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The relationship between morphological and behavioral mimicry in hover flies (Diptera: Syrphidae)[1]

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Abstract
Palatable (Batesian) mimics of unprofitable models could use behavioral mimicry to compensate for the ease with which they can be visually discriminated, or to augment an already close morphological resemblance. We evaluated these contrasting predictions by assaying the behavior of 57 field-caught species of mimetic hover flies (Diptera: Syrphidae), and quantifying their morphological similarity to a range of potential hymenopteran models. A purpose-built phylogeny for the hover flies was used to control for potential lack of independence due to shared evolutionary history. Those hover fly species that engage in behavioral mimicry (mock stinging, leg waving, wing wagging) were all large wasp mimics within the genera *Spilomyia* and *Temnostoma*. While the behavioral mimics assayed were good morphological mimics, not all good mimics were behavioral mimics. Therefore, while the behaviors may have evolved to augment good morphological mimicry, they have not been selected in all hover fly species.
Keywords: Batesian mimicry, imperfect mimicry, behavioral mimicry, deception, phylogenetic comparisons

Introduction

Batesian mimicry arises when members of a palatable species (mimics) evolve a resemblance to a noxious or otherwise defended prey species (models), and thereby gain protection from predators (Bates 1862). While Batesian mimics are often recognized through their close morphological similarity to defended models (see Ruxton et al. 2004 for a review), mimicry can also occur in other sensory modalities (Golding and Ennos 2005). Thus mimics can smell (Ceccarelli 2008; Ruxton 2009) and sound (Rowe et al. 1986; Young et al. 1999) like their models, and they may also adopt behaviors that are characteristic of their models. Behavioral mimicry has been documented in some of the best-studied mimetic complexes including the *Myrmarachne* spider-ant complex (e.g. Ceccarelli 2008), the syrphid-hymenopteran complex (e.g. Waldbauer 1970), lepidopteran complexes (e.g. Srygley 1994; Kitamura and Imafuku 2010) as well as intra-specific sexual mimics (Forbes et al. 1997) and masqueraders such as leaf or twig mimics (Bradburne 1995).

Resembling a model in more than one manner may increase the likelihood that predators will be deceived by the Batesian mimicry, and it may potentially dupe different predators that use different sensory modalities to detect their prey (Pekár et al. 2011). However, if behavioral mimicry is so beneficial then one might wonder why all mimics have not evolved such traits. Likewise, it is now widely recognised that not all Batesian mimics can be considered perfect or high fidelity mimics, and one might also wonder why natural selection has not improved this

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resemblance (e.g. Getty 1985; Edmunds 2000; Johnstone 2002; Sherratt 2002; Chittka and Osorio 2007; Kikuchi and Pfennig 2010; Penney et al. 2012).

Given the lack of data on behavioral mimicry at a comparative level, the relationship between morphological and behavioral mimicry is particularly unclear. Nevertheless, it has been proposed repeatedly that poor mimics can use behavior to compensate for their ease of visual discriminability (Howarth et al. 2004; Gilbert 2005; Pekár et al. 2011). By contrast, behavioral mimicry might provide little selective benefit if imperfect mimics are readily visually discriminated. Instead, behavioral mimicry might be restricted to cases of good morphological mimicry, reflecting overall stronger selection for mimetic fidelity on all levels. Of course, the apparent variation in morphological mimetic perfection may be illusory and/or misleading if the potential predators of mimics and humans discriminate mimics from models in different ways (Cuthill and Bennett 1993). So, a third possibility is that there is no relationship between morphological and behavioral mimicry. Of course, if the propensity to engage in behavioral mimicry was in some way associated with the perfection of morphological mimicry as judged by humans, then this would suggest that the human-based categorization of mimetic fidelity carries wider significance to predators in general, and is not simply a reflection of human perception, as this "eye of the beholder" hypothesis implies (e.g. see Cuthill and Bennett 1993).

In this study we set out to test for an association between the indices of behavioral and morphological mimicry in hover flies (Diptera: Syrphidae), a group well known for both phenomena. In the syrphid-hymenopteran mimicry system, behavioral mimicry takes a number of different forms, which may in part be model-dependent. At its simplest, behavioral mimicry can arise through the adoption of diurnal activity patterns which serve to increase the likelihood of mimics and models co-occurring (Howarth et al. 2004). However, mimicry can also involve actively simulating aspects of a model's behavior or morphology. For example, Golding et al. (2001) reported that the hover fly mimic *Eristalis tenax*, resembled its honeybee model in flight behavior. Moreover, wasp-mimicking hover flies have been observed to push the tip of their abdomens into the holder when grasped, a behavior that has been interpreted as an attempt to emulate a stinging insect (Waldbauer 1996). In addition, wasp-mimicking hover flies have been observed to fold their two wings and wag them in a manner similar to a four-winged hymenopteran, and wave their legs in front of their heads. This latter behavior seemingly enhances the resemblance of the fly to their hymenopteran models that have longer antennae (Waldbauer 1970), a feature now formally recognized to be important to birds when discriminating wasps from flies (Bain et al. 2007). Finally, it has been argued that hover flies sound like stinging Hymenoptera when attacked (e.g. Gaul 1952), although the evidence in both bee and wasp mimics is weak at best (Rashed et al. 2009).

The primary aim of this study was to provide a comparative test for an association between morphological and behavioral mimicry in one of the groups best known for it, using current available phylogenies to control for potential lack of independence due to shared evolutionary history. We then provide a more-nuanced analysis of the distributions of behavioral and morphological mimicry in hover flies, including an assessment of the role of model type in mediating this relationship.

