THE BEHAVIORAL ECOLOGY AND EVOLUTION OF KLEPTOPARASITISM IN AUSTRALIAN GALL THRIPS

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ABSTRACT

We used a combination of behavioral-ecological and molecular-phylogenetic data to analyze the origin and diversification of kleptoparasitic (gall-stealing) thrips in the genus Koptothrips, which comprises four described species that invade and breed in galls induced by species of Oncothrips and Kladothrips on Australian Acacia. The genus Koptothrips is apparently monophyletic and not closely related to its hosts. Two of the species, K. dyskritus and K. flavicornis, each appears to represent a suite of closely-related sibling species or host races. Three of the four Koptothrips species are facultatively kleptoparasitic, in that females can breed within damaged, open galls by enclosing themselves within cellophane-like partitions. Facultative kleptoparasitism may have served as an evolutionary bridge to the obligately kleptoparasitic habit found in K. flavicornis. Evidence from phylogenetics, and Acacia host-plant relationships of the kleptoparasites and the gall-inducers, suggests that this parasite-host system has undergone some degree of cospeciation, such that speciations of Koptothrips have tracked the speciations of the gall-inducers. Quantification of kleptoparasitism rates indicates that Koptothrips and other enemies represent extremely strong selective pressures on most species of gall-inducers. Although the defensive soldier morphs found in some gall-inducing species can successfully defend against Koptothrips invasion, species with soldiers are still subject to high rates of successful kleptoparasite attack. Gall-inducing thrips exhibit three main types of life-history adaptations that have apparently evolved in response to kleptoparasite pressure: (1) “fighters”, which exhibit long-lived galls and soldier morphs, (2) “runners”, which have quite short-lived galls, from which offspring disperse as second instar larvae, and (3) “hiders”, whose galls are long-lived, especially tight-sealing, and induced on a taxonomically-distinct group of Acacia host plants that is seldom attacked.

Key Words: Acacia, cladistics, gall thrips, kleptoparasites, sociality

RESUMEN

Para analizar el origen y la diversificación de trips cleptoparásitos del género Koptothrips, se estudiaron aspectos sobre su comportamiento, ecología, y filogenética molecular. Este género tiene cuatro especies descritas que invaden y se crían dentro de agallas inducidas por especies de Oncothrips y Kladothrips en Acacia spp. australianas. El género Koptothrips parece ser monofilético y no estrechamente relacionado con sus hospederos. Las especies de K. dyskritus y K. flavicornis parecen representar dos grupos de especies estrechamente relacionadas o de razas que difieren en sus hospederos. Tres de las cuatro especies de Koptothrips son cleptoparasiticas facultativas; las hembras pueden criarse dentro de agallas dañadas y abiertas protegiéndose con unos tejidos parecidos al papel celofán. El hábito de cleptoparasitismo facultativo parece haber servido como un puente evolutivo hacia el cleptoparasitismo obligado como el encontrado en K. flavicornis. La evidencia filogenética y de las relaciones entre la Acacia hospedera y los cleptoparasitós y los insectos agalleros, sugiere que este sistema de parasito-hospedero ha tenido algún grado de co-especiación ya que la espe-
ciación de Koptothrips ha copiado la especiación de los agalleros. La cuantificación de las proporciones de cleptoparasitismo indica que Koptothrips y otros enemigos naturales representan presiones selectivas sumamente fuertes sobre la mayoría de las especies de agalleros. Aunque los soldados defensivos que ocurren en algunas especies de trips agalleros pueden defender las agallas con éxito contra la invasión de Koptothrips, las especies con soldados de todos modos están sujetas a altas proporciones de ataque exitoso por cleptoparásitos. Los trips agalleros exhiben tres tipos principales de adaptación que han evolucionado al parecer en respuesta a la presión ejercida por los cleptoparásitos: (1) "peleadores", que producen agallas duraderas y soldados; (2) "escapadores", que producen agallas bastante efímeras y de las cuales la descendencia se dispersa durante los segundos instares larvales; y (3) "escondedores", cuyas agallas son duraderas, selladas firmemente, y que son inducidas en un grupo taxonómicamente distinto de plantas hospedera de Acacia que raramente son atacadas.

To survive, grow, and reproduce, all animals must engage in one or both of two strategies: utilize resources that they themselves obtain, or steal resources from others. Broadly construed, biological criminals include all predators, parasites, and parasitoids, but we usually consider parasitism as the primary example of illicit resource use. Natural selection for parasitic thievery might be expected to increase in strength with the value of the resources used, but so also would natural selection for defense. Indeed, defense against parasites has been considered the main selective pressure for much of the spectacular, beautiful, and stunningly-complex diversity of adaptation, including sex (Hamilton et al. 1990), sexual selection (Hamilton and Zuk 1982; Andersson 1994), social cooperation (Lin 1964; Lin and Michener 1972; Alexander 1986), immune systems, and even multicellularity itself (Frank 1994). How the evolutionary dynamics of host-parasite attack and defense play out for any set of species depends on the variation available for selection, the genetic bases of the variation, and the degree to which each party can control resource use. As such, in-depth analyses of the ecology and evolution of particular host-parasite systems are necessary to uncover the general principles underlying the forms and maintenance of these two strategies.

The purpose of this paper is to describe and test hypotheses for the origin, evolution, and behavioral ecology of kleptoparasites in Australian gall thrips, especially with reference to the evolution of various forms of defense against these enemies. Kleptoparasites are a special subset of natural enemies that usurp valuable physical resources from the creator or obtainer, and thus fit most closely with our human perspective on theft. We first describe the natural history and ecology of kleptoparasitic thrips and their victims, thrips that induce galls on species of Australian Acacia. Second, we present evidence from phylogenetics designed to uncover the evolutionary origins of the kleptoparasitic lifestyle, and the patterns in its diversification. Finally, we present data from behavioral-ecological studies that are focused on understanding the ecology and evolution of these host-kleptoparasite interactions, and we fit these data on processes into our phylogenetic framework.

