## Differences in the First Progeny of *Drosophila melanogaster* Strains in Interspecific Competition

A strong degree of frequency-dependence in progeny production[1,2] in mixed cultures of 2 species of *Drosophila* was verified by one generation tests[1]. A decrease or maintenance of progeny number in cultures with high and low parental density is also observed[1]. We attempted to investigate, in mixed cultures of two species, whether the increase in parental density of one species inhibits the first progeny of the other competitor species maintained with constant parental frequency.

We used 15 wild strains of *D. melanogaster*, 12 homozygous and 3 heterozygous for the 2nd chromosome by means of CyL/Pm technique. From each strain 3 replicates were made with 200 flies each and introduced into bottles of $^1/_4$ l, with fresh medium. These 3 replicates received respectively 200, 400, and 600 flies, *D. pseudoobscura*.

In this way, three different sets of populations were established, with species initial frequencies of 1:1, 1:2, and 1:3. The populations were kept at 25 °C and, after 7 days of egg-laying, the parental flies were discarded. 14 days later the progeny counts were made only once. The results are summarized in the Table. Comparisons by means of $\chi^2$ test between outcomes showed in the Table permit us to arrange the 15 strains of *D. melanogaster* in 5 groups with decrease of the first progeny: 1. in direction 1:1, 1:2, 1:3, strain M6; 2. in direction 1:3, 1:2, 1:1, strains M12 and M24; 3. in direction 1:1, 1:3, 1:2, strains M7, M8, M10, M15, M17, M27, POL 1, and POL 3; 4. in direction 1:3, 1:1, 1:2, strains M18 and M32; and 5. the strains M19 and POL 2, without statistically significative differences between the populations 1:1, 1:2, and 1:3.

Number, percentage, and values and $P$ (with 2df) of $\chi^2$-test of the first progeny of *Drosophila melanogaster* for the 3 types of populations

| Strains | 1:1 | | 1:2 | | 1:3 | | Values of $\chi^2$ and $P$ |
|---|---|---|---|---|---|---|---|
| | *n* | % | *n* | % | *n* | % | |
| M 6 | 18 | 86 | 2 | 9 | 1 | 5 | 26.01** |
| M 7 | 203 | 60 | 34 | 10 | 100 | 30 | 129.16** |
| M 8 | 239 | 66 | 28 | 8 | 97 | 26 | 190.79** |
| M 10 | 166 | 70 | 20 | 8 | 52 | 22 | 148.48** |
| M12 | 6 | 3 | 34 | 16 | 175 | 81 | 228.97** |
| M 15 | 202 | 88 | 4 | 2 | 23 | 10 | 312.70** |
| M 17 | 380 | 62 | 26 | 4 | 207 | 34 | 306.72** |
| M 18 | 121 | 42 | 26 | 9 | 142 | 49 | 79.32** |
| M 19 | 104 | 36 | 101 | 34 | 88 | 30 | 1.48 |
| M 24 | 27 | 23 | 40 | 33 | 53 | 44 | 8.46* |
| M 27 | 253 | 52 | 64 | 13 | 172 | 35 | 110.32** |
| M 32 | 24 | 11 | 3 | 1 | 194 | 88 | 297.82** |
| Mean | 145 | ±34 | 32 | ± 8 | 109 | ±20 | |
| POL 1 | 258 | 72 | 49 | 14 | 51 | 14 | 241.72** |
| POL 2 | 73 | 36 | 67 | 33 | 61 | 31 | 1.08 |
| POL 3 | 184 | 60 | 26 | 8 | 98 | 32 | 121.89** |
| Mean | 172 | ±54 | 47 | ±12 | 70 | ±14 | |

*, $P < 0.05$; **, $P < 0.01$.

In the 2nd group strains, the greater the parental density of *D. pseudoobscura* the greater is the first progeny of *D. melanogaster*, as was observed[3] in *D. pseudoobscura*. i.e., the carriers of the 3rd chromosome gene arrangement standard (ST) are superior in fitness over the carriers of the 3rd chromosome inversion Chiricahua (CH) in cultures with high larval density, and the opposite relation occurs in cultures with low larval density. In this sense, the larval competition between *D. melanogaster* and *D. pseudoobscura*, can be increased by extended egg-laying period, because these 2 species lay more eggs in longer oviposition periods[4].

