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Drosophila robusta 種群と *D. melanica* 種群の生殖器の比較研究

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Abstract

The male and female genitalia of the *Drosophila robusta* and the *D. melanica* species-groups were compared between and within the groups. The degree of similarity was analyzed by a character × species matrix using seven characteristics of the phallic organs. In this matrix, the ten drosophilid species examined here were divided into the following six groups; Group Ⅰ: *D. okadai, D. neokadai* and *D. unimaculata*; Ⅱ: an unknown *D. sp. 2* of the *robusta* group; Ⅲ: *D. lacertosa*; Ⅳ: *D. sordidula, D. pseudosordidula* and *D. robusta*; Ⅴ: *D. moriwakii*; and Ⅵ: *D. tsigana*. The results of this grouping support a chromosomal relationship between the Group Ⅳ flies (Narayanan, 1973) and a new systematic position of *D. unimaculata* (Beppu, 1988). Based on this matrix, clustering was performed to determine the systematic relationship between these groups. The dendrogram implies that the *Drosophila robusta* species-group is divided into three heterogeneous species-subgroups: the *okadai, lacertosa*, and *robusta* subgroups. The structure of the phallic organs of the *robusta* species-subgroup is more closely related to the *melanica* species-group than other member of the *robusta* species-group.

Introduction

The *Drosophila robusta* and the *D. melanica* species-groups belong to the *virilis* Section of the subgenus *Drosophila*. These groups are supposed to have emerged in the *virilis-repleta* Radiation, which might have occurred in the Old World during the Oligocene to early Miocene (Throckmorton, 1975). Most *robusta* group flies have been recorded in the temperate regions of the eastern Old World, while, except for a few species, the *melanica* group is restricted to the New World. From a genitalial observation, Beppu (1988) has recently transferred the Japanese *D. moriwakii* Okada et Kurokawa and North American *D. colorata* Walker from the *robusta* species-group to the *melanica* species-group, and the European *D. unimaculata* Strobl to the *robusta* group. However, not all genitalia of these group flies are described in the original papers. Further, an unknown species of the *robusta* group has been found in northern Japan (Watabe, 1986).

The aim of this paper is to examine the male and female genitalia of the *robusta* and *melanica* group flies and to consider the systematic relationship among and within the groups.

Materials and Methods

All of the *robusta* species-group flies, except two Chinese species *D. cheda* Tan *et al.* and *D. pullata* Tan *et* al., and two Japanese species of the *melanica* group were examined (Table 1). The genitalia were treated with 20% KOH solution for about 20 minutes, and were examined under a light microscope.

Results of Observations

1. Periphallic organs.

1. 1. Epandrium (Fig. 1) : The *robusta* and *melanica* group-flies have pubescent epandrium fused to the cercus at the middle. The epandrium is entirely dark brown, and its anterior margin of *D. neokadai* is characteristically black (Fig. 1B). The ventral portion is marginally roundish in most species. The anteroventral portion of *D. okadai* is nearly straight (Fig. 1A), that of *D. lacertosa* convex (Fig. 1E) and in *D. moriwakii* somewhat pointed (Fig. 1I). In most species the epandrium anterior is bare and posterior pubescent. In *D. okadai* and *D. neokadai* the upper half of the epandrium is pubescent (Figs. 1A, B), and in *D. unimaculata* only the lower 1/3 pubescent (Fig. 1C). The epandrium has about 15 to 25 long bristles, most of which are on the middle or lower portion and a few on the upper part. In *D. tsigana* bristles are restricted to the lower portion (Fig. 1J). In *D. neokadai* some bristles on the caudoventral margin are tooth-like.

Table 1. Species used for observations, collection data and distribution.

Species	Distribution	Collection data
The *Drosophila robusta* species-group		
D. okadai Takada, 1959	Japan	Mimmaya, Aomori Pref., 26-28. Ⅶ. 1987.
D. neokadai Kaneko *et* Takada, 1966[1]	Japan, China	*Ibid.*
D. unimaculata Strobl, 1893	e. and c. Europe[3]	Dobro Polje, Yugoslavia, 25-28. Ⅶ. 1984.
D. sp. 2[1]	Japan, China	Mimmaya, Aomori Pref., 26-28. Ⅶ. 1987.
D. lacertosa Okada, 1956	Japan, Korea, China, Burma, India, Nepal	*Ibid.*
D. sordidula Kikkawa *et* Peng, 1938	Japan, Korea	Misumai, Hokkaido, 14-22. Ⅶ. 1988.
D. pseudosordidula Kaneko, Tokumitsu *et* Takada, 1964	Japan	Nopporo, Hokkaido, 1987.
D. robusta Sturtevant, 1916	North America	Living stock in Tokyo Metropolitan Univ.
The *Drosophila melanica* species-group		
D. moriwakii Okada *et* Kurokawa, 1957	Japan	Misumai, Hokkaido, 5-16. Ⅹ. 1987
D. tsigana[2] Burla *et* Bloor, 1952	Japan, Korea, w. Europe[3]	Misumai, Hokkaido, 10-11. Ⅵ. 1988.

1). *Drosophila sp.* 2 (Watabe, 1986) will be described as a new species, *Drosophila* (*Drosophila*) *gani* Liang et Zhang (Watabe *et al.*, in press).
2). *D. pengi* Okada et Kurokawa is a junior synonym with *D. tsigana* (Watabe *et al.*, in press).
3). e : eastern. c : central. w : western.

1. 2. Surstylus (Figs. 1, 2) : The shape of the surstylus is quadrate or rectangular in both the *robusta* and *melanica* groups, but that of *D. sp. 2* is nearly semicircular with a small projection at the caudodorsal corner (Fig. 2D). The basal part connected to the epandrium is relatively narrow in these groups. The distal margin is nearly straight in four species, *D. sp. 2, D. pseudosordidula, D. robusta* and *D. moriwakii* (Figs. 2D, G-I), and in the remaining it is moderately concave.

