3.0		<i>Mallochohelea</i> sp.	Crt: Dip	0.7		+
<i>P. (Pd.) roseus</i>	Phialat	2.0		sp.	Cur: Col	0.5		++
				<i>Stomorhina obsoleta</i>	Cal: Dip	2.5		+++
				sp.	Lau: Dip	0.5		++
<i>P. marojejiensis</i>	Mt. Marojeji	4.0	4.0	<i>Epicephala</i> sp. 5 (on leaf)	Gra: Lep	0.3		+++

<i>Reverchonia</i>							
<i>R. arenaria</i>	New Mexico	3.0	sp.	Bru: Col	0.3		
			<i>Sphixapata trilineata</i>	Sar: Dip	1.0	+	
			<i>Lasioglossum</i> sp. 6	Hal: Hym	0.3	+	
<i>Breynia</i>							
<i>B. retusa</i>	Vientiane	2.0	<i>Clinodiplosis</i> sp.	Cec: Dip	4.5	++	
<i>Sauropus</i>							
<i>S. quadrangularis</i>	Vientiane	8.0	<i>Paragus</i> sp.	Syr: Dip	0.5	+++	
<i>S. brevipes</i>	Vientiane	4.0	—				
<i>S. androgynus</i>	Thakhaek	1.0	—				
<i>S. granulosis</i>	Vientiane	2.0	<i>Halictus</i> sp.	Hal: Hym	0.5	+++	

[†]Abbreviations for family and order names: Ant, Anthomyiidae; Api, Apidae; Arc, Arctiidae; Bru, Bruchidae; Cal, Calliphoridae; Cec, Cecidomyiidae; Cer, Cerambycidae; Crt, Ceratopogonidae; Coc, Coccinellidae; Cur, Curculionidae; Dro, Drosophilidae; Emp, Empididae; For, Formicidae; Gra, Gracillariidae; Hal, Halictidae; Ich, Ichneumonidae; Lau, Lauxaniidae; Mus, Muscidae; Nit, Nitidulidae; Nym, Nymphalidae; Oed, Oedemeridae; Pap, Papilionidae; Pha, Phasiidae; Pie, Pieridae; Pla, Plataspidae; Sar, Sarcophagidae; Sca, Scarabaeidae; Str, Stratiomyidae; Syr, Syrphidae; Ves, Vespidae; Col, Coleoptera; Dip, Diptera; Hem, Hemiptera; Hym, Hymenoptera; Lep, Lepidoptera.

[§]Visitation rate is given as the number of individuals collected per plant per hour for each insect species.

[‡]Amount of pollen load is assessed as: +, <10 grains; ++, 10–99 grains; +++, ≥100 grains.

Table S4. List of *matK* primers

Primer name [†]	Strand	Sequence (5' to 3')
phy-matk1	F	CTTHTNNTTTCGGGARTATATT
phy-matk6	F	TAAARCGTTTAATTACTIONCG
phy-matk3 [§]	F	GRTTYTTTCTTCACGAGTATTG
phy-matk4 [§]	R	CGAGRGAYTGYYTTCGATAATG
phy-matk2	R	ACAAAYAATATCMAAATACC

[†]Primers are located in this order from the 5'-end to the 3'-end.

[§]Used for sequencing only.

Table S5. GenBank accession numbers for the plant sequences used in this study.

