

Independent evolution of the same set of characters in fig flies (*Lissocephala*, Drosophilidae)

DANIEL LACHAISE and SHANE F. McEVEY

Laboratoire de Biologie et Génétique Evolutives, Centre National de la Recherche Scientifique, 91198 Gif-sur-Yvette Cedex, France

Summary

Comparison of species of African fig flies (*Lissocephala*, Drosophilidae) shows that an unusual phenotype has evolved independently when a particular environment prevails. Each time the adaptation is observed, associated morphological and behavioural characters also appear. Such convergence suggests that selection operates on sets of predetermined morphogenetic pathways.

Keywords: *Lissocephala*; *Ficus*; drosophilid behaviour; convergence.

Introduction

African fig flies (*Lissocephala* Malloch, Drosophilidae) are a group of highly specialized (Lachaise, 1977; Lachaise *et al.*, 1982) drosophilids with unusual morphology and behaviour. The group represents a case of convergence of a complex system involving both morphological and behavioural traits which have evolved only in Africa and are unknown among southeast Asian and Australasian species. Only African species breed in figs (Carson and Wheeler, 1973; Carson, 1974; McEvey, 1981; Bock, 1982; Okada, 1985).

The Afrotropical *Lissocephala* fauna is diverse, with 21 known species which have been reared from 16 species of *Ficus* (Lachaise and Tsacas, 1983). The host species include both cauliflorous savanna shrubs (e.g. *Ficus sur*) as well as tall, strangling, hemi-epiphytic, rainforest species (e.g. *F. macrosperma*). *Lissocephala* comprises two groups of approximately equal size: the *juncta* and *sanu* species groups. They can be recognized from male terminalia (Tsacas and Lachaise, 1979).

Certain traits combine in only two ways in two species groups of African *Lissocephala* (Fig. 1). Here we discuss the two alternate states of abdominal morphology, of egg morphology, of oviposition behaviour and of mating behaviour in the Cote d'Ivoire.

Methods

Fig flies were observed on the monoecious shrub *Ficus sur* during daylight for a total of 250 h. Four *Lissocephala* species coexisted on its syconia: three of the *juncta* species group (*L. disjuncta*, *L. ambigua* and *L. nigroscutellata*) and *L. sanu*. In the Taï rainforest, also in Cote d'Ivoire, *L. melanosanu* (*sanu* species group) was reared from the strangling, hemi-epiphytic *F. lyrata*. Apart from the oviposition behaviour of *L. disjuncta* (Lachaise, 1977) the biology of these species is little known.

Fig. 2 shows the relative lengths of female post-abdomens of different species: the short post-