The surstyli of *D. neokadai, D. sp. 2* and *D. moriwakii* are bare, those of *D. unimaculata* and *D. tsigana* entirely pubescent, and those of the remaining five species partially pubescent. The surstyli of *D. sordidula, D. pseudosordidula,* and *D. robusta* are very similar in shape and chaetotaxy (Figs. 2E-H). The surstylus of *D. okadai* is characterized by wart-like tiny hairs on the outer surface. The surstylus of all species has black primary teeth on the distal margin and several brown bristles at the caudoventral corner. The number of primary teeth in the *robusta* and the *melanica* species-groups is from 7 to 13, with intra-specific variations. The

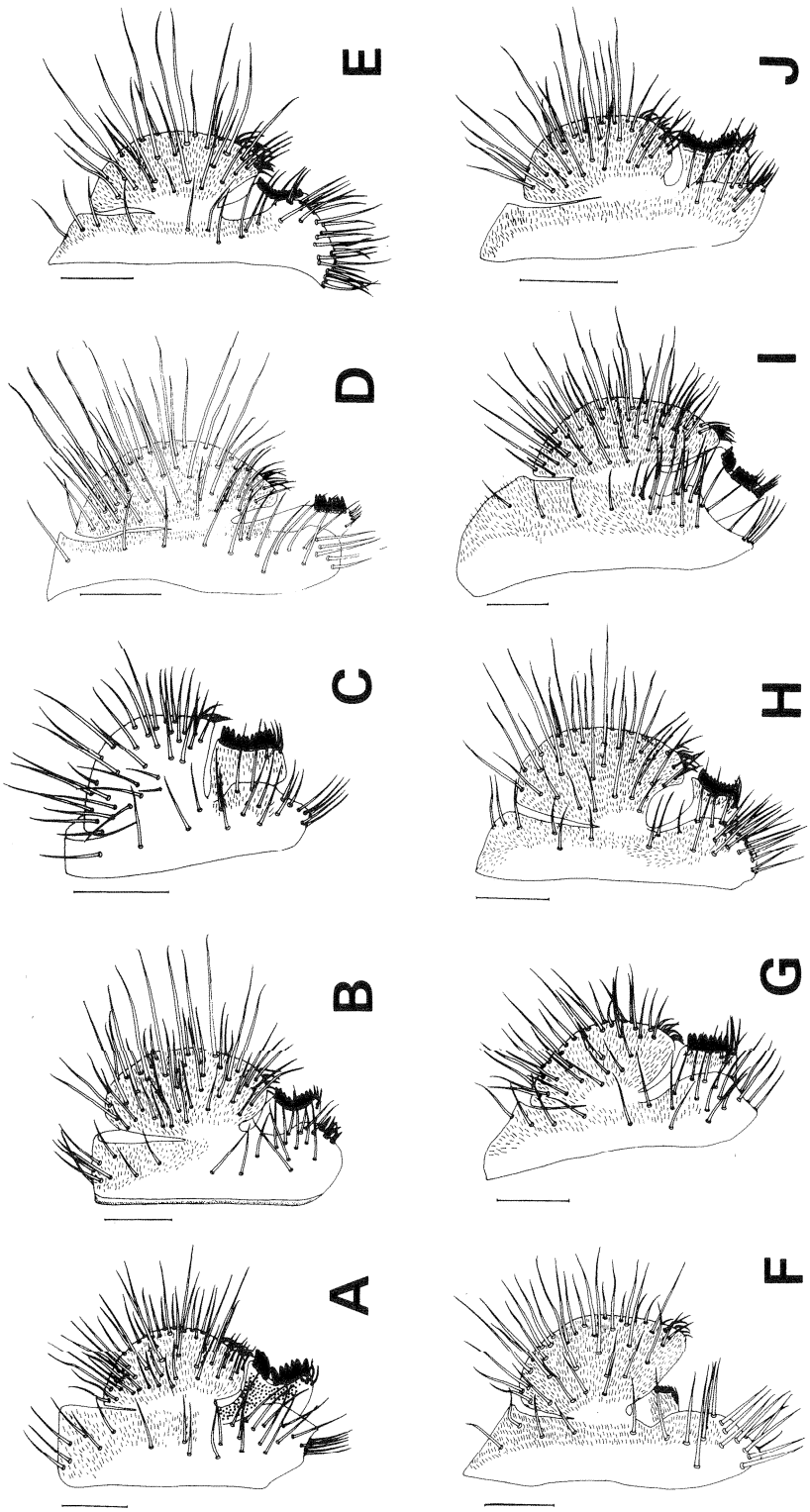

Fig. 1. Periphallic organs in the *Drosophila robusta* and the *D. melanica* species-groups. A : *D. okaaai*. B : *D. neokadai*. C : *D. unimaculata*. D : *D. sp.* 2. E : *D. lacertosa*. F : *D. sordidula*. G : *D. pseudoscrdidula*. H : *D. robusta*. I : *D. morivahii*. J : *D. tsigana*. Scale=0.1mm.