**Materials and Methods**

Natural history of Australian gall-inducing thrips and their kleptoparasites

A total of 21 described species of gall-inducing thrips in the genera Kladothrips, Oncothrips, and Onychothrips induce galls on phyllodes (petioles modified to function as leaves) of Acacia in the sections Plurinerves, Juliflorae, and Phyllodinae (Mound et al. 1996). Three described species, K. rugosus, O. habrus, and O. waterhousei, apparently
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each represents a suite of host-specific sibling species, inducing galls on different species of plant (Crespi et al. 1998). Galls are induced on young, actively-growing phylloides by single adult macropterous females, and in some Kladothrips species a male sometimes joins a female during gall initiation. During the period from gall initiation until closure, females of Oncothrips tepperi, O. habrus, Kladothrips rugosus, Onychothrips arotrum and Ony. tepperi have been observed to fight with one another over gall ownership, using their enlarged, armed forelegs (Crespi 1992a,b, Mound and Crespi 1995). All other known gall-forming species, except Oncothrips rodwayi and O. antennatus, have notably enlarged forelegs in the females which are also indicative of fighting. Males are also known to fight one another during gall induction in K. rugosus, and their enlarged forelegs suggest that males also fight in K. ellubus and K. acaciae.

After successfully inducing a gall, the enclosed female lays eggs on the inner surface of the gall, which develop into larvae that feed within the gall. Three main forms of life-history are exhibited in the gall-inducers. First, in some species (Kladothrips rugosus, K. acaciae, and K. ellubus), the length of time spent in the gall by developing larvae is short relative to other gall-inducers on Acacia, on the order of 1-2 months, and mature second-instar (i.e., last-instar) larvae apparently leave the gall prior to pupation in the soil (B. Crespi and B. Kranz, personal observation). Second, in some species (Oncothrips tepperi, O. habrus, O. waterhousei, O. morrisi, K. hamiltoni, and K. harpophyllae), some or all of the offspring produced by the foundress develop into wing-reduced or wingless “soldier” morphs, which have enlarged forelegs that they use to defend the gall against interspecific invaders (Crespi 1992b; Mound and Crespi 1995; Crespi et al. 1997; Crespi and Mound 1997). Third, some species (Oncothrips antennatus, O. schwarzi, Onychothrips arotrum, Ony. tepperi, Ony. zygus and K. auggonaxxos) do not exhibit soldier morphs, but the offspring of the foundress develop into a variant of this life cycle is exhibited by Oncothrips sterni, in which a cohort of wingless, larviform, non-soldier adults develop within the gall, and apparently breed and contribute to the production of winged dispersers (Mound et al. 1996, Crespi and Mound 1997).

The genus Koptothrips comprises four described species, K. xenus, K. zelus, K. dyskritus, and K. flavicornis (Mound 1971, Crespi 1992a, Crespi and Mound 1997), all of which kleptoparasitize galls of the gall-inducers. K. xenus and K. zelus are host-specific, attacking Kladothrips ellubus and K. acaciae respectively, whereas specimens that key to K. dyskritus attack the various sibling species of K. rugosus (and possibly other taxa), and specimens that key to K. flavicornis attack O. tepperi, O. habrus, O. waterhousei, K. rugosus, and several other species. Koptothrips are found in galls of all stages, from recently-founded by the gall-inducer to at least several months old. Galls are usually invaded by single females, which attack and attempt to kill the gall-forming thrips inside. If successful, a female Koptothrips produces a single brood of female and male offspring, which disperse from the gall as adults. In K. zelus and K. xenus, multiple females may invade a gall, in which case the females are each found in a small section of the gall which is partitioned from other sections by a cellophane-like material, apparently produced by the thrips (Crespi and Mound 1997). Females of K. dyskritus have also been collected from within cellophane-like partitions that they create with accessory gland secretions to enclose them within sections of damaged, otherwise-open galls of K. rugosus that no longer contain any of the gall-inducers (Crespi, unpublished data; see also Mound et al. 1997).

Encounters between females of K. flavicornis and K. dyskritus, and adults of various species of Oncothrips and Kladothrips, have been observed in galls in the laboratory (Crespi 1992b, Crespi and Mound 1997). Winged foundresses of the gall-inducers, and soldier morphs in the Oncothrips and Kladothrips with soldiers, will fight the Koptothrips, attempting to grasp and kill them with their enlarged forelegs, which are armed
at the apex with sharp, pointed fore-tarsal teeth. The *Koptothrips* usually fight back, also by grasping and stabbing with their forelegs and fore-tarsal teeth, and if they successfully pierce a gall-inducer with their fore-tarsal teeth, the pierced individual usually dies within a few minutes (Crespi and Mound 1997). Galls successfully invaded by any of the four *Koptothrips* taxa always contain a dead foundress or a dead foundress and founder male, which indicates that invading *Koptothrips* kill the gall-inducers.

Molecular-phylogenetic analysis

The first main goal of our phylogenetic analysis was to assess the taxonomic status of *Koptothrips flavicornis* and *K. dyskritus* collected from galls of host thrips species on different host plants. Each of these two described species may represent either a single, more or less panmictic, generalist species, or a suite of more or less specialized species, each attacking a different species of gall-inducing thrips. For *K. flavicornis*, Mound (1971) noted that specimens collected from the galls of different host thrips often vary considerably in color, ranging from bicolored with a reddish head and yellow abdomen, to brownish, to black. Although mitochondrial DNA sequence data cannot unambiguously indicate species status, levels of divergence between putative taxa can help in achieving this goal (Avise 1994).