Methods

Field work

Hover flies were captured as adults throughout the flying season (April to September) in 2010 between 9 am and 4 pm local time in Ontario (ON) and Quebec (QC), Canada. Our collection sites were Fletcher Wildlife Garden, Ottawa, ON (45°23'08"N, 75°42'15"W); Uxbridge, ON (44°14'30"N, 79°10'41"W); Queens Biological Field Station, Chaffey's Locks, ON (44°34'22"N, 76°20'05"W); Gatineau Park, Gatineau, QC (45°30'31"N, 75°48'59"W) and Mont Rigaud, Rigaud, QC (45°27'06"N, 74°19'33"W). To allow a wide comparison, a broad mixture of (morphologically) mimetic (including honey bee mimics, bumble bee mimics, and wasp mimics) and non-mimetic hover fly species were caught and tested, noting the presence or absence of key behaviors. For consistency, only behavioral data from captured flies were included in our comparative analysis, although the behavioral responses were so readily elicited that they were often evident in specimens both before and after capture.

On capture, specimens were placed in small, clear plastic vials and kept until they were evaluated (within 10 minutes on days above 30°C, but within 2 hours maximum on cooler days). On testing, each fly was placed in a mesh cage (30x30x30 cm), and was allowed 5 minutes to settle. After this period, no qualitative differences were evident between our field and cage-observed behaviors. Following acclimation, the hover flies were assayed by first presenting them with a simulated avian predator (a single stuffed blue jay head – *Cyanocitta cristata*), although even passing a shadow over the flies would often produce the same response. Specimens were then prodded with the beak of the jay up to five times (with a few seconds delay between prods), and all behaviors of the fly (both incidental and those that arose as direct response to the stimulus) were recorded.

The behavioral responses of the flies were classified as follows. A "mock sting" was defined as the specimen tapping the tip of the abdomen onto the beak or surface of the cage in an aggressive manner. If the fly did not exhibit the mock sting when faced with the avian predator, it was lightly grasped between the thumb and forefinger of a researcher (HDP in all cases, for consistency) in an attempt to elicit the behavior. "Wing wagging" was defined as holding and wagging the wings in a wasp-like rather than fly-like manner. "Leg waving" was defined as the specimen resting on their mid and hind legs and waving their fore legs in front of their head. The leg waving movement was clearly distinct from a washing movement. When observations were complete, the specimens were killed (using either cyanide or freezing), pinned, and identified. When possible, specimens were identified to species level using morphology or, in a few cases, genotyping (in genera such as *Syrphus* and *Sphaerophoria* identification to species in females is not possible with the current key).

Quantification of mimetic similarity

We obtained estimates of the mimetic fidelity of representatives of 56 of the 57 recognized hover fly species that were behaviorally assayed during the field season, to five different potential hymenopteran models (see below). Our approach employed a simple human ranking system which correlates well both with an avian assessment of hover fly mimetic similarity and a multidimensional measure of morphological similarity of hover flies to their hymenopteran

models (Penney et al. 2012). While humans and birds have different visual systems, there is no evidence of a widespread UV component to either wasp or hover fly mimetic patterns (Gilbert 2005), which may explain why quantifications of mimetic similarity by pigeons were broadly consistent when pigeons were presented with naturally lit specimens of hover flies (Green et al. 1999) compared to their projected images (Dittrich et al. 1993).

Those hover fly species assayed in the field that could only be identified to genera were represented by several congeneric species in our similarity assessments, ultimately generating mimetic fidelity estimates for 77 hover fly species. Although we could not reliably use these additional species when testing for an association between behavioral and morphological mimicry, we did use this information in our tests of rater consistency and model-dependent similarities (see below).

Photographs of the dorsal view of three different pinned individuals from each of the 77 hover fly species were taken from specimens stored at the Canadian National Collection of Insects, Arachnids and Nematodes (CNC) in Ottawa, ON. In addition, three photographs of members of the five separate hymenopteran species that may serve as models of these hover flies were taken: the honeybee *Apis mellifera*, the common wasp *Vespula alascensis*, the buff-tailed bumblebee *Bombus terrestris*, the eastern bumblebee *Bombus impatiens* and the bald-faced hornet *Dolichovespula maculata* (see Online Appendix 1). All of the photographs were taken using a Canon EO5 50D with a Canon macro lens (100 mm). Illumination was provided by an 80 LED microscope ring light (KD-200).

Estimates of the extent of mimetic fidelity of mimics to models were conducted in three participant blocks with 24 participants in block 1 and 10 participants in blocks 2 and 3 (44 in total). Each block involved projecting a photo of each of the 77 hover fly species (presented in random order) alongside one photo for each of the five models for comparison. The species photos for both models and mimics, and the order of presentation of mimics, were changed for each block. Human participants were asked to rank each hover fly photograph on a scale of 1 (very poor mimic) to 10 (very good mimic) separately for each of the 5 potential models shown. To produce a mimetic fidelity score, we first identified the model type to which the potential mimic bore the closest resemblance (based on overall mean score for images of that species). We then noted the extent of mimetic fidelity to that model based on the mean score. It can be extremely challenging to match mimic to model (if a match exists at all, Edmunds 2000) in systems where the phenotypic diversity of mimics and models is exceptionally high (Wilson et al. 2013). Our limitation of five models, albeit with relatively high phenotypic diversity, was therefore out of necessity rather than desire.

