

## **Electrophoretic and immunological analyses of almond (*Prunus dulcis* L.) genotypes and hybrids.**

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### **Abstract:**

Aqueous extracts from sixty almond samples representing various genotypes and interspecies hybrids of almond, including almond-peach, were analyzed for protein and peptide content using electrophoresis, Western immunoblotting, and enzyme-linked immunosorbent assay (ELISA). Nondenaturing nondissociating polyacrylamide gel electrophoresis (NDND-PAGE) of the aqueous extracts indicated that a single major storage protein (almond major protein - AMP or amandin) dominated the total soluble protein composition. Denaturing SDS-PAGE analyses of the aqueous extracts revealed that the AMP was mainly composed of two sets of polypeptides with estimated molecular masses in the range of 38-41 kDa and 20-22 kDa, regardless of the source; however, distinct variations in the intensity and electrophoretic mobility of some bands were noted between samples. In addition to AMP, several minor polypeptides were also present in all the genotypes, and variations were seen in these as well. Regardless of the genotype, AMP was recognized in Western blots by rabbit polyclonal anti-AMP antibodies, mouse monoclonal anti-AMP antibodies (mAbs), and serum IgE from patients displaying strong serum anti-almond IgE reactivity. As with protein staining results, antibody reactivity also revealed common patterns but displayed some variation between samples. An anti-AMP inhibition ELISA was used to quantify and compare aqueous extracts for various samples. All samples ( $n = 60$ ) reacted in this assay with a mean  $\pm$  standard deviation (oh) =  $0.82 \pm 0.18$  when compared to reference aqueous extract from Nonpareil designated as 1.0.