Comparing the mean productivity of *D. melanogaster*, first progeny of the homozygous wild strains, by means of $t$-tests, the following relations were found: 1:1 ≠ 1:2 ($t$ = 3,225; 22 df; $P < 0.01$), 1:1 = 1:3 ($t$ = 0,897; 22 df; $P > 0.05$), and 1:2 ≠ 1:3 ($t$ = 3,424; 22 df; $P < 0.01$). Moreover, for the heterozygous no differences were found. As is suggested by the results observed, homozygous and heterozygous for 2nd chromosome of *D. melanogaster* exhibit different outcomes in the production of the first progeny in interspecific competitive conditions.

*Resumen.* El resultado de la producción de la primera progenie de *D. melanogaster* fué diferiente para los tres tipos de poblaciones de competencia com *D. pseudoobscura* y también para las estirpes homo y heterocigotas de *D. melanogaster*.

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[1] P. W. Hedrick, Drosoph. Inf. Serv. *48*, 128 (1972).
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[3] L. D. Birch, Evolution *9*, 389 (1955).
[4] F. J. Ayala, Ecology *48*, 67 (1967).
[5] I am indebted to Dr. C. Abbade Mourão for his guidance and to Dr. C. Daghlian for helping with the manuscript.

## A Chromosome Mutation Affecting the Number of Nucleoli in *Xenopus borealis* Parker[1–3]

When crossed with a male of the same species, *Xenopus borealis* ♀ produces offspring whose number of nucleoli (*nu*) per cell is 1 or 2: the proportion of 2 *nu* nuclei varies from one embryo to another between 46 and 76%. It is interesting to report here the exceptional case of a couple of *X. borealis* from Lake Samburu area (Northern Kenya).

Embryos, squashed in toto at stages 10–11 and later at stage 46 (Nieuwkoop and Faber[4]) were examined by phase contrast microscopy. In some there were 1 or 2 nucleoli per nucleus, in others 1, 2 or 3: out of 53 embryos, 25 (47%) belonged to the first category, 28 (53%) to the second one. These proportions suggested a Mendelian distribution of this anomaly.

[1] Supported by a grant (No. 3.60.68) from the Fonds national suisse de la Recherche Scientifique and the George and Antoine Claraz donation.
[2] Unpublished research in this laboratory has shown that *Xenopus laevis borealis* (Parker) is a species on its own right and we propose to name it *Xenopus borealis* (Parker).
[3] Material collected by Fischberg and Kobel, who express their gratitude to the Fisheries and Game Dept. of Kenya for permission to collect frogs of the genus *Xenopus*.
[4] P. D. Nieuwkoop and J. Faber, *Normal Table of Xenopus laevis (Daudin)* (North Holland Publishing Company, Amsterdam 1956).

To test this hypothesis we squashed tailtips of 198 tadpoles at stages 47 and 48: 89 embryos (45%) presented 2 *nu* at most, 109 (55%) up to 3 nucleoli par cell. In the first group (2 *nu*) we found 17 to 49% 1 *nu* and 51 to 83% 2 *nu* cells; in the second one, 4–32% 1 *nu*, 29–72% 2 *nu* and 14–57% 3 *nu* cells. Embryos with less than 14% 3 *nu* nuclei were discarded.

ELSDALE, FISCHBERG and SMITH[5] in 1958, FISCHBERG and WALLACE[6] in 1960 have shown that in *X. l. laevis* (Daudin), the number of secondary constrictions per metaphase plate is identical to the maximal number of nucleoli during the interphase; they observe 2 chromosomes with terminal satellites in the normal *X. l. laevis* whereas the 1 *nu* mutant (Oxford mutant) presents only one of them. This relationship becomes quite clear in the light of biomolecular data, BIRNSTIEL et al.[7] having demonstrated that the secondary constrictions contain the ribosomal cistrons coding for the cytoplasmic r-RNA;

In *X. borealis* caryotype TYMOWSKA and FISCHBERG (in press) have shown 2 types of secondary constrictions borne by chromosomes 4 and 5 respectively. The same situation occurs in *X. mulleri* whose caryotype is nearly identical to the *borealis* one (TYMOWSKA and KOBEL[8]). If nucleic acid hybridizations have made it possible (PARDUE, in press) to demonstrate that, in *X. mulleri*, the r-DNA is localized on the terminal constriction of chromosome 4, this property ought to be valuable in the case of *X. borealis*.

