Floral volatiles, pollinator sharing and diversification in the fig–wasp mutualism: insights from *Ficus natalensis*, and its two wasp pollinators (South Africa)

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Combining biogeographic, ecological, morphological, molecular and chemical data, we document departure from strict specialization in the fig-pollinating wasp mutualism. We show that the pollinating wasps *Elisabethiella* stuckenbergi and *Elisabethiella socotrensis* form a species complex of five lineages in East and Southern Africa. Up to two morphologically distinct lineages were found to co-occur locally in the southern African region. Wasps belonging to a single lineage were frequently the main regional pollinators of several *Ficus* species. In South Africa, two sister lineages, *E. stuckenbergi* and *E. socotrensis*, pollinate *Ficus natalensis* but only *E. stuckenbergi* also regularly pollinates *Ficus burkei*. The two wasp species co-occur in individual trees of *F. natalensis* throughout KwaZulu-Natal. Floral volatile blends emitted by *F. natalensis* in KwaZulu-Natal were similar to those emitted by *F. burkei* and different from those produced by other African *Ficus* species. The fig odour similarity suggests evolutionary convergence to attract particular wasp species. The observed pattern may result from selection for pollinator sharing among *Ficus* species. Such a process, with one wasp species regionally pollinating several hosts, but several wasp species pollinating a given *Ficus* species across its geographical range could play an important role in the evolutionary dynamics of the *Ficus*-pollinating wasp association.

**Keywords:** mutualism; coexistence; specificity; host switch; volatile compounds

1. INTRODUCTION

*Ficus* species are pollinated by one or a few species of agaonid wasps that breed within the enclosed inflorescences called figs. These wasps are generally host-specific [1]. This led to the assumption that co-speciation of figs and fig-pollinating wasps is prevalent [2,3]. However, accumulating data demonstrate numerous exceptions to parallel cladogenesis [4–7]. Two or more agaonid wasps pollinating the same host *Ficus* has been documented [8–10], as well as cases of one wasp species pollinating several *Ficus* species [10]. Studies based on molecular markers have shown at local- [11], regional- [12] and in one case broad-scale [13], the coexistence of two or more genetically closely related pollinators on the same host. In some cases, coexistence of distantly related pollinators has been documented [14]. Genetic data on other species suggest that, while more than one wasp species may occur across the fig-distribution range, a single pollinator species is usually present locally [11,15,16]. In some cases, the two wasp species pollinating the same *Ficus* species are associated with different environments [8]. In other cases, one of the two sympatric species is a non-pollinating agaonid [17,18].

There are numerous observations of agaonid wasps associated with a *Ficus* species that sometimes also visit (and pollinate) another *Ficus* species [16]. The importance of such events in terms of the potential for genetic introgression among *Ficus* species should not be underestimated [7]. Data from Panama could correspond to such a case or alternatively to a situation of an agaonid wasp species that is a quantitatively important pollinator of two or more *Ficus* species [11,19]. At least two cases of a wasp being the major pollinator of two *Ficus* species have been confirmed by molecular markers: *Ceratozolen arabicus* is the sole pollinator of both *Ficus sycomorus* and *Ficus mucor* [20] and *Alfonsiella binghamii*, the sole...
pollinator of both *Ficus petersii* and *Ficus stuhlmannii* [21]. Finally, some molecular data suggest that this could also be the case in the species complex of *Elisabethiella stuckenbergi*–*Elisabethiella socotrensis*, which is associated with a number of *Ficus* species in East Africa [21]. Such situations may be evolutionarily relevant. They suggest that transient or persistent pollinator sharing may be an important facet of how the *Ficus*-pollinating wasp association has evolved and diversified.

Fig pollinators are attracted by volatile compounds emitted by receptive figs of their host *Ficus*, i.e. by figs at the stage when the wasps enter them to oviposit, and they are usually not attracted by the odours of other *Ficus* species [22–24]. The chemical message generally comprises several compounds and is differentiated among species [24,25]. The wasps are assumed to recognize a mixture of volatile organic compounds constituting the host species'-specific signature [23,25–27]. Data on two *Ficus* species have evidenced significant among-population variation of receptive fig floral odour within species [28]. As in other systems, intraspecific variation in the floral blend, forming floral 'chemotypes', could allow for more than one species of pollinator to be associated with the same host species [29,30]. Conversely, similarity in the floral blends produced by two *Ficus* species could facilitate their pollination by the same wasp species.

