

Scratching for food

An original feeding behavior in an African flower breeding *Drosophila*

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Drosophila suma is a flower breeding species widespread in the Afrotropical region. We describe an original and so far unique feeding behavior in that species. Strong black spines on the fore tarsus of both sexes are used for scratching the surface of the petals: the juice pouring out from the scratched cells is immediately ingested and this might be the only source of food. *D. suma* has been investigated on two kinds of flowers, Ipomoea and Crinum. In spite of the very different shape and color of these flowers, the feeding behavior of the flies is the same. Various morphological and anatomical adaptations are described, especially those related to laying a few very big eggs. The taxonomic position of *D. suma* has long remained and still is elusive. Further taxonomic and phylogenetic investigations would thus be needed to understand how adaptation to flower breeding has evolved.

Introduction

With almost 4,000 described species, the family Drosophilidae is remarkable by its large diversity, not only for size, shape and pigmentation, but also for its ecological niches. Most species are saprophytes, feeding on fungi developing in decaying plant materials, including fruits, leaves, bark and fungi, while some more bizarre niches are also known, related to parasitism or predation.¹

Flower breeding species are numerous in all tropical places and convergent adaptations mainly in the shape of the ovipositor (egg guide) have been observed.^{2,3} A general feature is the reduction or absence of egg filaments. Larvae feed in decaying flowers and can be reared, often abundantly, from such resources. Adult life history traits are far less known, and especially the feeding resources. It has sometimes been assumed that pollen could be a convenient protein source:³ this is unlikely for adult flies because the pollen diameter often exceeds the size of the esophagus, the diameter of which is about 25 µm (David JR, unpublished).

In Tropical Africa, flower-breeding Drosophilid species belong to the genera *Apenothecia* Tsacas, *Scaptodrosophila* Duda, *Zaprionus* Coquillett and *Drosophila* Fallen.^{4,5} In the latter genus, only species belonging to the subgenus *Sophophora* and a pair of closely related species, *D. spinipes* Lamb and *D. suma* Burla, are bred from flowers. The subgeneric classification of these two species is unknown^{6,7} but they can easily be identified by the presence, in both sexes of a row of 6–7 strong dark spines or pegs on the first two tarsomeres of the foreleg (Fig. 1H). These pegs are completely different from the male sex combs observed

in many species belonging to the subgenus *Sophophora*, such as *D. melanogaster*. Here, we describe the function of these spines in feeding behavior and discuss other possible morphological adaptations to flower breeding.

Results

Adult behavior or ecology. Up to now, the possible function of the characteristic tarsal pegs remained elusive. We report here that they permit a peculiar feeding behavior, which to our knowledge, is original among all Drosophilids.

Ecological field observations were made in three places: The Dimonika field station in Congo Republic, The rainforest in Ranomafana (Madagascar) and, to a lesser degree, Mauritius island. Two kinds of very different flowers were used by the flies: the small pink flowers of *Ipomoea* (an introduced species) in Congo and Mauritius and the large white flowers of *Crinum* (a native species) in Madagascar (Fig. 1). In spite of these very different resources, the behaviors of the flies were strikingly identical, as well as their morphology, and they probably belong to the same, widespread species *Drosophila suma*. Adults are black on the head and the dorsal parts of the thorax and abdomen while the lower part of the body and the legs are completely white (Fig. 1F and G). Such a contrast, also observed in the *Mycodrosophila* Oldenberg genus, might be a cryptic camouflage, making the overall shape of the fly more difficult to identify by a predator. However, we found that predation by spiders on the *Crinum* flowers was quite common (Fig. 1G).