The second main goal of our phylogenetic analysis was to evaluate the evolutionary relationships between the *Koptothrips* and their hosts. Alternative hypotheses for these relationships, all of which have been discussed with reference to other taxa (e.g. Bourke and Franks 1991, Choudhary et al. 1994, Ward 1996, Lowe and Crozier 1997, Morris et al. 1998) include: (a) monophyly of the kleptoparasites, and of the hosts, and a lack of sister-taxon status between the two; (b) monophyly, and sister-taxon status, of the kleptoparasites and hosts; (c) monophyly of the kleptoparasites, but paraphyly of the hosts with respect to the kleptoparasites, such that the parasites evolved from within the host lineage; or (d) sister-taxon relationships between each or most of the pairs of hosts and parasites, such that neither is monophyletic. As discussed below, these hypotheses imply different sets of ecological and behavioral mechanisms for the origin and diversification of the hosts and kleptoparasites.

To infer a phylogeny for the gall-inducing thrips, we used a combination of data from mitochondrial DNA sequence from the COI and 16S genes, adult morphological characters, and gall morphology characters (Crespi et al. 1998). For the kleptoparasites, we used about 450 base pairs of mitochondrial DNA sequence from the COI gene for all taxa, with about 250 base pairs of 16S for some taxa. Sequence data for most of the gall-inducers included here is given in Crespi et al. (1998), and all other data (e.g., for all of the *Koptothrips*) is described and analyzed below. Procedures used for DNA isolation, PCR, and sequencing, and sequence for the gall-inducing taxa, are described in Crespi et al. (1998). As described below, we used *Gynaikothrips ficorum* as our outgroup (Crespi et al. 1998), and maximum parsimony analysis and neighbor joining in PAUP 4.0 (Swofford 1998) to analyze the data. We used 500 bootstrap replicates with neighbor joining to assess the robustness of the neighbor joining tree; maximum parsimony bootstrapping was computationally impossible due to the large number of taxa in our data set.

Measurement of kleptoparasitism rates

We collected data on rates of successful kleptoparasitism in the field to evaluate the prevalence and patterns of kleptoparasitism in different host species. Galls were collected from numerous sites throughout Australia (Table 1) directly into 60-100%
ethanol. Any given site includes galls from one to several dozen Acacia trees, and we either collected all galls encountered (when galls were rare), or collected so as to obtain a representative sample of galls with respect to variation in size. For all species, hosts or kleptoparasites in galls from a given site are quite synchronized in their life cycle, as a result of breeding cued by either an annual cycle of new shoot growth, or rainfall.

We dissected galls in the laboratory and recorded the species of thrips inside, and their life-cycle stages present, as well as the presence of other invaders. The predominant non-thysanopteran invaders were lepidopteran larvae (mainly or all species of Lepidoptera in the family Cosmopterigidae, one to a gall), which eat gall tissue from the inside and lead to a drastic reduction in thrips numbers, but usually do not kill all of the gall inhabitants. In some galls, both Koptothrips and a lepidopteran larva were present, but Koptothrips and gall-forming thrips never coexisted alive, except in several galls of Oncothrips antennatus on Acacia adsurgens. We calculated percent Koptothrips invasion as the number of galls containing live Koptothrips divided by total gall number, and made similar calculations for non-thysanopteran invaders (lepidopteran and dipteran larvae). Thus, the kleptoparasitism data refer to rates of suc-

<table>
<thead>
<tr>
<th>Gall-inducing thrips species</th>
<th>Number of collections</th>
<th>Number of galls</th>
<th>% Koptothrips invaders</th>
<th>% Other invaders</th>
</tr>
</thead>
<tbody>
<tr>
<td>*Oncothrips morrisi</td>
<td>1</td>
<td>24</td>
<td>0.29</td>
<td>0</td>
</tr>
<tr>
<td>*Oncothrips waterhousei</td>
<td>2</td>
<td>68</td>
<td>0.32 (0.003)</td>
<td>0.44 (0.09)</td>
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<tr>
<td>*Oncothrips habrus1</td>
<td>5</td>
<td>240</td>
<td>0.23 (0.18)</td>
<td>0.17 (0.07)</td>
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<tr>
<td>*Oncothrips habrus2</td>
<td>2</td>
<td>38</td>
<td>0.08 (0.002)</td>
<td>0.51 (0.37)</td>
</tr>
<tr>
<td>*Oncothrips tepperi</td>
<td>10</td>
<td>423</td>
<td>0.31 (0.26)</td>
<td>0.29 (0.23)</td>
</tr>
<tr>
<td>Oncothrips rodwayi</td>
<td>3</td>
<td>125</td>
<td>0.07 (0.02)</td>
<td>0.30 (0.24)</td>
</tr>
<tr>
<td>*Kladothrips hamiltoni</td>
<td>6</td>
<td>215</td>
<td>0.25 (0.15)</td>
<td>0.36 (0.17)</td>
</tr>
<tr>
<td>*Kladothrips harpophyllae</td>
<td>1</td>
<td>20</td>
<td>0.25</td>
<td>0.35</td>
</tr>
<tr>
<td>Kladothrips rugosus1</td>
<td>6</td>
<td>237</td>
<td>0.19 (0.15)</td>
<td>0.24 (0.19)</td>
</tr>
<tr>
<td>Kladothrips rugosus2</td>
<td>6</td>
<td>171</td>
<td>0.04 (0.04)</td>
<td>0.03 (0.04)</td>
</tr>
<tr>
<td>Kladothrips rugosus3</td>
<td>4</td>
<td>133</td>
<td>0.03 (0.06)</td>
<td>0.11 (0.19)</td>
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<tr>
<td>Kladothrips ellobus</td>
<td>6</td>
<td>147</td>
<td>0.40 (0.26)</td>
<td>0.16 (0.21)</td>
</tr>
<tr>
<td>Kladothrips acacieae</td>
<td>4</td>
<td>84</td>
<td>0.35 (0.30)</td>
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<tr>
<td>Oncothrips antennatus1</td>
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<td>105</td>
<td>0.24 (0.04)</td>
<td>0.19 (0.01)</td>
</tr>
<tr>
<td>Oncothrips antennatus2</td>
<td>10</td>
<td>315</td>
<td>0.02 (0.05)</td>
<td>0.45 (0.25)</td>
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<tr>
<td>Onychothrips arotrum</td>
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<td>229</td>
<td>0</td>
<td>0.16 (0.13)</td>
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<tr>
<td>Onychothrips tepperi</td>
<td>1</td>
<td>11</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