In this study, we collected and compared wasp morphological and genetic data, fig-visitation patterns and fig volatile chemical profile data within and among localities to document and analyse a situation of non-pollination by the same wasp species. *Ficus* produced by two *E. socotrensis*–*E. stuckenbergi* [33] and *E. stuckenbergi–E. socotrensis* species complex of species using multiple hosts or a species could facilitate their pollination by the same wasp species. 

Figure 1. Co-occurrence of *Elisabethiella socotrensis* and *E. stuckenbergi* on *Ficus natalensis* throughout KwaZulu-Natal. Pie slices represent the proportions of figs per tree visited only by *E. socotrensis*, only by *E. stuckenbergi* or by both species together, at each site.

In order to address this conundrum of complex host associations, we used molecular tools to clarify the genetic clusters within the *E. stuckenbergi–E. socotrensis* species complex, and asked whether this clustering follows *Ficus* host identity or geographical distribution. Then, in KwaZulu-Natal, once the local correspondence between morphotype and genetic identity had been confirmed, we assessed which species colonized individual figs on a number of *F. natalensis* trees across several locations in order to establish (i) whether the two wasp species are effective pollinators of *F. natalensis*; (ii) whether the two wasp species frequently co-occur on the same tree, over a broad geographical area; and (iii) whether the two wasp species compete for receptive figs. Finally, we analysed the odours produced by receptive figs from the sampling sites in order to test (i) whether variation in the suite of volatile chemicals produced by *F. natalensis* could be associated with preferential visitation by one or the other pollinating wasp species and (ii) whether the volatile chemicals of receptive figs were similar between *F. burkei* and *F. natalensis*, facilitating wasp sharing.

### 2. MATERIAL AND METHODS

(a) *Ficus natalensis* and *Ficus burkei*

Both *F. natalensis* subspecies *natalensis* (henceforth, *F. natalensis* will be designated as *F. natalensis* subspecies *natalensis* except in parts of the text where ambiguity between subspecies is possible) and *F. burkei* belong to subgenus *Urostigma*, section *Galoglychia*, subsection *Chlamydodorae*. They are distributed from South Africa to Uganda and Kenya in East Africa. *Ficus natalensis* has a more coastal distribution than *F. burkei* [33]. In southeast Africa, they form a monophyletic species group with *Ficus cratostoma*, *F. petersii* and *Ficus lingua* [32] and subsection *Chlamydodorae* is further represented by *Ficus burtt-darbyi*, *Ficus fisheri* and *Ficus ilicina* [34]. The two *Ficus* species studied here are morphologically well-defined and are easily identified [33]. They are not sister species, but are closely related [32]. Although the two species mainly occupy different habitats (forest and rocky habitat for *F. natalensis* and savannah woodland and wet or dry forest for *F. burkei* [35]), they also co-occur locally [33].

In most of South Africa, *E. stuckenbergi* is the sole pollinator of *F. burkei*, with sporadic occurrences of *E. socotrensis*, but in Zambia, Tanzania, Kenya, Zimbabwe and Botswana, *F. burkei* is also regularly pollinated by *Alfonsiella brongersmai* with both wasp species present in the same crop (i.e. a cohort of figs produced by a single tree) [14]. In contrast, *F. natalensis* is pollinated by at least five fig wasp species [36]. In East Africa, it is pollinated by *E. socotrensis*, *A. brongersmai*, *Alfonsiella natalensis* and *Alfonsiella longiscapa*, which can all be present in the same crop of figs [21]. Only two of these pollinator species, *A. longiscapa* and *E. socotrensis*, are present in KwaZulu-Natal, where most *F. natalensis* trees are pollinated by *E. socotrensis* and by *E. stuckenbergi*. However, in the extreme north of KwaZulu-Natal, extending from Kosi Bay (figure 1) northwards into Mozambique, *A. longiscapa* replaces these two species as the pollinator of...
*E. socotrensis* [14]. Further, the figs of both *E. natalensis* and *F. burkei* are also entered by the syconecine non-pollinating wasps *Philocænus barbarus* Grandi and *Crosogaster odorans* Wiebes that gall ovules usually without providing a polination service [37], but in the population of *E. natalensis* pollinated by *A. longisca* these two syconecine species are replaced by another two species: *Philocænus medius* van Noort and *Crosogaster lurida* van Noort [37–40].

