Suitable crosses are presently being made to confirm this relationship, to further characterize the genetic relationships of the variants, and to establish any linkage relationships with the two catalase structural loci in maize.

John C. Sorenson and John G. Scandalios

Comparative biochemical properties of maize aminopeptidases

We have investigated some biochemical properties of maize aminopeptidase isozymes for comparative purposes. Maize aminopeptidases ("leucine" aminopeptidase, LAP) are the products of four diallelic loci (Scandalios, 1969, Biochem. Gen. 3:37-79), which are designated A, B, C, and D.

Figure 1 shows the relative heat stabilities of the aminopeptidases which have been separated by starch gel electrophoresis of immature endosperm extracts. The

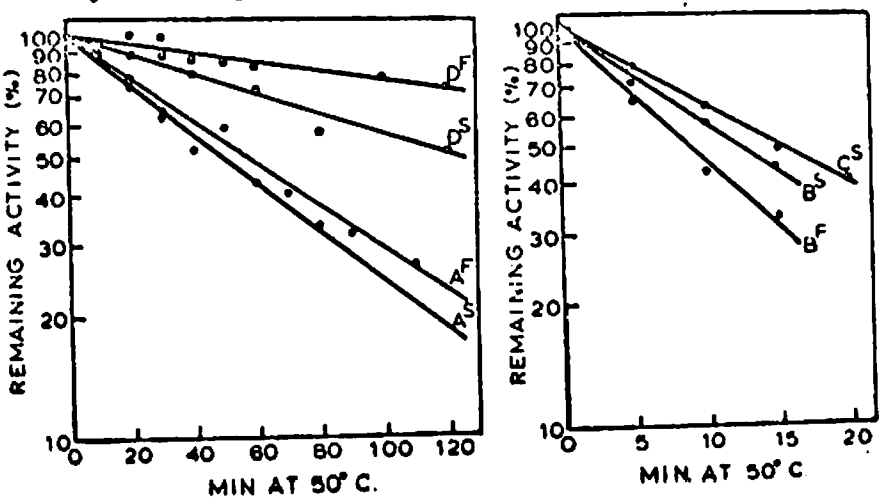

Figure 1. Relative heat stabilities of maize aminopeptidase isozymes at 50 C.

B^F, B^S, and C^S forms have half-lives of approximately 9, 12, and 15 minutes at 50 C, respectively. In contrast, the A and D forms are much more heat stable. $T_{½}$ for the A forms is in the range of 55 minutes while the half-lives for the D variants are over 120 minutes at 50 C.

We have previously reported on the substrate specificities and apparent Michaelis constants for the aminopeptidase isozymes with various amino acid-naphthylamide substrates (MNL 49:145, 1975). The molecular weights of the aminopeptidases have been determined utilizing the knowledge of their differential substrate specificities. Immature endosperm extracts were applied to a calibrated G-200 Sephadex column. Ten-drop (0.61 ml) fractions were collected to maximize resolution. Aliquots of the fractions were assayed with three substrates, arginine-, alanine-, and leucine-naphthylamide for aminopeptidase activities and with benzoyl DL-arginine-naphthylamide (BANA) for maize endopeptidase activity.

Figure 2 shows the resolution of the aminopeptidase activities. Based on knowledge of the substrate specificities, the alanine-NA peak represents the D isozyme (the D forms have high activities toward alanine-NA in addition to high activities with arginine-NA). The arginine-NA peak, which coincides with the alanine-NA peak, would be less specific. Its value would be contributed to by both the A and D forms. The B and C isozymes have highest activities with leucine-NA and would constitute this peak. Zymogram patterns of column fractions are consistent with this interpretation. They show that the A isozyme elutes ahead of the B and C forms. Based on this evidence, it is clear that the arginine-NA peak represents both the A and D isozymes, which have molecular weights of 71,500. The B and C forms are 63,500, and the endopeptidase is somewhat smaller, 58,000. While

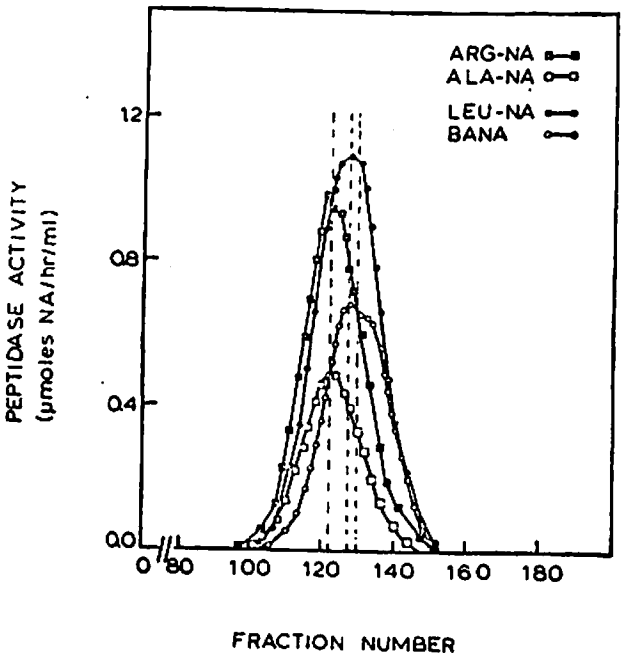

Figure 2. Molecular weight determination for maize endo- and aminopeptidases by elution from G-200 Sephadex. Dotted lines denote the peaks for the indicated activities.

the maize aminopeptidase isozymes do overlap in specificities, they are distinct enough to show the molecular weight differences.

Lila A. Ott and John G. Scandalios

Quantitative expression of alcohol dehydrogenase (ADH) and the ADH-specific inhibitor during germination

Quantitatively, ADH in any given tissue, including the scutellum, the root, the shoot and the endosperm, declines sharply after germination (Fig. 1). Ho and Scandalios demonstrated that the decline of ADH in the scutellum is due to the increase of an endogenous ADH-specific inhibitor (Plant Physiol.56:56, 1975). Further studies suggested the regulation of ADH in the root and in the shoot follow the same scheme: the control of the degenerative process of ADH activity relies on the buildup of a specific inhibitor to the enzyme. The inhibitor activity would then maintain at a certain level after the fourth day of germination (Fig. 1). In contrast, in the endosperm, the inhibitor activity is low and does not increase significantly after germination. Presumably, the inactivation of the enzyme in this tissue preceded the formation of the inhibitor prior to kernel maturation. In fact, the enzyme level in the endosperm of dry seeds is significantly lower than that in the milky endosperm stage. The decrease of ADH activity in the endosperm during kernel development has been reported to be due to inactivation of pre-existing enzyme (D. Fischer, MNL 47:55, 1973). Control of ADH activity after germination does not rely only on the inhibitor; when 6 day-old seedlings were subjected to anaerobic conditions, ADH was found to be increased in the root and the shoot but not in the scutellum and the endosperm. Whether it is due to de novo synthesis or other activating mechanisms for ADH protein molecules is not yet known. Correlations between the anaerobic "induction" and inhibitor levels are being investigated.

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