

about two hundred years earlier. Obviously, no one, not even the Government cares for such valuable monuments of nature. If the authorities concerned had cared the trees could have been carefully dug out with the roots and transplanted. In other countries old cycads are valued and protected wherever they grow. The Chinese and Japanese plant cycads in their temples and some of their famous oldest and largest trees are situated there, e.g. the Ryugeji Temple near Tokyo has many old trees of *Cycas revoluta*. Surely we could emulate this practice in our temples and churches instead of decorating them with cut leaves and crowns.

### Suggested methods for conservation of Indian cycads

The manner in which India could embark on a programme for the protection of its cycads is envisaged as follows:

1. A survey of the natural areas and

population counts of all our cycads in an All-India basis along with a search for new forms.

2. Protection of their habitats by declaring some of their habitat areas as 'Cycad Reserves'.

3. A programme should be embarked for the education of the public in the areas of our cycads and also elsewhere on the need of protecting our cycads wherever they grow, and, if necessary, we should embark on legislation for the protection of cycads in nature and old plants in cultivation. We should have a Cycad Society, if necessary subsidized by the Government, for education and research on cycads like the cycad societies of other countries. This could help us in developing our much needed research on different aspects of cycads.

4. India should have a few cycad gardens and special cycad sections in all botanical gardens. Indeed India needs many more botanical gardens in different parts of the country to preserve the diversity of its flora.

We could encourage cultivation of cycads in parks, road sides and private gardens by providing a subsidy for the purpose.

In this connection it is important to point out that Indian scientists sitting in positions of power in Government Departments should take effective measures for the protection of our endangered species like *Cycas beddomei* and not merely write articles about their imminent extinction. Such statements should also be backed by actual surveys of their distribution and numbers and by the establishment of 'plant sanctuaries'. India has a number of 'National Parks' for the protection of animals facing extinction but not a single one for the protection of its endangered plants. Perhaps, I could also blame myself for this situation since I repeatedly refused positions in Government Departments.

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## SCIENTIFIC CORRESPONDENCE

### Evolution of the nasuta-albomicans complex of *Drosophila*

During the last two decades, the *nasuta* subgroup of the *immigrans* group of *Drosophila* has attracted the attention of taxonomists, cytogeneticists, biochemists, molecular biologists and evolutionary biologists. The *nasuta* subgroup has evolutionary peculiarities, which include little morphological differentiation despite their distribution over an enormous territory, the ability to intercross in the laboratory, often producing fertile offspring and substantial chromosomal evolution<sup>1</sup>. The story started with the report of *D. nasuta* by Lamb<sup>2</sup> from Seychelles islands. Subsequently, morphologically almost similar forms found in different parts of southeast Asia were studied by Wilson *et al.*<sup>3</sup>. Further investigations by Indian<sup>4-7</sup> and Japanese researchers<sup>8,9</sup> resulted in establishing the 14 different species of the *nasuta* subgroup. The females of all the species are morphologically indistinguishable, while three types of males are recognized<sup>3,8</sup>. On the basis of the morphology of the frons and orbits of the males, three morphophenotypic complexes have been identified<sup>3-5,8</sup>. The males of

*D. nasuta*, *D. albomicans*, *D. kepulauan*, *D. kohkoa* and *D. neveifrons* with complete silvery frons constitute 'frontal sheen complex'. On the other hand, the silvery sheen is restricted to the sides of the frontal orbits in the males of *D. s. sulfurigaster*, *D. s. bilimbata*, *D. s. albostrigata*, *D. s. neonasuta* and *D. pulaua*, constituting the 'orbital sheen complex'. The third complex shows absence of pollinosity of frontal region and includes *D. pallidifrons*, taxons F, J, and I.

In the frontal sheen complex, *D. nasuta* and *D. albomicans* are a pair of chromosomal allopatric races (earlier they were treated as biologically valid reproductively isolated species, and hence were given different names) with  $2n=8$  and  $2n=6$ , respectively<sup>10</sup>. The difference in the diploid number is because the sex chromosomes and the 3rd autosome exist as independent acrocentric entities in *D. nasuta*, whereas in *D. albomicans*, these two components of the karyotype exist as one unit in the form of a metacentric chromosome. In spite of the difference in diploid number, there is similarity

between the corresponding chromosomes of *D. nasuta* and *D. albomicans*, as seen in the polytene chromosomes of  $F_1$  hybrid larvae<sup>11</sup>. They also show variation not only in chromosome number, but also in the quantum of heterochromatin present in different chromosomes<sup>12</sup>, and in the organization of micro (dot) chromosomes<sup>13</sup>. Because of this difference, the size of the respective chromosomes of *D. nasuta* and *D. albomicans* differ, and this helps to distinguish the chromosomes of these races in their hybrids. In spite of such karyotypic differences, the  $F_1$  hybrids with  $2n=7$ , (4 chromosomes of *nasuta* and 3 of *albomicans*) are fertile and the hybrid progeny can therefore be maintained for generations. The ability to identify the parentage of each chromosome in the hybrid karyotypes and to indefinitely maintain the hybrid populations have formed the basis for long range evolutionary studies of the present authors<sup>11,14</sup>.