(b) *The Elisabethiella socotrensis—Elisabethiella stickenbergi* species group

Both *E. socotrensis* and *E. stickenbergi* are widely distributed in East Africa and extend into South Africa. They do not seem to form distinct monophyletic groups [21]. In the original descriptions, the length–width ratio of the head is different [34,41], suggesting that they are adapted to different fig ostiole shapes. Indeed, head shape, among other diagnostic differences, was shown to be a function of ostiole length as determined by fig wall thickness for wasps associated with section *Galoglychia* [42]. Further diagnostic differences recorded for samples collected in South Africa include the proportional length to width shape of the mandibular appendage, number of rows of teeth on the appendage, shape and armature of the mandible itself, shape of the antennal pedicel, and the length of the post-marginal vein relative to the stigmal vein. These are all distinctive morphological traits that delimit species within the family Agaonidae.

(c) *Phylogenetic methods applied to pollinating wasps*

Phylogenetic relationships within the *E. socotrensis—E. stickenbergi* species group were examined using a Bayesian analysis of the mitochondrial gene cytochrome oxidase I (COI). Samples were obtained from multiple sites to assess genetic grouping (see electronic supplementary material, ESM1, for more details, method S1 and table S1). We estimated the mean percentage sequence divergence (p-distance) ± s.d. between and within the clades or lineages of *Elisabethiella*. A limited number of wasps belonging to the different lineages were examined morphologically to assess whether (i) locally coexisting wasp lineages were correctly separated and (ii) two consistent morphotypes were recognizable across the investigated range.

(d) *Local occurrence of the two pollinating wasp species on Ficus natalensis in KwaZulu-Natal*

To analyse local and regional co-occurrence of *E. stickenbergi* and *E. socotrensis* in *F. natalensis* across its distribution in KwaZulu-Natal, wasps were sampled from figs of 28 *F. natalensis* trees at seven sites (Vernon Crooks, Durban, Mtunzini, Tugela Mouth, St Lucia, Richard's Bay and Kosi Bay; see electronic supplementary material ESM2, table S4, for more details). Sampling was carried out in March–June 2008 and covered a range of 290 km (figure 1). At each site, 20–40 figs per tree were collected, either during fig receptivity when pollinator foundresses were present, or at the end of pollinator development just before wasp emergence. Figs collected at emergence stage were placed in closed vials and wasps were allowed to emerge naturally from their natal figs within the vials. Figs collected at receptivity stage were opened and pollinators were extracted from within the fig cavity. All figs were stored individually together with their pollinators in 70 per cent ethanol. In the laboratory, fig wasps were sorted to morpho-species under a dissecting microscope. For each sampled receptive fig, the numbers of foundress *E. socotrensis* and *E. stickenbergi* females trapped inside the fig cavity were recorded. For mature figs, presence–absence of the two wasp species was recorded. Furthermore, because receptive figs were collected close to the end of the receptive phase of the crops, but before abortion of unvisited figs, we could not only measure the relative proportion of receptive figs visited by each species but we could also provide an estimate of the absolute proportion, taking into account the proportion of still unvisited figs.

We tested for among-site variation of each of the two variables ‘proportions of figs per tree visited by *E. socotrensis* only and by *E. stickenbergi* only’ using a generalized linear model with a quasi-binomial error distribution (including only the three sites at which at least two trees were sampled: St Lucia, Mtunzini, Durban).

We also tested whether the two pollinator species competed for access to figs of a tree. Interspecific competition for available receptive figs will translate into presence of one species reducing the probability that the fig is visited by the other species. We tested this probability using continuity χ²-tests at the crop level and over the distributional range through KwaZulu-Natal. For all statistical analyses, we used R (v. 2.7.0; R Development Core Team; URL http://www.R-project.org).