The frequency of the 3 nucleoli mutation (nearly 50%) suggests that one of the parents possesses a supernumerary nucleolar organizer in the heterozygous state. To test this hypothesis, we crossed them with other frogs of the same population: despite many sollicitations with gonadotrophins, the mother (♀ 1) has so far obstinently refused to lay. The male parent (♂ 1) was the father of 1900 new embryos, 100 of which were fixed between stage 10 and 46. Except 4 ill-formed larvae with some 3 *nu* cells, they all presented 1 to 2 nucleoli per nucleus (normal number). Thus the origin of the mutation is not paternal but maternal (Figures 1 and 2).

*Cytological analysis. X. borealis* with 2 *nu*: 2 $n = 36$: the chromosomes can be distributed into 8 groups according to the classification proposed by TYMOWSKA (in press) for this species.

*X. borealis* with 3 *nu*: 2 $n = 36$: except the pairs 4, 5 and 7, the chromosomes do not differ from the homologous pairs of 2 *nu* individuals. Chromosomes of pair 4 are easily identified, although the terminal nucleolar organizer may be difficult to reveal. The association of the homologues of this pair observed in 50% of the 2 *nu borealis* metaphases is present in only 6% of the 3 *nu* metaphase plates analyzed.

To form pairs 5 and 7, there are 4 chromosomes, 1 submetacentric and 3 metacentrics bearing an intercalary secondary constriction. In order to characterize them precisely, we analyzed 22 metaphases from 2 ♂♂ with 2 *nu* and from 2 ♂♂ and 2 ♀♀ with 3 *nu*, and made the following measurements: 1. Centromeric Index (CI). 2. Length of the constriction in % of the lenght of the long arm which bears it. 3. Length of the chromatid segment between the centromere and the proximal end of the constriction, in % of the long arm's total length.

*Results* (figure 3). A) 2 *nu* individuals: 5a and 5b are the 2 elements with secondary constrictions; $7a^m$ (mutated) and 7b constitute pair 7. For these 2 pairs the CI values are 0.45 and 0.42 respectively. The constriction at 5a and 5b represents the 15.5% of the long arm's length and the distance separating the proximal extremity of the constriction from the centromere corresponds to 18.9% of the lenght of this same arm.

B) 3 *nu* individuals: 2 of the chromosomes with constrictions have a CI of 0.46, the constriction measuring 14.1% of the long arm and the centromereproximal end of the constriction distance 19.7% of the same. These numbers are very close to those found for 5a and 5b (individuals with 2 *nu*) and allow us to identify these 2

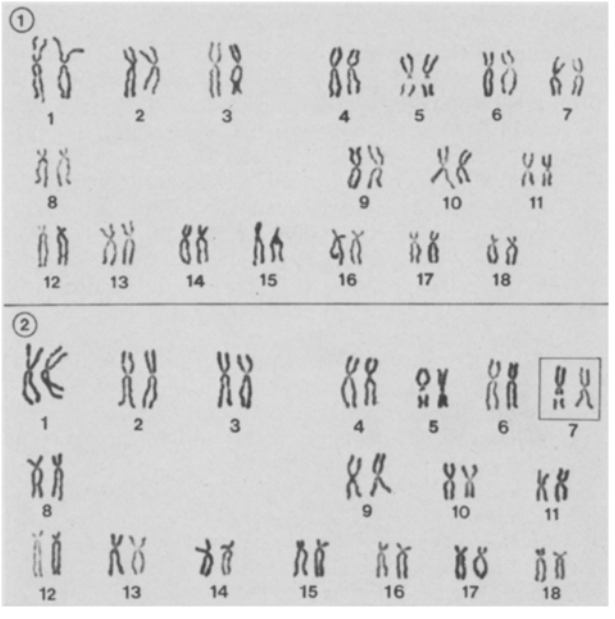


Fig. 1. *X. borealis* (2 *nu*): normal caryotype: 2 $n = 36$.
Fig. 2. *X. borealis* (3 *nu*): caryotype with mutation on 1 chromosome 7; 2 $n = 36$.

