Cyto-genetic Studies of *Drosophila montium* (Report I)

By

Tyozo Osima

Zoological Institute, Kyoto Imperial University

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1. Introduction

Since the end of 1937, I have devoted myself to genetical and cytological studies of *D. montium* under the direction of Prof. T. Komai. The results obtained so far are published in the present report. Before going further, it is my pleasure to record here my indebtedness to Prof. T. Komai for his kind guidance. I wish to express my hearty thanks also to Dr. M. Chino, Mr. S. Fujii and Mr. T. Takaku for their kindness in giving me valuable aids in many ways.

2. Material

D. montium resembles *D. melanogaster* in its general features, but can be distinguished from it in that the eye is considerably larger and the body is much slenderer, and especially that the sex-combs are found on both the first and second tarsal joints of the prothoracic leg. The species also resembles *D. auraria* very closely. The only marked difference consists in the presence in *D. auraria* and the absence in *D. montium* of snowy white sprinkling on the carina of male.

The species is widely distributed, very common in the tropical regions of the Eastern Hemisphere, and has been recorded from Africa, Formosa, Java, India, Samoa, Sumatra, etc. (De Meijere 1916, Duda 1924, 1926, Malloch 1934, Kikkawa & Peng 1938).

Kikkawa discriminated two races among the species inhabiting Japan according to the difference in chromosomes as mentioned in the next chapter.

3. Cytological findings

a. Gonial chromosomes

The oogonial chromosome group of *D. montium*, as stated by Kikkawa (1936), consists of four pairs of chromosomes, namely, two large V-shaped pairs, a rod-shaped pair and a pair of small chromosomes which are V-shaped in Race A and rod-shaped in Race B (Fig. 1, a, b).

In the spermatogonial group of Race B, one of the rod-shaped chromosomes is replaced by a small V-shaped chromosome (Fig. 1, e).

In the oogonial metaphase of the non-disjunctional female of Race B, a small V-shaped chromosome is always found in the neighbourhood of one pair of the rod-shaped chromosome (Fig. 1, d).

Fig. 1. Gonial chromosomes. ($\times 2300$). a-d. Oogonial chromosomes. e. Spermatogonial chromosomes. a. A-race. b. B-race. c. Hybrid of A & B. d. XXY female e. B-race (X, X-chromosome. Y, Y-chromosome. h, heteromorphic chromosomes.)

These findings show that the small V is the Y-chromosome and the rod is the X-chromosome. The X-chromosome may be recognized from the other small chromosome by having two constrictions. The latter chromosome which is rod-shaped in Race B and V-shaped in Race A, has only one constriction, at about one-fourth from one end of the rod and at the middle of V. These constrictions in the small autosomes seem to indicate the points of spindle-fiber attachment.

In the hybrid between the two races which has normal viability, the oogonial complex contains one heteromorphic pair composed of a V-shaped chromosome and a rod-shaped chromosome (Fig. 1, c).

b. Salivary gland chromosomes

In both races, there are six strands branching off from the chromocenter, of which five are long and one very short. One of the long strands represents the X-chromosome.

Fig. 2. Salivary gland chromosomes.

This strand exists in a slender haploid condition in the salivary gland nuclei of the male larva. The other four strands represent the arms of the long V-shaped chromosomes. The short strand is attached to the chromocenter with both proximal and distal ends and appears like a ring (Kikkawa 1936) (Fig. 2).

These observations apparently show that the small ring-like chromosome corresponds with the heteromorphic chromosome found in the meiosis, and its distal part, as well as its proximal end, is formed by inert substance.

No chromosomal aberration such as inversion and translocation is observed.

Among the strains of the species kept in this laboratory, those from Saipan (South-Sea Ids.), Matuyama (Sikoku), Yuasa, Simoda and Kabano (Honsyū) belong to B race, and the strains from Oita, Amakusa (Kyūsyū), Isigakizima (Ryūkyū Group) and Ninpo (China) belong to A race.