Hybrids of *D. nasuta* and *D. albomicans* have been maintained in our laboratory for over a decade. The  $F_1$  with  $2n=7$

breeds to yield  $F_2$  hybrid population, which is karyotypically heterogeneous. In addition to individuals with *nasuta*-like, *albomicans*-like, and  $F_1$ -like karyotypes, other combinations are also seen<sup>15</sup>. Such karyotypic mosaicism persists for many hybrid generations, while in some hybrid lineages, karyotypic variability disappears and the population has one type of stabilized karyotype. During the evolution of a stabilized karyotype, some of the parental chromosomes are eliminated, while others are retained. The time taken for a karyotype to stabilize ranges from 20 to over 150 generations and, in a few cases, heterogeneity is maintained without attaining karyotype stability (unpublished). Stabilized hybrid karyotypes are invariably a combination of chromosomes inherited from both the parents. Thus, a new karyotype, called cytorace was developed in our laboratory having chromosomes of both *nasuta* and *albomicans*. There are four such cytoraces<sup>16,17</sup>. The males and females of these four cytoraces along with *D. nasuta* and *D. albomicans* were intercrossed and they yielded 30 new hybrid lines, which are being maintained (unpublished). Periodically, the karyotypes of these hybrid populations passing through different hybrid generations are being analysed along with other features such as mating preference<sup>18</sup>, population fitness<sup>19,20</sup>, ecogenetic difference<sup>21</sup>, if any, among different hybrid lineages.

At present, as a result of interracial hybridization experiments for over a decade, we have created 16 different races derived from intermixing the genomes of *D. nasuta* and *D. albomicans* (Table 1) (unpublished). Details of the role of different chromosomes in the evolution of these races will be discussed elsewhere. In brief, all these hybrid populations have chromosomes derived from *nasuta* and *albomicans*. Therefore, cytologically these races are closely related to one another. They differ from one another in the hybridization path through which they have evolved and are evolving. These genetically diverged cytologically parsimonious races, evolved under laboratory conditions, constitute a new assemblage within the 'frontal sheen complex' of *Drosophila*, called the *nasuta*-*albomicans* complex. This new complex stands parallel to the three naturally evolved morphophenotypic complexes of the *nasuta* subgroup mentioned earlier.

Chang *et al.*<sup>1</sup> analysed mitochondrial DNA of the members of the *nasuta* subgroup and suggested that most of the lineages of this subgroup diverged from each other between 1 and 2 Myr. On the contrary, the newly established *nasuta*-*albomicans* complex has evolved/diversified within a span of one decade in the laboratory. The members of the three naturally evolved complexes of the *nasuta* subgroup exhibit genetic divergence without morphological differentiation. In this regard, the laboratory evolved *nasuta*-*albomicans* assemblage is comparable to

these natural groups. The emergence of this new assemblage is through interracial hybridization between *D. nasuta* and *D. albomicans*, followed by hybrid recombination and karyotype stabilization. The event of hybridization has hastened the process of the formation of new differentiating populations and it justifies the opinion of Dobzhansky *et al.*<sup>22</sup>, that hybridization is an evolutionary catalyst. It might have taken millions of years in nature to evolve a group of such differentiating races, but it has taken just a decade in the present experimental setup.

**Table 1.** Summary of the karyotypes of cytoraces along with their parental races under laboratory conditions. The superscripts 'n' and 'a' indicate *nasuta* and *albomicans* respectively