	<i>matK</i>	<i>ndhF</i>	<i>atpB</i>	<i>PHYC</i>
Ingroup				
<i>Margaritaria discoidea</i>	FJ235280	FJ235490	FJ235326	FJ235372
<i>Margaritaria indica</i>	FJ235279	FJ235489	FJ235325	FJ235371
<i>Flueggea jullienii</i>	FJ235278	FJ235488	FJ235324	FJ235370
<i>Flueggea suffruticosa</i>	FJ235276	FJ235486	FJ235322	FJ235368
<i>Flueggea virosa</i>	FJ235277	FJ235487	FJ235323	FJ235369
<i>Phyllanthus ussuriensis</i>	FJ235274	FJ235484	FJ235320	FJ235366
<i>Phyllanthus virgatus</i>	FJ235275	FJ235485	FJ235321	FJ235367
<i>Phyllanthus liukuensis</i>	FJ235272	FJ235482	FJ235318	FJ235364
<i>Phyllanthus pulcheroides</i>	FJ235273	FJ235483	FJ235319	FJ235365
<i>Phyllanthus reticulatus</i>	FJ235270	FJ235480	FJ235316	FJ235362
<i>Phyllanthus</i> sp.	FJ235271	FJ235481	FJ235317	FJ235363
<i>Phyllanthus flexuosus</i>	FJ235269	FJ235479	FJ235315	FJ235361
<i>Phyllanthus oligospermus</i>	FJ235268	FJ235478	FJ235314	FJ235360
<i>Phyllanthus tenellus</i>	FJ235267	FJ235477	FJ235313	FJ235359
<i>Phyllanthus amarus</i>	FJ235264	FJ235474	FJ235310	FJ235356
<i>Phyllanthus debilis</i>	FJ235265	FJ235475	FJ235311	FJ235357
<i>Phyllanthus lepidocarpus</i>	FJ235252	FJ235462	FJ235298	FJ235344
<i>Phyllanthus aeneus</i>	FJ235260	FJ235470	FJ235306	FJ235352
<i>Phyllanthus gneissicus</i>	FJ235262	FJ235472	FJ235308	FJ235354
<i>Phyllanthus guillauminii</i>	FJ235263	FJ235473	FJ235309	FJ235355
<i>Phyllanthus vulcani</i>	FJ235261	FJ235471	FJ235307	FJ235353
<i>Phyllanthus bourgeoisii</i>	FJ235256	FJ235466	FJ235302	FJ235348
<i>Phyllanthus chamaecerasus</i>	FJ235255	FJ235465	FJ235301	FJ235347
<i>Phyllanthus caudatus</i>	FJ235259	FJ235469	FJ235305	FJ235351
<i>Phyllanthus koniamboensis</i>	FJ235258	FJ235468	FJ235304	FJ235350
<i>Phyllanthus mangenotii</i>	FJ235257	FJ235467	FJ235303	FJ235349
<i>Phyllanthus acidus</i>	FJ235250	FJ235460	FJ235296	FJ235342
<i>Phyllanthus emblica</i>	FJ235251	FJ235461	FJ235297	FJ235343
<i>Phyllanthus roseus</i>	FJ235240	FJ235450	FJ235286	FJ235332
<i>Phyllanthus marojejiensis</i>	FJ235254	FJ235464	FJ235300	FJ235346
<i>Phyllanthus humbertii</i>	FJ235253	FJ235463	FJ235299	FJ235345
<i>Reverchonia arenaria</i>	FJ235266	FJ235476	FJ235312	FJ235358
<i>Sauropus androgynus</i>	FJ235247	FJ235457	FJ235293	FJ235339
<i>Sauropus brevipes</i>	FJ235248	FJ235458	FJ235294	FJ235340
<i>Sauropus granulatus</i>	FJ235246	FJ235456	FJ235292	FJ235338
<i>Sauropus quadrangularis</i>	FJ235249	FJ235459	FJ235295	FJ235341
<i>Breynia disticha</i>	FJ235241	FJ235451	FJ235287	FJ235333
<i>Breynia fruticosa</i>	FJ235245	FJ235455	FJ235291	FJ235337
<i>Breynia oblongifolia</i>	FJ235242	FJ235452	FJ235288	FJ235334
<i>Breynia retusa</i>	FJ235244	FJ235454	FJ235290	FJ235336
<i>Breynia vitis-idaea</i>	FJ235243	FJ235453	FJ235289	FJ235335
<i>Glochidion acuminatum</i>	FJ235239	FJ235449	FJ235285	FJ235331
<i>Glochidion lanceolatum</i>	FJ235235	FJ235445	FJ235281	FJ235327
<i>Glochidion obovatum</i>	FJ235238	FJ235448	FJ235284	FJ235330
<i>Glochidion rubrum</i>	FJ235236	FJ235446	FJ235282	FJ235328
<i>Glochidion zeylanicum</i>	FJ235237	FJ235447	FJ235283	FJ235329
Outgroup (Phyllanthaceae)				
<i>Actephila lindleyi</i>	AY552415		AY830190	AY579830
<i>Amanoa strobilacea</i>	AY830258	AY830289	AY830191	AY830365
<i>Andrachne aspera</i>	AY830259	AY830290	AY830192	AY830366