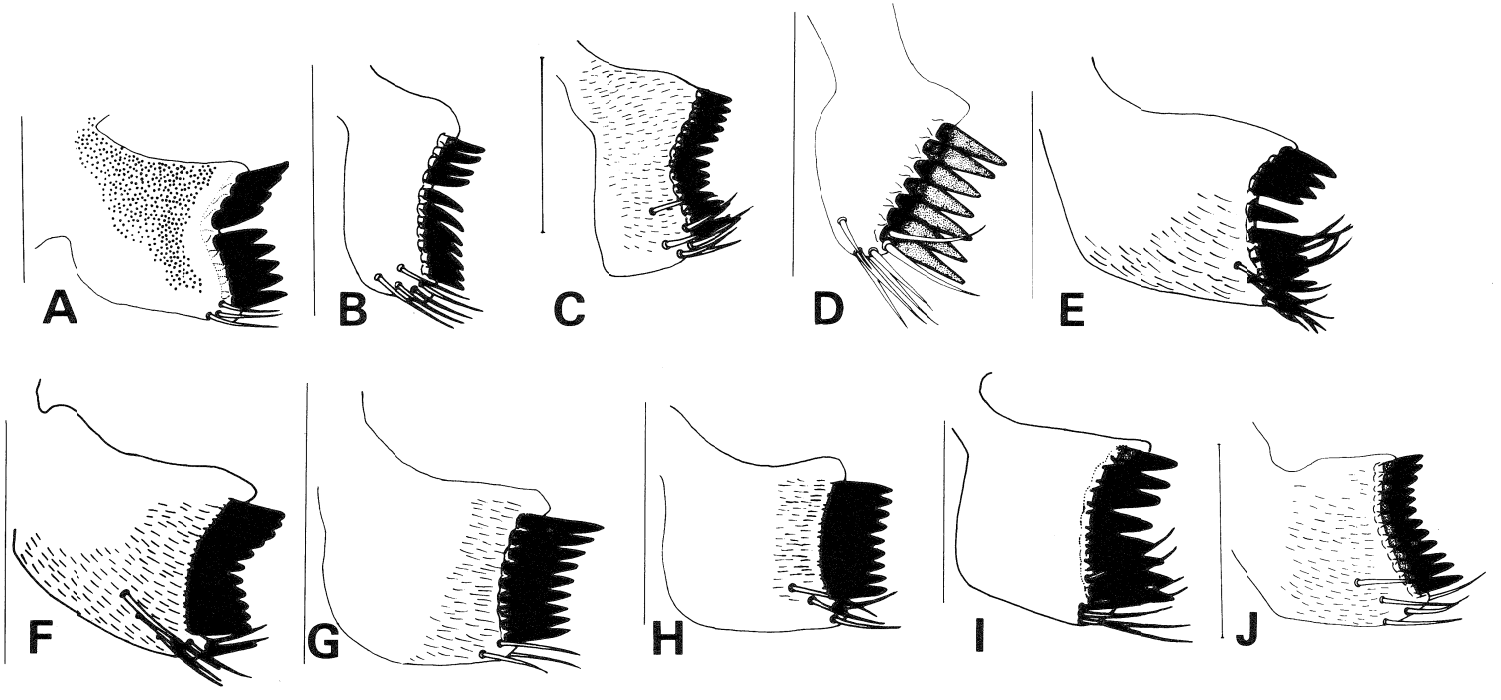

Fig. 2. Surstylus in the *Drosophila robusta* and the *D. melanica* species-groups. A: *D. okadai*. B: *D. neokadai*. C: *D.unimaculata*. D: *D. sp. 2*. E: *D. laceriosa*. F: *D. sordidula*. G: *D. pseudosordidula*. H: *D. robusta*. I: *D. moriwakii*. J: *D. tsigana*. Scale=0.1mm.

surstyli of *D. unimaculata* and *D. tsigana* have about 13 primary teeth, the largest number in the species-group, while *D. sp. 2* has only 7 primary teeth.

1. 3. Cercus (Fig. 1) : The cercus is very similar in both the *robusta* and the *melanica* groups, being dark brown, oval and entirely pubescent, with 30 to 40 long bristles and a tuft of several short bristles along the ventral margin. The only exceptional species is *D. unimaculata* which has a bare cercus (Fig. 1C), a diagnostic character to distinguish *D. unimaculata* from other species.

2. Phallic organs.

2. 1. Novasternum or hypandrium (Fig. 3) : In the *robusta* species-group, only *D. lacertosa* has one pair of submedian spines on the inner margin of the novasternum (Okada, 1956). *D. moriwakii* has two and *D. tsigana* one pair of submedian spines in the middle part (Okada and Kurokawa, 1957). The submedian spine has been determined in all *melanica* group flies, which makes it a diagnostic characteristic for this group. The shape of the hypandrium is somewhat flattened in *D. sordidula, D. pseudosordidula, D. robusta,* and two species of the *melanica* group, but in the remaining species it is gently curved and surrounds the aedeagus. The outer surface of hypandrium is pubescent in three species of the *robusta* group, *D. okadai, D. neokadai* and *D. unimaculata,* and in two species of the *melanica* group. In *D. neokadai* and *D. unimaculata* these hairs on the hypandrium are small and wart-like (Figs. 3B, C).

2. 2. Aedeagus and its accessary organs (Figs. 3, 4) : The aedeagal characteristics common to the *robusta* group flies are that a lateral view shows a broadening in its submedial portion, that its upper part curves ventrally to the axis of the aedeagal apodeme, and that anterior paramere is present (Beppu, 1988). In *D. sordidula, D. pseudosordidula* and *D. robusta* the center axis of aedeagus bends ventrally to the apodeme one time, and in the remaining species of the *robusta* group it does so two times, first dorsally slightly and second ventrally considerably. Therefore, an aedeagal cavity just above the vertical rod is deep in these *robusta* group species. The aedeagal curve of the *D. lacertosa* appears intermediate between these two groups.

In *D. okadai, D. neokadai, D. unimaculata,* and *D. sp. 2,* the aedeagus separates into two lateral lobes, and these lobes fuse ventrally at its base above the vertical rod (Figs. 3A-D). The aedeagus of *D. lacertosa* separates dorsally into two lobes at its tip and ventrally at its distal half (Fig. 3E). In *D. sordidula, D. pseudosordidula,* and *D. robusta* (Figs. 3F-H), the aedeagus separates dorsally at the proximal portion and ventrally over the entire length. The aedeagal apodeme of the *robusta* group is shorter than the aedagus.

In the *melanica* group the aedeagus is slender and straight till the axis of the apodeme, with notches on its dorsal margin. The aedeagal structures of *D. moriwakii* and *D. tsigana* are identical to those of *D. sordidula* and *D. robusta,* both dorsally and ventrally separated into two lateral lobes and fused only at the tip (Figs. 4E-F, Fig. 7). The aedeagal apodeme of the *melanica* group is subequal to aedeagus.

