New Species

Drosophila parapallidosa Tobari, sp. nov., is a new member of the *D. ananassae* species complex.

Matsuda, Muneo[1*], and Yoshiko N. Tobari[2]. [1]Kyorin University, School of Medicine - Tokyo, Japan, [2]The Research Institute of Evolutionary Biology - Setagaya, Tokyo, Japan; *Author for correspondence: matsudam@ks.kyorin-u.ac.jp

Introduction

The *ananassae* species subgroup belongs to the *melanogaster* species group, and includes 22 species. In this subgroup, there are three species complexes: *ananassae*, *bipectinata* (Tobari, 1993), and *ercepeae* (Lemunier *et al.,* 1996). In the *ananassae* complex, which includes 10 species (Tobari, 1993), Tomimura *et al.* (1993) reported that, among 20 isofemale lines established from wild caught females from Kota Kinabalu, four lines are distinguishable from others by the composition of their inversions. These four isofemale lines do not carry the ST arrangement in the 2nd chromosome but do have *In(2L)B*, *In(2L)C*, and *In(2R)A*. They also have *In(3L)E* and *In(3R)B*, which have never been found elsewhere in the *D. ananassae* populations. We tentatively called these 4 lines Taxon-K. To investigate the phylogenetic relationships in the *ananassae* complex, analyses of mitotic and meiotic chromosomes, genomic DNA, mtDNA variation, and sterility of hybrid males were carried out. This paper diagnosed and describes Taxon-K, *Drosophila parapallidosa,* Tobari, sp. nov.**,** as a new member of the *ananassae* complex of the *ananassae* species subgroup.

Results

Taxonomy

Drosophila (Sophophora) *parapallidosa* Tobari sp. nov.

Diagnosis

The external morphology of *D. parapallidosa* is indistinguishable from that of *D. pallidosa*. The phallic organs of the three species are shown in Figure 1.

Description

It is very hard to distinguish *D. parapallidosa* from *D. pallidosa*, but differences in several morphological characters are listed in Tables 1 - 3. All type specimens have been deposited in the National Science Museum, Tokyo, Japan (NSMT).

Holotype

This is a male from the isofemale line T184, which was collected in August 1979 at Kota Kinabalu, Malaysia by Y. Fuyama, F. Hihara, and T.K. Watanabe.

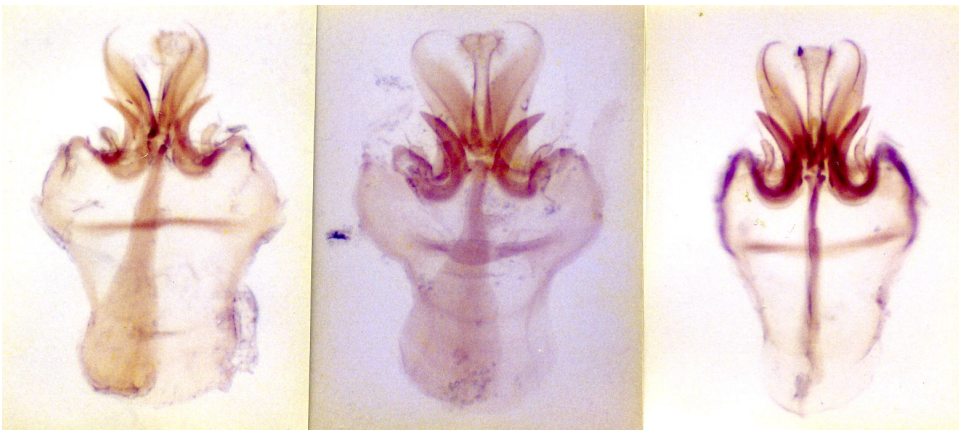


Figure 1. Phallic organs of three species, *D. parapallidosa*, *D. pallidosa*, and *D. ananassae.*

A. *D. parapallidosa* B. *D. pallidosa* C. *D. ananassae*

Table 1. Average number and teeth of the sex combs and their ranges in *D. parapallidosa* (T226), *D. pallidosa* (NAN24), and *D. ananassae* (HW).