(e) *The chemical signatures of Ficus natalensis and Ficus burkei*

Volatile organic compounds (compounds with usually fewer than 20 carbons) emitted by receptive figs from 15 *F. natalensis* trees from six different sites and five *F. burkei* trees from three sites (see electronic supplementary material ESM2, table S4) were collected using the headspace technique and subsequently analysed by GC–MS [24]. Details of methods are described in the electronic supplementary material ESM3, method S2. Preliminary tests confirmed that both pollinators were attracted by the blend of volatile organic compounds emitted by receptive figs of *F. natalensis* collected using the headspace technique, and eluted using dichloromethane (A. Cornille 2008, unpublished data).

Comparison of the composition of volatile compounds among samples was performed using multivariate analyses [43] using R with the Vegan package. First, data (using relative amounts of all the compounds with respect to total peak areas) were square root-transformed and standardized, then a data matrix of pairwise Bray–Curtis dissimilarity indexes [44] between samples was built. In order to scale intraspecific and interspecific variations among our samples, four other *Ficus* species from KwaZulu-Natal, analysed with the same methods and equipment, were included in the comparison: *Ficus sur* and *Ficus sycomorus* (both belonging to subgenus *Sycomorus*, section *Sycomorus*, subsection *Sycomorus*; data published in Proffit & Johnson [24]), three individuals of *Ficus glumosa* and three individuals of *F. stuhlmanni* (both belonging to subgenus *Urostigma*, section *Galoglychia*, subsection *Platyphyllae*, this study). We further compared our data with published Bray–Curtis index data for comparisons between populations and between species for two Asiatic *Ficus* species: *Ficus hispida* (subgenus *Sycomorus*, section *Sycomorus*) and *Ficus racemosa* (subgenus *Sycomorus*, section *Sycomorus*) [28]. Then, nonmetric multi-dimensional scaling (NMDS) was used to visualize similarities among all the samples from KwaZulu-Natal by finding the best two-dimensional representation of the distance matrix.

Within *F. natalensis*, permutational multivariate analysis of variance (permanova) of the Bray–Curtis index (100 000 permutations) was used to test variation in scent composition among populations and among trees visited mainly by one or
the other pollinator species (grouped as over 70% of figs per tree visited by *E. stuckenbergi* and less than 40% by *E. socotrensis*, or vice versa, or both wasp species present in more than 40% of the figs per tree). The same analysis was used to test for significant difference in the volatile profile of receptive figs, between species occurring in KwaZulu-Natal, and within and between the two Asian species. *p*-Values were adjusted for multiple comparisons using the false discovery rate [45].

The proportions of the seven main compounds present in the floral blends of the two *Ficus* species were compared using Student’s *t*-tests. Data were log(*x + 10*)-transformed to fit the assumption of normality (for all Shapiro tests, *p > 0.05*). For variables that failed the assumption of normality, a non-parametric Mann–Whitney rank sum test was used. *p*-Values were adjusted for multiple comparisons using the false discovery rate [45].

### 3. RESULTS

**a) Five molecular clades supported by morphology in the Elisabethiella stickenbergi–Elisabethiella socotrensis species complex**

In the Bayesian COI analysis, wasps identified as *E. socotrensis* and *E. stickenbergi* form a species complex presenting at least five lineages (figure 2). This is in agreement with *a posteriori* morphological investigation: no simple criterion allowed us to assign all the samples into only two classes of individuals (see electronic supplementary material ESM1, table S3). Nevertheless, within a geographical region, all individuals identified under the same name belonged to a single lineage. For instance, *E. stickenbergi* from Zambia, whatever their host, all belonged to clade E even though they were collected 600 km apart. Similarly, *E. stickenbergi* from South Africa, whatever their host, all belonged to clade B even though they were collected up to 590 km apart, and *E. socotrensis* from South Africa, whatever the host collected on, all belonged to clade C even though they had been collected up to 800 km apart. Furthermore, where two molecular lineages coexisted locally, they had been distinguished *a priori* on morphological grounds and samples were correctly attributed to distinct lineages, before molecular analysis. *A posteriori* morphological measurements, although performed on a limited number of specimens, provide a morphological basis to facilitate identification within regions (see electronic supplementary material ESM1, table S3). On the other hand, wasps collected from the same host in different parts of its range belonged to different clades.
Maximum intrACLade sequence divergences were generally one order of magnitude smaller than the minimum between-clade sequence divergences (see electronic supplementary material ESM1, table S2). Exceptions are the lower divergence between clades B and C, which co-occur in South Africa and are grouped into a monophyletic clade (94% bootstrap, 1.00 posterior probability) and between clades D and E.