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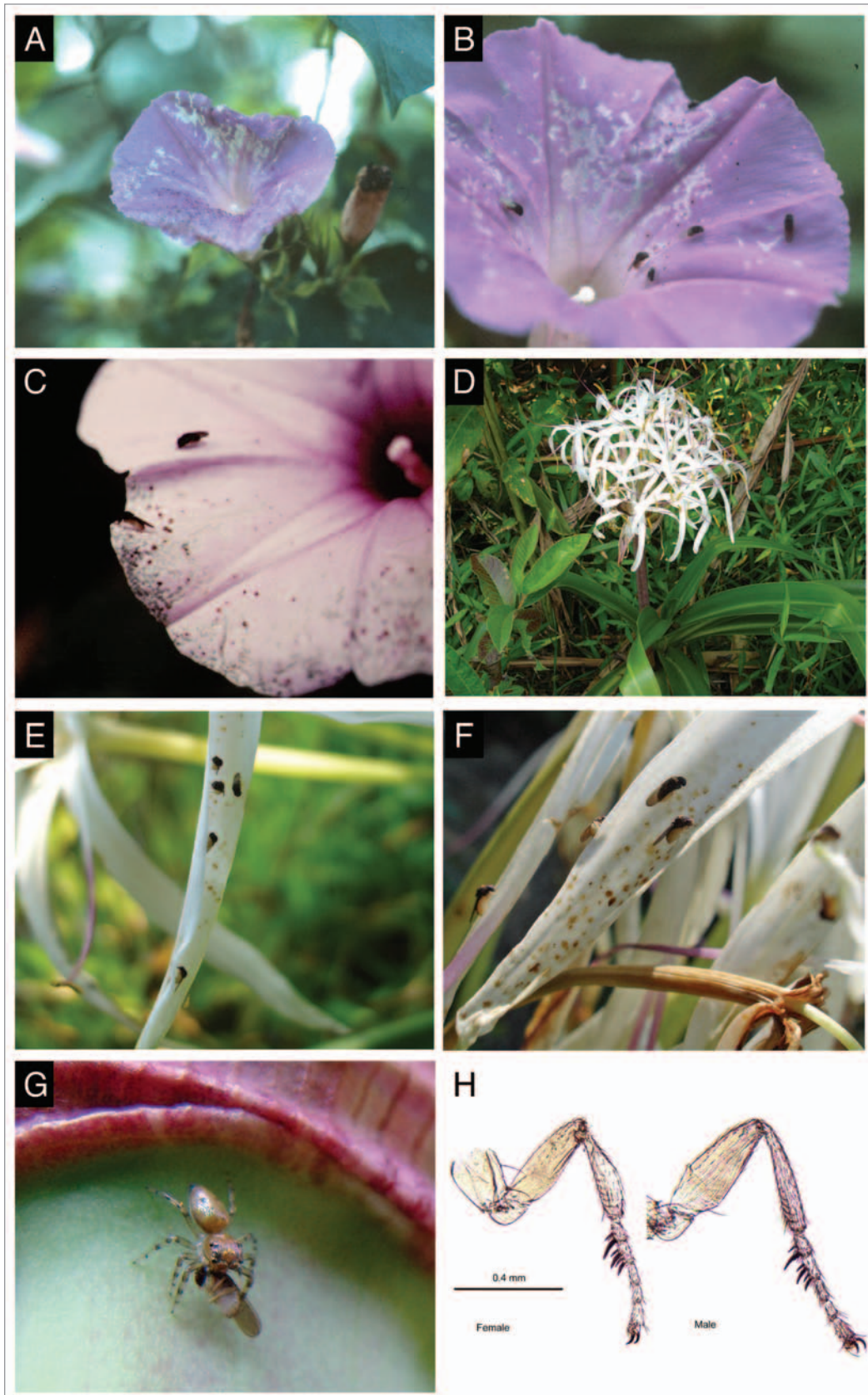


Figure 1. Various aspects of *D. suma* biology. (A) Ipomoea flower with traces of scratching on corolla; (B) idem with *D. suma* adults; (C) Ipomoea flower with scratching traces and droppings; (D) Crinum sp overall view of the flower; (E and F) adults of *D. suma* on a Crinum petal with orange droppings; (G) *D. suma* female captured by a salticid spider; (H) fore leg of *D. suma* with scratching spines on the tarsus; note the identical spines in female and male.

