Narise, S. Josai University, Sakado, Saitama, Japan. Substrate specificity of α- and β-esterase isozymes of D. virilis.

In gel electrophoretic techniques, non-biological substances, such as naphthyl derivatives, have been usually used as substrates of esterases. Purified α-esterases of D. virilis hydrolyze α- and β-naphthyl acetates at a similar rate, while β-esterases are active only on β-form. Neither esterases act on naphthylesters of long carbon chain fatty acids (Narise, S. 1973).

Activity of α- and β-esterase with aliphatic esters and glycerides

	α^3	β^A
Methyl acetate	0.0	35.1
Methyl propionate	12.6	193.7
Ethyl butyrate	50.2	71.2
Ethyl valerate	119.8	62.9
Ethyl caproate	55.5	14.8
Ethyl caprylate	28.4	11.9
Ethyl laurate	0.0	0.0
Ethyl stearate	0.0	--
Monoacetin	8.6	65.2
Monopropionin	47.2	146.7
Monobutyrin	--	--
Monolaurin	17.7	24.3
Monostearin	0.0	0.0
Triacetin	71.4	76.0
Tripropionin	139.4	214.1
Tributyrin	128.5	78.5
Trilaurin	0.6	0.3
Tristearin	0.0	0.3
α-naphthyl acetate	14.3	12.4
β-naphthyl acetate	17.4	27.1

The present work aims to get information on substrate or substrates in vivo of the esterase controlled by α- and β-esterase loci. Partially purified isozymes from homozygous stocks of each allele were used. Although the specific activity of these enzymes was about ten fold that of the crude extract, these samples were almost free from other esterases except that controlled by these loci. Esterase activities were assayed by the manometric technique at pH 7.4 with a 5% CO_2-95% N_2 mixture in the gas space. Substrates were added at a final concentration of 2.5×10^{-2}M. Specific activity was given as microliter of CO_2 produced per 30 min per mg of protein.

The activities of α^3 and β^A are shown in the Table. As presented in the Table, both enzymes attacked more actively methyl or ethyl esters of fatty acids having C^3 to C^8 than the naphthyl acetates. With the valeric acid ester as a substrate, the activity of α-esterase was highest, while that of β-esterase was best with the propionic acid ester. These two esterases also hydrolyzed more actively the lower mono- and tri-glycerides than the naphthyl acetates. However, they did not act on lauryl and stearyl esters except monolaurin. No significant difference was found in the enzyme activities among isoallelic isozymes.

Boström, G. and L.R. Nilson. University of Uppsala, Sweden. On tetrazoliumoxidases in Drosophila melanogaster.

Tetrazoliumoxidase isoenzymes in Drosophila melanogaster were first described and mapped by Jelnes, 1971. Starch gel electrophoresis of 19 wildtype stocks from our laboratory revealed that all strains except one were homozygous for the fast mobility allele To^F. The only strain homozygous for the slow allele To^S was Gruta. The strains homozygous for the To^F allele were Algeria, Amherst-3, Boa Esperanca, Canton-S, Crimea, Curitiba, Formosa, Hikone-R, Karsnäs, Kochi-R, Oregon-R, Salvador, San Miguel, Stäket, Tunnelgatan, Ultuna, Örebro and Knivsta.

Homogenates from Gruta (To^S/To^S) and Hikon-R (To^F/To^F) were subjected to iso-electric focusing (LKB 8100 Ampholine Electrofocusing Equipment) in a 100 ml column. The density gradient was formed by sucrose and the carrier ampholyte used was LKB Ampholine Ampholyte with pH-range 3-10. After focusing the column was bled in fractions of 1 ml with pH determination of each fraction. The carrier ampholyte was separated from the proteins by gel-filtration (Sephadex G-25) and starch gel electrophoresis was performed after concentration of the protein fractions. The isoenzyme pattern of To^S/To^S showed activity in the fraction of pH 4.5 and the To^F/To^F band was found at pH 4.8.

Tetrazoliumoxidases from mammals have been reported to catalyze the NADI-reaction and the oxidation of dichloroindophenol. The tetrazoliumoxidases from the strains studied by us did not catalyze the oxidation of dichloroindophenol. The NADI-reaction was more obscure (one, not reproducible, positive staining).

The isoenzymes did not loose their tetrazoliumoxidase-activity after incubation at 50°C for 45 min.