(b) Distribution and co-occurrence of pollinating wasp species in KwaZulu-Natal

Figs visited only by *E. stuckenbergi* or *E. sociotrensis* produced abundant seeds (qualitative records). Figs visited only by *Philocaenus barbatus* produced no seeds. Both *Elisabethiella* species are pollinators and *Philocaenus* is a parasite of the mutualism.

The two wasp species co-occurred within crops in all locations, across a range of 290 km (figure 1). The proportion of figs per tree visited by a single species did not differ among the three sample sites tested (binomial glm, respectively: *E. stuckenbergi*—$\chi^2 = 132.5$, d.f. = 2, $p = 0.69$; *E. sociotrensis*—$\chi^2 = 135.2$, d.f. = 2, $p = 0.29$; figure 1 and see electronic supplementary material ESM2, table S5). Indeed, for receptivity period figs, only 15 per cent of the collected figs contained the two species of wasps while only 2 per cent of the figs were unvisited.

(c) Competition between Elisabethiella sociotrensis and Elisabethiella stuckenbergi for figs

The two wasp species co-occurred within figs less frequently than expected by chance, within the whole dataset ($\chi^2 = 203.3$, d.f. = 1, $p = 2 \times 10^{-16}$), and within the 17 crops with over three figs sampled (see electronic supplementary material ESM2, table S5). Figs visited only by *Philocaenus barbatus* produced no seeds. Both *Elisabethiella* species are pollinators and *Philocaenus* is a parasite of the mutualism.

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(d) Chemical signature of Ficus natalensis

Receptive figs from the 15 *F. natalensis* trees emitted 38 volatile organic compounds, which all have fewer than 20 carbons (see electronic supplementary material ESM3, table S6). The mean number of compounds emitted by individual trees was as follows: St Lucia 10.3 $\pm$ 2.5, Durban 13.8 $\pm$ 6.1, Mnunzini 13.6 $\pm$ 6.3, Tugela Mouth 11, one individual, Kosi Bay 16, one individual and Vernon Crooks 11, one individual, and did not differ among sites (glm (Poisson): $\chi^2 = 20.5$, d.f. = 5, $p = 0.91$). We found no variation in the odour emitted by receptive figs of *F. natalensis* among sites (permanova: $F_{5,10} = 1.05$, $p = 0.38$) and no variation depending on which wasp species was more abundant among the foundresses (permanova: $F_{2,11} = 1.38$, $p = 0.11$).

(e) Comparison of the chemical signatures among species

Receptive figs from the five sampled *F. burkei* trees emitted 18 volatile compounds (see electronic supplementary material ESM3, table S6). The proportions of the main compounds emitted by receptive figs did not differ between *F. natalensis* and *F. burkei* (see electronic supplementary material ESM3, table S7). The number of compounds emitted per individual fig did not differ between the two species (glm (Poisson): $\chi^2 = 26.3$, d.f. = 1, $p = 0.07$): *F. natalensis* (12.9 $\pm$ 4.8), *F. burkei* (9 $\pm$ 2.2).

The NMDS plot suggests that odours of *F. natalensis* and *F. burkei* are more similar to each other than the odour of the four other *Ficus* species included in the analysis (figure 3). The odours produced by the two other species pairs of similar level of taxonomic proximity (subsection) did not overlap. This result was confirmed by the permanova: the composition of volatile organic compounds produced by receptive figs is significantly different among *Ficus* species ($F_{5,31} = 4.41$; $p_{\text{adj}} = 0.00003$) occurring in KwaZulu-Natal. The odour of *F. natalensis* was significantly different from those of all other African species except *F. burkei* (see electronic supplementary material ESM3, table S8). The two other within-*Ficus* subsection comparisons gave significant differences (subsection *Sycomorus*, between *Ficus sur* and *Ficus sycomorus*; subsection *Platyphylla*, between *Ficus glomosa* and *Ficus stuhlmannii*). For the two Asian species, the odours of receptive figs were significantly different between species but also among populations. Bray–Curtis indexes among populations within Asian species were higher than Bray–Curtis indexes between *F. natalensis* and *F. burkei* (see electronic supplementary material ESM3, table S8).