<i>Andrachne ovalis</i>	AY830260	AY830291	AY830193	AY830367
<i>Antidesma alexiteria</i>	AY552416	AY830292	AY830194	AY579832
<i>Apodiscus chevalieri</i>	AY552418	AY830294	AY830195	AY579834
<i>Aporosa frutescens</i>	AY552417	AY830295	AY830196	AY579835
<i>Astrocasia neurocarpa</i>	AY830261	AY830296	AY830197	AY830368
<i>Baccaurea javanica</i>	AY579878	AY830297	AY830198	AY579836
<i>Baccaurea lanceolata</i>	AY552419	AY830298	AY830199	AY579837
<i>Bischofia javanica</i>	AY552420	AY830299	AY830200	AY579838
<i>Blotia leandriana</i>	AY552440	AY830300	AY830201	AY579862
<i>Bridelia ferruginea</i>	AY830262	AY830303	AY830204	AY830370
<i>Bridelia insulana</i>	AY830263	AY830304	AY830295	
<i>Bridelia retusa</i>	AY552421	AY830305	AY830206	AY579841
<i>Cleistanthus oblongifolius</i>	AY552424	AY830307		AY579843
<i>Cleistanthus perrieri</i>	AY552425	AY830308	AY830207	AY579842
<i>Cleistanthus suarezensis</i>	AY830265	AY830309	AY830208	
<i>Croizatia brevipetiolata</i>		AY830310	AY788213	AY830371
<i>Discocarpus essequaboensis</i>	AY830267	AY830312	AY830209	
<i>Gonatogyne brasiliensis</i>	AY552429	AY830316	AY830213	AY579850
<i>Heywoodia lucens</i>	AY552430	AY830317	AY830214	AY579851
<i>Hieronyma oblonga</i>	AY830268	AY830318	AY830215	
<i>Hymenocardia acida</i>	AY830269	AY830319	AY830216	AY830373
<i>Lachnostylis bilocularis</i>	AY552431	AY830321	AY830218	AY579854
<i>Leptopus colchicus</i>	AY552434	AY830322	AY830220	AY579856
<i>Leptopus cordifolius</i>	AY552433	AY830323	AY830219	AY579855
<i>Leptopus diplopermus</i>	AY830271	AY830325	AY830221	
<i>Maesobotrya vermeulenii</i>	AY830273	AY830327	AY830224	
<i>Martretia quadricornis</i>	AY830275	AY830331	AY830228	AY830377
<i>Meineckia phyllanthoides</i>	AY552436	AY830332	AY830229	AY579857
<i>Petalodiscus bojerianus</i>	AY552439	AY830351	AY830247	AY579860
<i>Petalodiscus fadenii</i>	AY552437	AY830334	AY830231	AY579859
<i>Petalodiscus platyrachis</i>	AY552441	AY830335	AY830232	AY579861
<i>Poranthera corymbosa</i>	AY552447		AY830239	AY579870
<i>Poranthera microphylla</i>		AY830342	AY830240	AY830379
<i>Pseudolachnostylis maprouneifolia</i>	AY552448	AY830344	AY830241	AY579871
<i>Savia dictyocarpa</i>	AY552449	AY830352	AY830248	AY579873
<i>Securinea durissima</i>	AY830285	AY830353	AY830249	AY830382
<i>Spondianthus preussii</i>	AY830286	AY830354	AY830250	AY830383
<i>Thecacoris madagascariensis</i>	AY552452	AY830355	AY830251	AY579874
<i>Uapaca guineensis</i>	AY830287	AY830356	AY830390	AY830385
<i>Uapaca littoralis</i>	AY830288	AY830357	AY830252	AY830384
<i>Wielandia elegans</i>	AY552454	AY830358	AY830253	AY579875
<i>Zimmermannia capillipes</i>	AY552455	AY830359	AY830254	AY579876
<i>Zimmermanniopsis uzungwaensis</i>	AY552456	AY830360	AY830255	AY579877
Outgroup (Picrodendraceae)				
<i>Androstachys johnsonii</i>	AY552461	AY830361	AF209527	AY579831
<i>Hyaenanche globosa</i>	AY552462	AY830363		AY579852
Outgroup (Putranjivaceae)				
<i>Drypetes madagascariensis</i>	AY552458	AY830362	AY830256	AY579845
<i>Putranjiva zeylanica</i>	AY552459	AY830364	AY830257	AY830386

Table S6. GenBank accession numbers for the moth sequences used in this study.