Comparative analysis

The strength of the association between the average mimetic fidelity of a species and whether it engaged in any of our assayed behaviors was first evaluated using logistic regression. However, a group of species may all be good morphological mimics and all may engage in behavioral mimicry, but this commonality may arise from a shared common ancestor rather than as a result of independently driven relationships. We therefore analysed our results in the

context of the likely phylogenetic relationship among species, directly accounting for any lack of statistical independence arising from common ancestry. To do so, we first generated a molecular phylogeny for 55 of the 57 hover fly species tested for behavior using variation at the cytochrome oxidase c subunit I (COI) – see Penney et al. (2012). DNA extraction and sequencing were performed in house and at the Canadian Centre for DNA Barcoding (see Online Appendix 2 for GenBank accession numbers of specimens used to construct the phylogeny). The resultant phylogeny included most major syrphid clades (Figure 1). Reassuringly, all species fell within their prospective genera and our tree was largely congruent with other published results that used nuclear loci (Skevington and Yeates 2000; Ståhls et al. 2003; Mengual et al. 2008). We then tested for a phylogenetic signal in behavioral mimicry and mimetic fidelity (the highest rating given to each species) by comparing the phylogeny based on COI with a null model in which all branches were equal in a likelihood ratio test (see Hossie et al. 2013 for more details). Finally, to test for an association between mimetic fidelity and behavior while controlling for phylogeny we fitted Monte Carlo Markov Chain generalised linear models, using phylogenetic covariance matrices derived from the phylogeny described above. Model fitting was implemented using the MCMCglmm package in R (R: Development Core Team 2013) - for details on methods, see Hadfield (2010). Using the above complementary approaches, we can evaluate the impact of accounting for phylogeny on our findings and provide a more robust analysis of the data. Note that one fewer species was available for the phylogenetically controlled analysis (n=55, *vs.* n=56 for the logistic regression without phylogenetic control) due to the lack of COI sequence for *Temnostoma obscurum.*

Results

We caught and assayed 359 hover fly specimens. Of those specimens, 320 individuals were identified to species, giving 57 species in total. The remaining 39 specimens for which species-level identification was not possible were from the genera *Eupeodes* (n=1 specimen), *Melangyna* (n=1), *Platycheirus* (n=9), *Sphaerophoria* (n=10) and *Syrphus* (n=18). Online Appendix 3 lists the sample sizes of each identified species tested along with details of their behavior. There was no within-species variation of behavioral mimics: if one individual did or did not engage in the behavior, then all individuals of the species did the same. Of the 57 species assayed (and 39 additional specimens of known genera), there were only six species (*Spilomyia sayi*, *S. fusca*, *S. longicornis*, *Temnostoma alternans*, *T. barberi*, and *T. obscurum*) that exhibited any evidence of behavioral mimicry and all six of these hover fly species mimic wasps (as determined by our human ranking system). Five of the six mimetic species that engaged in behavioral mimicry exhibited all three behaviors, but *S. sayi* (13 specimens tested) did not engage in leg waving. The consistency in association between different forms of behavioral mimicry was highly significant. For example, of the 43 hover fly species ultimately classed as wasp mimics, the propensity to engage in wing wagging (6 species) was significantly associated with the propensity to engage in mock stinging ($G_1 = 34.754$, $P < 0.001$).

Intra-class correlation coefficients for ratings of similarity of 77 hover fly mimics to 5 hymenopteran models by all 44 human raters, were calculated using the icc function in the irr package (Gamer et al. 2012) in R. All tests showed significant ($P < 0.001$) consistency among raters (high ratings by one coder for a given hover fly species correspond with high ratings with

other) and agreement in absolute terms (Hallgren 2012) between raters (see Online Appendix 4). Hover fly species differed in their mean maximum similarity according to model, with hover flies classified as bumble bee mimics rated as the highest fidelity mimics (ANOVA $F_{3,73} = 5.68$, P = 0.0015, Online Appendix 5).

A comparison of the distributions of mimetic similarity and behavioral mimicry in both informative phylogenetic trees and control trees (where all branches are the same) showed that there was significant phylogenetic autocorrelation in behavioral mimicry (λ=1, p=0.002) but not in mimetic similarity (λ<0.001, p=1.000). Hence related species were more likely to share the same propensity to engage in behavioral mimicry, but not necessarily similar degrees of mimetic fidelity. Logistic regression models generally showed no association between behavioral mimicry and the mimetic fidelity of species to their most-similar model (Table 1, Figure 2). However, since only wasp mimics exhibited the behavior, the data were reanalysed to assess the effects of similarity to the nearest wasp model (*D. maculata* or *V. alascensis*). This second analysis was statistically significant, suggesting a greater likelihood of behavioral mimicry in species that more closely resembled wasps (Table 1), particularly when accounting for phylogeny. Finally, we consider the same relationship between behavioral and morphological mimicry using only those hover flies that were rated as being most similar to wasps (either *V. alascensis* or *D. maculata*, n=42 with phylogenetic control). Here, the trend for greater likelihood of behavioral mimicry at higher levels of morphological mimicry approached significance (p=0.077, Table 1).

Discussion

After systematically evaluating the behavior of a wide range of field caught hover flies and empirically evaluating their morphological similarity to a variety of hymenopteran models, we can confidently reject the hypothesis that the leg waving, wing wagging and mock-stinging behavior of hover flies is primarily selected to compensate for poor visual mimicry. Instead we conclude that there is evidence for a positive association between mimetic fidelity and these behavioral traits, even after controlling for potential lack of independence between species due to shared phylogeny. However, the pattern was only statistically significant when evaluating the extent of similarity of hover flies to wasps. Below, we discuss reasons why the relationship between morphological and behavioral mimicry might be clearer when considering only wasp mimicry, before going on to discuss the wider implications of the positive association.