4. DISCUSSION

*Ficus* are deeply rooted as a strict highly specialized mutualism albeit few recent molecular studies pinpointed the existence of multiple wasp species per host and their consequences for the evolutionary dynamics in this system [4,5,7,11]. Here, combining original polyvalent methodologies (biogeographic, ecological, morphological, molecular and chemical), we have provided the first consistent demonstration of a set of *Ficus* sharing their main pollinators.

The molecular data show that the *Elisabethiella sociotrensis–E. stuckenbergi* species complex encompasses at least five lineages (figure 2). No locally cryptic species was discovered and, within each region, specimens were consistently sorted *a priori* according to morphology into their genetic group. Geography rather than fig-host determined which clade was present in a location. A similar situation may be occurring in west Africa within the same *Ficus* species complex, but molecular data are lacking [10,34]. It may be noted for clade B that *F. n. natalensis* and *F. n. graniticola* are allopatric, and that they occupy different habitats from *F. burkei* [35]. Similarly, *F. lingua* and *F. burkei* also occupy different habitats [35]. This is also the case for another system of one pollinator–two *Ficus* species, namely the pollination of *Ficus mucuso* (forest species) and *Ficus sycomorus* (savannah species) by *Ceratosolen arabis* [8]. So it would seem that *Ficus* species occurring in the same region but in different habitats may share agaonid wasps that are their sole or a major pollinator. In that perspective, the most surprising situation is that of *F. petrii* (the sister species of *F. burkei*), which in South Africa is almost exclusively pollinated by the same wasp species (*A. binghami*) as *F. stuhlmannii*: the two *Ficus* species share the same habitat [35] and may grow side by side in South Africa. However, further north in the range of *F. petrii*, *E. stuckenbergi* has also been recorded as the
pollinator of this host fig in Zambia (S. van Noort 2006, personal observation). Pending further molecular and morphological investigation of the species group, we retain the names *E. stuckenbergi* for lineage C and *E. socotrensis* for lineage B in South Africa.

In South Africa, *F. burkei* and *F. natalensis* are both pollinated by *E. stuckenbergi* clade C, and *F. natalensis* is also abundantly pollinated by *E. socotrensis* clade B. However, in other parts of their range, both *Ficus* species are pollinated by other clades or even by a different genus. Further, clades B and C are sister clades restricted to South Africa. We may speculate that the observed zonation of pollinator distributions corresponds to climatic limits. A pollinating wasp adapted to the South African climate would have allowed the two host species to colonize South Africa and the wasp would be in the process of evolutionary diversification. A similar situation may occur in Australia, where cryptic species associated with a single host, *Ficus rubiginosa*, show some geographical separation, though the pattern seems less clear [13]. Range expansion following climatic modifications has been shown to shape species’ biology in an ant–plant species-specific insect–plant mutualism [46]. Climatic oscillations have also been shown to be important in driving the evolution of the *Ficus*–fig wasp mutualism, in particular [47]: they could be important for shaping mutualistic interactions in general.

The results show that *E. natalensis* is abundantly pollinated by two species of wasps over a range of 290 km. We found no evidence for variation in receptive fig odour that would correlate with more abundant visitation by one of the wasp species. Fig odour was also homogeneous among sites throughout KwaZulu-Natal (see electronic supplementary material ESM3, table S6). The odour of receptive figs of *F. burkei* was similar to that of *F. natalensis* figs (figure 3). These odours were much more similar to each other than are the odours produced by other pairs of species of similar taxonomic relatedness. They were even more similar than the reported variation among populations within species [28]. The type of analysis of odours presented here is not based on what compounds wasps respond to. It is rather a comparison emphasizing variation in molecules present in intermediate quantities. Nevertheless, the analysis seems to capture a biological reality: the odours produced by the two host species are similar but tend not to be strictly identical. This fits rather well with the biological result that one wasp is a major pollinator of both *Ficus* species while the other one is only a marginal pollinator of *F. burkei*. Two complementary processes may explain receptive fig odour similarity: one or both *Ficus* species may be selected to mimic the other species and/or phylogenetic inertia is involved. As odour variation among populations but within species for two Asian species was larger than the difference observed here between *F. burkei* and *F. natalensis*, we may suggest that phylogenetic inertia is not sufficient to explain the data.