On Ipomoea flowers adults are found on the internal part of the corolla (Fig. 1B and C). After landing, they start to scratch the plant epidermis with the spines of the foreleg tarsus. This behavior, easily observed in nature, produces a liquid pouring out the wounded cells, and this liquid is immediately swallowed by the proboscis. To get fresh juice, the fly moves slowly backwards, continuing to scratch and feed. After a few minutes, a scar is observed on the corolla, revealing by a color change on the petals following the movements of the fly (Fig. 1A and B). On Crinum flowers, the behavior was the same in spite of the fact that the flower aspect was very different, by its much larger size, white color and separate petals. Flying adults were easily observed landing on a petal and then starting to scratch. Flies were not equally distributed on all petals, but tended to concentrate on a few ones, as if there was a preference or a kind of social attraction. We also noticed that, when the flowers were in the shadow (along a river side), they stayed on the upper part of the petals, but moved down when the petals came in the sunshine. Scratching scars were not visible with a naked eye, since there was no color change in the white color of the flower. However, these scratches were easily observed with a binocular microscope. We also noticed, on both kinds of flowers, dry tiny drops of a material with a deep orange color. These drops were present only in Ipomoea flowers or on petals of Crinum where adult *D. suma* were abundant, as indicated by the presence of many scars on petals (see for example Fig. 1C and F).

In summary, *D. suma* adults ingest a large amount of liquid pouring out of the petals, and probably from the cell vacuoles. A major question remains: is this juice the only food? And is it sufficient, especially for the females, to provide the large amount of proteins needed for oogenesis? During the day, adult flies do not remain all the time on the flowers, and it is quite possible that they use another, more consistent but unknown, food.

By dissecting the females, we found that each ovary contained only one or two egg chambers at the end of vitellogenesis, while all other ovarioles contained only small, previtellogenic chambers. For that reason, we could not count precisely the ovariole number, but the only certitude is that it is very small, around five for each ovary. In one case, a female had a mature egg in the uterus, ready to be laid, and we were surprised by its very big size: 0.8 mm in length and 0.26 in width, without any filaments (Fig. 2E). Most fruit-breeding *Drosophila* species that are bred in the laboratory have much smaller eggs, for example the dimensions in *D. melanogaster* are 0.53 by 0.19 mm.⁹ Such a large egg, correlated with a small ovariole number and a low daily fecundity, reveals a strong parental investment in the survival of a few progeny.

We also noticed a remarkable morphological change of the posterior part of the abdomen, i.e., a very large size of the last, seventh segment (Fig. 2F and G). In *Drosophila* species like *D. melanogaster*, the sclerified tergite 7 is small, and the two lateral parts are not fused on the median dorsal line. By contrast, the seventh segment of *D. suma* is much bigger and the tergites are made of two parts: a lateral part having a triangular shape and harboring about 20 bristles, and an unusually very developed dorsal region making a large dorsal plate without any

bristles. We suggest this huge increase of the seventh segment is an adaptation for laying very big eggs.

We could not, unfortunately, identify the oviposition sites or the larvae. In *D. suma* female, the ovipositor exhibits strong and large spines (Fig. 2D) and this is generally considered as an adaptation for laying in a fairly hard substrate, such as plant tissues. Concerning the identity of the substrate, and the nature of the larval breeding site, we face a puzzling problem. When Ipomoea flowers are exploited, it seems likely that eggs are laid in the corolla. The flower remains open, and accessible to the flies, for half a day only, then, it shrinks and falls on the soil and eventually decays due to the proliferation of yeast and bacteria. This fallen, rotting corolla is the larval resource for many other flower breeding species, in Africa and America.^{2,10} In the case of Crinum, the floral biology is very different. First, the flower remains open several days before fading. Second, the petals do not fall down on the soil, but generally dry out on the flower. However, the basis of the flower, or its peduncle, often rots due to the attack of borer caterpillars. This rotten material might be convenient for larval development. Several such decaying flowers were collected but no *Drosophila* emerged from them.