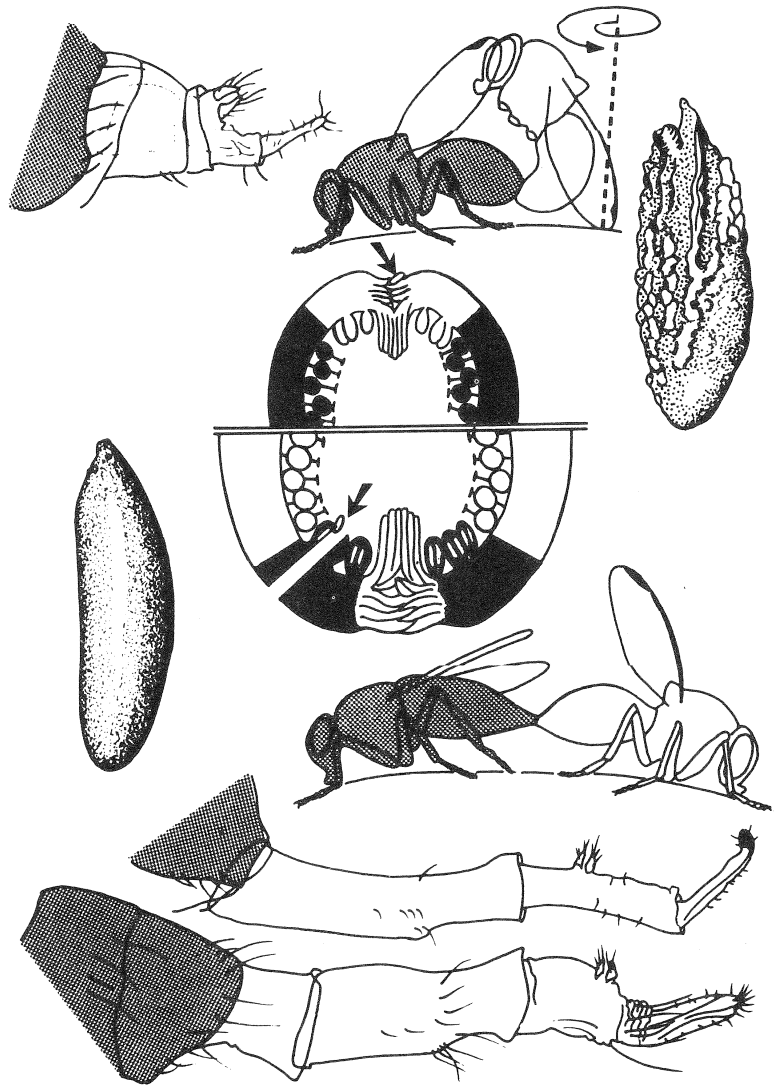


Figure 1. Alternative suites of morphology and behaviour in African fig flies (*Lissocephala*). Two stages of maturation of a host fig (*Ficus sur.* Moraceae) are represented in cross-section (centre); inward facing female flowers (marked in black above dividing line) mature early when pollinating female wasps force their way inside through compacted ostiolar bracts; male flowers around the ostiole (black below) mature later when male wasps bore exit-tunnels (lower left). One suite of fig fly characters (e.g. *L. disjuncta*, above) – short female post-abdomen, ‘waltz’ mating, embossed egg-shell and oviposition on ostiole (arrowed) – is associated with the early fig maturation stage. The other suite of characters (below) – long female post-abdomen, tail-to-tail mating, smooth egg-shell and oviposition deep in wasp tunnels (arrowed) – is associated with the late fig phase. *L. ambigua* (*juncta* group) and *L. sanu* (*sanu* group) both have the latter suite of characters, note their long post-abdomens (*sanu* below).

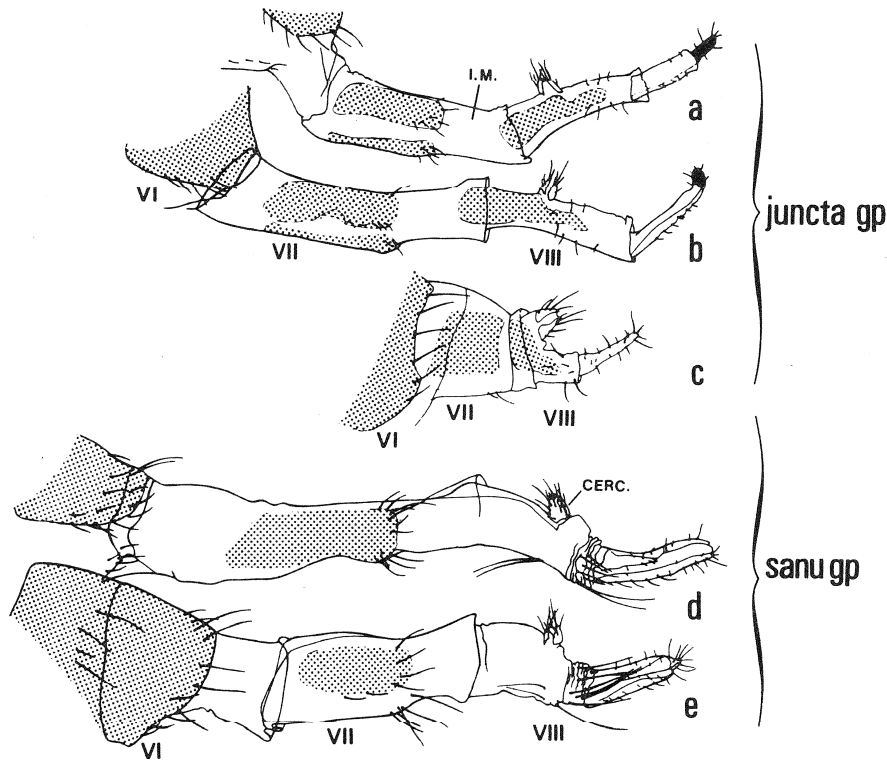


Figure 2. Female post-abdomens of five African fig flies belonging to two lineages: the *Lissocephala juncta* and *L. sanu* species groups. a) *L. nigroscutellata*; b) *L. ambigua*; c) *L. disjuncta*; d) *L. melanosanu*; e) *L. sanu*; cerc., cercus; i.m., intersegmental membrane. Extended post-abdomens fall into two classes: a-b and d-e; specific characters are: sclerotized VIII-segmental plates, setation of VII and VIII segments, pigmentation of ovipositor apically, orientation of cerci and post-abdominal breadth relative to length.

abdomen of *L. disjuncta* and the long post-abdomens of species belonging to the *juncta* and *sanu* species groups. The extension of the last segments is not permanent. At eclosion the female's post-abdomen is extended. After drying and hardening, retraction draws it into collapsed, telescopic form. It is everted again during, and for a short time after, oviposition and during the rotational movement of mating.

