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Relationship between D. funebris and D. multispina.

D. funebris which is known to be a cosmopolitan species has been recorded from both Hokkaido and Honshu in Japan. On the other hand, D. multispina Okada which closely allied to the former species has been collected in Hokkaido only. In some localities in Hokkaido, these two species appear to have close habitats though they discrete microecologically. D. multispina prefers to inhabit forested areas while D. funebris is apt to inhabit domestic environments.

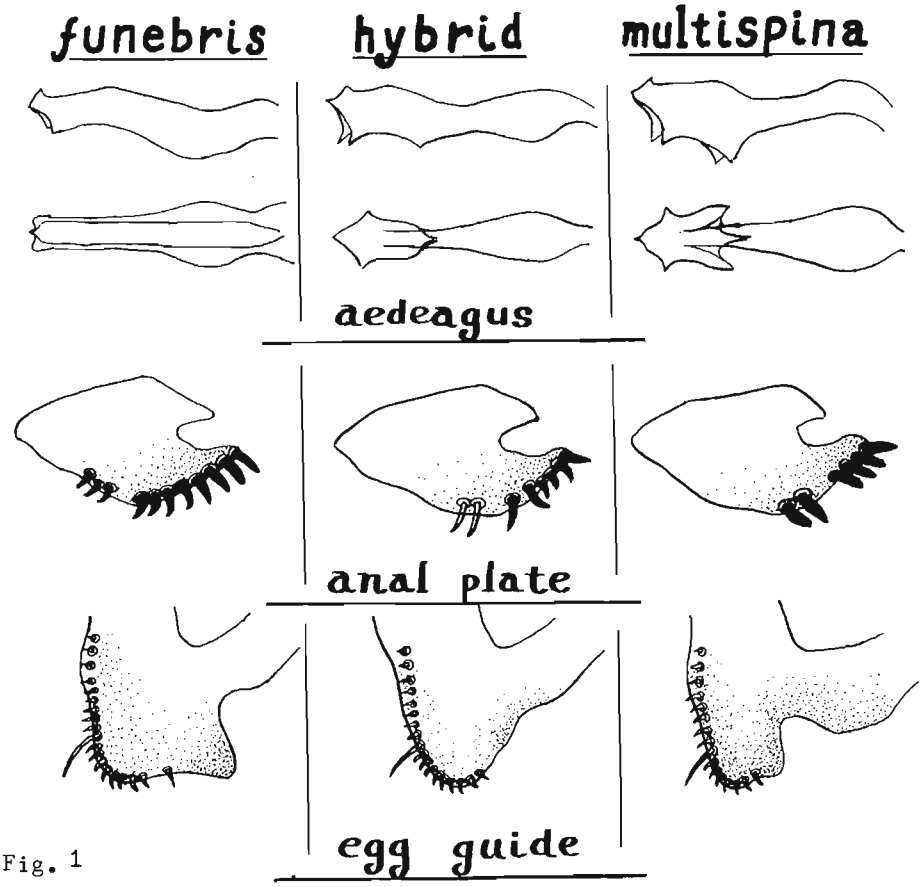

Fig. 1

They are also morphologically similar to each other (Okada, 1956). Only differences in the characters, such as, the shapes of male and female genitalia, and the abdominal sternites were shown (Figure 1). Any difference concerning the ganglionic metaphase chromosomes could not be detected between the two. Sexual isolation experiments were carried out at different temperatures by using male multiple choice technique. The results were summarized in Table 1. In the experiment at 23°C, none of funebris female could be inseminated with multispina male, while a few number of the females carrying alien sperms were seen in the lower temperature at 19°C. It is clear that strong sexual isolation has precluded gene exchange between two species.

Cross experiments between two species were made (Table 2). Hybrid males seemed to be sterile because subsequent backcross experiments using these males were not successful.

Table 1.

