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Genetic control of color dimorphism in *Drosophila punjabiensis* of montium subgroup.

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Abstract

Drosophila punjabiensis, belonging to the montium species subgroup of melanogaster group, was examined for abdominal melanisation. Females show dimorphism dark or light in coloration, whereas males exhibit monomorphic abdominal melanisation, *i.e*., all are dark. The color dimorphism is regulated by two alleles of a single autosomal locus, and the light allele is dominant. Thus, *D. punjabiensis* exhibits color dimorphism controlled by a single locus, but its ecological significance is not clear.

Introduction

Abdominal melanisation is a conspicuously variable adaptive trait in many insects including *Drosophila* (Wittkopp *et al.,* 2003; True, 2003; Rajpurohit *et al.,* 2008). In different insect taxa, there are diverse patterns of body melanisation, *i.e.,* (a) several black species of Collembola occur in temperate regions, *i.e.,* Pyrenees, Swiss Alps, and Himalayas (Mani, 1968; Rapoport, 1969); (b) in *D. melanogaster*, a cosmopolitan species, the extent of melanism varies with geographical location (Pool and Aquadro, 2007; Parkash *et al.,* 2008a,b); (c) discrete melanic and non-melanic morphs occur as genetic polymorphism in species of the montium species subgroup (Ohnishi and Watanabe, 1985). The color polymorphism in abdominal tergites was first reported by da Cunha (1949) for *D. polymorpha.* Later on color variations in montium species subgroup *D. rufa* (Oshima, 1952), *D. kikkawai* (Freire-Maia *et al.,* 1954), *D. auraria* (Lee, 1963), and *D. jambulina* (Parkash and Sharma, 1978; Parkash *et al.,* 2009) were described.

The present work is a first report showing the genetic basis of color dimorphism in *D. punjabiensis* of montium species subgroup, through Mendelian crosses between dark selected and

light selected strains. Our results exhibit color dimorphism for the last two abdominal segments in females; and the light morph is dominant over the dark morph.

Material and Methods

Cultures

Wild individuals of *D. punjabiensis* (n = 130–160) were collected from lowland localities (~28.5ºN; 219 m) of subtropical parts of the Indian subcontinent. The collections were made in September- October with net sweeping and bait traps from fruit markets and godowns, as well as from nurseries. Based on the T_{ave} data of the sites of origin of populations, cultures were maintained at 25ºC. Density was kept low (30–40 eggs per vial) by limiting the egg laying period for 6–8 h. Climatic data for the sites of origin of populations were obtained from the Indian Institute of Tropical Meteorology (IITM; www.tropmet.res.in).

Dark and light strains

In *D. punjabiensis*, body color polymorphism is limited to females only and is not evident in males. For obtaining true breeding dark and light strains, isofemale lines were established from the wild-caught females (60–80) of each population, and the progeny of each line were checked for 8 successive generations for dark and light phenotypes. Many of the isofemale lines showed segregation for dark and light strains. We isolated virgin flies from such laboratory cultures and made several single pair matings (40–50). The crosses that gave all dark or light progeny were considered as true breeding strains.

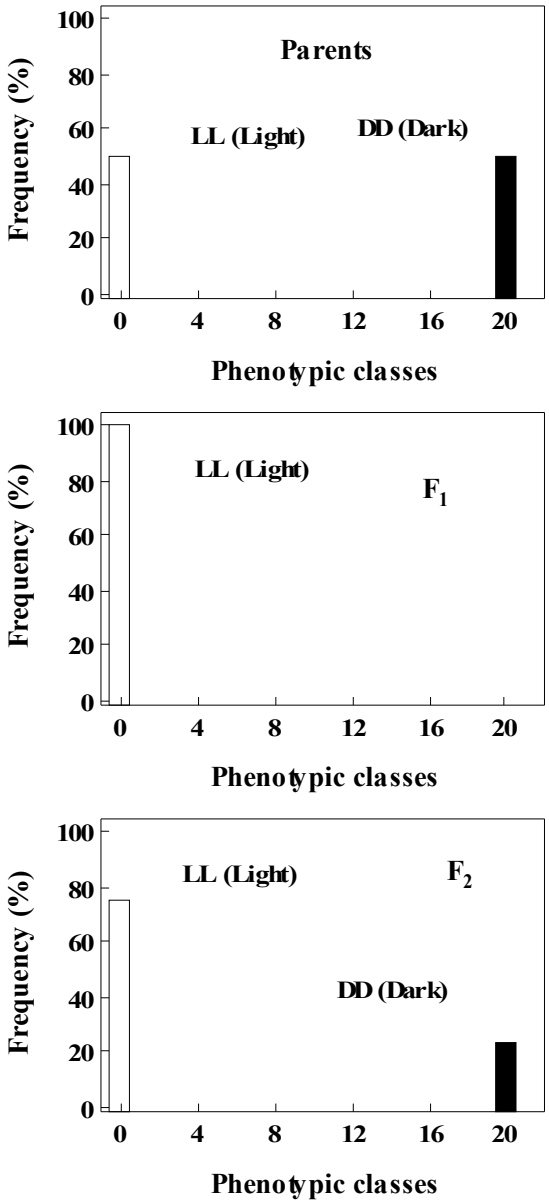
Genetic basis of color dimorphism

In female individuals of *D. punjabiensis,* there are variations in percent melanisation for the last two abdominal segments (6^{th} and 7^{th}), *i.e.,* either these segments are totally dark or light which correspond phenotypically to dark or light morphs. However, males do not show such variations for

Table 1. Results of F_1 and F_2 genetic crosses (5 replicates) between homozygous light and dark strains of *D. punjabiensis* for female progeny scored. Only females demonstrate body color polymorphism.