Developmental elongation involves transformation of two segments. The female terminalia in *Drosophila* are derived from the A8 primordia (Nöthiger *et al.*, 1977; Dübendorfer and Nöthiger, 1982) in which case the segments in *Lissocephala* would be A7 and A8. A comparison of species illustrated in Fig. 2 shows that the ventral expansion is greater than the dorsal expansion because the cercus has rotated forward. *Lissocephala sanu* is exceptional in that in addition to elongation of A7 and A8, A6 is also involved. Telescopic retraction is possible because of the wide intersegmental membranes between the tergal and sternal plates of A7 and A8.

Among the southeast Asian and Australasian species of *Lissocephala* and *Mulgravea*, the telescopic post-abdominal condition is unknown (Okada, 1985; 1987). However, within the *Drosophilidae* the condition has arisen independently in several unrelated genera. For example,

in *Amiota filipes* and *A. alboguttata* the female post-abdomen is elongate and extensively membranous between plates (Máca, 1980). Their breeding sites are unknown. In *Drosophila xiphiphora* females have a long post abdomen with which they insert eggs deep in flower buds (Pipkin *et al.*, 1966); this is true also of *Zygothrica florinjecta* (Grimaldi, 1987) and is known in *Zapriothrica* (Wheeler, 1959).

There are two forms of eggs in the African species of *Lissocephala*. The eggs which are laid in fig ostioles (i.e. receptacular entrances closed off by outer bracts) have a thickened, embossed chorion (*L. disjuncta*). This hardens the eggshell; unlike most other drosophilid eggshells they do not collapse after hatching. In contrast, those eggs laid in tunnels (*L. ambigua*, *L. nigroscutellata* and *L. sanu*) have thin, smooth chorion and collapse once empty. Both egg forms lack filaments and hence are similar to the unusual eggs of most flower-breeding *Drosophila*, *Lissocephala powelli* is not fig-breeding and has eggs with filaments (Carson and Wheeler, 1973).

On a monoecious fig there are two places where *Lissocephala* eggs are laid: the ostiole when the fig is small; and later in wasp exit-tunnels when the figs are larger. The almost impenetrable superficial bracts of the ostiole yield to the fig wasp (Agaonidae), which enters and pollinates. Later, male wasps bore exit-tunnels, freeing their sibling females (Wiebes, 1979; Michaloud *et al.*, 1985; Bronstein and McKey, 1989). There are thus two passages to a fig's interior, each exploited by different *Lissocephala* species. *L. disjuncta* lays eggs in the ostiole the day after the pollinating fig wasp enters. The three other species (*L. ambigua*, *L. nigroscutellata* and *L. sanu*) oviposit deep in wasp exit-tunnels a few weeks later. Females insert their abdomen into tunnels while they rest suspended by their wings and legs in the mouth of the hole. The female's post-abdomen is as long as the fig wall is thick, therefore oviposition occurs at the tunnel's inner opening. Females may withdraw and reinsert, but they do not turn *in situ*.

Mating occurs on the surface of figs. There are two mating behaviours, each associated with one or other of the female post-abdominal morphologies. We follow the terminology of McAlpine (1981) to describe mating in the Diptera. The initial coupling position is similar to *Drosophila* species and to most other Diptera. Both sexes are unidirectional and vertically upright, the male on top of the female. However, duration and final mating positions differ greatly.

In *L. disjuncta* (the females of which have short post-abdomens) mating is exceedingly brief, lasting just 3–5 sec. After mounting, the male leans backwards, his wing-tips touch the surface and form a pivot on which the pair turns rapidly through several full circles. We call this curious behaviour 'waltzing'. The final mating position involves no further change in the relative orientation of the partners (Fig. 1).

In *L. sanu*, *L. ambigua* and *L. nigroscutellata* (the females of which have long post-abdomens) mating lasts for 15 to 60 sec. The final mating position is tail-to-tail, the mating partners facing opposite directions and remaining vertically upright. The terminalia of each partner are locked together, they do not slip and apparently cannot twist. The tail-to-tail position involves rotation of the female's post-abdomen through 180° bringing the female terminalia to a dorsal position.