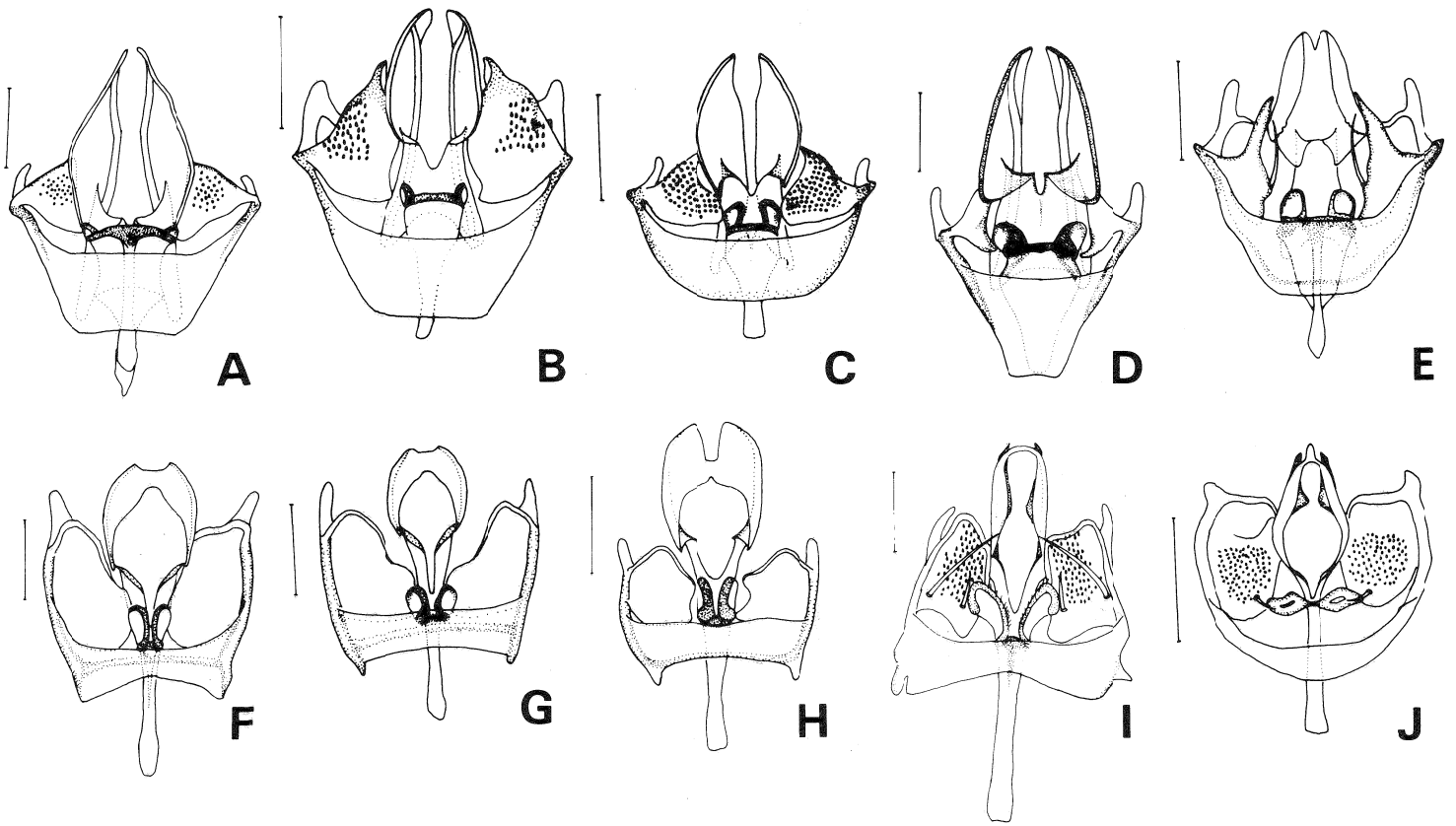

Fig. 3. Phallic organs in the *Drosophila robusta* and the *D. melanica* species-groups. A: *D. okadai*. B: *D. neokadai*. C: *D. unimaculata*. D: *D. sp. 2*. E: *D. lacertosa*. F: *D. sordidula*. G: *D. pseudosordidula*. H: *D. robusta*. I: *D. moriwakii*. J: *D. tsigana*. Scale=0.1mm.

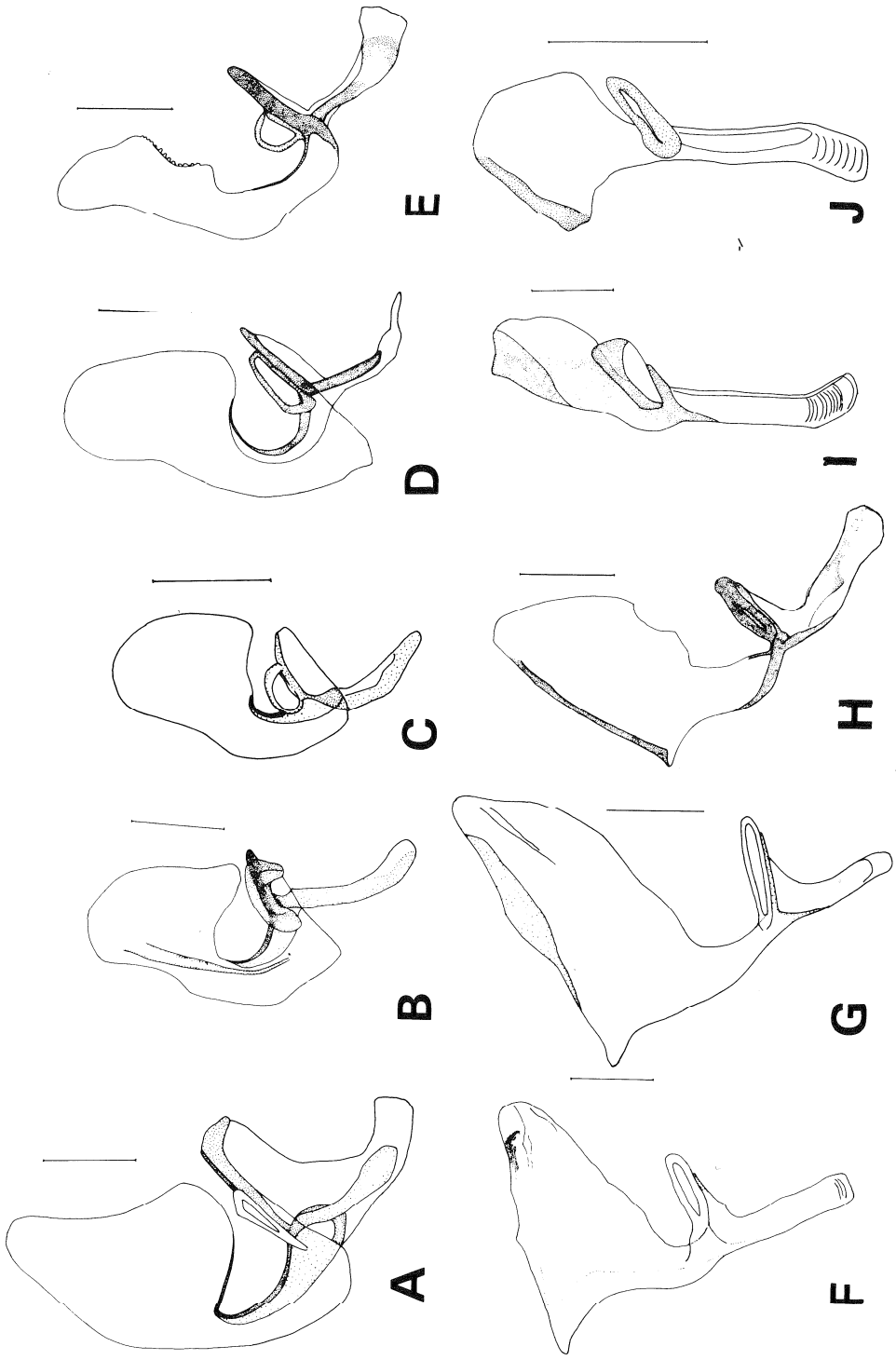

Fig. 4. Aedeagus (lateral view) in the *Drosophila robusta* and the *D. melanica* species-groups. A: *D. okadai*. B: *D. neokadai*. C: *D. unimaculata*. D: *D. sp. 2*. E: *D. lacertosa*. F: *D. sordidula*. G: *D. pseudosordidula*. H: *D. robusta*. I: *D. moriwakii*. J: *D. tsigana*. Scale=0.1mm.

