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Kaytes, P. & D.L. Hartl. Washington University School of Medicine, St. Louis, Missouri. Note on electrophoretic mobility and tissue localization of  $\beta$ -glucuronidase.

In the hope of using  $\beta$ -glucuronidase as a model for studying genetic regulation of enzyme activity, we have attempted to identify putative structural gene(s) by surveying isofemale and chromosome substitution lines of *Drosophila melanogaster* for variation in electrophoretic mobility. Mass homogenates were prepared by

sonicating 40 flies in sonication buffer (.1N sodium acetate, pH 5.0, 10% sucrose) for 3 15-second bursts from a Heat Systems sonicator at 35% duty cycle, output setting of 5 with intermittent ice cooling. After pelleting cellular debris by centrifugation in a Sorvall SS-34 rotor, 50 microliters of the supernatant was applied to a vertical gel run in the system of Clarke (1964) at 10 mA/gel for 2½ hours and stained by the method of Hayashi (1963) with the modification that gels were pre-incubated in the acetate buffer without sucrose for ½ hour to eliminate background staining. The stain is sufficiently sensitive that single-fly bands can be obtained by sonicating in 50 microliters of buffer and applying all of the supernatant to the gel. Enzyme activity was localized to a single insoluble red band. (A diffusely staining area not sharply banded was also observed but not studied in detail.) The sharp band was eliminated by the inclusion in the staining mixture of saccharolactone, a competitive inhibitor of  $\beta$ -glucuronidase. In all, 124 lines from 8 geographical locations were examined, but no mobility variation could be detected. In contrast,  $\beta$ -glucuronidase extracted from *D. simulans* exhibited a significantly slower ( $R_f$  = .91 relative to *D. melanogaster*) form of the enzyme. Isoelectric focusing by the method of Righetti and Drysdale (1971) showed the *melanogaster* enzyme to be slightly more acidic; mixing experiments failed to show interconversion of forms. We conclude that, under our electrophoretic conditions, the sharply banded form of  $\beta$ -glucuronidase is monomorphic in *Drosophila melanogaster*, but that interspecific variation does exist.

Tissue distribution studies were also carried out on the enzyme. Adult flies were mounted and thin frozen sections were taken as in Kankel and Hall (1976). The sections were stained for activity as for gels without pre-incubation; no fixation was necessary. The greatest level of activity could be seen in male reproductive structures, particularly the accessory glands, and also in the ejaculatory bulb and testes. The presence of  $\beta$ -glucuronidase in the male reproductive system was further confirmed by hand dissection and staining. Slight amounts of activity could be seen in the digestive tract, particularly the stomodeal valve and intermittently in the Malpighian tubules. All activity staining was abolished in the presence of saccharolactone.

References: Clarke, J.T. 1964, Ann. New York Acad. Sci. 121:428-436; Hayashi et al. 1963, J. Histochem. Cytochem. 12:293-297; Kankel & Hall 1976, Dev. Biol. 48:1-14; Righetti & Drysdale 1971, Biochim. Biophys. Acta 236:17-28.

Kekic, V., R. Hadziselimovic & Z. Smit. University of Belgrade and University of Sarajevo, Yugoslavia. *Drosophila* fauna of artificial microhabitats in Bosnia and Herzegovina, Yugoslavia.

During the fall of 1969, we collected *Drosophila* flies at 29 localities in Bosnia and Herzegovina, covering the heights from 90 to 1031 meters above sea level (see Figure).

Collecting was carried out at man-made microhabitats--in the immediate vicinity of barrels in which plums, prepared for home

distillation of plum-brandy, were fermenting; vials with a small amount of fermenting plums were set out and after a certain time (every 3 hours) closed. Caught flies were taken out by means of aspirator, then fixed and kept in 70% ethanol until the time of identification.