Title Purification and biochemical characterization of polygalacturonase produced by *Penicillium expansum* during postharvest decay of 'Anjou' pear
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Citation Phytopathology 100 (1): 42-48. 2010.
Keywords host specificity; maceration

Abstract

A polygalacturonase (PG) was extracted and purified from decayed tissue of 'Anjou' pear fruit inoculated with *Penicillium expansum*. Ammonium sulfate precipitation, gel filtration, and cation exchange chromatography were used to purify the enzyme. Both chromatographic methods revealed a single peak corresponding to PG activity. PG enzyme activity from healthy and wounded pear tissue was undetectable, which supports the claim that the purified PG is of fungal origin. The purified enzyme had a molecular mass of 41 kDa and a pI of 7.8. Activity of the PG was not associated with a glycosylated protein. The enzyme was active over a broad pH range from 3 to 6, with optimal activity at 4.5 in sodium citrate and sodium acetate buffers. The optimal temperature for activity was 37°C but the enzyme was also active at 0, 5, 10, 20, and 50°C. Thin-layer chromatographic analysis of PG hydrolysis products showed that the enzyme exhibits endoand exo-activity. The purified enzyme macerated tissue in vitro causing \approx 30% reduction in mass of pear plugs compared with \approx 17% reduction for apple. Additionally, it produced 1.5-fold more soluble polyuronides on pear than apple tissue. This work shows for the first time the production of a PG by *P. expansum* during postharvest decay of pear fruit is different from the previously described PG produced in decayed apple fruit by the same pathogen.