Anterior parameres are present in both the *robusta* and the *melanica* group, with inter-specific variation. The anterior paramere of *D. okadai* is smallest and that of *D. lacertosa* is largest in the *robusta* group. The parameres of *D. sordidula, D. pseudosordidula* and *D. robusta* are more developed than the vertical rod, while the opposite is the case for other *robusta* species. The anterior parameres of *D. lacertosa, D. unimaculata* and *D. sp.* 2 are semicircular in the lateral view, while, in *D. sordidula, D. pseudosordidula* and *D.robusta*, they are rod-shaped and very similar to the parameres of *melanica* group flies (Figs. 4F-J).

There are inter-specific differences in the degree of development of vertical rods. In general, the vertical rod of the *robusta* group is more developed than that in the *melanica* group. Within the *robusta* group, the vertical rods of the first five species (Figs. 4A-E) are more developed than those of the last three species(Fig. 4F-H). The vertical rod of *D. moriwakii* is a rudiment with tiny hairs on its margin, and it is absent on *D. tsigana* (Figs. 4I, J). The vertical rods of *D. sordidula, D. pseudosordidula,* and *D. robusta* are very slender and rod-shaped, and seem to be intermediate between the plate-shaped vertical rod in the first five species of the *robusta* group and the degenerated one in the *melanica* group.

2. 3. Ventral fragma (Fig. 3) : The ventral fragmae of the first five species of the *robusta* group are arch-shaped and narrow distally (Figs. 3A-E), while those of the last three species and *D. moriwakii* are rectangular and slightly convex on the distal margin (Figs. 3F-I). *D. tsigana* has a roundish ventral fragma (Fig. 3J)

3. Female reproductive oragans.

3. 1. Ovipositor (Fig. 5) : The structure of the ovipositors is very similar for all species, except for *D. okadai* with a black ultimate marginal tooth (Fig. 5A). Two chitinized lobes attach to the basal isthmus, with discal teeth, with marginal teeth in a regular row along the margin, and with a bristle-like subterminal hair at the caudoventral corner. The number of discal teeth is from 3 to 7 and there are 16 to 30 marginal teeth.

3. 2. Spermatheca (Fig. 6) : The spermathecae of the *robusta* group flies except *D. unimaculata,* are large and cylindric in the lateral view, and those of the *melanica* group and *D. unimaculata* small and bell-shaped. As explained below, the spermatheca in the *robusta* group is species-specific, and it is a reliable diagnostic character for the identification, especially between two closely related species (Kaneko *et al.*, 1964 ; Kaneko and Takada, 1966).

D. okadai (Fig. 6A) : Constricted at *ca.* 1/6 from the base and wrinkled here, without apical indentation; introvert relatively shallow in this group, *ca.* 3/5 of the height of the outer capsule.

D. neokadai (Fig. 6B) : Narrowing basally, with a small apical hollow, and with oblique wrinkles on *ca.* 2/5 of the basal portion of the outer surface; introvert very deep, just below the ceiling of the capsule.

D. unimaculata (Fig. 6C) : Very small, without constriction; introvert middle, *ca.* 2/3 of the height of the capsule; duct without coiling outside capsule.