4. Genetical Findings

So far thirty mutant characters have been obtained. Many of these were found among the progeny F_1 , F_2 , F_3 or F_4 of the flies treated with X-rays. Twenty-five of these have been assigned to three linkage groups. From the cytological observations stated above, four linkage groups are expected, so that there must be one group none of whose mutant genes has been discovered as yet. Presumably this missing group corresponds with the small heteromorphic chromosome indicated above.

a. Linkage data and chromosome maps

The map distance was determined by the recombination value between the two genes and the order of genes by means of the customary three point experiment. In some cases where no three point experiment had been performed, the order was determined by consulting various data concerning the two point experiments. Of the chromosome maps prepared in this manner those of the first and third linkage groups are relatively more complete than that of the second linkage group where it has been difficult to ascertain the loci of the mutant genes on account of the similarity of the phenotypes of many of them (Fig. 3).

b. Description of the mutant characters

Chromosome I (X-chromosome)

rl—rolled. (1–0.0) 39b28 Spontaneous in *vermilion*. Wing edges rolled downwards. RK1.

Pw—*Pointed wing*. (1–6.4) 39b15 Wings short and narrow. Wings of heterozygous female narrowed slightly at tip. Dominant hom. fully viable. RK2(♂).

w—*white*. (1–17.4) 38j18 Induced by X-rays. Eye color white and ocelli colorless. Rk1.

w^a—*apricot*. (1–17.4) 39f20 Allel. of white. Eye color yellowish pink. *w^a*. v. more yellowish. RK1.

Fig. 3. The chromosome maps.

N—*Notch*. (1–20.1) 38e2 Ind. by X-rays. Wings notched at tips and often along edges. *L2* and *L5* thickened. Hom. ♀ and ♂ lethal. RK2.

Ro—*Rough*. (1–44.9) 38f11 Spon. in *Notch*. Eye surface rough with facets irregular in size and in arrangement. Eyes of hom. slightly smaller than wild-type. RK2.

sb—*small bristles*. (1–45.5) 38d13 Spon. in wild stock. Bristle length 1/3 of wild-type. Late hatching. RK3.

v—vermilion. (1-49.8) 38j29 Spon. in *Notch*. Eye color bright scarlet, not translucent; ocelli colorless. RK1.

Ds—Dominant short. (1-52.4) 39f5 Spon. in wild stock. *L5* shortened. The inner margin of the wings often excised. Dominant hom. ♀ and ♂ fertile. RK1.

Chromosome II

M2a—Minute 2a. (2-0.0) 38f6 Ind. by X-rays. Minute bristles. Hom. lethal. Late hatching. RK3.

Co—Complex. (2-17.1) 38i28 Ind. by X-rays. Post-cross vein thickened like in *Plexate*. Hom. lethal. RK3.

ex—extra. (2-17.1) 38b5 Spon. in wild stock. Allel. of *Complex*. Post-cross vein with an extra vein, occasionally overlaps wild. RK3.

Cf—Confluent. (2-20.5) 38h9 Ind. by X-rays. *L2* and *L5* thickened and knotted. Wings slightly narrowed, often with a vesicle. Hom. lethal. RK2.

Cf²—Confluent.² (2-20.5) 39d1 Ind. by X-rays. Allel. cf *Cf*; like *Cf*. RK2.

Sv—Short veins. (2-26.8) 38c20 Spon. in wild stock. Veins shortened like in *Hairless*. Hom. lethal. RK1.

cu-b—curled-b. (2-29.0) 39c9 Spon. in *Short veins*. Wings curved upwards, occasionally throughout their length and slightly divergent. Postscutellars often erect and crossed. RK2.

H—Hairless. (2-?) 37i26 Spon. in wild stock. Many scutellars and dorsocentrals gone. Vein shortened, especially *L3* and *L5*. Hom. lethal. RK1.

Chromosome III

cu—curled. (3-0.0) 38e6 Spon. in wild stock. Wings curved upwards like *curled-b*, but postscutellars normal. RK2.

Bd-b—Beaded-b. (3-7.5) 39d16 Ind. by X-rays. Wings with excisions on anterior margin, intensified by *Notch*. Hom. lethal. RK1.

Pt—Plexate. (3-48.7) 38i28 Ind. by X-rays. Posterior cross-vein thickened. Hom. lethal. RK1.

se—sepia. (3-81.8) 39e16 Spon. in curled. Eye color light brownish on emergence, darkening to sepia. Viability poor. RK4.

px—plexus. (3-89.9) 38h9 Ind. by X-rays. Extra veins in the neighbourhood of post-cross vein. RK2.

ba—balloon. (3-?) 38c8 Ind. by X-rays. Wings inflated on the inner side, narrower than in wild-type and often divergent. RK3.

wo—white ocelli. (3-?) 38b19 Spon. in wild stock. Ocelli colorless. Eye color normal. RK4.