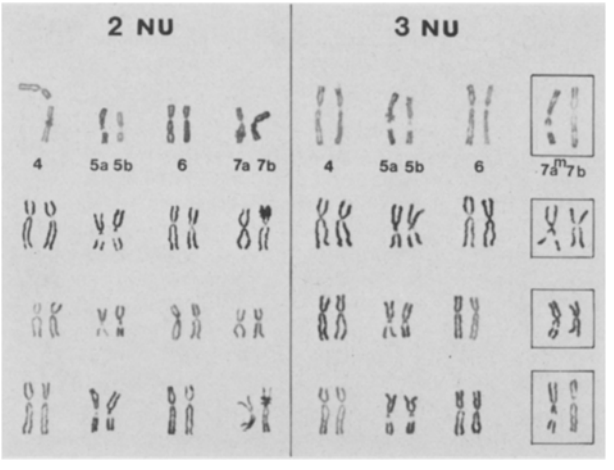


Fig. 3. 2 *nu X. borealis* (normal) and 3 *nu X. borealis* (mutated): comparison of chromosome pairs 4, 5, 6 and 7. 5a and 5b are the two homologues of pair 5.

[5] T. R. ELSDALE, M. FISCHBERG and S. SMITH, Expl. Cell Res. *14*, 642 (1958).
[6] M. FISCHBERG and H. WALLACE, in *The Cell Nucleus* (Ed. J. S. MITCHELL; Academic Press, New York 1960), p. 30.
[7] M. L. BIRNSTIEL, H. WALLACE, J. L. SIRLIN and M. FISCHBERG, Natn. Cancer Inst. Monogr. *23*, 431 (1966).
[8] J. TYMOWSKA and H. R. KOBEL, Cytogenetics *11*, 270 (1972).

