## Repression of enzymatic browning of shredded vegetables by using enzyme inhibitors and NaCl

Masatsune Murata and Seiichi Homma Department of Nutrition and Food Science, Ochanomizu University

## Summary

When shredded vegetable is stored at refrigerator, it gradually turns brown. Last year we used shredded lettuce and examined such factors of this enzymatic browning during cold storage as amount of phenolics, phenylalanine ammonia lyase (PAL) activity and polyphenol oxidase (PPO) activity. PPO catalyzes the oxidation of o-diphenols to corresponding quinones. o- Quinones are yellowish compounds and are easily polymerized to form brown pigments. PAL is a key enzyme of the biosynthesis of phenolic compounds.

Shredded lettuce turned brown after 3 or 4 days of cold storage. PPO activity was almost constant during storage, while PAL activity intensively raised at 3 days after cutting. There was no relation between browning and phenolics and between browning and PPO activity, while there was a significant correlation between browning and PAL activity. This result shows that PAL activity is induced in shredded lettuce as the response against injury, newly synthesized phenolics are successively oxidized by PPO and that shredded lettuce turns brown.

Next the effect of NaCl on browning of shredded lettuce was examined. NaCl and other chlorides inhibited PPO activity of shredded lettuce. However the inhibitory activity of NaCl against lettuce PPO was weak (Ki=0.11 M) and NaCl did not inhibit the browning of shredded lettuce.

This year we examined the effect of the inhibitors of polyphenol biosynthesis on the browning of shredded lettuce during cold storage. We further examined the synergistic effect of NaCl and the inhibitors on the browning. We here used 2-aminoindan-2-phosphonic acid, a PAL inhibitor, and glyphosate, an enolpyruvylshikimate 3-phosphatre synthase inhibitor. AIP competitively inhibited lettuce PAL (Ki=115nM) against phenylalanine (Km=125  $\mu$  M). When shredded lettuce was treated with 10  $\mu$  M AIP or 1 mM glyphosate solutions, the browning was significantly inhibited. The PAL activity of lettuce treated by AIP was about a half of the activity of untreated lettuce at 3 days of storage. The amount of polyphenols at 8 days of storage in the treated lettuce was 30% of that of non-treated lettuce. NaCl did not show the synergistic effect on the inhibition of browning by AIP and glyphosate.

We here showed the usefulness of the inhibitors of polyphenol biosynthesis to prevent the browning of shredded lettuce. We hope the development of safe PAL or EPSPS inhibitors derived of natural products.