Abstract

Polygalacturonase-inhibiting proteins (PGIPs) are believed to aid in plant defense against fungal pathogens by inhibiting polygalacturonases (PGs) secreted by the invading organism. In an effort to better understand this type of plant-pathogen interaction in cucurbits, we have isolated a cantaloupe PGIP (CmPGIP) from 5 to 15 day postanthesis cantaloupe fruit. CmPGIP inhibited crude extracts of PG from two of four fungal pathogens of cantaloupe that were tested. Results from assays for PG activity that utilized rate of substrate viscosity reduction or rate of reducing group formation were consistent with CmPGIP inhibition of endo-PG activity. The M(r) of CmPGIP by sedimentation equilibrium or MALDI-TOF MS was 38,500. The pI of CmPGIP was approximately 8.2, and its absorptivity at 280 nm was 0.93 ml/mg. The circular dichroism spectrum of native CmPGIP exhibited strong negative ellipticity in the near UV and possessed a far UV spectrum indicative of beta-sheet periodic structure. Amino acid sequences of the N terminus and a cyanogen bromide peptide were used to construct oligonucleotide primers for polymerase chain reaction sequencing. The sequenced open reading frame predicts a mature protein of 307 amino acids with up to 68% identity to other PGIP molecules. Northern blot analysis revealed differential expression during fruit development. The isolation and structural information obtained for CmPGIP by this investigation provide a foundation for the development of molecular strategies for pre- and postharvest crop protection.