Genetic Crosses	Type/ Replicate	♀ flies scored* (n)	Light ♀ Morph (Freq. %)	Dark ♀ Morph (Freq. %)	Mendelian Ratio	χ^2 test
(A) Light ♀ * ♂	F_1	200	200 (100 %)	0 (0 %)	----	----
F_{1A} ♀ * F_{1A} ♂	F_2: 1.	125	96 (76.80%)	29 (23.20%)	3.31:1	ns
	2.	146	109 (74.66%)	37 (25.34%)	2.96:1	ns
	3.	112	83 (74.11%)	29 (25.89%)	2.86:1	ns
	4.	134	101 (75.37%)	33 (24.63%)	2.97:1	ns
	5.	128	97 (75.78%)	31 (24.22%)	3.13:1	ns
(B) Dark ♀ * ♂	F_1	200	200 (100 %)	0 (0 %)	----	----
F_{1B} ♀ * F_{1B} ♂	F_2: 1.	130	97 (74.62%)	33 (25.38%)	2.94:1	ns
	2.	152	115 (75.66%)	37 (24.34%)	3.11:1	ns
	3.	140	106 (75.71%)	34 (24.29%)	3.12:1	ns
	4.	115	86 (74.78%)	29 (25.22%)	2.97:1	ns
	5.	129	98 (75.97%)	31 (24.03%)	3.16:1	Ns

F_{1A} = Light ♀ * ♂: F_{1B} = Dark ♀ * ♂, ♂'s appear phenotypically similar; ns = nonsignificant, no significant difference is observed between expectations and the observed numbers through χ^2 test.



the last two abdominal segments. The reason for this sexual dimorphism is not known. Thus, only female progeny of crosses were scored for genetic analysis. Furthermore, in order to ascertain the genetic basis as well as allelic dominance, we carried out Mendelian crosses (F_1 and F_2 crosses) with these true breeding dark and light strains of *D. punjabiensis*. We made 18 single pair matings using 1 ♂ and 1 ♀, of each morph type for obtaining F_1 progeny. We randomly scored 200 ♀ flies (F_1) and all were found to be of light morph. Further, out of the pooled F_1 progeny, 100 ♂ and 100 ♀ were randomly selected and ten replicates with 10 pairs each were used for F_2 progeny. Therefore, the data on male progeny were not included in the tables.

Figure 1. Frequency distributions of phenotypic classes on the basis of the last two abdominal segments (6^{th} and 7^{th}) for percent melanisation in dark and light females in F_1 and F_2 crosses of *D. punjabiensis*. For parents, equal numbers of dark and light flies were taken to perform crosses.

Results and Discussion

Table 1 shows the results of crosses between true breeding strains for dark and light morphs for the female color dimorphism in *D. punjabiensis*. Crosses helped to analyze genetic basis as well as allelic dominance for body color polymorphism of the last two abdominal segments in females. Reciprocal crosses for F_1 progeny always yielded light morph (Table 1). In reciprocal F_2 crosses ($F_1 \times F_1$), dark morph reappeared, and phenotypic ratios of light: dark morphs were close to the Mendelian F_2 ratio of 3:1 (Table 1; Figure 1). Dark and light morphs were discrete and followed Mendelian inheritance of a single locus with two alleles. The data from F_1 and F_2 progeny clearly evidenced lack of any intermediate morph (Table 1; Figure 1). Our results concur with similar genetic analyses of southeast Asian populations of *D. jambulina* (Ohnishi and Watanabe, 1985; Parkash *et al.,* 2009). The dominance of light allele over dark allele may help this species in adaptation to warm and humid environments in the tropics (Parkash *et al.,* 2009). Thus, like its sibling species *D. jambulina*, we hope to find similar evidence in *D. punjabiensis* of montium species subgroup and definitely this study needs further investigation.

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Reduced mating activity and fitness of *Drosophila ananassae* on exposure to valproic acid.

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Abstract

Over the last few decades, *Drosophila* has been used as model for the study of toxic effect of drugs. The drugs play major implication as stress molecules that reduce the overall fitness in general. The present study is aimed to address the exposure of *D. ananassae* to variable doses of Valproic acid an anti epileptic drug (AED). In order to ascertain the biological potentiality in terms of life history traits such as mating propensity, fecundity, fertility and life span mating propensity, fecundity, fertility and life span. Interestingly the observation reveals that flies exposed to the higher dose of VAL have experienced increased courtship duration with low fecundity and fertility, while the differences are insignificant for longevity.

Key words: Valproic acid, *D. ananassae*, Mating propensity, Fecundity, Fertility, life span.

Introduction

Valproic acid (VAL) is an anticonvulsant drug used in the treatment of epilepsy and bipolar disorder, across AEDs, valproate was associated with the highest risk of reproductive toxicity. VAL has the highest risk of birth defects of any of the commonly used antiepilepileptic drugs. However, some epilepsy can only be controlled by valproate, and also the risk of birth defects with valproate is two to five times higher than other frequently used anti-epileptic drugs (Sander, 2010).

D. ananassae is used as a model organism for genetic studies because of its excellent viability, high mutability, and certain peculiarities in its cytological and genetic behavior. With the progress of research, it has become clear that it is unique among the species in the genus *Drosophila* (Singh and Chatterjee, 1985; Singh, 2010; Sisoda and Singh, 2006, 2009). *Drosophila* has emerged as one of the most powerful models for human diseases and toxicological research (Chowdhuri *et al*., 2005). VAL can affect each life stage in a different manner and may depend on a specific stage of life history traits. Pharmacological tools have introduced behavioral alterations in *Drosophila* reminiscent of human behavior. The fly can effectively be used for low- to high-throughput drug screens (Pandey and Nichols, 1982).