The hover flies that were ultimately classified as bee mimics by human observers did not engage in any of the behavioral traits we assayed for (leg waving, wing wagging, mock sting). This observation is consistent with Waldbauer (1970)'s earlier proposal that hover flies which mimic bees neither have, nor give the appearance of having, long antennae. Hymenopteran models clearly vary in appearance, but a simple reason for the absence of leg waving in bee mimics may be that many of the common bee models lack the long antennae of wasps, so there may be less value in portraying long antennae. In addition, many bees do not wag their wings in quite the same manner as wasps, and there is little evidence that birds find the sting of a bee a significant deterrent (Gilbert 2005). Collectively therefore, there may have been little additional selection pressure to adopt behavioral mimicry in this group of mimics, because their models do

not exhibit sufficiently characteristic behaviors and/or because the behaviors themselves do not evoke aversive responses in observers.

Our work also confirms, to human eyes at least, that bee mimics are among the highest fidelity mimics (Gilbert 2005). This close mimicry of bee mimics may well have arisen as a consequence of the models' lack of averseness which requires close similarity before a reasonable high degree of protection from predators is achieved (Sherratt 2002; Gilbert 2005). However, bumblebee models and their mimics both have a hairy texture (Online Appendix 1), and simply sharing this characteristic may go some way to rendering a close visual similarity. Whatever the reason for the close similarity, given the lack of behavioral mimicry in bee mimics and their high fidelity, it is not surprising that the relationship between behavioral and morphological mimicry is most evident when we consider wasp mimicry alone.

Here we have employed a standardised field-based assay to generate an extensive list of hover fly species that engage in three specific forms of behavioral mimicry, but also those hover fly species that do not behave in this way. Our general, albeit tentative, evidence that the propensity to engage in behavioral mimicry is related to the extent of mimetic fidelity to wasps confirms earlier qualitative suggestions of both Nicholson (1927) and Waldbauer (1970) who noted that behavioral mimicry is generally found in those species in which the overall mimetic resemblance to Hymenoptera is the most highly developed. As in our study, Waldbauer (1970) reported mimicry of antennae through leg waving in just two hover fly genera namely *Spilomyia* (*S. hamifera, S. fusca, S. longicornis, S. quadrifasciata*) and *Temnostoma* (*T. balyras, T. pictulum, T. trifasciatum*). Hover flies from the genera *Spilomyia* and *Temnostoma* are generally regarded as good mimics and there were no examples of a behavioral mimic that received a low human mimetic fidelity ranking. To humans therefore, behavioral mimics were always good or intermediate mimics, but not all good or intermediate mimics were behavioral mimics. For example, the hover fly *Syrphus ribesii* appears as a good mimic (Online Appendix 1) and yet despite testing 26 field-caught individuals, none showed any form of behavioral mimicry.

Our three distinct behaviors (leg waving, wing wagging, mock stinging) were all significantly associated with one another, suggesting the evolution of an adaptive suite of behaviors as a response to selection. The one exception to this pattern was *S. sayi* which wagged its wings and mock stung, but did not leg wave. Intriguingly, *S. sayi* had longer antennae (0.437 cm ± 0.008 (SE)) than the other members of the genus that showed leg waving (*S. fusca* 0.266cm ± 0.002; *S. longicornis* 0.318cm ± 0.007), suggesting that the behavior may be of less value to *S. sayi* because it already has relatively large antennae. Nevertheless, the six behavioral mimics all shared a common trait in the dark pigmentation of the tibia and tarsi of the forelegs, which were a different colour to the mid- and hindlegs (Figure 3). While other species in the study exhibit wide variation in the colour of legs (both different legs and segments within legs), the *Spilomyia* and *Temnostoma* species were the only species of our 57 surveyed to exhibit this particular contrasting pattern. Waldbauer (1970) likewise noted that in each of the four *Spilomyia* species he investigated the front tarsi and a part of the front tibiae were dark. Given that the antennae of models are frequently dark, it is not unreasonable to suggest that the

morphology and behavior are associated. Indeed, a morphological examination might provide one of the first clues as to whether a species is likely to engage in behavioral mimicry.

Overall, we find that behavioural mimicry does not compensate for poor morphological mimicry in hover flies but it may supplement good morphological mimicry in at least some species. One might expect mimetic behaviours to readily evolve as an additional line of defence in any mimetic species if it serves to inhibit predator attack, perhaps by further convincing predators they are unprofitable. Therefore, it is an open question as to why not all good morphological mimics have evolved such behavioral traits.

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Table 1. Relationships between behavioral mimicry and mimetic similarity (either the greatest similarity to any hymenopteran model, or the greatest similarity to either of the two wasp models, "wasp similarity"). MCMCglmm models incorporate phylogenetic autocorrelation and logistic regression models do not. In each of the three analyses, the logistic regression without phylogenetic information incorporated one additional species, *Temnostoma obscurum*, which was assayed for behavioral mimicry and mimetic similarity but for which genetic data were not available. The removal of this species from the logistic regression to give the same set of species in both analyses does not qualitatively affect the results (max similarity: z=1.289, p=0.198; wasp similarity: z=1.861, p=0.063; wasp mimics only: z=1.547, p=0.122).

	Method	Predictor	Parameter estimate	P
Overall similarity	MCMCglmm	Intercept	-5379	0.020
	(n=55)	Max similarity	528	0.152
	Logistic regression	Intercept	-4.830	0.093
	(n=56)	Max similarity	0.444	0.327
Wasp similarity	MCMCglmm	Intercept	-4778	0.002
	(n=55)	Wasp similarity	570	0.019
	Logistic regression	Intercept	-7.166	0.021
	(n=56)	Wasp similarity	0.859	0.082
Wasp mimics only	MCMCglmm	Intercept	-4613	0.010
	(n=42)	Wasp similarity	541	0.077
	Logistic regression	Intercept	-5.903	0.075
	(n=43)	Wasp similarity	1.287	0.198