	COI	ArgK	EF-1a	Wg	18S rDNA
Ingroup (<i>Epicephala</i>)					
<i>E. sp. ex Flueggea suffruticosa</i>	FJ235373	FJ235392	FJ235491	FJ235206	FJ235416
<i>E. sp. ex Phyllanthus ussuriensis</i>	FJ235387	FJ235411	FJ235510	FJ235225	FJ235435
<i>E. sp. ex Phyllanthus reticulatus</i>	FJ235383	FJ235407	FJ235506	FJ235221	FJ235431
<i>E. sp. ex Phyllanthus sp.</i>	FJ235382	FJ235406	FJ235505	FJ235220	FJ235430
<i>E. sp. ex Phyllanthus amarus</i>	FJ235388	FJ235412	FJ235511	FJ235226	FJ235436
<i>E. sp. ex Phyllanthus lepidocarpus</i>	FJ235386	FJ235410	FJ235509	FJ235224	FJ235434
<i>E. sp. ex Phyllanthus aeneus</i>	AY269393	FJ235401	FJ235500	FJ235215	FJ235425
<i>E. sp. ex Phyllanthus gneissicus</i>	FJ235376	FJ235398	FJ235497	FJ235212	FJ235422
<i>E. sp. ex Phyllanthus guillauminii</i>	AY269399	FJ235399	FJ235498	FJ235213	FJ235423
<i>E. sp. ex Phyllanthus vulcani</i>	FJ235377	FJ235400	FJ235499	FJ235214	FJ235424
<i>E. sp. ex Phyllanthus bourgeoisii</i>	AY269404	FJ235395	FJ235494	FJ235209	FJ235419
<i>E. sp. ex Phyllanthus chamaecerasus</i>	AY269411	FJ235397	FJ235496	FJ235211	FJ235421
<i>E. sp. ex Phyllanthus caudatus</i>	FJ235375	FJ235396	FJ235495	FJ235210	FJ235420
<i>E. sp. ex Phyllanthus koniamboensis</i>	FJ235374	FJ235394	FJ235493	FJ235208	FJ235418
<i>E. sp. ex Phyllanthus mangenotii</i>	AY269397	FJ235393	FJ235492	FJ235207	FJ235417
<i>E. sp. ex Phyllanthus marojejiensis</i>	FJ235378	FJ235402	FJ235501	FJ235216	FJ235426
<i>E. sp. ex Phyllanthus humbertii</i>	FJ235379	FJ235403	FJ235502	FJ235217	FJ235427
<i>E. sp. ex Breynia disticha</i>	FJ235381	FJ235405	FJ235504	FJ235219	FJ235429
<i>E. sp. ex Breynia fruticosa</i>	FJ235380	FJ235404	FJ235503	FJ235218	FJ235428
<i>E. sp. ex Breynia oblongifolia</i>	FJ235384	FJ235408	FJ235507	FJ235222	FJ235432
<i>E. sp. ex Breynia vitis-idaea</i>	FJ235385	FJ235409	FJ235508	FJ235223	FJ235433
<i>E. sp. ex Glochidion acuminatum</i>	DQ298955	DQ298844	DQ299051	FJ235229	FJ235439
<i>E. sp. ex Glochidion lanceolatum</i>	DQ298967	DQ298858	DQ299065	FJ235227	FJ235437
<i>E. sp. ex Glochidion obovatum</i>	DQ299009	DQ298903	DQ299110	FJ235230	FJ235440
<i>E. sp. ex Glochidion rubrum</i>	DQ299027	DQ298924	DQ299131	FJ235228	FJ235438
<i>E. sp. ex Glochidion zeylanicum</i>	DQ299039	DQ298937	DQ299144	FJ235231	FJ235441
Outgroup					
<i>Cuphodes diospyrosella</i>	FJ235389	FJ235413	FJ235512	FJ235232	FJ235442
<i>Stomphastis labyrinthica</i>	FJ235390	FJ235414	FJ235513	FJ235233	FJ235443
<i>Melanocercops ficuvorella</i>	FJ235391	FJ235415	FJ235514	FJ235234	FJ235444

Table S7. Phyllanthaceae fossil records. Fossils indicated in bold were used to constrain node ages in the dating analysis.

