

fig.3

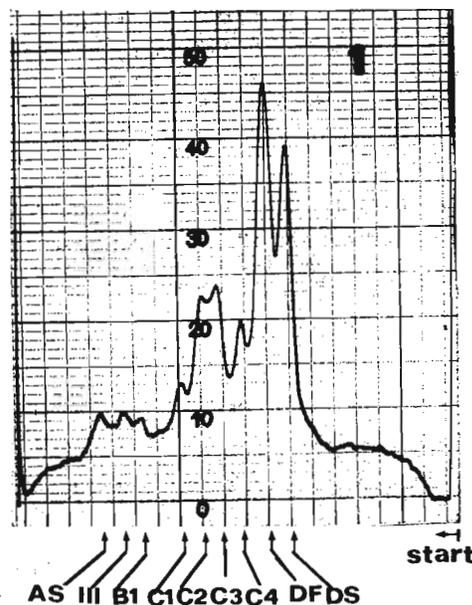


fig.4

Figure 3 shows the typical isozyme patterns of the leucineaminopeptidase (LAP) of 4 late pupae.

In Figure 4 the second sample from the left has been scanned.

References: Beckman, L. and F.M. Johnson 1964, *Hereditas* 51:212-230; Muhs, H.-J., Frequency of phenotypes (LAP, Aph and Est) seldom observed in laboratory strains of *D.m.* (in preparation); Poulik, M.D. 1957, *Nature* 180:1477.

Mulley, J.C. University of Sydney, Australia. Successive enzyme staining on acrylamide gels.

The small body size of *Drosophila* limits the number of samples with sufficient enzyme concentration that may be applied to gels. This report considers the employment of an agarose overlay for successive staining of a density

gradient acrylamide slice. This increases the number of markers detectable in the one individual which is useful in reducing labour and speeding up the analysis of many loci or getting the most information possible from populations of low numbers.

The following method was used for the detection of phosphoglucosmutase (PGM) in *D. buzzatii*. The sample was halved, electrophoresed and the two gels sliced yielding four identical electrophoretograms. Different staining procedures were applied to each slice. One slice was stained for PGM using a mixture of 25 ml tris-HCl (0.2 M, pH 8.5), 25 mg glucose-1-phosphate, 0.25 mg glucose-1,6-diphosphate, 3.75 mg NADP, 5 units glucose-6-phosphate dehydrogenase, 12.5 mg nitroblue tetrazolium, 1 mg PMS and 25 ml 4% agarose in 0.025 M MgCl₂ at 60°C. This solution was poured onto the gel in a 14 cm diameter petri dish and incubated at 37°C in darkness.

The concentrations of the components in this mixture are far in excess of the requirements for PGM detection. Rapid band development proceeds in the agarose as it hardens. Little or no stain deposits within the acrylamide avoiding interference to bands of succeeding assays. Within 10 minutes from commencement of incubation samples were typed for PGM, the agarose removed and the slice rinsed lightly in distilled water. The slice was then restained in the conventional manner for esterase, aldehyde oxidase or any other system. In this way the desired information was available from 4 instead of 5 slices.

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