*Oncothrips habrus1 = *O. habrus on Acacia melvillei; *O. habrus2 = on A. pendula; *K. rugosus1 = on A. pendula; *K. rugosus2 = on A. melvillei; *K. rugosus3 = on A. tephrina; *O. antennatus1 = on A. adsurgens; *O. antennatus2 = on A. aneura.
cessful attack and invasion. We have no information concerning what proportion of attacks by *Koptothrips* is unsuccessful, except to note that dead *Koptothrips* adults are occasionally found within galls containing live gall-forming thrips (Crespi 1992a, Mound and Crespi 1995). Many attacks could be aborted, however, before the invader fully enters the gall.

**Results**

**Molecular Phylogenetics**

The percent sequence divergence in the COI gene between *K. flavicornis* collected from different host thrips taxa and *Acacia* species ranged from 0 to 5.6% (with most of the values between 2.5 and 5.6%), and divergences for *K. dyskritus* ranged from 0.5 to 6.9% (with one value of 0.5%, and five values between 5.5% and 6.9%). For *K. flavicornis*, divergences were especially low between specimens collected from two different host thrips species on the same host plant (0.2% for specimens collected from *K. rugosus* and *O. waterhousei* on *A. loderi*, and 0.2% for specimens collected from *K. rugosus* and *O. waterhousei* on *A. ammophila*).

Maximum parsimony analysis of the mitochondrial DNA and morphology data yielded six shortest trees of length 1484. The strict consensus of these trees was well resolved (Fig. 1a) and indicated that the gall-inducing thrips, and the *Koptothrips*, each forms a monophyletic group. Within the *Koptothrips*, each described species was also monophyletic, with *K. xenus* and *K. zelus* most basal, *K. zelus* forming the sister-group to *K. dyskritus*, and (*K. zelus + K. dyskritus*) as sister-group to *K. flavicornis*.

Neighbor-joining analysis of the COI and 16S mitochondrial DNA data yielded a tree that was closely-similar to the maximum parsimony tree, the main differences being the monophyletic status of (*Koptothrips dyskritus + K. xenus + K. zelus*), and sister-taxon status of *Koptothrips xenus* and *K. zelus*, in the neighbor joining tree (Fig. 1b). About half of the nodes in the neighbor-joining bootstrap tree were well-supported by the bootstrap (with support of 70% or higher) (Fig. 1c). In particular, the bootstrap provided strong support for monophyly of the gall-inducers, and monophyly of the kleptoparasites, but it provided relatively weak support for the placements of *K. zelus* and *K. xenus*.

Consideration of the host plants inhabited by the gall-inducing thrips and their kleptoparasites, with respect to their positions in the maximum-parsimony phylogeny, reveals a notable pattern: the relatively basal kleptoparasite species *K. xenus* and *K. zelus* attack the relatively-basal host species *Kladothrips acaciae* and *K. ellobus* respectively. Moreover, all four of these species are host-insect and host-plant specific, and all are quite morphologically distinct from their closest relatives (Mound 1971). This finding suggests that, as described in detail below, the diversification of *Koptothrips* and their gall-inducing hosts has involved some degree of cospeciation. Further evidence for cospeciation is provided by two additional patterns. First, *K. dyskritus*, which apparently descended from the ancestor of (*K. xenus + K. zelus*), attacks primarily *K. rugosus*, which apparently descended from the ancestor of *K. ellobus* and *K. acaciae*. Second, *K. flavicornis*, which is sister-taxon to the other *Koptothrips* taxa, mainly attacks gall-inducing thrips in the (*Oncothrips morrisi + O. waterhousei + O. habrus + O. tepperi + O. rodwayi*) clade, which forms the sister-group to the *Kladothrips* species.