Name	Karyotypes
<i>D. nasuta</i>	(♂ & ♀) - 2n = 8 - X <sup>n</sup> X <sup>n</sup> /Y <sup>n</sup> 2 <sup>n</sup> 2 <sup>n</sup> 3 <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> )
<i>D. albomicans</i>	(♂ & ♀) - 2n = 6 - X3 <sup>a</sup> X3 <sup>a</sup> /Y3 <sup>a</sup> 2 <sup>a</sup> 2 <sup>a</sup> 4 <sup>a</sup> 4 <sup>a</sup> )
Cytorace 1	(♂ - 2n = 7 - 2 <sup>n</sup> 2 <sup>n</sup> X3 <sup>a</sup> Y <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> ; and ♀ - 2n = 6 - 2 <sup>n</sup> 2 <sup>n</sup> X3 <sup>a</sup> X3 <sup>a</sup> 4 <sup>n</sup> 4 <sup>n</sup> )
Cytorace 2	(♂ - 2n = 6 - 2 <sup>n</sup> 2 <sup>a</sup> X3 <sup>a</sup> Y3 <sup>a</sup> 4 <sup>n</sup> 4 <sup>a</sup> ; and ♀ - 2n = 6 - 2 <sup>n</sup> 2 <sup>a</sup> X3 <sup>a</sup> X3 <sup>a</sup> 4 <sup>n</sup> 4 <sup>a</sup> )
Cytorace 3	(♂ - 2n = 8 - 2 <sup>n</sup> 2 <sup>n</sup> X <sup>n</sup> Y <sup>n</sup> 3 <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> ; and ♀ - 2n = 8 - 2 <sup>n</sup> 2 <sup>n</sup> X <sup>n</sup> X <sup>n</sup> 3 <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> )
Cytorace 4	(♂ - 2n = 7 - 2 <sup>n</sup> 2 <sup>a</sup> Y3 <sup>a</sup> X <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> ; and ♀ - 2n = 8 - 2 <sup>n</sup> 2 <sup>a</sup> X <sup>n</sup> X <sup>n</sup> 3 <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> )
Cytorace 5	(♂ - 2n = 7 - 2 <sup>n</sup> 2 <sup>a</sup> X3 <sup>a</sup> Y <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> ; and ♀ - 2n = 6 - 2 <sup>n</sup> 2 <sup>a</sup> X3 <sup>a</sup> X3 <sup>a</sup> 4 <sup>n</sup> 4 <sup>n</sup> )
Cytorace 6	(♂ - 2n = 7 - 2 <sup>n</sup> 2 <sup>a</sup> Y3 <sup>a</sup> X <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> ; and ♀ - 2n = 8 - 2 <sup>n</sup> 2 <sup>a</sup> X <sup>n</sup> X <sup>n</sup> 3 <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> )
Cytorace 7	(♂ - 2n = 7 - 2 <sup>n</sup> 2 <sup>a</sup> X3 <sup>a</sup> Y <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> ; and ♀ - 2n = 6 - 2 <sup>n</sup> 2 <sup>a</sup> X3 <sup>a</sup> X3 <sup>a</sup> 4 <sup>n</sup> 4 <sup>n</sup> )
Cytorace 8	(♂ - 2n = 7 - 2 <sup>n</sup> 2 <sup>a</sup> X3 <sup>a</sup> Y <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> ; and ♀ - 2n = 6 - 2 <sup>n</sup> 2 <sup>a</sup> X3 <sup>a</sup> X3 <sup>a</sup> 4 <sup>n</sup> 4 <sup>n</sup> )
Cytorace 9	(♂ - 2n = 6 - 2 <sup>n</sup> 2 <sup>a</sup> X3 <sup>a</sup> Y3 <sup>a</sup> 4 <sup>n</sup> 4 <sup>n</sup> ; and ♀ - 2n = 6 - 2 <sup>n</sup> 2 <sup>a</sup> X3 <sup>a</sup> X3 <sup>a</sup> 4 <sup>n</sup> 4 <sup>n</sup> )
Cytorace 10	(♂ - 2n = 8 - 2 <sup>n</sup> 2 <sup>a</sup> X <sup>n</sup> Y <sup>n</sup> 3 <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> ; and ♀ - 2n = 8 - 2 <sup>n</sup> 2 <sup>a</sup> X <sup>n</sup> X <sup>n</sup> 3 <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> )
Cytorace 11	(♂ - 2n = 6 - 2 <sup>n</sup> 2 <sup>a</sup> X3 <sup>a</sup> Y3 <sup>a</sup> 4 <sup>n</sup> 4 <sup>n</sup> ; and ♀ - 2n = 6 - 2 <sup>n</sup> 2 <sup>a</sup> X3 <sup>a</sup> X3 <sup>a</sup> 4 <sup>n</sup> 4 <sup>n</sup> )
Cytorace 12	(♂ - 2n = 6 - 2 <sup>n</sup> 2 <sup>a</sup> X3 <sup>a</sup> Y3 <sup>a</sup> 4 <sup>n</sup> 4 <sup>n</sup> ; and ♀ - 2n = 6 - 2 <sup>n</sup> 2 <sup>a</sup> X3 <sup>a</sup> X3 <sup>a</sup> 4 <sup>n</sup> 4 <sup>n</sup> )
Cytorace 13	(♂ - 2n = 6 - 2 <sup>n</sup> 2 <sup>a</sup> X3 <sup>a</sup> Y3 <sup>a</sup> 4 <sup>n</sup> 4 <sup>n</sup> ; and ♀ - 2n = 6 - 2 <sup>n</sup> 2 <sup>a</sup> X3 <sup>a</sup> X3 <sup>a</sup> 4 <sup>n</sup> 4 <sup>n</sup> )
Cytorace 14	(♂ - 2n = 7 - 2 <sup>n</sup> 2 <sup>a</sup> Y3 <sup>a</sup> X <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> ; and ♀ - 2n = 8 - 2 <sup>n</sup> 2 <sup>a</sup> X <sup>n</sup> X <sup>n</sup> 3 <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> )
Cytorace 15	(♂ - 2n = 8 - 2 <sup>n</sup> 2 <sup>a</sup> X <sup>n</sup> Y <sup>n</sup> 3 <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> ; and ♀ - 2n = 8 - 2 <sup>n</sup> 2 <sup>a</sup> X <sup>n</sup> X <sup>n</sup> 3 <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> )
Cytorace 16	(♂ - 2n = 8 - 2 <sup>n</sup> 2 <sup>a</sup> X <sup>n</sup> Y <sup>n</sup> 3 <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> ; and ♀ - 2n = 8 - 2 <sup>n</sup> 2 <sup>a</sup> X <sup>n</sup> X <sup>n</sup> 3 <sup>n</sup> 3 <sup>n</sup> 4 <sup>n</sup> 4 <sup>n</sup> )

