Racial divergence in body weight: A study in the four members of newly evolved nasuta-albomicans complex of *Drosophila*

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In the laboratory, we have generated various Cytoraces as the newly evolved products of interracial hybridization between Drosophila nasuta nasuta and Drosophila nasuta albomicans. These races showed divergence from their parents in many features, including reduced body size and increased fertility. Here we present data on racial divergence in body weight in two of the hybrid Cytoraces and the two parental races. Analysis reveals that Cytorace 2 has the highest body weight and D. n. nasuta has the lowest body weight. The results suggest that body size and body weight may not be correlated because Cytorace 2 has higher fitness, maximum weight but reduced body size.

THE process and pattern of speciation is an important event in understanding the evolutionary genetics of any organism. Speciation genetics is largely concentrated on populations or races that have been separated from each other only recently and have not yet attained the status of species. The more advanced the stage of speciation of two diverging populations, the more difficult it is to delineate the genetic events that have set the process into motion. Thus it will not be possible to understand the process of speciation by looking at the finished products. Drosophila nasuta nasuta (2n = 8) and Drosophila nasuta albomicans (2n = 6) are a pair of cross-fertile allopatric chromosomal races of the nasuta subgroup of Drosophila with little morphological differentiation. The genetics and cytogenetics of these two races have been extensively studied²⁻¹⁰. Interracial hybridization between these two races in the laboratory has resulted in the evolution of two new karyotypic strains called Cytorace 1 and Cytorace 2 (ref. 7). Cytorace 1 has 2n = 7 in males $(2^{n}2^{u} \times 3^{u} Y^{n} 3^{n} 4^{n} 4^{n})$ and 2n = 6 in females $(2^n 2^a \times 3^a \times 3^a 4^n 4^n)$. Both males and females of Cytorace 2 have $2n = 6 (2^n 2^u \times 3^u \times 3^u / Y 3^u 4^u 4^u)^7$. These Cytoraces along with their parental races constitute a new assemblage, the nasuta-albomicans complex of Drosophila8. These Cytoraces have also been referred to as members of a laboratory hybrid zone of Drosophila with allosympatric populations¹¹. Studies on cytogenetic differentiation^{7,8}, mating preference^{1,11}, body size¹², sternopleural bristles¹³, and certain parameters of fitness such as fecunEvolutionary biologists have long been fascinated by the diversity in *Drosophila* species ^{16,17}, in which genetic variation has been observed virtually for every quantitative trait examined in natural populations ^{18–20}. However, there are contradictory reports about the relationship between the body size and body weight in *Drosophila*. In view of this we report here a comparison of body weight and body size in the four races of the *nasuta–albomicans* complex of *Drosophila*.

Drosophila stocks used in the present experiments were -D. n. nasuta (Coorg, India), D. n. albomicans (Okinawa, Texas Collections, USA, 3045.11), Cytorace 1 (ref. 7), Cytorace 2 (ref. 7).

It took approximately 20 generations for each Cytorace to stabilize its karyotype and to breed true. At the time of the present experiment, each of these Cytoraces had passed through 340 generations since hybridization. These stocks were cultured in an uncrowded condition with standard wheat cream agar medium at 22 ± 1°C. Fifty virgin females and males were isolated soon after the emergence (±4 h) and placed in fresh food media vials supplemented with yeast grains. After 48 h of emergence, individual flies were etherized and total fresh body weight was weighed using Mettler Toledo balance. Then the same set of flies were reared for 6 more days in fresh food media vials with three successive transfers once in two days, and on the eighth day after emergence, individual flies were etherized and weighed again. The average mean body weight was calculated for all the four races of nasuta-albomicans complex of Drosophila for each sex and subjected to analysis of variance (ANOVA) followed by Duncan's Multiple Range Test (DMRT) for multiple comparisons. Also correlation was applied between body size and body weight traits using statistical presentation system software for MS Windows.

Body size is one of the central features of any organism - physiologically, ecologically and evolutionarily. It is correlated with many physiological, ecological and life history traits and can be used to characterize many evolutionary patterns²¹. It is often one of the major traits utilized in evolutionary studies²². Body size has been measured in a number of ways, namely wing length, thorax length, wing width, front leg segments length and/or face width in *Drosophila*^{12,23-25}, and brain size in mammals²⁶. A number of workers have demonstrated that temperature²⁷, competition and larval density²⁸ influence body weight in Drosophila. In the present study, we have made two assessments of body weight on 2-day-old and 8days-old adult flies (Tables 1 and 2). All the four races studied have shown significantly increased body weight in 8day-old flies than in 2-day-old flies which is consistent with previous observations on many Drosophila species

dity, egg to adult rate of development, egg to adult viability, adaptedness and ability to utilize different sugar resources 14.15 among the parental races and Cytorace 1 and Cytorace 2 have shown appreciable racial divergence.