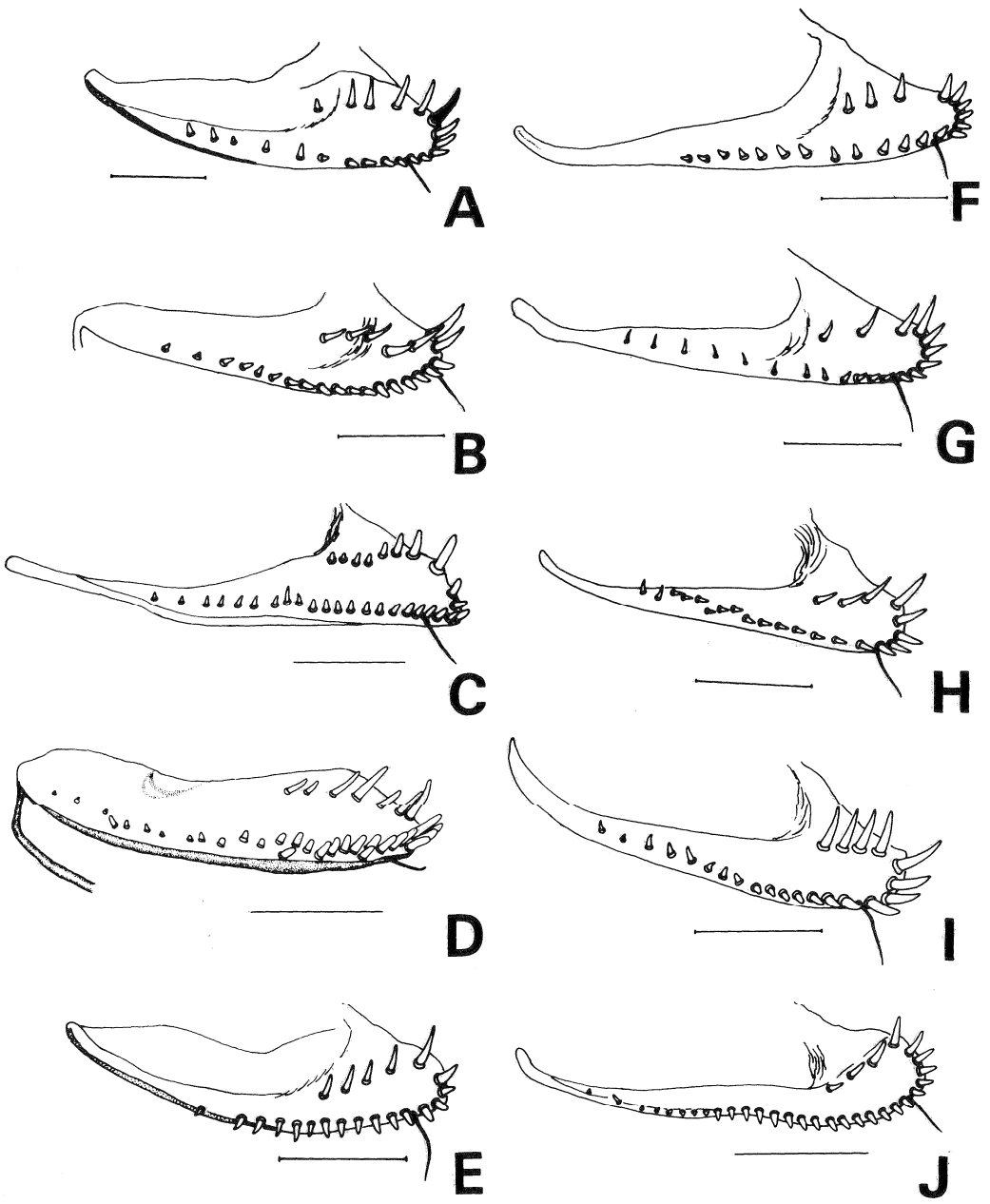

Fig. 5. Lobes of ovipositor of the *Drosophila robusta* and the *D. melanica* species-groups. A: *D. okadai*. B: *D. neokadai*. C: *D. unimaculata*. D: *D. sp. 2*. E: *D. lacertosa*. F: *D. sordidula*. G: *D. pseudosordidula*. H: *D. robusta*. I: *D. moriwakii*. J: *D. tsigana*. Scale=0.1mm.

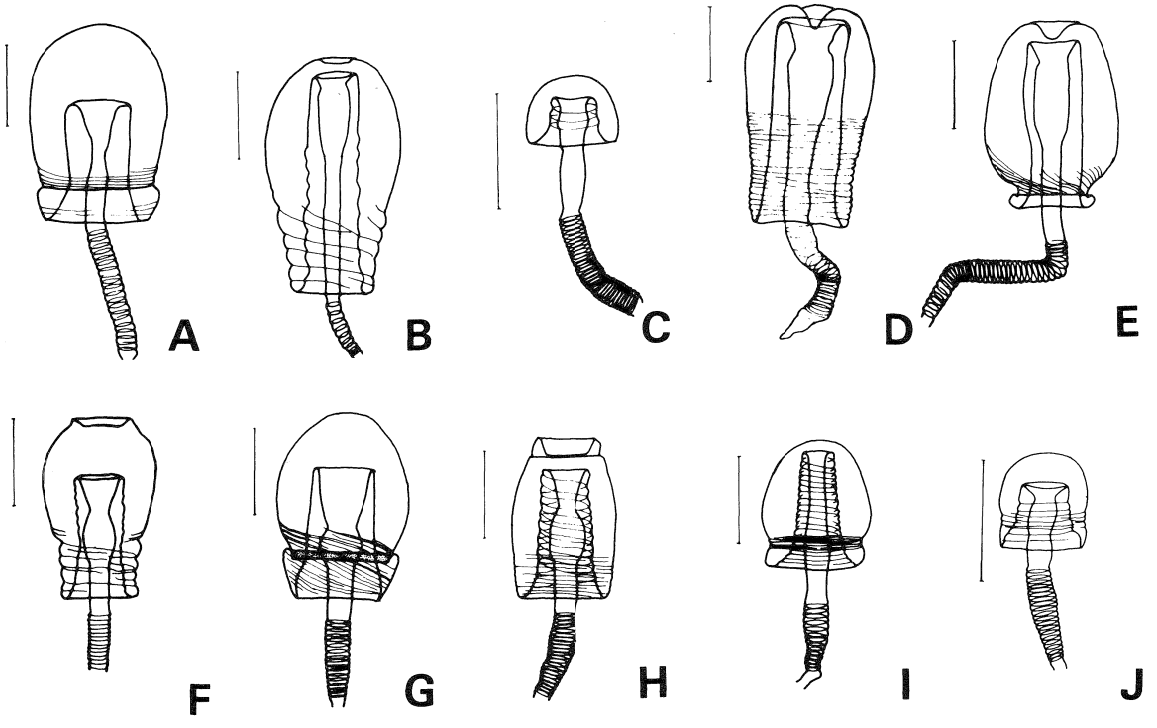

Fig. 6. Spermathecae in the *Drosophila robusta* and the *D. melanica* species-groups. A: *D. okadai*. B: *D. neokadai*. C: *D. unimaculatc*. D: *D. sp. 2*. E: *D. lacertosa*. F: *D. sordidula*. G: *D. pseudosordidula*. H: *D. robusta*. I: *D. moriwakii*. J: *D. tsigana*. Scale=0.1mm.