Sex comb	Row	Species		
		D. parapallidosa	*D. pallidosa*	*D. ananassae*
metatarsus	1			1.0(0-3)
	2		1.4(0-2)	2.5(0-4)
	3	0.2(0-3)	4.0(1-6)	4.1(1-6)
	4	2.9(1-6)	5.1(3-7)	5.5(3-7)
	5	4.1(2-7)	4.9(4-7)	6.0(3-8)
2nd tarsus	1			
	2		0.1(0-1)	0.6(0-2)
	3	0.4(0-2)	2.2(1-3)	2.8(0-4)
	4	3.2(2-5)	3.5(3-5)	5.0(2-6)
	5	3.4(2-5)	4.0(3-5)	4.0(3-6)
No. of flies tested		140	160	50

Data from Appendix Table of Matsuda et al., (2009)

Paratypes

This group is comprised of 9 males and 10 females from the T184 line and 10 males and 10 females from the T226 line that was collected in August, 1979 at Kota Kinabalu, Malaysia by Y. Fuyama, F. Hihara, and T.K. Watanabe.

Distribution

KOTA KINABALU, Malaysia, LANYU, Taiwan, and OKINAWA, Japan.

Etymology

The name indicates the phenotypic similarity to *D. pallidosa*.

Table 2. Average values and ranges of taxonomic indexes of wings in *D. parapallidosa*, *D. pallidosa*, and *D. ananassae.*

Indexes	Species		
	D. parapallidosa	*D. pallidosa*	*D. ananassae*
C	1.3 (1.1-1.6)	1.4 (1.2-1.9)	1.4 (1.3-1.6)
4V	2.5 (2.0-3.0)	2.2 (1.7-2.8)	2.4 (2.0-3.1)
4C	2.0 (1.6-2.4)	1.7 (1.5-2.1)	1.8 (1.6-2.4)
5X	2.1 (1.5-3.0)	2.1 (1.4-2.7)	2.0 (1.5-2.6)
C3fg	0.54 (0.51-0.58)	0.52 (0.47-0.57)	0.51 (0.47-0.53)`
No. of flies tested	60	62	75

Data from Appendix Table of Matsuda et al., (2009)

Table 3. Major components of the cuticular hydrocarbons in *D. parapallidosa*, *D. pallidosa*, and *D. ananassae.*

Species	Major cuticular hydrocarbon
*D. parapallidosa**	C31: (Z,Z)-5,25-hentriacontadiene [(Z,Z)-5-25-$C_{31:2}$]
*D. pallidosa***	C33: (Z,Z)-5,27-tritriacontadiene [(Z,Z)-5,27,$C_{33:2}$]
*D. ananassae****	C31: (Z,Z)-5,25-hentriacontadiene [(Z,Z)-5-25-$C_{31:2}$]

* Data from Matsuda et al., (2009)
** Data from Oguma (1993) and Nemoto et al. (1994)
*** Data from Doi et al. (1997)

Morphological Characteristics

Sex combs

The number of teeth of the sex combs in *D. parapallidosa* is less than those in *D. ananassae* and *D. pallidosa* (Table 1).

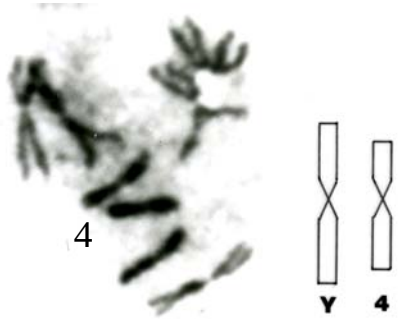
Male terminalia

Periphallic and phallic organs of *D. parapallidosa* are difficult to distinguish from *D. pallidosa* males, as shown in Figure 1.

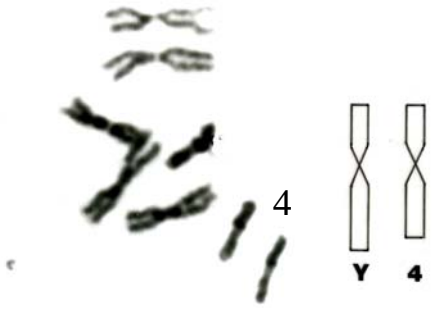
Wing index

Ranges of four traits: Costal index, 4V, 4C, and 5X are shown in Table 2. C3fg values overlap completely among species, ranging from 0.42 to 0.63; average values are 0.51, 0.52, and 0.54 in *D. ananassae*, *D. pallidosa*, and *D. parapallidosa*, respectively. Other indices overlap as well in all three species, although the mean values are slightly different.

D. parapallidoas



D. pallidosa



D. ananassae

Figure 2. Male mitotic chromosome configurations of three species, *D. parapallidosa, D. pallidosa,* and *D. ananassae*.