Temperatures	Crosses ♀♀ ♂♂	Homogamic N	Homogamic %(+)	Heterogamic N	Heterogamic %(+)	$K_{1,2}$ or $K_{2,1}$	K_1 and K_2	χ^2	P
23°C	f,m x f	100	74.0	98	21.4	0.702	0.851	57.95	<0.001
	f,m x m	98	69.4	100	0.0	1.000		109.79	<0.001
19°C	f,m x f	84	97.6	73	28.8	0.833	0.869	82.09	<0.001
	f,m x m	76	90.8	75	10.7	0.905		95.17	<0.001

K...Isolation Coefficient
f...funebris, m...multispina

Table 2.

Crosses 5♀♀		5♂♂	Pairs tested	No. of offspring ♀♀	♂♂
f	x	f	60	3140	3128
m	x	m	60	1224	1303
f	x	m	200	0	0
m	x	f	200	677	635
$(m \times f)F_1$	x	f	100	2298	1792
$(m \times f)F_1$	x	m	50	365	301
f	x	$(m \times f)F_1$	50	0	0
m	x	$(m \times f)F_1$	50	0	0

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Effect of prolonged CO_2 exposure on flight.

In the course of studies of energy metabolism with the Canton S strain of D. melanogaster, we have had occasion to employ thoracic sarcosomes of the adult and have made some observations which may be of interest to any worker employing CO_2 as an anesthetic. Thoraces have been obtained by removing head and abdomen of adults with watchmakers forceps. The procedure can be carried out either with well chilled flies in a cold room or with CO_2 anesthetized flies at room temperature. For the latter method, flies are placed in a dry plugged vial into which is inserted a small glass tube connected to a CO_2 generator consisting of a stoppered sidearm flask containing dry ice. It was noticed that chilled flies returned to room temperature seemed unharmed by the experience, whereas a large number of those exposed to CO_2 intermittently or continuously for periods of 45 minutes or more were unable to fly after recovery from anesthesia. Although the wings could be lifted, flight was not attained even when flies were shaken out of a vial in mid air. Walking and hopping movements, however, were normal. In most such individuals, the condition appeared to be permanent since the inability to fly was still evident the following day and as long as observed thereafter. Microscopic examination showed no abnormalities of thorax or wings. Fertility of males and females was not seemingly affected and the condition was not inherited by subsequent generations. To determine if the effect on flight was induced by anoxia alone, rather than by some more specific effect of CO_2, flies were exposed to a nitrogen atmosphere. The results were similar to those obtained with CO_2 indicating that extended periods of anoxia are a reasonable cause of the deleterious effect on flight. Since walking and body movements were normal, presumably the relevant musculature of the appendages was also normal. In contrast, the more highly specialized flight muscle seemed sensitive to anoxia. One likely site of injury in flight muscle is the sarcosome and thus the possibility that these specialized mitochondria were no longer able to effect coupled oxidation of an appropriate substrate was tested. Sarcosomes were isolated from untreated and CO_2 treated adults and P/O ratios were obtained by conventional Warburg techniques using α-glycerophosphate as substrate. Methodology followed that of Gregg et all (Biochim. Biophys. Acta, 45 (1960) 561). Results of 10 experiments with CO_2 treated flies gave a mean P/O ratio of 1.54 as compared to 1.49 in an equal number of experiments with controls. Oxygen uptake in µatoms/mg protein was 2.42 and phosphate esterified as ATP was 3.73 µmoles/mg protein in the experimental while corresponding values for controls were 2.24 µatoms O/mg and 3.42 µmoles P/mg. Thus, no significant differences in the ability of sarcosomes to effect oxidation and coupled phosphorylation were revealed in treated and untreated groups. It is obvious that the site of injury has not been identified. By analogy with mammalian systems, it is possible that inability to fly after prolonged anoxia may be of neurological origin. In any event, the use of CO_2 or other gases as anesthetic agents may result in physiological side effects requiring assessment in some types of experiments.

(Supported by research grant HD 01240 from NIH).