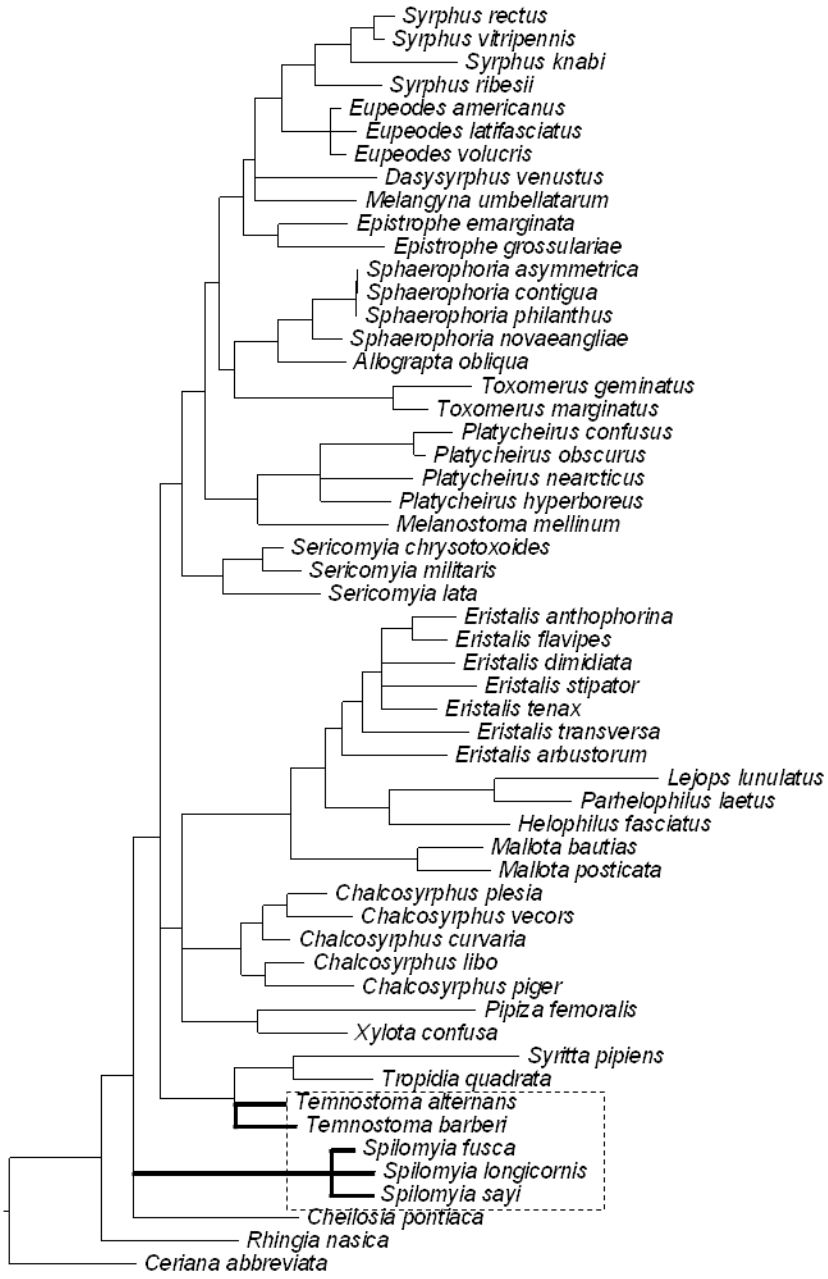

Figure 1. Phylogenetic tree of 55 field-tested hover fly species with behavioral mimics (those that wing wag, leg wave or mock sting) highlighted in bold within the dotted box. Note that while the tree has been oriented to show behavioral mimics within one box, the genera *Temnostoma* and *Spilomyia* are not sister taxa (e.g. see Ståhls et al. 2003).