Chromosome unknown

Bd—*Beaded*. 38k14 Spon. in *Notch*. Wings with excisions on anterior margin. Extremely variable and often overlaps wild. Hom. lethal. RK4.

br—*broad*. 38? Spon. in *curved*. Wings broadened and rounded at tip. RK4.

c—*curved*. 38d13 Spon. in wild stock. Wings curved downwards, more pronounced in ♀. RK4.

i—*interrupted*. 39c1 Ind. by X-rays. P.c. absent or reduced, more pronounced in ♀. RK4.

wp—*warped*. 38d22 Spon. in wild stock. Wings divergent, small narrow and dusky, curved or warped. RK4.

Table 1. Summary of linkage data

The first chromosome

Loci tested	Crossovers	Total flies	Recombination percentage
rl — w	34	195	17.4
w — N	54	2560	2.1
w — sb	218	829	26.2
N — sb	162	658	24.6
w — Ro	40	213	18.7
Ro — sb	3	650	0.4
Ro — v	30	377	7.9
v — Ds	27	1366	1.9
Ro — Pw	536	1712	31.3
w — Pw	188	1712	10.9
wa — Ro	210	1153	18.2

The second chromosome

Loci tested	Crossovers	Total flies	Recombination percentage
Cf — M2a	6	547	1.0
Cf — cu-b	6	190	3.1
Cf — Sv	59	891	6.6
Cf ² — cu-b	17	265	6.4
Co — cu-b	65	602	10.7
Sv — ex	47	484	9.7
M2a — ex	92	444	17.1
Sv — cu-b	23	1019	2.2
Cf ² — M2a	36	231	15.5
Sv — M2a	133	566	23.4

The third chromosome

Loci tested	Crossovers	Total flies	Recombination percentage
Pt — cu	266	678	39.2
Bd-b — px	168	342	49.1
Bd-b — cu	163	2203	7.3
Bd-b — Pt	920	2203	41.7
Bd-b — se	48	102	47.0
Pt — se	119	359	33.1

5. Non-disjunction

Females of Race B homozygous for *white* or *vermilion* (both sex-linked genes) were treated with X-rays (50KV. 4 ma. 20 cm. nonfilter 30 minutes), and mated with wild-type males. As shown in Table 2, the percentages of the exceptional females and males produced in F_1 are not significantly different between treated and control strains.

Table 2. (Primary non-disjunction)

Mating		Regular offspring		Non-disjunctions		Total
X-ray treated		♀	♂	♀	♂	
	$\frac{w}{w} \times +$	606	520	0	5	1131
	$\frac{v}{v} \times +$	922	824	2	7	1746
Total		1528	1344	2	12	2886
%				0.13	0.89	
Control		2104	1932	3	19	4058
%				0.14	0.98	

Table 3. (Secondary non-disjunction)

Mating		Regular offspring		Non-disjunctions		Total
XXY $\times$ + (vermilion)		♀	♂	♀	♂	
		638	537	1	4	1216

Among the 1216 flies produced by the XXY mother, five were exceptionals, the frequency being 0.75% in the male and 0.16% in the female, and the female to male ratio 1:4. Among the daughters of such a mother, the same number of XX's and XXY's are expected. Cytological examinations showed that, of 27 tested, 10 were XX's and 17 were XXY's.

6. Summary

1) *Drosophila montium* is one of the tropical species distributed widely in the Eastern Hemisphere: Africa, Formosa, Java, India, Samoa, Sumatra and southern regions of Japan and China.

2) Two races, A and B, may be distinguished in this species by the difference in chromosome complex. The oogonial metaphase plate includes four pairs of chromosomes, namely, two large V-

shaped pairs, a rod-shaped pair (X's) and a pair of small chromosomes, which are V-shaped in Race A and rod-shaped in Race B. In the male, one of the rod-shaped X's is replaced by a small V-shaped Y. In the salivary gland nucleus, six strands radiate from the chromocenter. One of these is much shorter than others and is attached to the chromocenter with both ends like a ring. This apparently corresponds with the short rod and small V found in the metaphase complex.

3) Thirty mutants have been discovered, of which twenty-five are assigned to three linkage groups. The mutants are described briefly.

4) Percentages of spontaneous non-disjunctions and of those obtained by X-ray treatment are given.

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