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Table 1. Mean body weight (in mg) of two-day-old flies of four members of nasuta-albomicans complex of Drosophila (Values are means ± SE of 50 flies for each sex)

Race	Total	Male	Female
D. n. nasuta	1.04 ± 0.02^{1}	0.97 ± 0.03 ^a	1.12 ± 0.02°
D. n. albomicans	1.13 ± 0.02^2	1.03 ± 0.02^{b}	1.24 ± 0.02^{f}
Cytorace 1	1.12 ± 0.02^3	1.05 ± 0.02^{c}	1.20 ± 0.02^{g}
Cytorace 2	1.29 ± 0.01^4	1.12 ± 0.01^{d}	1.41 ± 0.02^{h}

Analysis of variance – For both males and females; F = 15.96; DF = 7; 392; P < 0.001, DMR test; Except comparison of the mean values between 2 and 3 all other comparisons are significant at 5% level.

For males, F = 4.982; DF = 3; 196; P < 0.002; DMR test, Except comparison of the mean values between a/b and b/c all other comparisons are significant at 5% level.

For females, F = 5.648; DF = 3; 196; P < 0.01; DMR test, Except comparison of the mean values between f/g all other comparisons are significant at 5% level.

Table 2. Mean body weight (in mg) of eight-day-old flies of four members of nasuta-albomicans complex of Drosophila (Values are means ± SE of 50 flies for each sex)

Race	Total	Male	Female
D. n. nasuta	1.24 ± 0.01^{I}	1.09 ± 0.01^{a}	1.39 ± 0.02^{c}
D. n. albomicans	1.33 ± 0.02^2	1.18 ± 0.01^{b}	$1.48 \pm 0.03^{\rm f}$
Cytorace 1	1.32 ± 0.01^3	$1.23 \pm 0.01^{\circ}$	1.40 ± 0.01^{8}
Cytorace 2	1.45 ± 0.01^4	1.33 ± 0.02^{d}	1.57 ± 0.01^{h}

Analysis of variance – For both males and females; F = 19.017; DF = 7; 392; P < 0.001, DMR test, Except comparison of the mean values between 1/3 and 2/3 all other comparisons are significant at 5% level.

For males, F = 7.445; DF = 3; 196; P < 0.001; DMR test, Except comparison of the mean values between a/b; b/c and c/d all other comparisons are significant at 5% level.

For females, F = 5.336; DF = 3; 196; P < 0.01; DMR test, Except comparison of the mean values between e/g and f/g all other comparisons are significant at 5% level.

(Figure 1). Females of all the four races were heavier than males at both 2 and 8 days after eclosion. D. n. nasuta has the lowest body weight and Cytorace 2 has the highest body weight, which is statistically significant. This also reveals that the Cytorace 2 has shown differences in body weight with D. n. nasuta whereas Cytorace 1 and D. n. albomicans are almost intermediate to them. Based on DMRT the order of ranking for the body weight in these four races is as follows:

For 2-day-old flies: Males – Cytorace 2 > Cytorace 1 = D. n. albomicans = D. n. nasuta; Females – Cytorace 2 > D. n. albomicans = Cytorace <math>1 > D. n. nasuta.

For 8-day-old flies: Males – Cytorace 2 >Cytorace 1 = D. n. albomicans > D. n. nasuta; Females – Cytorace 2 > D. n. albomicans =Cytorace 1 > D. n. nasuta. (> indicates significant at 5% level and = indicates statistically insignificant at 5% level).

We have measured the body size of the four races under study by using the mean wing length, wing width, and front leg length¹². We have reported that the parental races D. n. albomicans and D. n. nasuta have greater body size than the Cytoraces. The order of ranking of body size in these four races for both males and females is the same and as follows: D. n. albomicans > D. n. nasuta > or = Cytorace 2 > Cytorace 1.

DMRT also reveals that D. n. nasuta and Cytorace 2 have almost identical body size. In the present study, D. n. nasuta has the lowest mean values and Cytorace 2 has the highest mean values for body weight. When body size and body weight are compared in these four races (1) D. n. nasuta has greater body size but lowest body weight than the Cytoraces; (2) D. n. albomicans has increased body size than all other races with lower body weight than