Races with 2n = 7 in males show a few sterile aneuploids. Even though some of the races appear to have similar chromosomal complement, the hybridization path and the phylogenetic history of chromosomes differ.

The present investigations can be considered as an evolutionary experimentation through hybridization under laboratory conditions. These races which are at different stages of evolutionary divergence offer a rare opportunity to study the process of riation (speciation?), under laboratory conditions. The *nasuta-albomicans* complex of *Drosophila*, the members of which are in the process of evolving, is a unique model system to witness the process and the pattern of sibling riation/speciation as well as the analysis of chromosomal and molecular basis of riation.

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## Evidence for positive assortative mating within *Drosophila bipectinata*

Sexual (or ethological) isolation, which is a premating barrier to gene exchange between Mendelian populations, is the most important class among the different ways of reproductive isolation in animal species. This plays an important role in evolution. The phenomenon of sexual isolation has been extensively studied in the genus *Drosophila* and has been found to be widespread<sup>1,2</sup>. Incipient reproductive isolation occurring between geographic strains of the same species has been reported in many *Drosophila* species, which corroborates the hypothesis that incipient isolation originates as a side-effect of genetic divergence<sup>1-3</sup>. Although a lack of assortative mating between mutant and wild type *D. melanogaster* has been reported<sup>4-6</sup>, selective mating has also been found in some cases<sup>7,8</sup>. Rendel<sup>9</sup> found selective mating (non-random) between yellow mutant and wild type with yellow males.

*Drosophila bipectinata* is a member of the *bipectinata* species complex of the

*ananassae* subgroup of the *melanogaster* species group. Population and behaviour genetical studies in this species have been initiated in our laboratory<sup>10-14</sup>. *D. bipectinata* shows incomplete sexual isolation with other members of the *bipectinata* complex<sup>15</sup>. It is also characterized by incipient sexual isolation between different geographic strains<sup>11</sup>. Spontaneous mutations have been detected in this species<sup>16-18</sup>. Effects of mutations on mating propensity and pattern of mating have also been tested<sup>19-21</sup>. We detected flies with bilateral outgrowths on thorax which is a unique phenotypic change in *D. bipectinata*<sup>17</sup>. A separate stock of mutant with outgrowths could be established. During the present study, we tested the pattern of mating between wild type and mutant (possessing bilateral thoracic outgrowths) *D. bipectinata* and the results are reported here.

During the present study, the wild type (TD) and mutant (OG) stocks of *D. bipectinata* were used. In mutant stock, flies

possess bilateral outgrowths on thorax. Originally, this unique phenotypic change was detected in sepia eye colour mutant stock<sup>17</sup>. By making cross between sepia mutant with outgrowths and wild type, flies with red eye and outgrowths were obtained and a separate stock of red-eyed flies with outgrowths was established, which was used in mating propensity tests.

In mating propensity tests, multiple-choice method was used and mating success was studied by direct observation in an Elens-Wattiaux mating chamber kept in a room maintained at approximately 24°C under normal laboratory light conditions between 7 and 11 AM. Virgin females and males were collected from both the stocks and flies were aged for seven days. In multiple-choice experiment, 15 flies of each sex were used and five trials were run. Fifteen females of each of the two stocks were introduced into the mating chamber with 15 males of each of the two strains and were