## Racial divergence in sternopleural bristles among the parental races and the newly evolved Cytoraces 1 and 2 of the nasuta-albomicans complex of Drosophila

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Interracial hybridization between a pair of cross fertile races, Drosophila nasuta (2n=8) and Drosophila albomicans (2n=6) has resulted in the evolution of new karyotypic strains, called Cytoraces, under laboratory conditions. These Cytoraces have inherited chromosomes from both the parents. Here we report the racial divergence in sternopleural bristles among the parental races and the newly evolved Cytorace 1 and 2. This analysis has revealed that the parental race, D. nasuta has lesser number of bristles than D. albomicans, Cytorace 1, and Cytorace 2. These newly evolved Cytorace 1 and Cytorace 2, with more number of sternopleural bristles, show better fitness.

The nasuta subgroup of Drosophila has certain evolutionary peculiarities which include little morphological differentiation, the ability to intercross in the laboratory, often producing fertile offspring, and the substantial chromosomal evolution. These features make this subgroup as one of the potent systems to study the genetics of speciation in Drosophila. D. nasuta (2n = 8) and D. albomicans (2n = 6) are a pair of sibling allopatric chromosomal races of the nasuta subgroup of Drosophila which are morphologically indistinguishable<sup>2-4</sup>.

The cytological distinctness of these two races has been extensively studied<sup>2-10</sup>. Interracial hybridization between D. nasuta and D. albomicans followed by the maintenance of hybrid populations for over 20 generations has resulted in the emergence of two new karyotypic strains; Cytorace 1 and Cytorace 2 (ref. 7). The Cytorace 1 is the product of interracial hybridization between the males of D. nasuta and the females of D. albomicans. It has 2n = 7 in males  $(2^n2^a \times 3^a \times 1^n \cdot 3^n \cdot 4^n \cdot 4^n)$  and 2n = 6 in females  $(2^n2^a \times 3^a \times 3^a \cdot 4^n \cdot 4^n)$  respectively (n = D. nasuta; a = D. albomicans). While the Cytorace 2 is the outcome of interracial hybridization between the females of D. nasuta and the males of D. albomicans. Both the males and females of Cytorace 2 have  $2n = 6 (2^n2^a \times 3^a \times 3^a / 3^$ 

These newly created cytoraces along with their parental races constitute a new assemblage, the nasuta-albomicans complex of Drosophila<sup>8</sup>. These cytoraces are also called

as the members of hybrid zone of *Drosophila* with 'allo-sympatric populations under laboratory conditions'<sup>11</sup>. Earlier studies on cytogenetic differentiation<sup>7,8</sup>, mating preference<sup>12</sup>, body size<sup>13</sup> and certain parameters of fitness<sup>14,15</sup> among the parental races (*D. nasuta*, *D. albomicans*) and the cytoraces (Cytorace 1 and Cytorace 2) have shown appreciable racial divergence.

Sternopleural bristle number in natural and laboratory populations of *Drosophila* has been observed as a quantitative trait<sup>16-22</sup>. A number of studies have demonstrated the association of sternopleural bristle with several components of fitness, which indicates that sternopleural bristle number is an adaptive trait<sup>17-21</sup>. In view of this, we have made an attempt to record the racial divergence in the evolving cytoraces, on the basis of their sternopleural bristle numbers.

## Materials and methods

The *Drosophila* stocks used in the present experiments were (i) *D. nasuta* (Coorg, India); (ii) *D. albomicans* (Okinawa, Texas collections, USA, 3045.11); (iii) Cytorace  $1^{(7)}$  and (iv) Cytorace  $2^{(7)}$ .

It took approximately 20 generations for each Cytorace to stablize its karyotype and to breed true. At the time of the present experiment, each of these cytoraces were passing through 300 generations. These stocks were cultured in standard wheat cream agar medium in an uncrowded condition at  $22 \pm 1$ °C. The adult males and females of all the above-mentioned four stocks were etherized and their sternopleural bristles recorded under stereomicroscope. The sternopleural bristles are present on the lateral side of the flies on a triangular-shaped sternum. The number of sternopleural bristles in 150 flies of both the males and females was recorded separately for all the four stocks. The mean values of the number of bristles of both the males and females of all the four stocks were subjected for analysis of variance and Duncan multiple range test to ascertain the differences among them. The program used was statistical presentation system software for MS windows.