Morphology and anatomy. In addition to the tarsal pegs, another interesting trait of *D. suma* is the shape of the proboscis (Fig. 2A). Compared to most *Drosophila* species, the proboscis is remarkably short, broad and with a very large labellum: such a structure might be considered as an adaptation for swallowing the large quantity of liquid made by the scratches. On the thorax sides, only two large sternopleural bristles are present.

No special characteristic was noticed on the intestine, but the Malpighian tubules revealed apparently a unique feature, as already indicated by Burla.¹¹ The usual two groups of tubules, one anterior and one posterior and with a yellow color, were easily observed. But, in supplement, a pair of narrower, colorless and very long tubules was found along the intestine; the connection between these 'surpernumerary' tubules with the normal Malpighian tubules could not be observed. A hypothesis is that these supernumerary tubules might increase the filtration capacity of the digestive tract, in relation with the very high water content of the food.

Male genitalia are shown in Figure 2B and C. The overall structure is quite simple, with no special differentiation of the cerci and of the epandrium. The surstily are elongated with a row of teeth at the internal margin. The hypandrium is triangular and bears the two median bristles which are found in most *Drosophila* species. The copulatory organ is made of a bifid phallus covered with tiny bristles and is fused to a basal phallapodeme.

Special morphological features were also observed in the female and were discussed in the Ecology section. These included very big egg size, low number of ovarioles, enlargement of tergite 7 and modifications of the oviscapts with an attachment of the two valves (Fig. 2D–G). In addition, we found by dissection that the spermathecae were very small, difficult to observe and probably non-functional, as stated by Burla.¹¹ The seminal receptacle for sperm storing had a length of 0.7 mm. In the male, testes were yellow, as usual in many *Drosophila* species, with a length of 1.1 mm.