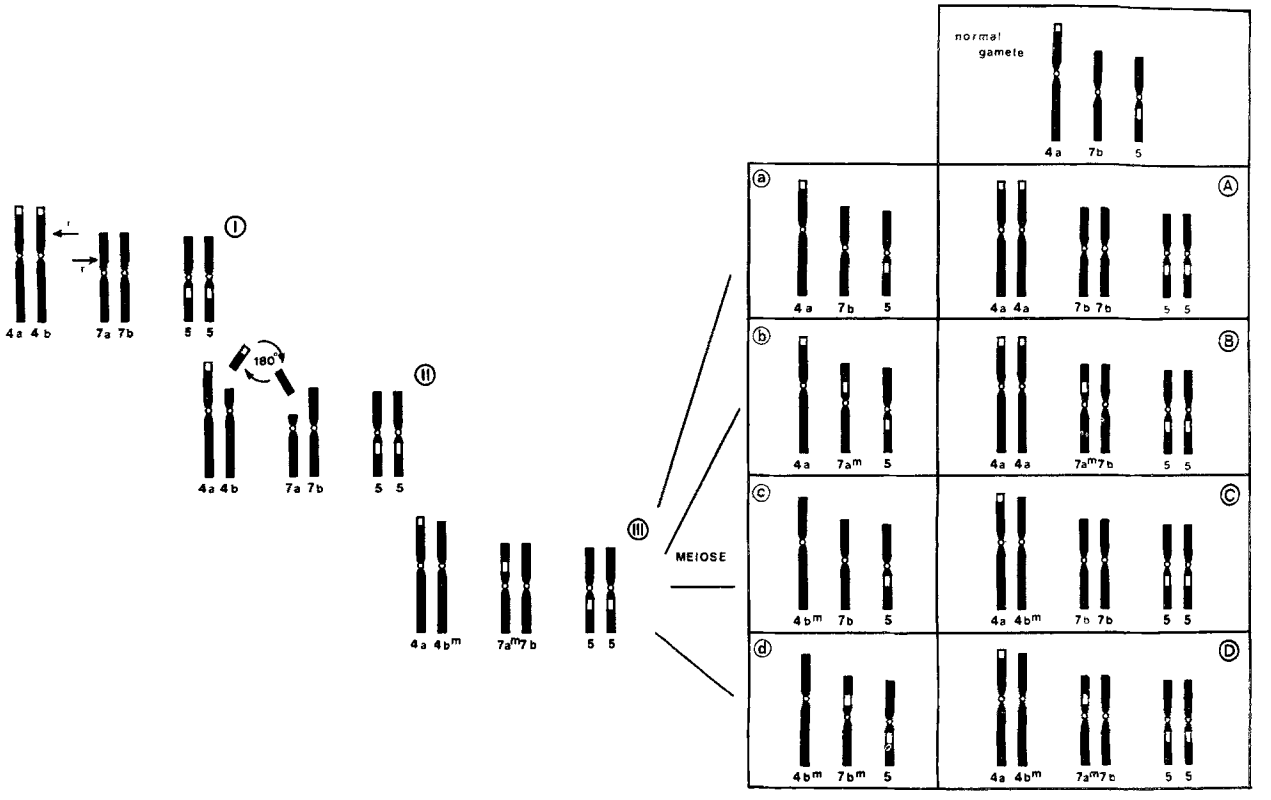


Fig. 4. Presumptive origin of the mutated $7a^m$ chromosome. 4a and 4b, 5 and 5, 7a and 7b: respective homologues of pairs 4, 5 and 7; $4b^m$ and $7a^m$: translocated 4b and 7a; a, b, c and d: the gametes of a type III individual; A, B, C and D: the 4 possible recombinants following fertilization of a, b, c or d by a normal gamete.

chromosomes as pair 5 elements. The $7a^m$ is quite different because of its CI of 0.48, of the relative shortness of its secondary constriction (13.2% of the long arm) but mainly because of the position of the former the distance of its origin to the centromere, comprizing 34.7% of the long arm's length. One chromosome is left: its CI of 0.40 shows that it belongs to pair 7 (7b).

*Origin of the mutated* $7a^m$. The presence of 3 nucleoli in the interphase nuclei implies the existence of 3 nucleolar organizers. If 2 of them are borne by the homologues of pair 4, the 3rd one must be located on the constriction seen in the altered chromosome of pair 7. This may be explained by a reciprocal translocation between 2 chromosomes, one belonging to pair 4, the other to pair 7. In the scheme of Figure 4 we added to these the elements of pair 5 that should not be mistaken for the mutated chromosome.

Let us imagine 2 breaks (*r*), one affecting the short arm of one element of pair 4, that we shall call 4b, and the other on pair 7, in the short arm of chromosome 7a, for instance. After having rotated 180° these segments will be exchanged: the end of 4b fuses with 7a, that of 7a coupling with 4b. The result is 2 heterozygous pairs 4a-$4b^m$ and $7a^m$–7b.

The moment of the occurrence of this translocation remains unknown. For reasons of clarity, Figure 4 does not take into account the theoretical necessity of a telomere at the distal end of the translocated arm. We must postulate a second break near the apex of the short arm of chromosome 4b and 7a: this will give 2 telomeres 4b and 7a that will each fuse with the extremity of the other chromosomes.

Let us go back to Figure 4 where we show the 4 types of gametes (a, b, c and d) from an individual with type III chromosomal pattern. These gametes, eggs or spermatozoans, fusing with a normal gamete will give rise to embryos of 4 types: A, B, C and D. The caryotypes we have got up to now being exclusively of type A (2 *nu*) or type B (3 *nu*), C and D never appearing, we feel authorized to conclude that *X. borealis* $\varphi_1$ inheredited the mutation from one parent and therefore bears the mutation in the heterozygous state.

*Résumé.* L'examen cytologique de la descendance de 2 *Xenopus borealis* révéla 50% d'individus dotés de noyaux à 1 et 2 nucléoles (situation normale) et 50% à 1, 2 et 3 nucléoles. Le caryotype de ces derniers démontra la présence de 3 organisateurs nucléolaires, au lieu de 2, l'origine de l'élément supplémentaire relevant d'une translocation réciproque entre 4 et 7.

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