Immediately after initial coupling the male springs backwards drawing out the female's telescopic post-abdomen. In the same movement the male twists in order to become vertically upright. Consequently the intersegmental membrane of the female's post-abdomen rotates between segments 7 and 8. Females withdraw their extended post-abdomens when mating terminates.

While flies are mating tail-to-tail they appear to move away from predaceous ants (*Oecophylla longinoda*) as freely as do single individuals with which they form aggregations. The mating pair can move in any direction synchronously and at various speeds. The tail-to-tail, vertically upright position lends itself to predator avoidance while not interrupting fertilization.

This is the first report of tail-to-tail mating as a normal position within the Drosophilidae although it is well known in other Diptera (McAlpine, 1981). Tail-to-tail mating has not been observed as a normal mating position in Hawaiian species (Kaneshiro, pers. comm.; Carson, pers. comm.; Spieth, 1952). In *Lissocephala* the final mating position is species specific. In this respect it contrasts with the sex-limited mutant *stuck* (*sk*) of *D. melanogaster* which results in the failure of males to disengage at the end of copulation (Beckman, 1970). While they tug against each other the *sk* male and female occasionally assume a transient tail-to-tail position. This condition is therefore aberrant, prolongs copulation and can be fatal (Hall *et al.*, 1980). It is not rare in *Drosophila* (Grossfield, 1972), and is not due to a morphological change (Hall *et al.*, 1980). In *Lissocephala* the tail-to-tail position is associated with morphological and behavioural changes in both females and males.

Flies have either: a short post-abdomen and rough, resistant eggshells and oviposit on fig ostioles; or a long, telescopic post-abdomen and smooth eggshells and oviposit in wasp exit-tunnels. The different mating behaviours accompany these characters: flies with short post-abdomens do not rotate during mating, while flies with telescopic post-abdomens rotate so that mates face away from one another. In the *juncta* lineage *L. disjuncta* and a new undescribed species have the former suite *a* characters and *L. ambigua* and *L. nigroscutellata* have the latter. Only the latter has been observed in the *sanu* lineage (viz. *L. sanu*).

Discussion

In the fig-dwelling *Lissocephala* a particular behaviour is always associated with the same morphology and microhabitat. The same suite evolves in independent phylogenetic lineages. What can be concluded from the existence of two different and complex character suites (*a* and *b*) in a single monophyletic lineage (*Lissocephala juncta* species group) when one (*b*) is found also in a separate lineage (*L. sanu* species group)?

It is critical to establish whether the *b* character suite has evolved once or twice. The simplest interpretation would be that it evolved once in the *juncta* group and was then inherited by the derived *sanu* group. We reject this hypothesis. The assumption that the *b* suite is ancestral is improbable, outgroups (*Lissocephala* from southeast Asia, drosophilid species in general) strongly imply that it is derived. Morphologically and in other details the *b* suite of the *juncta* group is not exactly the same as that in the *sanu* group. *Nigroscutellata* and *ambigua* on the one hand, and *melanosanu* and *sanu* on the other, have achieved only superficially similar long post-abdomens which differ in morphological detail (Fig. 2) and hence probably have independent origins. Finally, male terminalia are strikingly similar within either the *sanu* or *juncta* species groups and strongly different between them. There is a general consensus that male terminalia is of more importance in insect systematics than female post-abdominal size, egg morphology or behaviour; the latter may be largely adaptive. On this premise the *a* suite is ancestral and the *b* suites are both derived and hence may be viewed as convergent.

Parallelism and convergence of single traits suggest that a common external or internal environment of organisms is involved. If the constant factor is a narrow environmental constraint there will be convergence in similar habitats (as observed here). Simply offering plausible explanations for each convergent character does not explain how the evolved traits arise always together. Evolution depends on the raw material available, on a limited set of things 'with which nature tinkers' (Jacob, 1977). Internal developmental or morphogenetic constraints is probably always involved in convergence or parallelism: natural selection acts on a restricted range of possibilities, so that a restricted and perhaps convergent outcome is to be expected. The conjunction of behaviour and form seen here implies developmental co-ordination. Discrete

environmental opportunities (e.g. fig ostioles or fig tunnels) will narrow the range of behavioural and morphological phenotypes even further.