## Result and discussion

Kitagawa et al.22 have studied variations in a number of morphophenotypic traits in different natural populations of 10 members of D. nasuta subgroup of Drosophila. While the number of sternopleural bristles in various natural populations of D. nasuta males and females ranges from 8.20 to 10.36 and 8.52 to 13.52, respectively, the sternopleural bristles in different natural populations of D. albomicans males and females varies from 10.4 to 11.76 and 11.04 to 13.40, respectively. In the present study we used D. nasuta and D. albomicans, which have been bred in the laboratory for more than 20 years. While the males and females of laboratory-bred D. nasuta have 11.65 and 11.82 sternopleural bristle numbers respectively, the males and females of laboratory-bred D. albomicans have 12.91 and 13.40 sternopleural bristle numbers, respectively (Table 1). The analysis revealed that D. nasuta has significantly lesser number of sternopleural bristles than D. albomicans. The females of D. nasuta and D. albomicans have more number of sternopleural bristles than the males, which is in agreement with the Kitagawa et al. 22 observations.

A number of earlier studies have demonstrated that while D. nasuta is the ancestral form having 2n = 8, D. albomicans (2n = 6) is derived from D. nasuta karyotype<sup>10,23</sup>. Based on the karyotypic phylogeny, the ancestral member, D. nasuta, has lesser number of sternopleural bristles than the derived member, D. albomicans, suggesting that the derived member acquired more number of sternopleural bristles during the course of its evolution.

The comparative analysis of the sternopleural bristles in all the four races of the present study revealed the following (Figure 1 and Table 1):

(i) In D. nasuta, both the males and females have lesser number of sternopleural bristles than D. albomicans as well as the Cytorace 1 and Cytorace 2.

Table 1. Mean sternopleural bristle number of four members of nasuta-albamicans complex of Drosophila (values are means ± SE of 150 flies in each sex)

Races	Total	Males	Females	
Drosophila nasuta	$11.74^{1} \pm 0.11$ $13.16^{2} \pm 0.12$	11.65 ± 0.11"	11.82 ± 0.10°	
D. albomicans		12.91 ± 0.12"	13.40 ± 0.13′	
Cytorace 1 Cytorace 2	$12.89^3 \pm 0.12$	$12.70 \pm 0.12^{\circ}$	13.08 ± 0.11*	
	$13.35^4 \pm 0.11$	$13.38 \pm 0.10^{\circ}$	13.33 ± 0.11*	

Analysis of variance:

For both males and females: F = 80.14; DF = 7;1192; P < 0.0000.

DMR test: Except the comparison of the mean values between 2/4, all other comparisons are significant at 5% level.

For males: F = 40.18; DF = 3,596; P < 0.0000.

DMRT test: Except the comparison of the mean values between b/c, all other comparisons are significant at 5% level.

For females: F = 42.24; DF = 3,596; P < 0.0000.

DMRT test: Except the comparison of the mean values between g/h, f/g, f/h all other comparisons are significant at 5% level.

- (ii) The differences in sternopleural bristle number between the males of *D. albomicans* and Cytorace 2 as well as between the males of Cytorace 1 and Cytorace 2 are significant. All other comparisons are insignificant. (iii) In all the four races, the females have higher number of sternopleural bristles than the males.
- (iv) Further, more divergence in the number of sternopleural bristles is seen among the males of these four races.
- (v) The ranking of sternopleural bristle number in these four races are: males: Cytorace 2 > D. albomicans = Cytorace 1 > D. nasuta; females: D. albomicans = Cytorace 2 = Cytorace 1 > D. nasuta; both males and females: Cytorace 2 = D. albomicans = Cytorace 1 > D. nasuta.

There are reports that females have higher bristle number than males, which has been correlated with body size, and that under high competitions flies are smaller with fewer bristles 16.22. Recently we have reported smaller body size of Cytorace 1 and Cytorace 2 than the parental race, D. nasuta and D. albomicans. In the present study, Cytorace 1 and Cytorace 2 show higher number of sternopleural bristles than D. nasuta, and almost the same number as that of D. albomicans. Hence the body size and bristle number are not correlated in the present study.

