Najera, C. & J.L. Mensua. University of Valencia, Spain. The evolution of artificial populations of eye colour mutants of <u>Drosophila melanogaster</u> in mediums with and without alcohol.

The adaptation of D. melanogaster to environments with high levels of alcohol is a question of interest to several authors. McKenzie & Parsons (1974) find that flies from wine cellar populations have a higher tolerance to alcohol

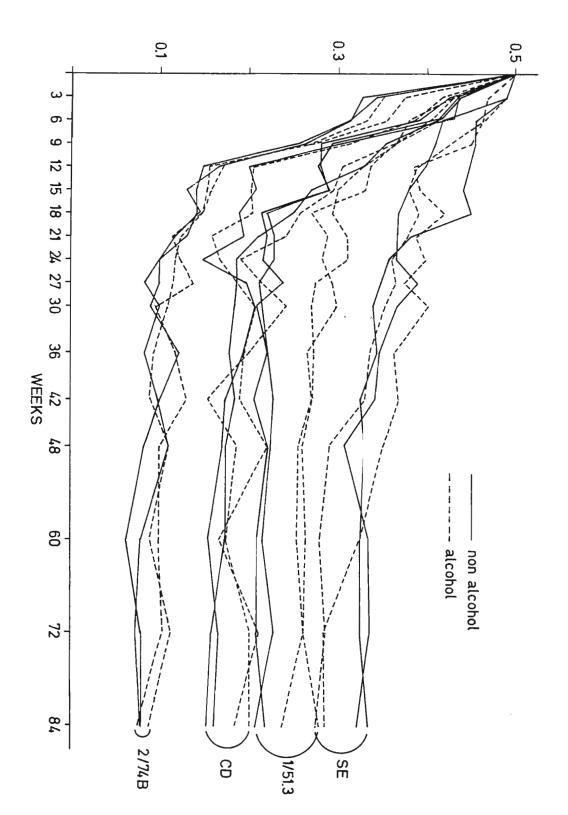


Figure 1. Evolution of gene frequencies.

than those from other populations. Najera & Mensua (1979) analyzed two cellar populations and they found that flies from these populations had a greater number of eye colour mutations than flies from other non cellar populations.

In order to explain this fact we tested the behavior of some wine cellar mutants against their wild allele in artificial populations, comparing two culture mediums, one supplemented with alcohol at 10% and the other without alcohol. We chose four eye colour mutants for their phenotype: two of light colour and two of dark colour, which we named 2/54A (allele of cardinal), 2/74B (strain segregating cardinal and cinnabar mutants), 2/58A (allele of sepia), and 1/51.3 (dark eye not yet identified).

The Buzzati-Traverso (1955) serial exchange technique was used to study the action of natural selection and to follow the population dynamics. The populations were started with 100 heterozygotic individuals, obtained from crossing each mutant with a wild wine cellar stock descending from a female which did not give any variability in  $F_2$  of eye colour mutants. The initial frequency of both alleles was, then, p=q=0.5.

Two replicae for each mutant population in each medium (alcohol and non-alcohol) was made (making a total of sixteen populations). The culture temperature was 19±1°C and the exchanges to new bottles were carried out every week. All individuals were counted every three weeks at the beginning, every six weeks afterwards, and every twelve weeks at the end of the experiment. Figure 1 shows, in graphic form, the evolution of all populations.

Each mutant attained different gene frequency at equilibrium. Equilibrium was attained approximately 300 days from starting. There were no differences between the normal and the alcohol experiment except in the 1/51.3 mutant, in which the gene frequency was clearly higher in the alcohol medium.

It seems that the different gene frequencies attained are correlated with the grade of colour from darker to lighter.

References: Buzzati-Traverso, A.A. 1955, Heredity 9:153-186; McKenzie, J.A. & P.A. Parsons 1974, Genetics 77:385-394; Najera, C. & J.L. Mensua 1979, IV Bienal. Real Sociedad Espanola de Historia Natural 65Z.

Narise, S. Josai University, Saitama, Japan. Activity difference among acid phosphatase allozymes from D.virilis.

Starch gel electrophoregrams at pH 7.0 of crude extracts from D. virilis showed the different activity in acid phosphatase (acph) among homozygotes for each  $Acph^{1}$ ,  $Acph^{2}$  and  $Acph^{4}$  allele (Narise 1976) at Acph locus which

presumably corresponds to Acph-1 described by MacIntyre (1971). As shown in Fig. 1, acph migrated to cathode under this condition and some activity was found near the origin in Acph¹ and Acph² strains, but not in Acph⁴. Extraction of the enzyme with 0.5% Triton from Acph¹ and Acph² flies resulted in increase in activity of the main band and decrease in activity near the origin. However, no effect of Triton was observed in Acph⁴. These facts indicate that the activity near the origin is partly due to the enzyme in particle fractions. Acph in D. melanogaster has been found to be localyzed to lysosomes (Sawicki & MacIntyre 1978). On the basis of these findings, biochemical study to search the factor(s) causing the activity difference has been conducted.

Adult flies, one day old, from the three allozyme strains were separately homogenized in 0.25M sucrose buffered with 20 mM Tris pH 7.0 in a Potter's homogenizer. The slurry was squeezed through two layers of gause. The crude extract was then centrifuged at 15,000g for 30 min. Acph activity in the supernatant was compared with that in the crude extract (Table 1). 72% of activity in the crude extract from Acph $^4$  strain was found in supernatant, while 31% from Acph $^1$  and 45% from Acph $^2$ . Thus, the activity difference in supernatant among three strains is greater than that in crude extract.

In order to examine intracellular distribution of acph, cell fractions (nuclei, motichondria and lysosomes, microsomes, and supernatant) were prepared by means of differential centrifugation and acph activity of each fraction was determined. Distribution of  $Acph^1$  activity among these four fractions (26, 29, 4 and 31%) was similar to that of  $Acph^2$  activity, whereas the distribution of  $Acph^4$  activity was 13, 17, 4 and 62%. These results suggest that  $Acph^4$  enzyme is easily released to supernatant from cell particles to (or in) which acph is attached or contained. Evidence for this was obtained using sucrose gradient fractionation. A combined cytoplasmic particle fraction prepared by centrifugation at 100,000g after removal