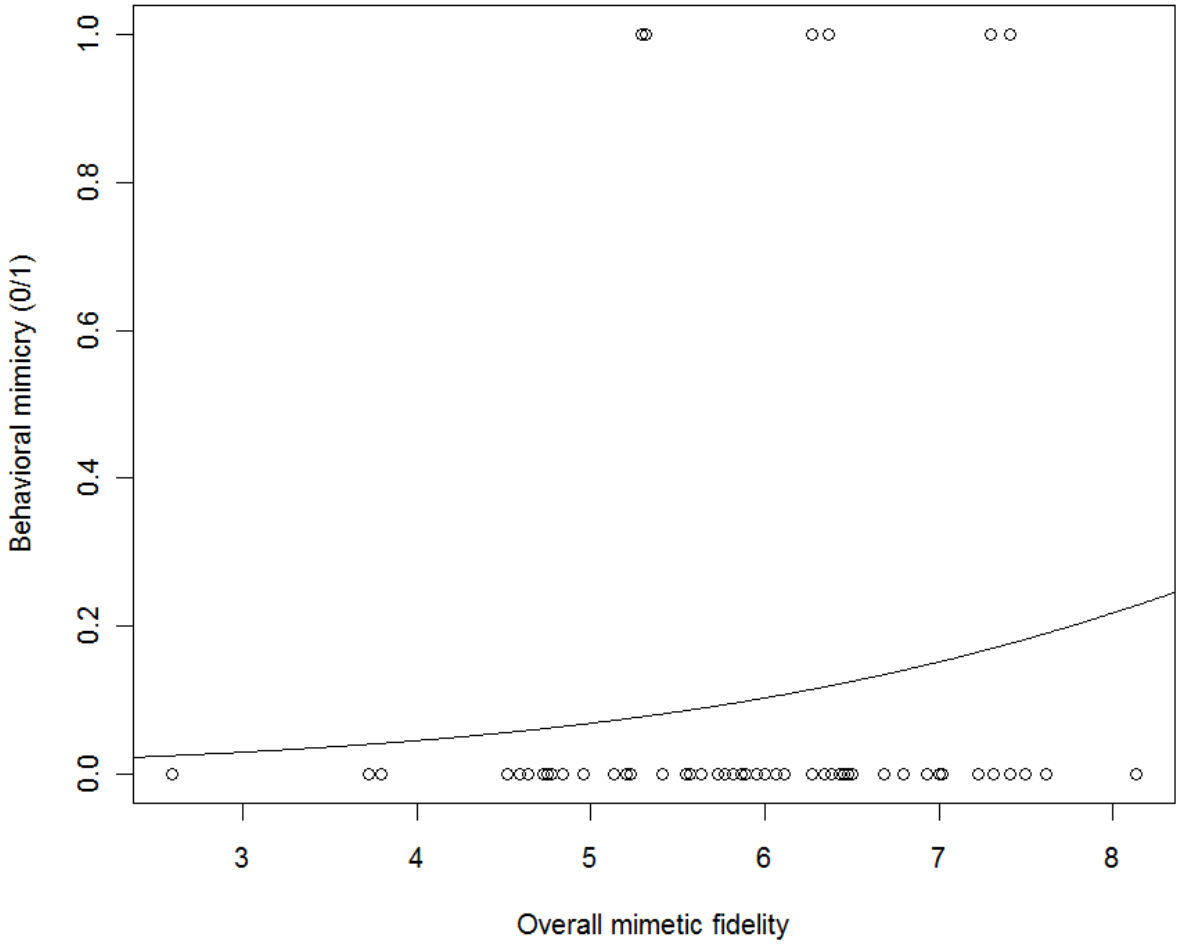

Figure 2. The relationship between the mean mimetic fidelity of a species to its closest model and whether or not it is a behavioral mimic. Behavioral mimics had relatively high mimetic fidelity (to wasps), but there were species with high mimetic fidelity that did not exhibit the behavior. Line shows the fit of a logistic regression model assuming phylogenetic independence.

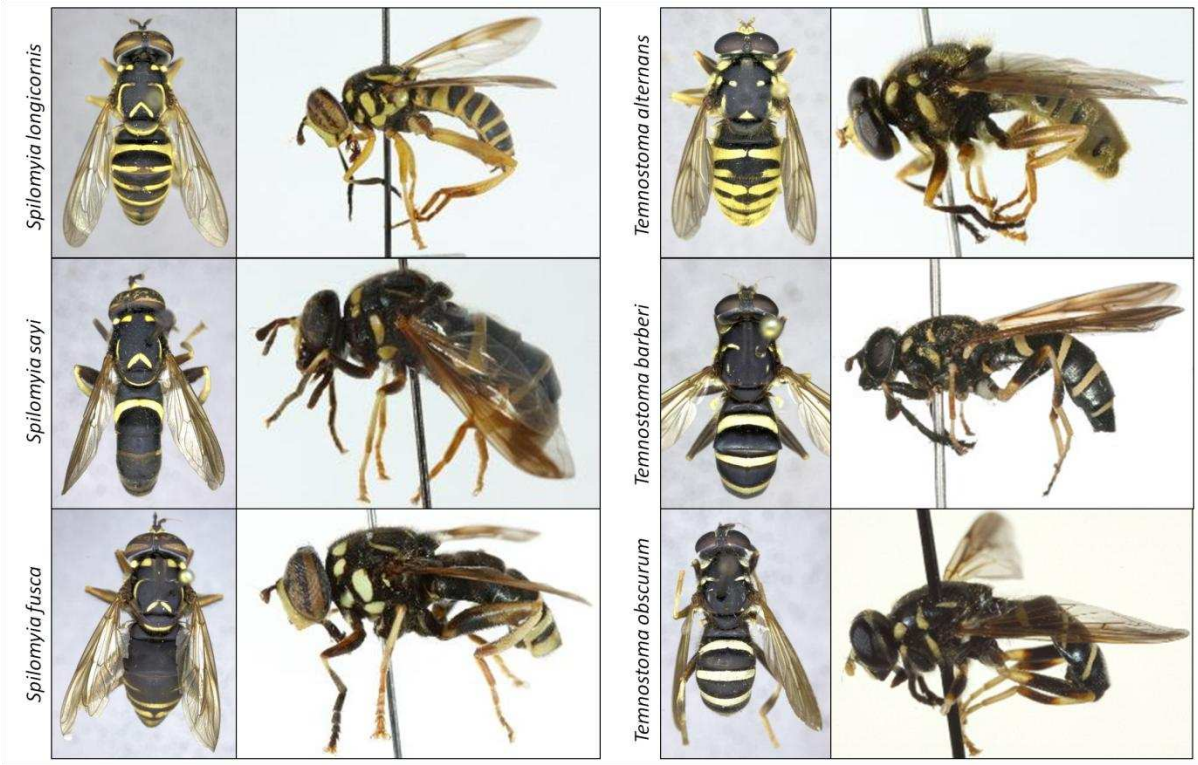

Figure 3. Proposed morphological adaptations to behavioral mimicry in six hover flies (Diptera: Syrphidae) exhibiting behavioral mimicry. Note in each of the lateral views that all behavioral mimics possess heavily-pigmented forelegs. Dorsal view photographs are by Brent Lamborn, lateral view photographs are by CNC/BIO Photography Group, Biodiversity Institute of Ontario.

Online Supplementary Appendices

The relationship between morphological and behavioral mimicry in hover flies (Diptera: Syrphidae)

Heather D. Penney, Christopher Hassall, Jeffrey H. Skevington, Brent Lamborn & Thomas N. Sherratt

Appendix 1. A standard presentation of a hover fly (*Syrphus ribesii*) along with five potential models (left to right: *Apis mellifera*, *Bombus impatiens*, *Bombus terrestris*, *Dolichovespula maculata*, *Vespula alascensis*).





