Mechanism for the suppressive effect of salts on polyphenol oxidase activity of vegetables

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Summary

Polyphenol oxidase (monophenol dihydroxyphenylalanine:oxygen oxido-reductase :EC 1.14.18.1; PPO) is widely distributed in plants, and responsible for the browning reactions during handling, storage, processing, and cooking of vegetables. The browning reaction is initiated by the oxidation of phenolics to quinones by PPO. The browning reaction affects the nutritional quality and appearance of vegetables and fruits. Peeled apple fruits and potato tubers were usually soaked in NaCl solution to prevent the browning reaction on the surface of them. To elucidate the mechanism for suppressive effect of salts on PPO activity of vegetables, we purified PPO from mung bean sprouts by ammonium sulfate fractionation, butyl Toyopearl column chromatography and chromatofocusing. Six isozymes of PPO were detected from mung bean sprouts by SDS-polyacrylamide gel electrophoresis and chromatofocusing. PI of an isozyme, which has the highest activity, was 6.7. The PPO isozyme was purified as a single protein and the molecular mass was determined as 35 kDa by SDS-polyacrylamide gel electrophoresis. The apparent Km for chlorogenic acid was 1.3 mM. Inhibition by substrate was observed at higher concentration of chlorogenic acid than 5.0 mM. NaCl inhibited PPO activity from apple fruits, potato tubers and mung bean sprouts. Lineweaver-Burk plots of the purified PPO (pI 6.7) isozyme in the absence and presence of NaCl showed that inhibition by NaCl was non-competitive with the substrate chlorogenic acid. Ki value for NaCl was 0.22M (1.3%), that is the concentration of NaCl used for preventing the browning reaction of vegetables. These results suggest that NaCl inhibits non-competitively PPO activity, and results in preventing of enzymatic browning of vegetables.