The two *Koptothrips* taxa that appear derived with respect to *K. xenus* and *K. zelus*, *K. dyskritus* and *K. flavicornis*, each attacks multiple species of gall-inducing thrips, and the gall-inducing thrips that they attack, primarily *K. rugosus*, *O. waterhousei*, and *O. habrus*, also each appears to represent a set of closely-related species
Fig. 1. (a) Strict consensus of six most-parsimonious trees of length 1484, inferred using heuristic searching in PAUP 4.0. For the gall-inducing species comprising sibling species or host races on different host plants, the thrips species name is followed by a code for the species name of the Acacia. For the Koptothrips, specimens of K. flavicorne and K. dyskritus collected from galls from different host-thrips species are given unique letter codes. Complete host localities and other collection information is available from BJC.
Fig. 1. (b) Neighbor-joining tree, inferred from the mitochondrial COI and 16S DNA data. For the gall-inducing species comprising sibling species or host races on different host plants, the thrips species name is followed by a code for the species name of the *Acacia*. For the *Koptothrips*, specimens of *K. flavicornis* and *K. dyskritus* collected from galls from different host-thrips species are given unique letter codes. Complete host localities and other collection information is available from BJC.
Fig. 1. (c) Neighbor-joining bootstrap tree (500 replicates). For the gall-inducing species comprising sibling species or host races on different host plants, the thrips species name is followed by a code for the species name of the Acacia. For the Koptothrips, specimens of *K. flavicornis* and *K. dyskritus* collected from galls from different host-thrips species are given unique letter codes. Complete host localities and other collection information is available from BJC.
on different Acacia host-plants (Crespi et al. 1998). However, whereas K. dyskritus primarily attack species of Kladothrips rugosus, K. flavicornis commonly attack O. waterhousei and O. habrus, O. tepperi, O. rodwayi, and sometimes K. rugosus. These data suggest that the diversification of Koptothrips has involved an expansion of host range if K. flavicornis represents a single species, or a radiation involving diverse hosts if it represents a suite of closely-related sibling species.

Mapping of the behavior of Koptothrips onto the maximum-parsimony phylogeny reveals another notable result: Koptothrips xenus, which is one of the three Koptothrips taxa known to be facultatively kleptoparasitic, is basal with respect to the lineage giving rise to the obligately-kleptoparasitic taxon K. flavicornis. Thus, facultative kleptoparasitism is inferred as ancestral for the genus Koptothrips, with one inferred shift to obligate kleptoparasitism. This finding supports the hypothesis that kleptoparasitism in Koptothrips originated as a facultative alternative, and became obligate in association with the speciation event that gave rise to K. flavicornis.

In our neighbor-joining tree, the three facultatively-kleptoparasitic Koptothrips taxa form a monophyletic group, which is sister-taxon to K. flavicornis, and (K. xenus + K. zelus) also forms a monophyletic group. This phylogeny is also broadly compatible with a cospeciation model, in that the two main lineages of hosts, Oncothrips and Kladothrips, are attacked respectively by the two main lineages of kleptoparasites, K. flavicornis and (K. dyskritus + K. zelus + K. xenus), and the K. zelus and K. xenus lineages appear old and divergent relative to the other two Koptothrips species. The weakness of the bootstrap support for the positions of K. xenus and K. zelus in this tree indicates that we cannot consider the results of the maximum-parsimony and neighbor-joining analysis incompatible. These analyses also tell us that additional data from a more slowly-evolving molecule would help to resolve the positions of these two species.

Kleptoparasitism Rates

Data on rates of successful kleptoparasitism, and successful invasion by lepidopterans and dipterans, are summarized in Table 1. Four patterns are notable in these data. First, the highest kleptoparasitism rates are found in the two species, Kladothrips acaciae and K. ellobus, that are attacked by the host-specific, morphologically-specialized invaders Koptothrips zelus and K. xenus. Second, rates of kleptoparasitism are also quite high in five of the six species of gall-inducing thrips with soldier castes, with on the order of one-quarter to one-third of galls successfully invaded. Indeed, rates of Koptothrips invasion are about twice as high overall in species with soldiers as in species without soldiers. Third, some of the lowest rates of kleptoparasitism are exhibited by three species, Onychothrips arorum, Onychothrips tepperi, and Oncothrips antennatus on Acacia aneura, that are related phylogenetically, all being found in the same monophyletic group. Moreover, this clade is unusual in that all of its species induce galls on Acacia species in the section Juliflorae, whereas all of the other species induce galls on Acacia in the section Plurinerves. Finally, rates of invasion by non-thysanopterans, mainly lepidopterans and dipterans, do not show the same clear interspecific patterns as those for kleptoparasites; instead, almost all of the gall-inducers are heavily beset by these enemies.

DISCUSSION

The main goal of this study is to understand the evolutionary and behavioral-ecological dynamics of the kleptoparasite-host relationships found in Koptothrips and gall-inducing thrips on Australian Acacia. To achieve this goal, we have (1) used mi-
to chondrial DNA data to assess the taxonomic status of two taxa, *Koptothrips flavicornis* and *K. dyskritus*, that attack multiple species of gall-inducing thrips on multiple host plant species; (2) tested hypotheses for the evolutionary origin of kleptoparasitism in these insects, (3) analyzed the phylogenetic and behavioral-ecological patterns of diversification of *Koptothrips*, with respect to the diversification of their hosts, and (4) used data on rates of kleptoparasitism in different species to draw inferences concerning its importance as a selective pressure. Our hypothesis for the origin and diversification of *Koptothrips* and their hosts is depicted and summarized in Fig. 2, and described in detail below.