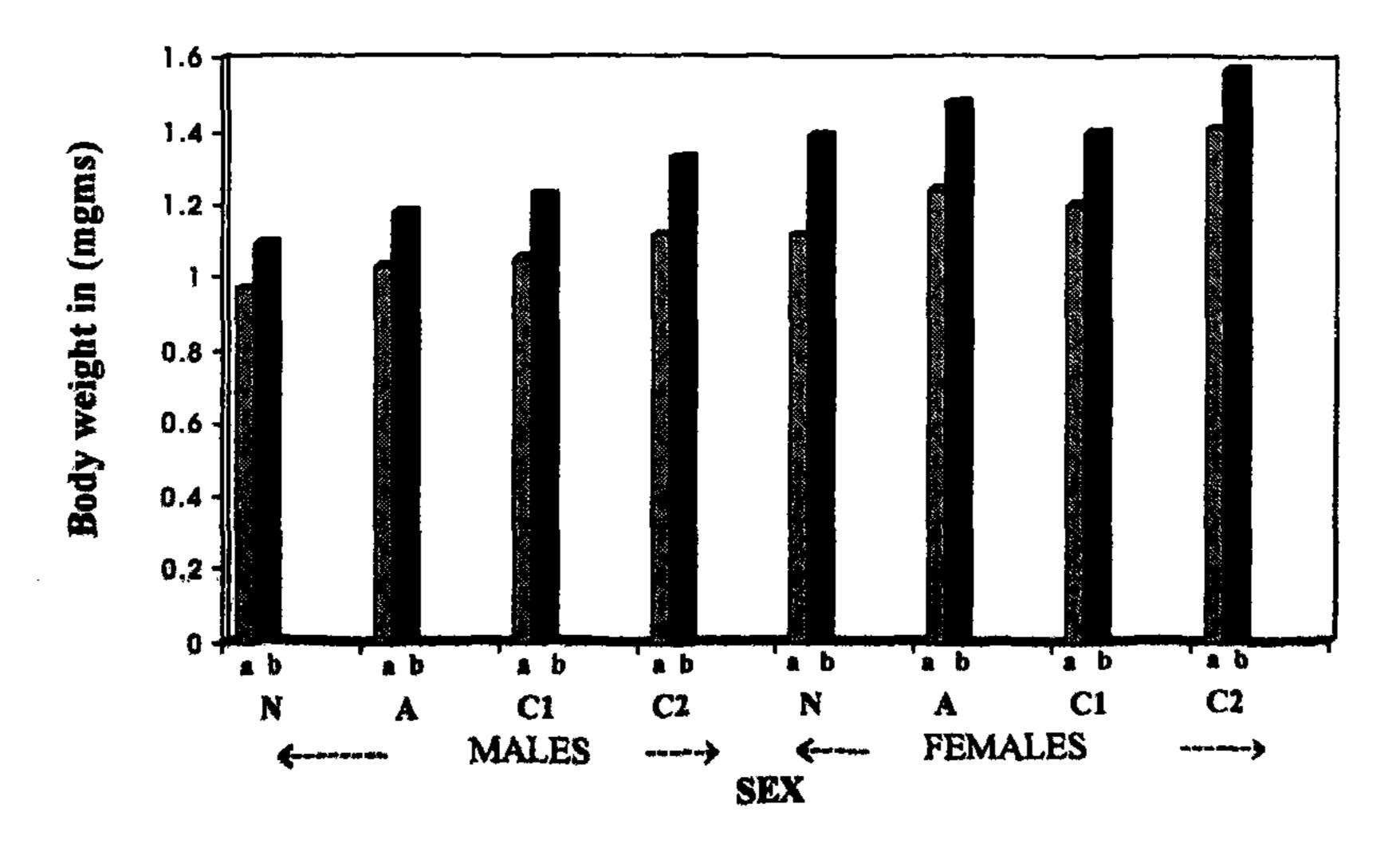


Figure 1. Body weight of 2-day-old and 8-day-old flies of four members of the nasuta-albomicans complex of Drosophila. N. Nasuta; A. Albomicans, Cl. Cytorace 1; C2, Cytorace 2; a, 2-day-old flies; b, 8-day-old flies.

Table 3. Correlation between body size and body weight of four members of nasuta-albomicans complex of Drosophila

Body size	Body weight				
	D. n. nasuta	D, n, albomicans	Cytorace I	Cytorace 2	
D. n. nasuta	- 0.224	~ 0.142	- 0.021	- 0.109	
D. n. albomicans	0.030	~ 0.091	-0.226	- 0.166	
Cytorace 1	0.054	0.053	-0.184	-0.144	
Cytorace 2	0.085	~ 0.210	-0.176	- 0.025	

Body size and body weight are negatively correlated at 5% level.

Data pertaining to the body size of these races is reported by us in Genetica 1999, 105, 1-6. The mean values of the wing length, an index of the body size of D. n. nasuta, D. n. albomicans, Cytorace 1, and Cytorace 2 are 242.83 ± 1.55 , 248.54 ± 1.73 , 233.09 ± 1.86 , and 243.88 ± 1.77 , respectively.

Cytoraces; (3) Cytorace I has smaller body size than all other races with increased body weight than D. n. nasuta; (4) Cytorace 2 has reduced body size than parental races with maximum body weight than all other races. Table 3 clearly suggests that the body size and body weight traits are not correlated. We have also recorded the higher fertility and ovarioles number in Cytorace 2 than other races. It appears that the greater body weight of Cytorace 2 could be due to better fertility and ovarioles number 12. D. n. nasuta has lesser sternopleural bristles¹³ and reduced body weight than the other races. Hence these two traits could be correlated. Thus this study presents the subtle morphophenotypic divergence among the parental races and the newly evolved Cytoraces. It also suggests that the body size and body weight are two different traits and are not correlated.

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ACKNOWLEDGEMENTS. We thank Prof. H. A. Ranganath, Droso-phila Stock Centre, Department of Studies in Zoology, for his encouragement, and the Chairman of our Department for providing facilities. B.P.H. is grateful to University of Mysore for awarding research fellowship. We thank Dr Lancy D'Souza for statistical analysis.

Received 6 August 1999; revised accepted 12 November 1999