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Genetic load affecting chromosomes II and III in two natural populations of *Drosophila melanogaster* from Mexico.

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A great many studies on the genetic load in natural populations of *Drosophila* have been carried out by many authors with reference to the frequency of lethal and semilethal genes found in one of the chromosomes in the genome. There exist fewer studies in which two or more chromosomes are analyzed at the same time (*e.g*., Pavan *et al.*,1951; Dobzhansky and Spassky, 1953; Spiess and Allen, 1961; Dobzhansky *et al.*, 1963; Band and Ives, 1963; Spassky, Dobzhansky and Anderson, 1965; Hoenigsberg and Navas, 1965; Wallace, Zouros and Krimbas, 1966; Paik, 1966; and Allen, 1966, 1969). As a group these authors found information on natural populations of *D. melanogaster*, *D. persimilis*, *D. pseudoobscura* and *D. willistoni*, and the information has been reported by them in reference to the amount of the load found in different species and populations. This load could be restricted to a single chromosome or in some cases to two of them. In special situations they deal with particular phenomena, such as epistatic effects (Spassky, Dobzhansky and Anderson, 1965) or to the measurement of the variability (Dobzhansky and Spassky, 1951; Spiess and Allen, 1961), or other aspect of interest for the author.

Among the studies dealing with the load due to interaction between two or more chromosomes, we have those of Wallace, Zouros and Krimbas (1966) and Paik (1966) as representatives of this kind of analysis.

All of the above studies were done outside of Mexico. Those investigations dealt principally with an analysis of frequencies of deleterious chromosomes in different localities and at different seasons. In the present case, we are dealing with the effect of genetic load in two natural populations of *D. melanogaster* from Mexico.

Material and Methods

Samples of *D. melanogaster* from two localities (Coyoacan, D.F., Mexico City, with an altitude above sea level of 2250 m, and Veracruz City in the State of Veracruz located at sea level) were analyzed using the *Cy L* / *Pm* ; *Ubx* / *Sb* technique.

Groups of 20 males were studied weekly. Each male was crossed with three virgin females of the marker stock SM1 al^2 *Cy* sp^2 / *Pm*, In(3RL) ; Ubx^{130}, Ubx^{130} e^s / *Sb*. A single F1 *Cy L* / + ; *Sb* / + male was back-crossed with three virgin females from the marker stock. In the F2, five pairs of flies of the *Cy L* / + ; *Sb* / + constitution were mated *inter se*. The flies were allowed to oviposit during four days, transfered to a second bottle for an additional four day period and then discarded. Starting 12 days after the initiation of the culture, emerged flies were counted every other day until no more appeared. It is expected that, in the absence of a significant genetic load, four kinds of F3 flies will have been produced and in the proportions: 4 *Cy L* / + ; *Sb* / + : 2 *Cy L* / + ; + / + : 2 +/+ ; *Sb* / + : 1 +/+ ; +/ +. All the

cultures were kept at 25 ± 1°C, using as a food the regular corn meal - sugar - agar culture medium employed at the laboratory.

In the case of the population from Coyoacan located at a distance of about 5 km from the laboratory, males were collected each week prior to the crosses being done. Since Veracruz is approximately 400 km from the laboratory, a four-day collection of 314 males and 264 females was made. The flies were then introduced into a population cage and allowed to increase in number for two generations. Then 20 males were extracted for analysis every week. A minimum of 200 males were tested from each population. Analysis of 135 genomes from Coyoacan and 148 from Veracruz, were carried out.

Results

When all the counts were completed, we assigned chromosomes to different classes according to their relative viabilities. The classification system is shown in Table 1.

Differences in classification (column 2 vs. column 3) are due to the fact that when a chromosome is analyzed independently for its viability, the expected ratio with respect to the control *Cy L* / + ; *Sb* / + heterozygotes is 2 :1, and when interactions are measured this proportion is 4:1.

Table 2 shows the corresponding data when the total number of flies is summarized according to genotype and a corresponding Mendelian proportion. This allows us to evaluate the overall viability of the populations from the two different areas.

Table 3 shows the corresponding data when the total number of flies is summarized according to genotype and a corresponding Mendelian proportion. This allows us to evaluate the overall viability of the populations from the two different areas.

The numbers and frequencies of chromosomes II and III carrying deleter-ious genes and II-III interactions found in each population are shown in Table 3.

Table 1. Classification of the different categories of chromosomes.

Category of Chromosomes	Chromosomes II and III, independently, % of expected wild-type flies	Interaction of chromosomes II and III, % of expected wild-type flies
Lethal	0	0
Extreme semilethal	1-3	1-2
Semi-lethal	4-17	3-8
Sub-vital	18-25	9-16
Normal	26-40	17-23
Super normal	>40	>23

Table 2. General behavior in the viability of the populations. Observed and expected numbers of flies according to the mendelian proportions of the experimental crosses.

Population		Cy L /+;Sb/ + 4	+ /+ ; Sb / + 2	Cy L / + ; +/+ 2	+ / + ; + / + 1
Coyoacan	Observed	22012	8549	7213	2527
	Expected	17911	8955	8955	4477
Veracruz	Observed	23609	6982	7429	2093
	Expected	17828	8914	8914	4457

* all comparisons are different at P<0.0001

The actual number of lethals found per chromo-some and their interaction for each population studied is shown in Table 4.