Association between sternopleural bristles and certain components of fitness have been demonstrated in D. melanogaster<sup>17,24</sup>. Singh and Mathew<sup>21,25</sup> have reported

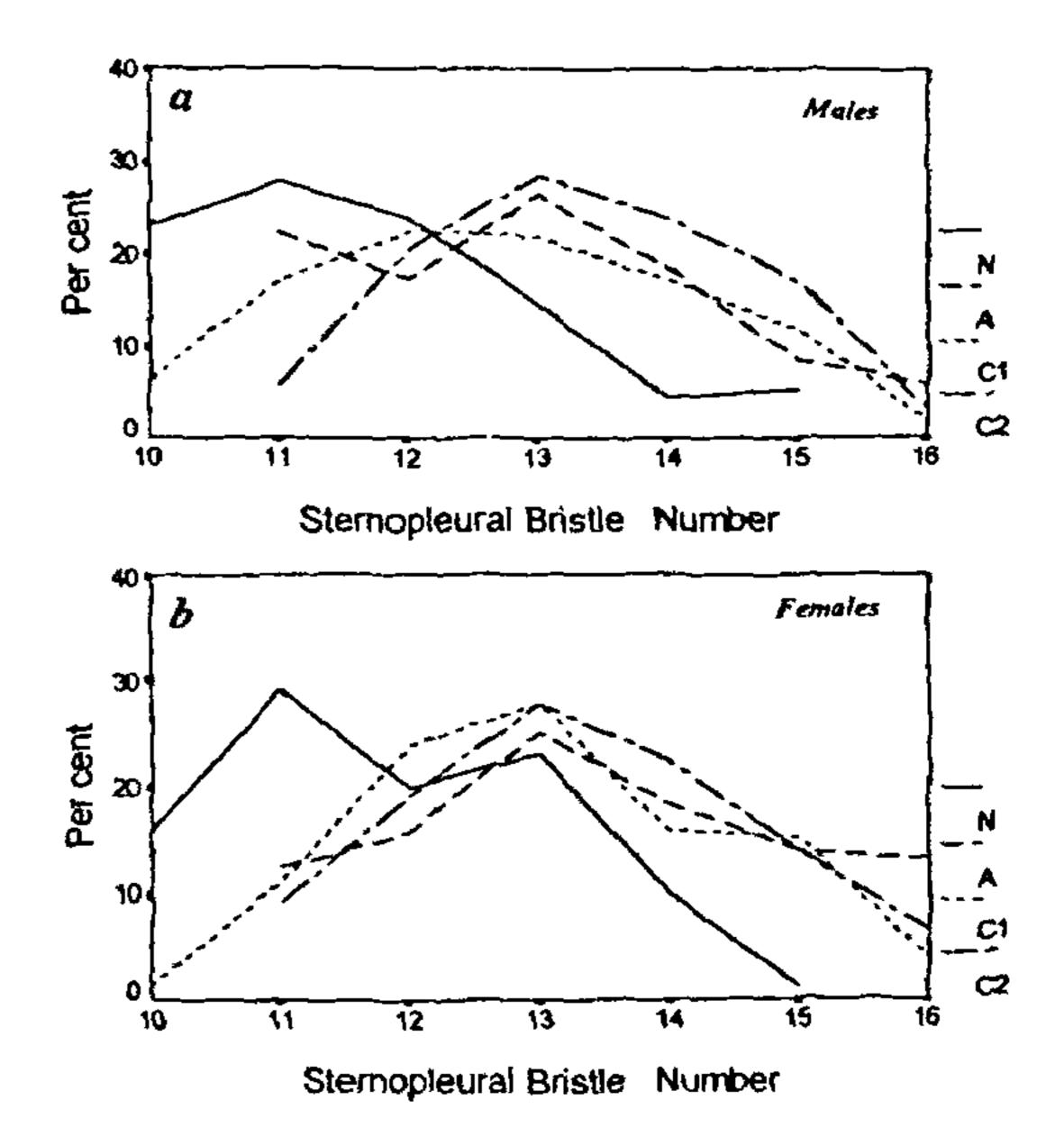


Figure 1. Pattern of sternopleural bristle distribution in D. nasuta, D, albomicans, Cytorace 1 and Cytorace 2 of nasuta-albomicans complex of Drosophila (a) males, (b) females (N = D. nasuta; A = D. albomicans; C = Cytorace 1; C = Cytorace 2).

greater mating success and fertility of *D. ananassae* flies possessing high number of sternopleurals. We have reported that Cytorace 1 and Cytorace 2 have greater fertility<sup>13</sup>, and faster rate of development<sup>14</sup> than *D. nasuta* and *D. albomicans*. This suggests that these cytoraces with more number of sternopleural bristles, show better fitness for certain parameters. Thus, the present study is an important parameter to quantify subtle morphophenotypic divergence among the parental races and the newly evolved/evolving cytoraces of the *nasuta-albomicans* complex of *Drosophila*.

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