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References

- Beckman, C. (1970) *sk stuck*. *Dros. Inf. Serv.* **45**, 36.
- Bock, I. R. (1982) Drosophilidae of Australia V. Remaining genera and synopsis (Insecta: Diptera). *Aust. J. Zool., Suppl. Ser. No.* **89**, 1–164.
- Bronstein, J. L. and McKey, D. eds (1989) The comparative biology of figs. Multi-author Review. *Experientia* **45**, 599–680.
- Carson, H. L. (1974) Three flies and three islands: parallel evolution in *Drosophila*. *Proc. Natl. Acad. Sci. USA* **71**, 3517–21.
- Carson, H. L. and Wheeler, M. R. (1973) A new crab fly from Christmas Island, Indian Ocean (Diptera: Drosophilidae). *Pacific Insects* **15**, 199–208.
- Dübendorfer, K. and Nöthiger, R. (1982) Clonal analysis of cell lineage and growth in the male and female genital disc of *Drosophila melanogaster*. *Wilhelm Roux's Arch. dev. Biol.* **191**, 42–55.
- Grimaldi, D. (1987) Phylogenies and Taxonomy of *Zygothrica* (Diptera: Drosophilidae). *Bull. Am. Mus. Nat. Hist.* **186**, 103–268.
- Grossfield, J. (1972) Decapitated females as a tool in the analysis of *Drosophila* behaviour. *Anim. Behav.* **20**, 243–51.
- Hall, J. C., Siegel, R. W., Tomkins, L. and Kyriacou, C. P. (1980) Neurogenetics of courtship in *Drosophila*. *Stadler Symp. University of Missouri, Columbia* **12**, 43–82.
- Jacob, F. (1977) Evolution and tinkering. *Science* **196**, 1161–6.
- Lachaise, D. (1977) Niche separation of African *Lissocephala* within the *Ficus* drosophilid community. *Oecologia (Berl.)* **31**, 201–14.
- Lachaise, D. and Tsacas, L. (1983) Breeding-sites in tropical African drosophilids. In *The Genetics and Biology of Drosophila*. (M. Ashburner, H. L. Carson and J. N. Thompson Jr, eds) pp. 221–332, vol. 3d. Academic Press, London and New York.
- Lachaise, D., Tsacas, L. and Couturier, G. (1982) The Drosophilidae associated with tropical African figs. *Evolution* **36**, 141–51.
- Máca, J. (1980) European species of the subgenus *Amiota* s.str. (Diptera, Drosophilidae). *Acta ent. bohemoslov.* **77**, 328–46.
- Mayr, E. (1983) How to carry out the adaptationist program? *Am. Nat.* **121**, 324–34.
- McAlpine, J. F. (1981) Morphology and terminology – adults. In *Manual of Nearctic Diptera*. (J. F. McAlpine et al eds.) pp. 9–63, vol. 1. Agriculture Canada Monograph No. 27. Biosystematics Research Institute, Ottawa, Ontario.
- McEvey, S. F. (1981) Drosophilidae (Insecta: Diptera) of three Torres Strait islands with description of a new species of *Drosophila*. *Aust. J. Zool.* **29**, 907–19.
- Michaloud, G., Michaloud-Pelletier, S., Wiebes, J. T. and Berg, C. C. (1985) The co-occurrence of two pollinating species of fig wasp and one species of fig. In *Proceedings of the Koninklijke Nederlandse Akademie van Wetenschappen*, Series C, **88**, 93–119.
- Nöthiger, R., Dübendorfer, K. and Epper, F. (1977) Gynandromorphs reveal two separate primordia for male and female genitalia in *Drosophila melanogaster*. *Wilhelm Roux's Arch. dev. Biol.* **181**, 367–73.
- Okada, T. (1985) The genus *Lissocephala* Malloch and an allied new genus of southeast Asia and New Guinea (Diptera, Drosophilidae). *Kontyu, Tokyo* **53**, 335–45.

14 OCT 1991

- Okada, T. (1987) Note on the genus *Mulgravea* (Diptera, Drosophilidae). *Kontyu, Tokyo* **55**, 187.
- Pipkin, S. B., Rodríguez, R. L. and León, J. (1966) Plant host specificity among flower-feeding neotropical *Drosophila* (Diptera: Drosophilidae). *Am. Nat.* **100**, 135-56.
- Spieth, H. L. (1952) Mating behavior within the genus *Drosophila* (Diptera). *Bull. Amer. Mus. Nat. Hist.* **99**, 395-474.
- Tsacas, L. and Lachaise, D. (1979) La radiation africaine des *Lissocephala* inféodées aux *Ficus* (Dipt. Drosophilidae). *Annls Soc. ent. Fr. (N.S.)* **15**, 589-603.
- Wheeler, M. R. (1959) Three new species of *Zapriothrica* Wheeler (Diptera, Drosophilidae). *Southwestern Nat.* **4**, 83-7.
- Wiebes, J. T. (1979) Co-evolution of figs and their insect pollinators. *Ann. Rev. Ecol. Syst.* **10**, 1-12.