Figure 3. Non-metric multi-dimensional scaling of the odours of receptive figs of six African *Ficus* species based on Bray–Curtis dissimilarity index (stress = 0.021). *Ficus natalensis* and *F. burkei* produce odours that are more similar to each other than they are to those of other *Ficus* species from KwaZulu-Natal. na, *F. natalensis*; bu, *F. burkei*; gl, *F. glumosa*; st, *F. stuhlmannii*; sy, *F. sycomorus*; su, *F. sur*. The second letter indicates collection site for the species (D, Durban; S, St Lucia; M, Mtunzini; V, Vernon Crooks; T, Tugela Mouth, K, Kosi Bay, P, Pietermaritzburg).
To answer the question, we will need to investigate *Ficus petersii*, a close relative of *F. burkei* [34, 48]. It has the same pollinator as *F. stuhlmannii* [21] and receptive fig odour of *F. stuhlmannii* is different from that of *F. burkei* (figure 3). Interestingly, a single genetic lineage of the fig-entering wasp *Philocaenus barbarus* is associated with *F. burkei* and *F. natalensis*, with another lineage associated with *F. petersii* and *F. stuhlmannii*: odour similarity at receptivity may also result in sharing of non-pollinator fig wasp species (S. van Noort 2008, unpublished data).

Our observations that fewer than the expected number of figs within crops were visited by both species suggest strong interspecific competition for access to receptive figs. This raises a question as to how these two species can coexist within the same fig crop, but generally not in the same fig. The origin of species coexistence is a complicated issue [49–51]. In our case, we surmise that the same fig. The origin of species coexistence is a complicated issue [49–51]. In our case, we surmise that the same fig. The origin of species coexistence is a complicated issue [49–51]. In our case, we surmise that the same fig. The origin of species coexistence is a complicated issue [49–51]. In our case, we surmise that the same fig.

The plasticity of the interaction between agaonid wasps and *Ficus* evidenced here could have important implications for *Ficus* evolution. Indeed, in order to reproduce in a *Ficus* species, wasps must (i) be attracted by the odours of receptive figs [42], (ii) be capable of getting into the fig through the ostiole [42], (iii) present an ovipositor length compatible with the length of the styles through which they oviposit [1, 54], and (iv) be capable of initiating the transformation of the ovule into a gall on which they will feed [6, 55]. Hence, we may surmise that host use by fig pollinators is constrained by a number of factors that are generally more similar among related hosts. As a result, pollinators using several hosts can be predicted to use related hosts, a feature that should favour fig hybridization, and hence genetic introgression among related fig species [7]. Natural interspecific hybridization commonly occurs in flowering plants, and is thought to play an important role in their evolution [56]. It is as yet unclear how much genetic introgression occurs within *Ficus*, but reticulate evolution could be at work in some cases [7].

The situation illustrated here for a group of African *Ficus* may occur in several other generally highly species-specific pollination systems. For instance, in orchids of genera *Ophrys* and *Pseudorchis*, situations of flower odour convergence associated with local use of the same pollinator by two plant species have been demonstrated [57]. Similarly, the receptive flowers of *Glochidion ovatum* and *Glochidion rubrum*, two parapatric species pollinated by the same two species of *Epicephala* moths, produced similar odours, more similar than those produced by co-occurring species of *Glochidion* pollinated by other species of *Epicephala* [58]. We may suggest that there is sometimes sufficient overlap in the chemical messages produced by different host species and in the chemicals to which different pollinator species respond for selection to lead to odour convergence among species that use the same pollinator(s) species. Imperfect chemical differentiation may facilitate a dynamic reticulate evolution of host–pollinator associations. As such, it may facilitate ecological and evolutionary plasticity of species that were previously considered to be locked into hyper-specialization.

A. Cor. performed acquisition of data, analysis, interpretation of data, drafted the article and incorporated all comments from revisions. M.P. conceptualized and designed the study, performed acquisition of data and analysis, interpretation of data and gave final approval of the version to be published. F.K. and M.H.K. contributed to analysis, interpretation, writing and gave final approval of the version to be published. J.G.U. and A.Cr. gave final approval of the version to be published. K.A.T. did acquisition of phylogenetic data, and gave final approval of the version to be published. S.D.J. contributed to the writing and interpretation of data and gave final approval of the version to be published.

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