**Fig. 2.** Hypothesized scenario for the broad-scale evolutionary relationships between Australian gall-inducing thrips on *Acacia* and their *Koptothrips* kleptoparasites. (1) Origin of the genus *Koptothrips*, via a host shift onto *Acacia*, attacking the ancestor of the (*Oncothrips* + *Kladothrips*) lineage of gall-inducers. (2) Ancestor of (*Oncothrips* + *Kladothrips*) splits into two genera, and, as a result, *Koptothrips* splits into two lineages, one giving rise to (*K. xenus* + *K. zelus* + *K. dyskritus*) (which attack *Kladothrips*), the other giving rise to *K. flavicornis* (which attack *Oncothrips*). (3) *Oncothrips* lineage and *Koptothrips flavicornis* lineage diversify. (4) *Kladothrips acaciae* and *K. ellobus* descend from the ancestral *Kladothrips* lineage, leading to the evolution of their host-specific *Koptothrips*, *K. zelus* and *K. xenus*. (5) Next, *Kladothrips rugosus* originates along the *Kladothrips* lineage, leading to the evolution of *Koptothrips dyskritus*. *K. rugosus* and *K. dyskritus* diversify together, onto different species of *Acacia* in the section Plurinerves. (6) Some *Koptothrips flavicornis* lineages expand their host range by attacking *Kladothrips rugosus* that are on the same host plant as their ancestral *Oncothrips* hosts. By plausible alternative phylogenies, *Kladothrips acaciae* and *K. ellobus* may be sister-taxa, and *Koptothrips zelus* and *K. xenus* may also be sister taxa. We stress that this diagram represents a hypothesis that, although consistent with our available data, requires additional testing.
The levels of mitochondrial DNA sequence divergence found between specimens of *K. flavicornis* and *K. dyskritus* collected from different species of host thrips on different *Acacia* species range up to 7% and average about 3%. These values are consistent with the hypothesis that each of these named species actually comprises a set of multiple closely-related sibling species or host races. However, we also note that some of the pairwise divergences within these taxa are very low, below 0.5%, and that two of these low divergence values are for samples of *K. flavicornis* collected from different species of host thrips (*K. rugosus* and *O. waterhousei*) each on the same host plant (*A. loderi* or *A. ammophila*). These findings strongly suggest that whereas some *K. flavicornis* are sufficiently genetically divergent from others that high levels of gene flow are unlikely to be occurring between them, the *K. flavicornis* attacking different host thrips species on the same host plant may well be conspecific. Further analysis of the systematic status of *K. flavicornis* and *K. dyskritus* requires quantification of genetic variation both between and within putative conspecific populations, and experimental transfer of *Koptothrips* between host thrips and host plants.

Our phylogenetic analyses indicate that the genus *Koptothrips*, and its gall-inducing host species, are each monophyletic. These results falsify the hypothesis that *Koptothrips* arised from within the lineage of gall-inducers on *Acacia*, ostensibly via intraspecific kleptoparasitism during gall induction (Crespi 1992a). Because our phylogeny does not yet include genera of thrips on Australian *Acacia* other than *Oncothrips*, *Kladothrips*, *Onychothrips*, and *Koptothrips* (Mound 1971), we must turn to other information to assess whether or not *Koptothrips* and their gall-inducing hosts are (1) sister-taxa, such that they share a common ancestor, or (2) not closely related, such that kleptoparasitism arised via a host-plant shift. Using a cladistic morphological analysis of Australian Phlaeothripines, Morris et al. (1998) have shown that the latter hypothesis is supported. By their analysis, the genus *Koptothrips* is not closely-related to its gall-inducing hosts, nor is it found in a clade of thrips that inhabits *Acacia*; instead, it appears to be related to species of *Teuchothrips*, which induce leaf-roll or curl galls on a variety of plant taxa. Since *Koptothrips* are not known to attack *Teuchothrips*, these results suggest that the genus *Koptothrips* originated, and evolved its kleptoparasitic habit, in conjunction with a major host-plant shift onto *Acacia* (Morris et al. 1998). This hypothesized scenario for the origin of *Koptothrips* is depicted as stage 1 in Fig. 2. Whether the progenitors of *Koptothrips* were gall-inducers like *Teuchothrips*, or non-galling plant feeders, cannot be inferred from the phylogenetic information available to date.

What might be the ecological basis and evolutionary significance of kleptoparasitism originating in association with a host-plant shift? Morris et al. (1998) suggest that the habit of invading galls could have facilitated host-shifting because galls provide a highly favorable microhabitat, especially in a climate like that of arid-zone Australia. Thus, the advantages of using galls as domiciles could have helped offset the disadvantages of adapting to live on a novel host plant. Moreover, in the same way that host-plant shifts by phytophagous insects can be facilitated by escape from natural enemies (Brown et al. 1995, Feder 1995, Shorthouse and Brooks 1998), we suggest that host-insect shifts by incipient enemies could be facilitated by a lack of evolved defenses of their hosts. Support for a hypothesis of host-shifting coinciding with the origin of kleptoparasitism comes from remarkably parallel situations in two taxa unrelated to gall thrips: (1) in *Eriosoma* aphids, kleptoparasitism of galls has also apparently originated via a host-plant shift (Akimoto 1981, 1989), and (2) in yucca moths, phylogenetic evidence indicates that non-pollinating ‘cheater’ species originated in association with host-plant shifts (Pellmyr et al. 1996). Our hypothesis of evolutionary cheating arising as a result of ecological-phylogenetic saltation could be tested further by designing phylogenetic studies to uncover the ecological habits of
the closest honest relatives of such cheaters, rather than focussing just on the parasites and hosts when analyzing the origin of the parasites.

Phylogenetic and behavioral data suggests that, in addition to involving a host-plant shift, the origin of kleptoparasitism in *Koptothrips* may also have involved a facultative stage. This hypothesis is supported by the observation that *K. xenus*, *K. zelus* and *K. dyskritus* can create cellophane-like partitions in damaged, open galls bereft of gall-inducers, and breed successfully inside. By contrast, the obligately-kleptoparasitic *K. flavicornis* apparently cannot do so. We suggest that the primordial *Koptothrips* used damaged, open galls for breeding, as do some *Grypothrips* and *Katothrips* (Crespi et al. 1997), that facultative kleptoparasitism evolved via selection for obtaining a better, larger, and fresher resource for breeding, and that obligate kleptoparasitism evolved in the *K. flavicornis* lineage via evolutionary refinement of usurpation behavior (see Field 1992 for description of similar patterns in some Hymenoptera). This hypothesis fits with West-Eberhard's (1986) model of evolutionary transitions arising from facultative alternative behaviors, and it could be tested via more-detailed study of *Koptothrips* morphology and behavior in the context of their phylogeny. In particular, we need better resolution and support for the phylogenetic placements of *Koptothrips xenus* and *K. zelus*, for which neighbor-joining and maximum-parsimony yield differing, albeit weakly-supported, results.