Appendix 2. GenBank accession numbers and Canadian National Collection unique identifiers for specimens used to construct the phylogeny.

Species	Unique Identifier	GenBank Number
Allograpta obliqua	CNC DIPTERA 11743	KC900491
Ceriana abbreviata	CNC DIPTERA 37873	KC900433
Chalcosyrphus curvaria	CNC DIPTERA 38022	KC900488
Chalcosyrphus libo	CNC DIPTERA 647	KC900498
Chalcosyrphus piger	JSS-15350	KC900460
Chalcosyrphus plesia	CNC DIPTERA 40866	KC900448
Chalcosyrphus vecors	CNC DIPTERA 40974	KC900454
Cheilosia pontiaca	CNC DIPTERA 101552	KC900496
Dasysyrphus venustus	CNCD 30893	HQ577348
Epistrophe emarginata	CNC DIPTERA 103074	JN991975
Epistrophe grossulariae	CNC DIPTERA 105425	KC900427
Eristalis anthophorina	CNC DIPTERA 101997	KC900462
Eristalis arbustorum	CNCD6652	KC900447
Eristalis dimidiata	CNC DIPTERA 67123	KC900429
Eristalis flavipes	CNC DIPTERA 56765	KC900476
Eristalis stipator	CNC DIPTERA 1592	KC900461
Eristalis tenax	JSS-16952	KC900459
Eristalis transversa	CNC DIPTERA 102012	KC900430
Eupeodes americanus	CNC DIPTERA 11194	KC900495
Eupeodes latifasciatus	CNCD9199	KC900451
Eupeodes volucris	HP211	KC900434
Helophilus fasciatus	CNC DIPTERA 39802	KC900443
Lejops lunulatus	CNC DIPTERA 44807	KC900442
Mallota bautias	JSS18768	KC900487
Mallota posticata	CNC DIPTERA 45552	KC900456
Melangyna umbellatarum	CNC DIPTERA 105726	KC900446
Melanostoma mellinum	CNCD9349	KC900435
Parhelophilus laetus	CNC DIPTERA 47694	KC900472
Pipiza femoralis	CNC DIPTERA 48205	KC900441
Platycheirus confusus	CNC DIPTERA 25175	KC900449
Platycheirus hyperboreus	CNC DIPTERA 26022	KC900439
Platycheirus nearcticus	CNC DIPTERA 27310	KC900428
Platycheirus obscurus	CNCD11206	KC900463
Rhingia nasica	JSS18895	KC900465
Sericomyia chrysotoxoides	CNC DIPTERA 66	KC900450
Sericomyia lata	CNC DIPTERA 33901	KC900477
Sericomyia militaris	CNC DIPTERA 30237	KC900484
Sphaerophoria asymmetrica	CNC DIPTERA 101522	KC900483
Sphaerophoria contigua	CNC DIPTERA 75676	KC900440
Sphaerophoria novaeangliae	CNC DIPTERA 76214	KC900486
Sphaerophoria philanthus	HP177	KC900490

Spilomyia fusca	CNC DIPTERA 49700	KC900493
Spilomyia longicornis	CNC DIPTERA 49784	JN992036
Spilomyia sayi	CNC DIPTERA 50934	KC900444
Syritta pipiens	CNC DIPTERA 106560	KC900458
Syrphus knabi	CNC DIPTERA 78728	KC900475
Syrphus rectus	CNC DIPTERA 80057	KC900464
Syrphus ribesii	CHU06-SYR-103	KC900426
Syrphus vitripennis	CNC DIPTERA 105797	KC900478
Temnostoma alternans	CNC DIPTERA 54108	JN992045
Temnostoma barberi	CNCD-2203	KC900489
Toxomerus geminatus	CNC DIPTERA 106058	KC900492
Toxomerus marginatus	CHU06-SYR-074	KC900453
Tropidia quadrata	CNC DIPTERA 55141	KC900467
Xylota confusa	CNC DIPTERA 103101	KC900468

Appendix 3. Species list (n=57) for hover flies (Diptera: Syrphidae) that were assayed for behavioral mimicry. Also shown are (i) the hymenopteran model which each species most closely resembled (AM=*Apis mellifera*, VA=*Vespula alascensis*, BT=*Bombus terrestris*, DM=*Dolichovespula maculata*, or BI=*Bombus impatiens*), (ii) the similarity to that model (out of 10) with standard error, (iii) the greatest similarity to either of the wasp models (*Vespula alascensis* or *Dolichovespula maculata*) with standard error, (iv) the sample size ("N"), and whether or not the species exhibited behavioral mimicry. Behaviors are designated as "ww" for wing-wagging, "lw" for leg-waving, and "ms" for "mock sting".