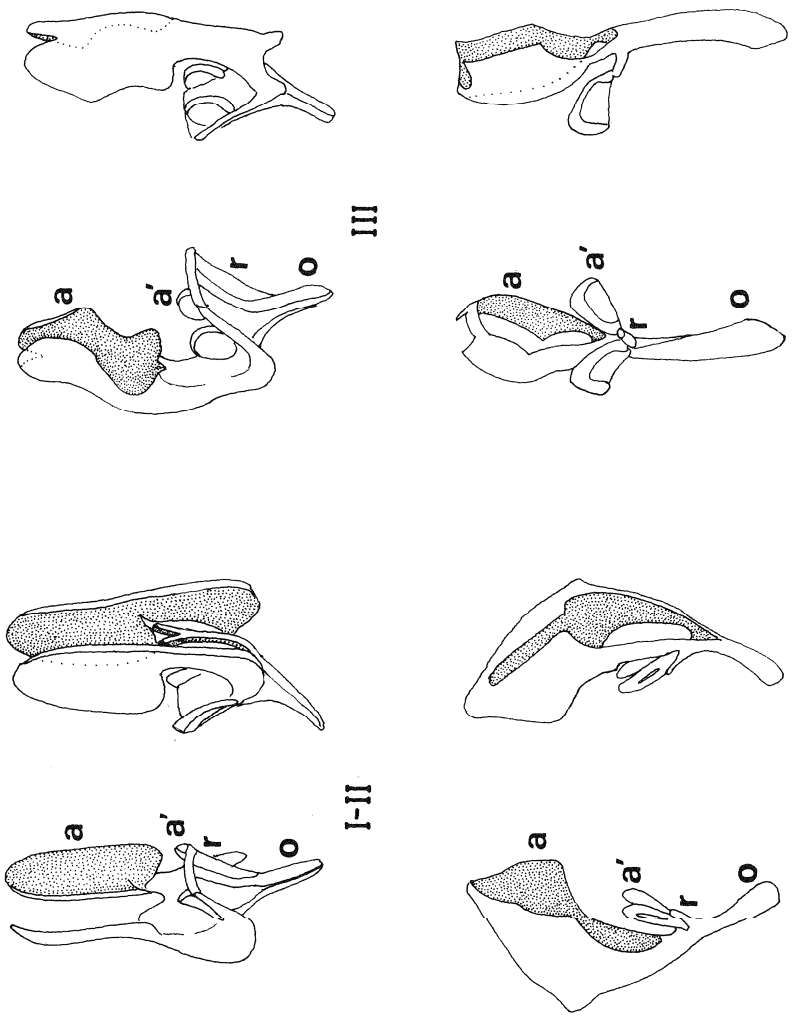

Fig. 7. Four views of phallic organs in the *Drosophila robusta* and the *D. melanica* species-groups. The Roman numerals correspond to the group numbers in Table 2. Left: postero-lateral view. Right: anterodorsal view. a: aedeagus. a': anterior paramere. o: apodeme. r: vertical rod.

D. sp. 2 (Fig. 6D) : Slightly constricted at *ca.* 1/4 of the basal portion, with apical hollow and horizontal wrinkles on *ca.* 1/2 of the basal portion; introvert very deep, *ca.* 11/12 of the height of the outer capsule.

D. lacertosa (Fig. 6E) : Folded basally, constricted heavily at *ca.* 1/8 portion, with apical indentation and oblique wrinkles on *ca.* 1/5 of the basal portion.

D. sordidula (Fig. 6F) : Constricted at *ca.* 2/5 portion, with wrinkles below the submedial constriction, and with large apical hollow; introvert middle, *ca.* 2/3 of the height of the capsule.

D. pseudosordidula (Fig. 6G) : Constricted heavily at *ca.* 1/4 basal portion, with obliqus wrinkles on *ca.* 1/3 basal portion ; introvert *ca.* 2/3 of the height of the capsule.

D. robusta (Fig. 6H) : Constricted *ca.* 1/17 from the tip, with large apical hollow, and with horizontal wrinkles on *ca.* 1/4 basal portion; introvert *ca.* 4/5 of the height of the capsule, with spiral wrinkles.

D. moriwakii (Fig. 6I) ; Constricted at *ca.* 1/6 portion and wrinkled around there; introvert *ca.* 3/4 of the height of the capsule, with spiral wrinkles.

D. tsigana (Fig. 6J) : Slightly wrinkled on *ca* 2/3 basal portion, without apical indentation; introvert *ca.* 3/4 of the height of the capsule.

Discussion

With a few exceptions, the male peripallic organs and female ovipositors, are very similar in both the *robusta* and *melanica* groups, while the male phallic organs and female

Table 2. Character ($n=7$) × species ($t=10$) matrix for 10 species of the *Drosophila robusta* and the *D. melanica* species-groups.

Group	Species (t)	Characteristic codes (n)[1] A	B	C	D	E	F	G	Diversity index
I.	*D. okadai*	0	0	0	0	0	0	1	1.0
	D. neokadai	0	0	0	0	0	0	1	1.0
	D. unimaculata	0	0	0	0	0	0	1	1.0
II.	*D. sp. 2*	0	0	0	0	0	0	0	0.0
III.	*D. lacertosa*	0	1	0.5	0	0	1	0	2.5
IV.	*D. sordidula*	0.5	0.5	1	0	0.5	0	0	2.5
	D. pseudosordidula	0.5	0.5	1	0	0.5	0	0	2.5
	D. robusta	0.5	0.5	1	0	0.5	0	0	2.5
V.	*D. moriwakii*	1	0.5	1	1	0.5	1	1	6.0
VI.	*D. tsigana*	1	0.5	1	1	1	1	1	6.5

1. These characteristis and the values are explained in the text.

spermathecae vary among species. The spermatheca is highly species-specific and cannot be included in considerations of the systematic relationship among these groups, despite its function to store sperm and without relation to the copulation itself. The male phallic organs, the aedeagus with its apodeme, anterior paramere, vertical rod, and hypandrium or novasternum, were used for the study of the systematic relationship.

Seven characteristics of the aedeagus and its accessary organs were chosen for the similarity matrix, and each was divided into two states, 0 for a presumed ancestral state and 1 for a presumed derivated (Okada, 1967). When necessary, an intermediate state of 0.5 was assigned. Plesimorph or apomorph characteristics were determined by a comparison with characteristics of alien species-groups, mainly the *D. polychaeta* group (Toda and Peng, 1989), as this group is considered to have emerged earlier than any other group of the *virilis* Section (Throckmorton, 1975). For example, a curved aedeagus is assumed to be primitive, since such an aedeagus has been found in *polychaeta* group flies. An entirely bifid aedeagus is assumed primitive with less certainty, as Okada (1954) has pointed out that the aedeagus tends to be easily separable into lateral lobes or lobes that are easily fused.

Table 3. Similarity matrix based on the matching coefficient in the male phallic organs of the *robusta* and the *melanica* species-groups.

Group	I	II	III	IV	V	VI
I		++++	++	+	±	±
II	0.86		++	++	±	±
III	0.43	0.57		+	±	±
IV	0.29	0.43	0.29		++	+
V	0.14	0.00	0.14	0.43		++++
VI	0.14	0.00	0.14	0.29	0.86	

0.0≦±<0.2≦+<0.4≦++<0.6≦+++<0.8≦++++<1.0

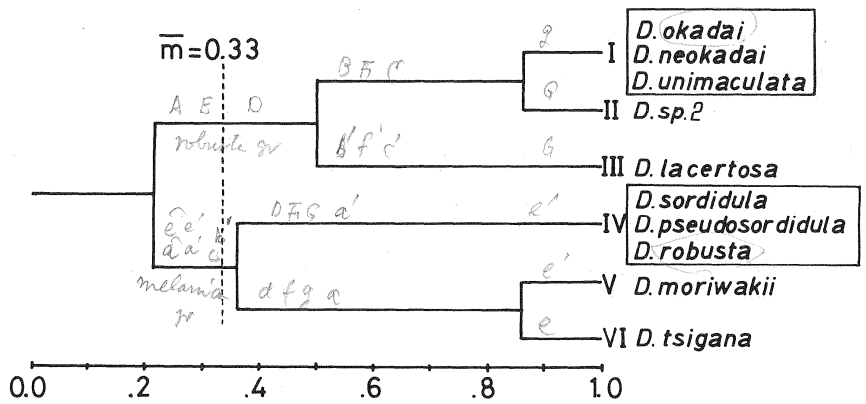

Fig. 8. A dendrogram of the *Drosophila robusta* and *D. melanica* species-group flies, derived from taximetric analysis of the phallic organs.

