Title Pheophytinase activity and gene expression of chlorophyll-degrading enzymes relating to UV-B treatment in postharvest broccoli (*Brassica oleracea* L. Italica Group) florets
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Abstract

Pheophytinase (PPH) activity and gene expression of chlorophyll (Chl)-degrading enzymes relating to UV-B treatment in postharvest broccoli (*Brassica oleracea* L. Italica Group) florets were determined. PPH is involved in the dephytylation of Mg-free Chl *a*, pheophytin (Phy) *a*. However, *in vitro* chlorophyllase (Chlase, EC.3.1.1.14) also uses Phy *a* as a substrate to produce pheophorbide (Pheide) *a* by dephytylation. For an accurate determination of PPH activity, the PPH protein fraction was separated from Chlase protein by ammonium sulfate precipitation. The protein precipitated by 45–60% saturated ammonium sulfate included a little bit of Chlase activity and was suitable for PPH determination. PPH activity in broccoli florets treated with a UV-B dose of 19 kJ m⁻² was repressed for the first 2 d of storage at 15 °C, whereas it increased gradually with senescence of control broccoli florets. The expression level of *BoCLH1* was reduced in broccoli florets on day 4 of storage, while *BoCLH2* and *BoCLH3* were up-regulated with UV-B treatment. A high *BoPAO* expression level was found in senescent broccoli florets, and the up-regulation of this gene was delayed by UV-B treatment. The highest expression level of *BoPPH* was found in the control, and its expression was clearly repressed by UV-B treatment on day 2 of storage. We suggest that the up-regulation of Chl-degrading enzyme genes could be delayed by UV-B treatment, resulting in the suppression of floret yellowing in stored broccoli.