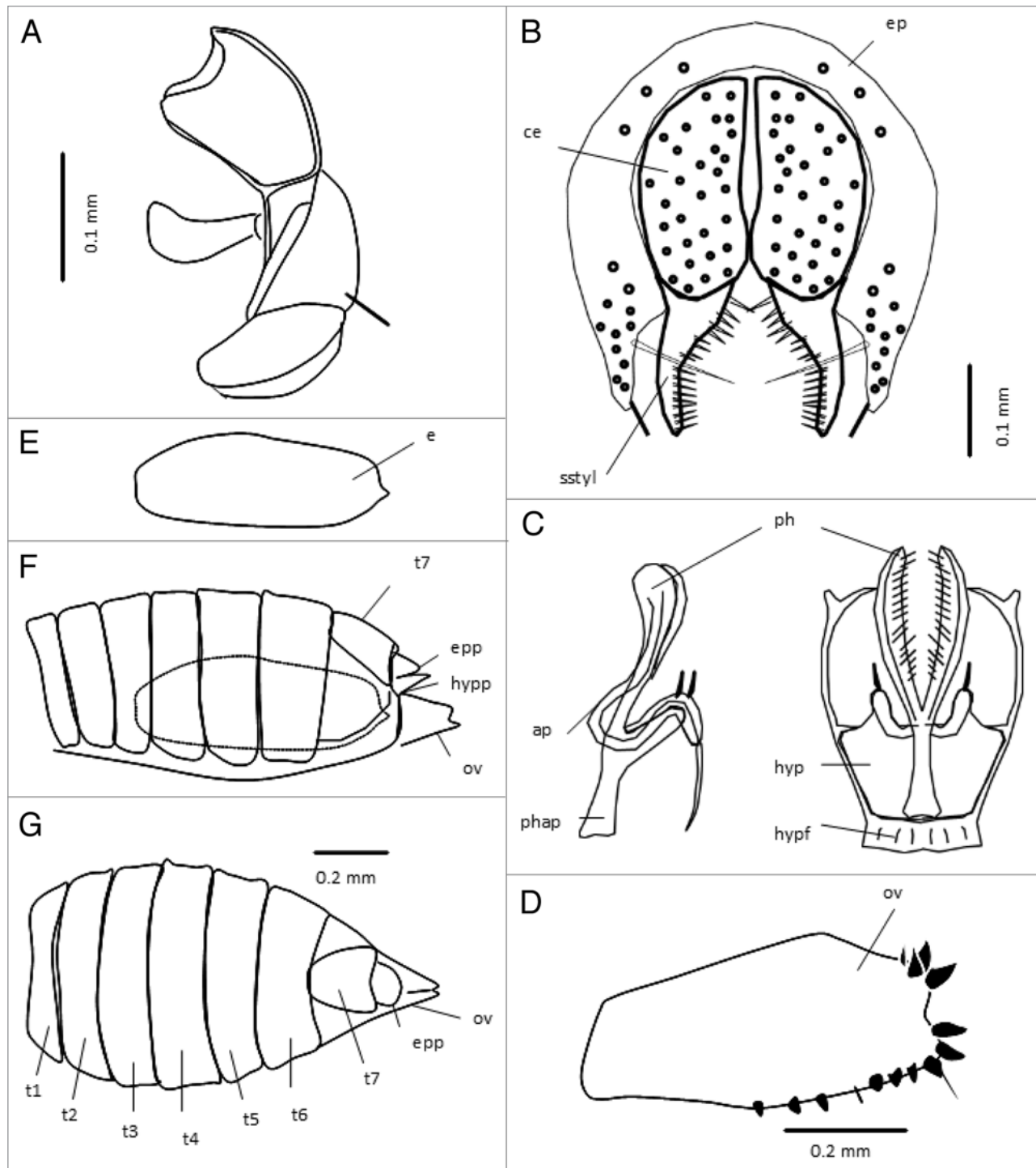


Figure 2. Morphology of body parts in *D. suma*. (A) proboscis, lateral view; (B) dorsal parts of male terminalia; (C) ventral parts of male genitalia; (D) oviscapt, lateral view; (E) egg; (F) female abdomen, lateral view; (G) female abdomen, dorsal view. ap: anterior parameres; ce: cercus; e: egg; ep: epandrium; hyp: hypandrium; hypf: hypandrial phragma; ov: oviscapt; ph: phallus; phap: phallapodeme; t1-t7: tergites 1-7.

Discussion

The major findings of this paper are the description of the feeding behavior of *D. suma* and the presence of some morphological and anatomical adaptations to this behavior and the species ecology. Most notable, is the presence of strong pegs

on the foreleg tarsomeres. Males and females of two other Drosophilid genera are also known to bear spines on their forelegs: *Colocasiomyia* de Meijere and *Laccodrosophila* Duda.³ Although species of these genera are also flower-breeding, they appear not to be related to *D. suma* on the basis of male genitalia morphology.

In spite of a relatively significant literature on the flower-breeding ecology of *Colocasiomyia*,¹²⁻¹⁷ the function of the tarsal pegs in the feeding behavior reported here in *D. suma* have not been described in the Oriental species. The scratching feeding behavior of *D. suma* on the petals of various flowers might be unique in the whole family Drosophilidae. Moreover, we are not aware of any similar behavior in any insect species. Special differentiations on the fore tarsus are frequent in Drosophilids, but they are generally restricted to the male (e.g., sex combs, brushes or tarsal dilatations) and involved in sexual behavior. The tarsal pegs of *D. suma*, *Colocasiomyia* and *Laccodrosophila* are unique because they are identical in both sexes and involved, at least in *D. suma*, in nutrition. The very large amount of ingested liquid pouring out of the cell vacuoles raises an interesting question: is this liquid the only source of food? If so, a very large amount must be ingested in order to cover the nutritional needs of basal metabolism, flight energy and especially egg production which requires a large amount of proteins. Other insects which ingest only a liquid food, such as aphids, are known to have special anatomical and physiological features, such as a gut adapted to filtration and symbiotic bacteria. In the case of *D. suma*, we have two observations in agreement with the liquid as the only food: first the large quantity of droppings observed on the petals; second, the special anatomical feature of ‘supernumerary’ Malpighian tubules which could speed up the rate of filtration and concentrate nutrients. Worth mentioning is the fact that the liquid food ingested comes from the cell vacuoles and not from the sap as in aphids, so that the proportion and quality of the proteins may be adequate for oogenesis, in case the liquid were the sole source of food. Obviously these observations and hypotheses require new field and chemical investigations.