Extant taxon	Fossil taxon	Fossil type	Epoch, stage	Age (mya) [†]	Location	Reference
<i>Glochidion</i>	<i>Glochidion miocenica</i>	Leaf	Middle Miocene	11.6	Kathgodam, Uttar Pradesh, India	Prasad (1994)
<i>Glochidion</i>	<i>Glochidion palaeohirsutum</i>	Leaf	Middle Miocene	11.6	Darjeeling, West Bengal, India	Antal and Prasad (1996)
<i>Phyllanthus</i>	<i>Phyllanthus wuhanensis</i>	Wood	Pliocene	1.8	Wuhan, Hubei, China	Yang et al. (1998)
<i>Phyllanthus</i>	<i>Phyllanthus palaeoreticulatus</i>	Leaf	Miocene	5.3	Suraikhola, Nepal	Prasad and Awasthi (1996)
<i>Phyllanthus</i>	<i>Phyllanthus palaeoreticulatus</i>	Leaf	Miocene	5.3	Tamil Nadu, India	Agarwal (2002)
<i>Phyllanthus</i>	<i>Phyllanthus securinegiformis</i>	Seed	Miocene	5.3	Lower Lusatia, Germany	Mai (1996)
<i>Phyllanthus</i>	<i>Phyllanthus sexseminata</i>	Seed	Late Miocene, Tortonian	7.2	Gdow Bay, Poland	Lancucka-Srodoniowa (1966)
<i>Phyllanthus</i>	<i>Phyllanthus mioreticulatus</i>	Leaf	Middle Miocene	11.6	Koilabas, Nepal	Prasad et al. (1999)
<i>Phyllanthus</i>	<i>Phyllanthus koilabesensis</i>	Leaf	Middle Miocene	11.6	Koilabas, Nepal	Prasad et al. (1999)
<i>Phyllanthus</i>	<i>Phyllanthus siwalica</i>	Leaf	Middle Miocene	11.6	Kathgodam, Uttar Pradesh, India	Prasad (1994)
<i>Phyllanthus</i>	<i>Phyllanthus</i> type	Leaf	Early Miocene	16.0	Mampui, Mizoram, India	Agarwal and Mandaokar (2002)
<i>Phyllanthus</i>	<i>Phyllanthus kireevskiana</i>	Seed	Oligocene	23.0	Kireevsk, Russia	Dorofeev (1960)
<i>Phyllanthus</i>	<i>Phyllanthus triquetra</i>	Seed	Oligocene	23.0	Kozyulino, Tomsk, Russia	Dorofeev (1959)
<i>Phyllanthus</i>	<i>Phyllanthus compassica</i>	Seed	Late Oligocene, Chattian	23.0	Rezhenka, Tomsk, Russia	Dorofeev (1958)
<i>Phyllanthus</i>	<i>Phyllanthus</i> type	Pollen	Late Eocene	33.9	Cape Verde Rise, Atlantic Ocean	Zaklinskaja (1978)
<i>Phyllanthus</i>	<i>Reverchonia</i> type	Pollen	Late Eocene	33.9	Mont Pagnotte, Oise, France	Gruas-Cavagnetto and Köhler (1992)
<i>Phyllanthus</i>	<i>Phyllanthus</i> type	Pollen	Early Eocene	48.6	New Haven, East Sussex, UK	Gruas-Cavagnetto and Köhler (1992)
Poranthereae	<i>Actephila</i> type	Pollen	Late Eocene	33.9	Celas, Gard, France	Gruas-Cavagnetto and Köhler (1992)
<i>Bridelia</i>	<i>Bridelia wuhanensis</i>	Wood	Pliocene	1.8	Wuhan, Hubei, China	Yang et al. (1998)
<i>Bischofia</i>	<i>Bischofia polycarpa</i>	Wood	Pliocene	1.8	Wuhan, Hubei, China	Yang et al. (1998)
<i>Bischofia</i>	<i>Bischofia javanoxyla</i>	Wood	Early Miocene	16.0	Kweishan, Taoyuan, Taiwan	Li et al. (2003)
<i>Bischofia</i>	<i>Bischofia</i> type	Pollen	Middle Eocene, Bartonian	37.2	Manche, France	Gruas-Cavagnetto and Köhler (1992)

[†]Oldest reliable age of the fossil given as the upper boundary of each epoch/stage (geologic times based on Gradstein et al., 2004).

Table S8. Results of selective exclusion experiments in *Phyllanthus lepidocarpus*

	Plants	Flowers per plant	Fruit set [†]
Control	15	20–32	99.4 ^a
Bagged with fine mesh	15	26–38	0.0 ^b
Only ants allowed to forage	15	22–39	99.1 ^a
Hand self-pollinated	5	15	100.0 ^a

[†]Fruit sets in the four treatments were significantly different (Kruskal-Wallis test, $\chi^2 = 40.01$, $df = 3$, $P < 0.001$). Letters with different superscript letters indicate significant difference.