Once a parasitic habit has evolved, diversification of parasite lineages can proceed via two main mechanisms: cospeciation, such that the parasites simply track the speciations of their hosts (Page 1994), and host-shifting, such that parasites move between host species more or less respective of the phylogenetic affinities of their current and future hosts. Our phylogenetic data indicates that the oldest split in the gall-inducers on *Acacia* in the section Plurinerves was between the genera *Oncothrips* and *Kladothrips*, and it suggests that this split was mirrored by *Koptothrips* kleptoparasites, as they diversified into two clades, *K. flavicornis*, which attack mainly *Oncothrips*, and (*K. zelus + K. xenus + K. dyskritus*), which attack species of *Kladothrips* (stage 2 in Fig. 2). Moreover, within the *Kladothrips* lineage, *K. acaciae* and *K. ellobus* are the most-basal species, and they are attacked by the two *Koptothrips* species that appear relatively old and basal, *K. zelus* and *K. xenus*. These relationships are also supported by the similarity in levels of divergence in mtDNA between *K. xenus* and *K. zelus* (14%), and between *K. ellobus* and *K. acaciae* (13.3%), which suggests that the two pairs of lineages may be of similar ages.

What of *K. dyskritus* and *K. flavicornis*? *K. flavicornis*, which our phylogeny identifies as the sister-taxon to the other *Koptothrips* species, attack species of gall-inducers in the sister-taxon of (*K. acaciae + K. ellobus + K. rugosus*), which comprises species of *Oncothrips*. This pattern suggests that the ancestor of *K. flavicornis* attacked the ancestor of (*O. morrisi + O. waterhousei + O. habrus + O. rodwayi + O. tepperi*) (stage 3 in Fig. 2), and has diversified by some combination of cospeciation, host shifting, and perhaps independent speciation. The descent of *K. dyskritus* from the ancestors of *K. xenus* and *K. zelus* is compatible with the observation that *K. dyskritus* primarily attack *Kladothrips rugosus*, a lineage that has descended from the ancestor of *K. acaciae* and *K. ellobus* (stages 4 and 5 in Fig. 2). Thus, our phylogeny is consistent with the hypothesis that cospeciation was also involved in the evolution of *K. dyskritus*. Finally, the observation that some *K. flavicornis* attack *K. rugosus*, as well as species of *Oncothrips*, suggests that some *K. flavicornis* lineages have undergone a host-insect range expansion, to include *K. rugosus* in their list of victims (stage 6 in Fig. 2). Our data also indicate that the *Acacia* host plants have mediated the putative expansion of host range: in both cases where *K. flavicornis* attack both *Oncothrips* and *Kladothrips*, the two species of gall-inducers are on the same host-plant species.
Further analyses of the evolution of across trophic-level interactions in *Koptothrips* and their hosts requires: (1) more thorough sampling of the *Koptothrips* from different host thrips and host plants, (2) more detailed elucidation of the taxonomic status of the *K. flavicornis* and *K. dyskritus* attacking different hosts, (3) statistical analysis of cospeciation models (Page 1994), (4) better understanding of the mechanisms responsible for cospeciation and host-shifting, (5) better support for the phylogenetic positions of *Koptothrips zelus* and *K. xenus*. However, the data presented here suggest that *Koptothrips* and their hosts have evolved together via some combination of cospeciation and host shifting, which will make them especially useful for analyzing the causes of these disparate processes.

Within lineages, kleptoparasitism evolves as a consequence of natural selection on both the parasites and their hosts, and our data on rates of kleptoparasitism allows us to assess its importance as a selective pressure in host species that differ in various aspects of their life history. Our quantification of rates of parasitism by *Koptothrips* and non-thysanopterans has uncovered four main patterns.

First, the highest rates of *Koptothrips* kleptoparasitism occur in *Kladothrips ellobus* and *K. acaciae*, the only two species with host-specific kleptoparasites, *Koptothrips xenus* and *K. zelus*. These high kleptoparasitism rates may be due in part to highly-developed parasite specialization, if higher kleptoparasite efficiency has evolved as a consequence of adaptation to single rather than multiple hosts (see Bernays and Graham 1988). Similarly high rates of successful attacks by specialist enemies have been reported in a subsocial pentatomid bug (Eberhard 1975), and in some species of gall aphids (e.g., Itô 1989, Moffett 1989, Stern and Foster 1996).

Second, rates of successful *Koptothrips* parasitism are also quite high in almost all species with soldiers. At first glance, this result might appear paradoxical, because behavioral evidence from numerous species with soldiers indicates that soldiers fight, and often kill, invading *Koptothrips* (Crespi and Mound 1997, Crespi unpublished data). However, such defense, though often spectacular, is frequently impossible or ineffective: *Koptothrips* sometimes invade galls before any soldiers have eclosed (Crespi and Mound 1997), and *Koptothrips* are often victorious in their fights with soldiers. Moreover, if high rates of kleptoparasitism were important in selecting for the origin of soldier castes (Crespi 1996, Crespi et al. 1998), then such high rates should not surprisingly be instrumental in maintaining soldiers.