Chromosome Configurations

Mitotic chromosomes

The Y and 4th chromosomes are slightly shorter in *D. parapallidosa* than those in *D. pallidosa*. Both chromosomes are sub-telocentric in *D. parapallidosa,* while the Y chromosome is submetacentric in *D. ananassae* and metacentric in *D. pallidosa* (Figure 2).

Polytene chromosomes

D. parapallidosa has no Standard arrangement on 2L and 2R, but has *In(2L)B, In(2L)C,* and *In(2R)A*. *D. parapallidosa* also has the *In(3L)E* arrangement on the 3rd chromosome, which has not been found in either *D. ananassae* or *D. pallidosa* (Tomimura *et al*., 1993. Matsuda *et al*., 2009).

Table 4. Hybrid male sterility and insemination success (%) among three species, *D. parapallidosa*, *D. pallidosa*, and *D. ananassae.*

Female \ Male	*D. parapallidosa*	*D. pallidosa*	*D. ananassae*
D. parapallidosa	-	F (68%)	F (44%)
D. pallidosa	S (82%)	-	F (67%)
D. ananassae	S (8%)	F (2%)	-

F: fertile, S: sterile (Data from Matsuda et al., 2009)

Cuticular Hydrocarbons

The main cuticular hydrocarbon of *D. parapallidosa* is the same as that of *D. ananassae*, but different from that of *D. pallidosa,* as shown in Table 3.

Interspecific Hybridization

Hybrid females are fertile in all interspecific crosses. F_1 males from crosses between *D. ananassae* or *D. pallidosa* females and *D. parapallidosa* males are sterile, while those from the reciprocal crosses are fertile (Table 4). Mating success between species is described in Matsuda *et al*. (2009) and summarized in Table 4. There are large differences in the reciprocal crosses between *D. ananassae* and *D. parapallidosa* (44% *vs.* 8%), but only slight differences in the reciprocal crosses between *D. pallidosa* and *D. parapallidosa* (82% *vs* 68%). Both pre-mating and post-mating isolations are found between *D. parapallidosa* and *D. ananassae* or *D. pallidosa*.

Table 5. Typical gene arrangements of three species, *D. parapallidosa*, *D. pallidosa*, and *D. ananassae.*

Gene arrangements	*D. parapallidosa*	*D. pallidosa*	*D. ananassae*
XLST	+	-	+
XLA	-	+	-
XRST	+	+	+
2LST, 2LA, 2LJ	-	-	+
2L(C+B)	+	-	-
2L(CD+B)	-	+	-
2RST	-	+	+
2RA		+	-
2RAB	-	+	-
3LST	+	+	+
3LA	-	-	+
3LE	+	-	-
3RST	+	-	+
3RA	-	-	+
3RB	+	+	-

Data from Tomimura et al., (1993), and Matsuda et al., (2009)
+ : present, - : absent

Discussion

The external morphology of *D. parapallidosa* is similar to *D. pallidosa* Bock and Wheeler, but they can be distinguished by the number of teeth of the sex comb, chromosome arrangements, karyotypes, hybrid sterility, cuticle hydrocarbons, and molecular variations. Although *D. parapallidosa* was first found in 1971 from Kota Kinabalu, Malaysia, and recurrently found in 1979 from the same area, it was not found in other populations until 1998. Since 1998, we have found *parapallidosa* in Ishigaki-jima, Iriomote-jima, and Hateruma-jima, Okinawa, Japan, and in Lanyu, Taiwan. Apparently, *D. parapallidosa* has recently migrated from a tropical area north to a sub-tropical area and expanded its habitat.

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References: Doi, M., T. Nemoto, H. Nakanishi, Y. Kuwahara, and Y. Oguma 1997, J. Chem. Ecol. 23: 2067-2078; Lemunier, F., S. Aulard, M. Arienti, J.M. Jallon, M.L. Cariou, and L. Rsacas 1996, Ann. Entomol. Soc. Am. 90: 28-42; Matsuda, M., C-S. Ng, M. Doi, A. Kopp, and Y.N. Tobari 2009, Fly 3: 157-169; Nemoto, T., M. Doi, K. Oshio, H. Matsubayashi, and Y. Oguma 1994, J. Chem. Ecol. 3029-3037; Oguma, Y., 1993, In: *Tobari, 1993*, pp. 199-207; Tobari, Y.N., 1993, Drosophila ananassae: *Genetical and Biological Aspects*. Tokyo, Japan Scientific Society Press; Tomimura, Y., M. Matsuda, Y.N. Tobari, M.L. Cariou, J.L. Da Lage, W. Stephan, and C.H. Langley 1993, In: *Tobari, 1993*, pp. 139-198.

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