RELATIVE POSITION OF Adh-1 AND Gal-3 ON CHROMOSOME 3

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Previous mapping studies have identified three Loci which code (or specific enzymes (Lap-1, Lap-2, and Acp-3) on chromosome 1 (1, 2). These loci span most of the known linkage map ol this chromosome and should be useful markers for further mapping studies. In this communication we report the position of two additional enzyme loci, Adh-1 and Gal-3, relative to those already mapped and to the two morphological markers St and B.

Starch gel electrophoresis was performed on seed, young leaf, and flooded root tissue as described previously (2, 3, 4). Seed extracts were primarily used to obtain acid phosphatase (AcP), beta-galactosidase (Gal) and leucine aminopeptidase (Lap) phenotypes. Alcohol dehydrogenase phenotypes were determined using extracts from root tissue which had been submerged in water for 12 hours. Young leaf tissue was used to confirm the Acp, Gal and Lap phenotypes. Only small amounts of tissue were required for each sampling so that a single plant could be assayed for all isozyme systems and still be grown to maturity in the greenhouse.

The results of our analysis of five different F2 populations are presented in Table 1. All results are consistent with a gene order of

Lap 2 — Gal-3 — (Adh-1 , Acp-3) -- St -- (Lap-1, B).

Despite the significant recombination frequency observed between Adh-1 and Acp-3 in crosses #3 and #4, we were not able to determine which of these loci was closer to St. We therefore place the two 1oci together within parentheses to indicate that their relative sequence is unknown. The loci Lap-1 and B were treated similarly although previous work suggests that Lap-1 should be on the St side of B (2).

The map distances calculated between loci were relatively reproducible when results from different crosses were compared. The St, — (Adh-1, Acp-3) was calculated as 10, 20, and 10 recombinant units in crosses 1, 4, and 5, respectively. Similar consistency was observed for the St — Gal-3 and Gal-3 — (Adh-1, Acp-3) map distances. Ihe greatest source of error probably was in the scoring of the Acp 1 phenotypes, for these are faint in leaf tissue extracts and relatively poorly resolved in seed extracts. Thus Adh-1 would appeal to be a better genetic marker than Acp-3 for this particular region of chromosome 3.

1. Almgard, G. and K. Ohlund. 1970. PNL 2:9.
2. Weeden, N. F. and G. A. Marx. 1984. I. Hered. 75: 365-370.
3. Weeden, N. F. 1985. PNL 17:76-78.
4. Weeden, N. F. and E. Pagowska. 1985. PNL 17:79+80.

Table 1. Joint segregation analysis of loci on chromosome 3.

Cross	Loci	N	Linkage Chi-square	Recomb. Fract.	Std. Error
1	Lap-1 : St	83	27.0	20	±5
A283-19-24	Lap-1 : Acp-3	72	9.8	34	±5
	Lap-1 : Gal-3	78	2.0ns	-	
	St : Acp-3	66	22.0	19	±5
	St : Gal-3	78	1.9ns	-	
	Acp-3 : Gal-3	68	13.0	25	±6
2	St : B	67	5.1	31	±11
C283-562-564	St : Gal-3	68	13.0	25	±6
	B : Gal-3	65	1.1ns	-	
3	Lap-2 : Adh-1	25	8.1	31	±8
A284-290-292	Lap-2 : Gal-3	14	2.2ns	-	
	Adh-1 : Gal-3	14	2.8ns	-	
4	St : Adh-1	51	14.0	22	±6
A284-303-307	St : Acp-3	46	15.0	20	±6
	St : Gal-3	41	8.6	28	±13
	Adh-1 : Acp-3	46	44.0	11	±3
	Adh-1 : Gal-3	41	14.0	19	±7
	Acp-3 : Gal-3	37	10.0	20	±7
5 (11b x J136)	St : Adh-1	59	17.0	21	±6

IDENTIFICATION AND PARTIAL CHARACTERIZATION OF 3 BETA-GALACTOS1DASE ISOZYMES IN PEA LEAVES

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Enzymes catalyzing the hydrolysis of beta-D-galactose units from the non-reducing ends of B-galactosides may be conveniently visualized after electrophoresis on starch gels by flooding the gel with a solution containing 4-methylumbelliferyl galactoside (Sigma) and observing the fluorescence of the 4-methyl umbelliferone product under long-wave ultraviolet light (1). Analysis of pea leaf extracts revealed three zones of beta-galactosidase activity (Fig. 1). One zone migrated anodally on a pH 8.1 gel while the other two migrated cathodally on a pH 6.1 gel (extraction buffers and gel systems were as described in [2]). Sharp, well defined bands of activity were produced by the anodally migrating enzyme, which showed maximum activity at alkaline pH (assay solution: 0.1 M Tris pH 8.5 containing 2 mg 4-methylumbelliferyl-beta-D-galactoside which had been dissolved in 1 ml acetone). The cathodal enzymes exhibited an acid pH optimum (assay solution 0.1 M sodium citrate pH 4.5 containing 2 mg substrate as described above) and formed broader zones of activity which were often very faint (Fig. 1). Young leaves proved to be the most convenient tissue to sample for beta-galactosidase activity; however, seed and root extracts also gave similar phenotypes.