Species	Genetic data	Closest model	Max similarity	Max similarity SE	Wasp similarity	Wasp similarity SE	N	Behavioral mimicry
Allograpta obliqua	Yes	*VA*	3.795	0.238	3.795	0.238	7	No
Ceriana abbreviata	Yes	*DM*	6.068	0.231	6.068	0.231	1	No
Chalcosyrphus curvaria	Yes	*DM*	4.955	0.294	4.955	0.294	5	No
Chalcosyrphus libo	Yes	*DM*	2.591	0.226	2.591	0.226	1	No
Chalcosyrphus piger	Yes	*DM*	4.750	0.246	4.750	0.246	3	No
Chalcosyrphus plesia	Yes	*DM*	6.386	0.232	6.386	0.232	1	No
Chalcosyrphus vecors	Yes	*DM*	6.000	0.265	6.000	0.265	1	No
Cheilosia pontiaca	Yes	*DM*	5.886	0.285	5.886	0.285	6	No
Dasysyrphus venustus	Yes	*VA*	6.432	0.246	6.432	0.246	2	No
Epistrophe emarginata	Yes	*VA*	7.614	0.183	7.614	0.183	13	No
Epistrophe grossulariae	Yes	*VA*	6.341	0.225	6.341	0.225	7	No
Eristalis anthophorina	Yes	*BI*	7.409	0.156	3.295	0.205	2	No
Eristalis arbustorum	Yes	*AM*	5.818	0.194	3.909	0.220	17	No
Eristalis dimidiate	Yes	*AM*	5.864	0.219	5.068	0.300	1	No
Eristalis flavipes	Yes	*BI*	7.318	0.180	2.114	0.171	11	No
Eristalis stipator	Yes	*AM*	5.773	0.250	4.114	0.241	1	No
Eristalis tenax	Yes	*AM*	7.500	0.206	5.023	0.235	12	No
Eristalis transversa	Yes	*AM*	5.545	0.223	4.773	0.221	7	No
Eupeodes americanus	Yes	*VA*	7.023	0.178	7.023	0.178	5	No
Eupeodes latifasciatus	Yes	*VA*	6.455	0.186	6.455	0.186	4	No
Eupeodes volucris	Yes	*VA*	6.795	0.175	6.795	0.175	5	No
Helophilus fasciatus	Yes	*VA*	6.477	0.232	6.477	0.232	3	No
Lejops lunulatus	Yes	*VA*	6.273	0.239	6.273	0.239	3	No
Mallota bautias	Yes	*BI*	5.568	0.238	3.045	0.247	1	No
Mallota posticata	Yes	*BI*	8.136	0.157	2.318	0.157	4	No
Melangyna umbellatarum	Yes	*VA*	7.000	0.248	7.000	0.248	2	No
Melanostoma mellinum	Yes	*DM*	4.523	0.251	4.523	0.251	4	No
Parhelophilus laetus	Yes	*AM*	6.795	0.180	5.795	0.193	6	No
Pipiza femoralis	Yes	*DM*	5.955	0.224	5.955	0.224	1	No
Platycheirus confusus	Yes	*DM*	5.864	0.240	5.864	0.240	1	No
Platycheirus hyperboreus	Yes	*VA*	3.727	0.231	3.727	0.231	1	No
Platycheirus nearcticus	Yes	*VA*	5.409	0.239	5.409	0.239	2	No

Platycheirus obscurus	Yes	*VA*	5.545	0.202	5.545	0.202	16	No
Rhingia nasica	Yes	*AM*	5.227	0.236	4.023	0.211	1	No
Sericomyia chrysotoxoides	Yes	*VA*	6.114	0.209	6.114	0.209	7	No
Sericomyia lata	Yes	*VA*	4.841	0.211	4.841	0.211	1	No
Sericomyia militaris	Yes	*DM*	5.568	0.249	5.568	0.249	2	No
Sphaerophoria asymmetrica	Yes	*VA*	5.205	0.225	5.205	0.225	1	No
Sphaerophoria contigua	Yes	*VA*	5.136	0.271	5.136	0.271	1	No
Sphaerophoria novaeangliae	Yes	*VA*	4.727	0.253	4.727	0.253	3	No
Sphaerophoria philanthus	Yes	*VA*	4.773	0.214	4.773	0.214	7	No
Spilomyia fusca	Yes	*DM*	6.273	0.255	6.273	0.255	2	Yes – LW, WW, MS
Spilomyia longicornis	Yes	*VA*	7.409	0.176	7.409	0.176	16	Yes – LW, WW, MS
Spilomyia sayi	Yes	*DM*	6.364	0.258	6.364	0.258	13	Yes – WW, MS
Syritta pipiens	Yes	*VA*	4.591	0.200	4.591	0.200	16	No
Syrphus knabi	Yes	*AM*	6.455	0.232	6.205	0.214	1	No
Syrphus rectus	Yes	*VA*	6.500	0.240	6.500	0.240	4	No
Syrphus ribesii	Yes	*VA*	7.227	0.232	7.227	0.232	26	No
Syrphus vitripennis	Yes	*VA*	6.932	0.222	6.932	0.222	10	No
Temnostoma alternans	Yes	*VA*	7.295	0.142	7.295	0.142	7	Yes – LW, WW, MS
Temnostoma barberi	Yes	*VA*	5.295	0.177	5.295	0.177	4	Yes – LW, WW, MS
Temnostoma obscurum	No	*DM*	5.318	0.262	5.318	0.262	1	Yes – LW, WW, MS
Toxomerus geminatus	Yes	*VA*	5.636	0.221	5.636	0.221	22	No
Toxomerus marginatus	Yes	*VA*	5.727	0.211	5.727	0.211	9	No
Tropidia quadrata	Yes	*AM*	4.636	0.259	3.705	0.206	2	No
Volucella bombylans	No						6	No
Xylota confusa	Yes	*DM*	6.682	0.225	6.682	0.225	2	No

Appendix 4. Intra-class correlation coefficients for ratings of the similarity of 77 hover fly mimics to 5 Hymenoptera models by 44 human raters, showing the consistency (relative rating of mimetic similarity irrespective of absolute judgements) and agreement (the absolute ratings) between observers. All coefficients are significant at $p<0.001$.

Model	Consistency	Agreement
Apis mellifera	0.409	0.361
Vespula alascensis	0.454	0.411
Bombus terrestris	0.424	0.342
Dolichovespula maculata	0.316	0.251
Bombus impatiens	0.504	0.416

Appendix 5. Box and whisker plots showing similarity distribution of species to their overall closest ranked model (*Bombus terrestris* was never the closest ranked model). Median and interquartile ranges are shown, along with means (squares), the maxima and minima (whiskers) and outliers (stars).