The hypothesis that parasite pressure has been an important cause of the origin and maintenance of sociality could be analyzed further by comparing parasite and predator pressure between non-thysanopteran taxa with and without soldiers or other types of worker that defend. At present, such data are available for only two taxa (see also Crespi and Choe 1997). Moran (1993) compared rates of successful predation by a gall-specializing fly larva between a *Pemphigus* gall aphid species with soldiers, and a *Pemphigus* species without soldiers, for galls taken from the same tree. The aphid species with soldiers had lower predation rates than the species without soldiers for most of the season, but rates of unsuccessful attack on the species with soldiers are unknown. Schwarz (1994) found higher rates of parasitism by encyrid wasps and cuckoo bees, and higher levels of cooperative nests and nest aggregation, in montane populations of the alldapine bee *Exoneura robusta* (= bicolor) than in heathland populations of the closely related species *Exoneura nigrescens*. Rates of parasitism by encyrids did not covary with colony site at either location, but higher numbers of females may help in defense against cuckoo bees. By contrast, larger colony sizes have been inferred to enhance protection of brood against ants in *Allodapula melanophas* (Michener 1971), in *Exoneura robusta* (Schwarz 1986, 1994) and in *Exoneura nigrescens* (Bull and Schwarz 1996). Among alldapines in general, species exhibiting eusociality ap-
pear more beset by inquiline bees than are species with only solitary nests, but whether high inquilinism rates are a cause or effect of sociality is not yet known.

Third, some species of gall-inducing thrips, notably some *K. rugosus*, *O. antennatus* from *Acacia aneura*, *Onychothrips arotrum*, and *Onychothrips tepperi*, exhibit very low rates of *Koptothrips* parasitism. This interspecific variation in kleptoparasitism rates may be due in part to the different life-histories of the lineages involved. In some species, such as *K. rugosus* and *K. ellobus*, the life-cycle appears to be quite short relative to the other gall-inducers (several months, compared to over nine months in other taxa), and the offspring of the foundress apparently disperse from the gall as second-instar larvae and pupate in the soil (B. Crespi and B. Kranz, personal observation). As a result, some of these taxa may keep kleptoparasitism rates low by escaping in time (i.e., minimizing the time that they are vulnerable to invasion). By contrast, species of *Onychothrips*, and *O. antennatus*, inhabit galls that appear to be more tightly sealed than those of other species. The galls are long-lived in these species, and offspring of the foundress all eclose within the gall. In addition, all of the gall-inducing thrips in the lineage containing *Onychothrips*, *O. antennatus*, *O. sterni*, and *K. augon-saxxos* are found on *Acacia* in the taxonomic section Juliflorae, whereas all other gall-inducing thrips inhabit *Acacia* in the section Plurinerves (Mound et al. 1996). Of all of the gall-inducers in this lineage, only *O. antennatus* on *A. adsurgens* suffers from much *Koptothrips* kleptoparasitism, and we have never found *Koptothrips* in galls of *O. sterni*, *K. augonsaxxos*, *O. torus*, or *Ony. pilbara* (Crespi, unpublished data).

Taken together, these observations suggest that gall-inducing thrips have three distinct strategies for reducing the impact of *Koptothrips*: 'hiding' (living in tightly-sealed galls, on host plants that may be less suitable for the kleptoparasites), 'fighting' (maintaining soldier morphs), or 'running' (escaping in time, via an accelerated life cycle). These latter two strategies are provided a striking parallel in gall aphids, some of which also exhibit soldiers and long-lived galls, while others lack soldiers and develop relatively rapidly (Moran 1993, Stern 1998). Our data also indicate that, in gall-inducing thrips, each of the three strategies is more or less successful in different species.

The fourth pattern shown in our kleptoparasitism data is that, in contrast to the results for *Koptothrips* kleptoparasites, almost all species of gall-inducing thrips are heavily attacked by species of parasites in other insect orders, primarily lepidopteran and dipteran larvae (see also Mound et al. 1996). Some of these species feed upon gall tissue, while others feed upon thrips eggs, and all cause a major reduction in thrips numbers, if not total reproductive failure. Behavioral observations indicate that soldiers are ineffective against lepidopteran larvae within their galls, because the larvae remain within silken, frass-covered tunnels with only their sclerotized head capsule exposed (Crespi, personal observation). Moreover, the caterpillars bite at soldiers that approach, usually removing their antennae in the process, in a behavior that, if it represents an adaptation, could be aptly termed ‘sensory castration’. Analysis of the influences of the various non-thysanopteran enemies of gall thrips on their life-histories and behavior must await in-depth study of these enemies and their mechanisms of subversion.

What ecological factors might have ultimately led to the high kleptoparasitism rates, and the differences among gall-inducing thrips species in kleptoparasitism rates? Among Hymenoptera, factors promoting a high incidence of interspecific and intraspecific kleptoparasitism include high host synchrony and density, and concomitant highly seasonal environments (Wcislo, 1987; see also Petanidou et al. 1995). In accordance with Wcislo’s hypothesis and results for Hymenoptera, the life cycles of almost all gall-forming thrips are normally highly synchronized by being tied to the synchronous production of new foliage, which occurs either annually in the spring, or, in highly-arid regions, unpredictably, after rare heavy rainfalls.
What can our analyses of kleptoparasitism in gall thrips tell us about host-parasite interactions in social insects in general? One of the main findings of this study, that gall thrips taxa with soldiers suffer rates of Koptothrips kleptoparasitism at least as high as those of related taxa without soldiers, compels reconsideration of the idea that the presence of a complex social adaptation coincides with a high success rate of that adaptation (see also Tallamy and Schaeffer 1997). Rather than viewing social behavior as an evolutionary pinnacle (e.g., Wilson 1971, 1990), perhaps it sometimes actually represents a relatively low and local adaptive peak, eroding and barely maintained as soldiers and workers fight their way uphill under an onslaught of enemies. And although we might consider this pattern as leading to a pessimistic view of life, we must recall that without such thieves, there might be no such beautiful an adaptation as cooperation in an insect so otherwise-ignoble as a thrips.

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REFERENCES CITED