Discussion

Analysis of Table 2 indicates that the populations tend to carry appreciable loads of genetic variability, since virtually all observed values are higher than the expected, in terms of the number of flies counted in each category.

Summing the lethals, extreme semi-

lethals (considered by some people as lethals; Wallace, 1970), and semilethals for each chromosome and their interactions, we note that the value for the population from Coyoacan concerning chromosome II is 25.91 as shown in Table 3**,** which is in the range of those found by the majority of the authors that have studied this chromosome (*i.e*., 12 to 30%).

In the case of the population from Veracruz, the corresponding value is 40.52%, quite high according to the values from other studies. If semilethals are excluded, the value is 24.31% and in the range of those populations previously studied by other authors, suggesting the possibility that the difference is primarily due to the high frequency of semilethals found in the present population of Veracruz. If the same assumption is made for the population from Coyoacan (semilethals omitted) the value of the genetic load is 10.37% and in the very low end of those reported previously.

Table 3. Number and frequency of deleterious chromosomes II and III in each population and their interaction.

Kind	Populations					
	Coyoacan 135 genomesChromosome			Veracruz 148 genomesChromosome		
	II	III	II and III	II	III	II and III
Lethals	(6) 4.44	(4) 2.96	(27) 20.00	(19) 12.83	(0) 0.0	(36) 24.32
Extreme semilethal	(8) 5.92	(4) 2.96	(10) 7.40	(17) 11.48	(0) 0.0	(18) 12.16
Semilethal	(21) 15.55	(28) 20.74	(27) 20.00	(24) 16.21	(32) 21.62	(27) 18.24
Subvitals	(12) 9.88	(19) 14.07	(37) 27.40	(17) 11.48	(51) 34.45	(42) 28.24
Normals	(73) 54.07	(76) 56.29	(29) 21.48	(60) 40.54	(59) 39.86	(19) 12.83
Super normals	(15) 11.11	(4) 2.96	(5) 3.70	(11) 7.43	(6) 4.05	(6) 4.05

When similar analyses are carried out on data from chromosome III, the Coyoacan population yields a value of 26.66% and Veracruz 21.62%. In both cases the corresponding frequencies for lethals and semilethals together are included in the range of values reported in the literature. An interesting finding in the population from Veracruz (which we cannot explain yet) is the apparent absence of lethals/extreme semilethals in chromosome III (none in 148 chromosomes analyzed as opposed to 36 in 148 chromosome II analyzed). The statistical significance of it is shown in Table 4. Here the differences are significant. In contrast, lethals and extreme semilethals were found in both chromosomes II and III in the population from Coyoacan 14/135 = 10.37% in the second chromosome and 8/135 = 5.92% in the third chromosome. They are also shown in Table 4.

Table 4. Differences for the number of lethas found between the two populations for chromosomes II, III and their interaction II-III.

	Chromosome II	Chromosome III	Interaction II and III
Veracruz n=148	36	0	54
Coyoacan n=135	14	8	37
X^2	7.78*	8.77*	1.88

∗ P<0.001

Finally results repor-ted by others are compared with those from the present report as shown in Table 5. Here we must make reference only to those studies done in *D. melanogaster* (Band, 1963; Hoenigsberg and Navas, 1965; Wallace, Zouros and Krimbas, 1966; and Paik, 1966). These authors reported for chromosome II values that varied from 15.20% in Korea up to 35.90% in USA. In Colombia, depending on the population, they ranged from 25.34 to 48.09%, with an average value of 36.71%. As could be seen, our populations show values that fall into the limits reported by those authors. It is important to mention that the differences in the two populations here reported, with respect to the load for the second chromosome, is significant as shown in Table 4.

With respect to the third chromosome also, the values found for us are included within the range reported by those authors, since they found values that varied from as low as 18.40% in Korea up to a highest of 40.10% in the USA. For this chromosome the values found for us are quite similar and not significant.

Table 5. Percentage of deleterious (lethal plus semiletals) reported for chromoses II, III and their interaction in several populatios of Drosophila as reported by different authors.

Population	II Chrom.	Species	II Chrom.	Interaction	Reference
Coyoacan	25.9	*D. melanogaster*	26.7	47.4	This report
Veracruz	40.5	*D. melanogaster*	21.7	54.7	This report
USA	35.9	*D. melanogaster*	40.1	----	Band, 1963
Colombia	36.7	*D. melanogaster*	----	----	Hoenigsberg, 1965
Colombia	26.6	*D. melanogaster*	26.6	32.8	Wallace, 1966
Korea	15.2	*D. melanogaster*	18.4	16.8	Paik, 1966

As for the interaction, the values found by Wallace, Zouros and Krimbas (1966) in Colombia and the one by Paik (1966) in Korea are our points of reference for comparisons. In Colombia the value is 32.80% and in Korea 16.80%. Both values are quite a bit lower and, with respect to our findings, statistically different. Our populations also are different among them.

Of course the biological significance of it could be only ascribed to the very particular background of the populations in which the environment has much influence, and it is reflected in the differences found either by us or by the other authors.

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