Seven Characteristics for Proximity Analysis

Four patterns of the male phallic organs are demonstrated in Fig. 7, for a better elucidation of characteristic states.

A : aedeagus curved ventrally and broadened at the distal half (0, I-III in Fig. 7), curved ventrally and narrowing distally (0.5, IV), or straight to apodeme (1, V-VI).

B : aedeagus entirely bilobed (0, I-II), bilobed except at the tip (0.5, IV-VI), or ventrally bilobed at the distal half (1, III).

C : anterior paramere small and semicircular (0, I-II), relatively large and oval (0.5, III), or developed and rod-shaped (1, IV-VI).

D : apodeme shorter than (0, I-IV) or subequal to the aedeagus (1, V-VI).

E : vertical rod well-developed and plate-like (0, I-III), short and rod-like (0.5, IV-V), or absent (1, IV, see for Fig. 4J).

F : novasternun without (0, I-II, IV) or with submedian spines (1, III, V-VI).

G : hypandrium bare (0, II-IV) or pubescent (1, I, V-VI, see Fig. 3).

The character×species matrix in Table 2 was constructed with these seven characteristics. The *robusta* and the *melanica* group flies were divided into six groups: Group I involves *D. okadai, D. neokadai* and *D. unimaculata*; II is *D. sp. 2;* III is *D. lacertosa*; IV is *D. sordidula, D. pseudosordidula* and *D. robusta*; V is *D. moriwakii*, and VI is *D. tsigana*. This grouping partly supports the chromosomal relationship between Group IV species (Narayanan, 1973) and the new systematic position of *D. unimaculata* (Beppu, 1988). From the diversity index, the sum of code values, the *melanica* group is more derived than the *robusta* group. Within the *robusta* group, *D. sp. 2* is more primitive than the other species.

The proximity analysis for the groups was made by the similarity coefficient. There are several different similarity coefficients, and here the matching coefficient of Sokal and Michener (1958) was adopted.

The matching coefficient is: $Ssm = Njk/N$, where N is the number of characteristics chosen, and Njk is the number of matching characteristics between two species j and k. This formula gives the ratio of the number of matching characteristics/that of all characteristics. Both positive and negative matchings were adopted for the clustering, because too few characteristics were available. This analysis is suitable for closely related species as those under discussion here, although the results may not always reflect real phylogeny or direction of speciation when comparing higher taxonomical groups such as families and genera, since the positive matching is treated in equivalence to the negative matching (cf. Russel and Rao, 1940 ; Sokal and Michener, 1958; Okada, 1967, 1981; Sneath and Sokal, 1973).

The clustering was made by the average linkage method, on the basis of the matrix of coefficient association (Table 3), and the result is shown in the dendrogram (Fig. 8). The dendrogram shows that the unknown *D. sp. 2* is nearest to Group I *(D. okadai, D. neokadai* and *D. unimaculata*). This agrees well with the external morphology and wing index (Watabe and Nakata, unpubl.). *D. moriwakii* is close to *D. tsigana*, 0.86, which supports the new systematic

position of *D. moriwakii* proposed by Beppu (1988). *D. lacertosa* relates to Groups I-II at a relatively low level (0.50), suggesting a somewhat distant relationship between these groups. The three speies of group IV, *D. sordidula, D. pseudosordidula* and *D. robusta*, relates to the two species of the *melanica* group at only 0.36. This suggests that the structure of phallic organs of *D. sordidula* and its two relatives are more closely related to the *melanica* group than any other member of the *robusta* group.

The results imply that the *robusta* species-group are divided into three species-subgroups: the *okadai* subgroup of *D. okadai, D. neokadai, D. unimaculata,* and *D. sp. 2*; the *lacertosa* subgroup of *D. lacertosa* and probably three new species with submedian spines from southern China (Watabe *et. al.*, in press); and the *robusta* subgroup of *D. sordidula, D. pseudosordidula,* and *D. robusta.*

Sturtevant (1942) established the *robusta* species-group by three species, *D. robusta, D. sordidula* and *D. colorata* Walker, under the following diagnostic characteristics : *"Large black species; posterior Malpighian tubes apposed, not with a continuous lumen; costal index about 4.0; arista with about 9 branches; horn 2/5 length of puparium"*. Since then, six species have been added to the group under this brief diagnosis. As mentioned previously, Beppu (1988) has transferred *D. moriwakii* and *D. colorata* to the *melanica* group. Nayaranan (1973) postulated these two to be an ancestral form of the *robusta* group. Therefore, the phylogenetic relationship between and within the *robusta* group and allied groups is still not settled. Toda and Peng (1989) state that a newly establised *D. quadrisetata* species-group, which is probably intermediate between the *polychaeta* group and the *robusta* group, resembles *D. okadai* and *D. neokadai* in body color, morphology of aedeagus, and the large value of the costal index. Further, Watabe *et al.* (in press) have found three new species of the *robusta* group with submedian spines, common to the *melanica* and *virilis* species-groups. This suggests that the *okadai* subgroup may be related to the *quadrisetata* species-group, and the *lacertosa* and/or the *robusta* subgroup to the *melanica* and *virilis* species-groups.

Sturtevant (1942) also established the *melanica* species-group by three North American species, and this group is considered to have emerged in the Old World and then to have diverged to the New World via the Bering Strait. Until recently, only two *melanica* species, *D. tsigana* and *D. afer* Tan *et al.*, have been recorded from the Old World (Tan *et al.*, 1949; Okada and Kurokawa, 1957; Levitan, 1983). Toda (1988) has found two new species of the *melanica* group in Burma, and now five species are known in the Old World. This is about two-fifth of the total number of the *melanica* group (Levitan, 1983).

Stalker (1966) examined the chromosomes of the *melanica* and Narayanan (1973) the *robusta* group flies, and proposed phylogenetic trees for each group. Levitan (1983) reviews the inter-and intra-specific relationships of these flies, and points to a need for further study. Recent reports indicate that the evolutionary aspects of the *robusta* and *melanica* species-groups should be reconsidered including new or chromosomally unknown species. This is also important in considerations of the origin of Hawaiian drosophilids, since *D. primaeva* Hardy et Kaneshiro, a primitive member of the Hawaiian "picture wing flies", is chromosomally closer to *D. colorata* than to any other species of continental drosophilids (Stalker, 1972).

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