From an ecological point of view, we consider that a female can produce a maximum of one or two eggs per day, revealing a very strong K strategy in the life history.^{18,19} Species with a very low reproductive rate must compensate this disadvantage by increasing the duration of the laying period, i.e., a long survival of adults. We did not observe, however, adults which seemed very old. Moreover, we noticed an apparently high level of predation by spiders (Fig. 1). The very big size of the egg suggests that, in agreement with a K strategy, there is a strong parental investment in the survival of the progeny, hence a low larval mortality. In spite of a very specialized niche and a low capacity to proliferate, *D. suma* appears as a very successful species which has now a widespread distribution on African mainland and Indian Ocean islands. How a very specialized niche persists in such a widespread species appears a demographic puzzle.

Understanding the evolution of flower-breeding adaptations in *D. suma* is also convoluted by its ambiguous taxonomic and phylogenetic positions within the Drosophilidae. Its relationship to another species with similar tarsal spines, *D. spinipes* Lamb,²⁰ has never been fully resolved. *Drosophila spinipes* is only found in the Seychelles islands but its breeding site is unknown. In the

original description of *D. suma* from Côte d’Ivoire, Burla¹¹ noted that the main difference between the two species was related to body pigmentation which was lighter (yellow) in *D. spinipes* while darker (black) in *D. suma*. Burla also identified flowers of *Aframomum* and *Crinum*, both Afrotropical plants, as the breeding sites of *D. suma*. The species was also bred from *Ipomea* flowers by Graber²¹ in the Kivu region in the northeast of the Democratic Republic of Congo and by Buruga and Olembo²² in Uganda. Lachaise²³ mentioned both *D. spinipes* and *D. suma* from Côte d’Ivoire but later only *D. spinipes* was included from Côte d’Ivoire in a taxonomic review of the Afrotropical breeding sites.^{4,24} Tsacas²⁵ also reported only *D. spinipes* from South Africa. It seems thus that these authors considered *D. suma* to be a junior synonym of *D. spinipes* without justifying such taxonomic decision. Lachaise et al.⁶ reported *D. spinipes* from the islands of the Indian Ocean other than Seychelles, namely Madagascar, Mauritius and La Réunion. Interestingly, they also mentioned a species, dubbed *D. cf. suma*, from Madagascar and Mauritius. Finally, Cariou et al. stated that: “*Together these species form a natural group and several lines of evidence suggest that a new genus should be erected to accommodate them*”. Although some authors placed both species in the genera *Hirtodrosophila*^{25,26} and *Mycodrosophila*¹¹ at some times, we did not find support for these hypotheses based on molecular sequences of the 28S rRNA gene (Yassin A, unpublished data). Future investigations in Tropical Africa, including molecular, morphological and ecological data will certainly resolve the taxonomic boundaries in this group and shed light on the evolution of the feeding behavior in *D. suma*.

Material and Methods

Wild living adults were collected with a net and put in vials containing a *Drosophila* food. This method permitted a survival of several days but neither eggs nor larvae were observed. We also collected decaying flowers that were attacked by the adults but no progeny emerged. Some live adults were dissected, and most of the material was preserved, either in pure alcohol for molecular studies or in dilute alcohol with acetic acid and glycerol for morphological studies. Some specimens were also pinned to be conserved in the collection of the Paris museum (M.N.H.N.).

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

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