

# Characterization and Subcellular Localization of Chlorophyllase from *Ginkgo biloba*

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Chlorophyllase (Chlase) catalyzes the initial step of chlorophyll (Chl)-degradation, but the physiological significance of this reaction is still ambiguous. Common understanding of its role is that Chlase is involved in de-greening processes such as fruit ripening, leaf senescence, and flowering. But there is a possibility that Chlase is also involved in turnover and homeostasis of Chls. Among the de-greening processes, autumnal coloration is one of the most striking natural phenomena, but the involvement of Chlase during autumnal coloration is not clear. Previously, it was shown that Chlase activity and expression level of the Chlase gene were not increased during autumnal coloration in *Ginkgo biloba*, indicating that Chlase does not work specially in the de-greening processes in *G. biloba*. In this study, we characterized the recombinant Chlase and analyzed its subcellular localization to understand the role of the cloned Chlase of *G. biloba* (*GbCLH*). *GbCLH* exhibited its highest activity at pH 7.5, 40 °C. Kinetic analysis revealed that *GbCLH* hydrolyzes pheophytin (Pheo) *a* and Chl *a* more rapidly than Pheo *b* and Chl *b*. Transient expression analysis of 40 N-terminus amino acids of *GbCLH* fused with GFP (green fluorescent protein) and subcellular fractionation showed that *GbCLH* localizes within chloroplasts. Together with our previous results, property of *GbCLH* and its location within the chloroplasts suggest that *GbCLH* plays a role in the turnover and homeostasis of Chls in green leaves of *G. biloba*.

**Key words:** Chlorophyllase, *Ginkgo biloba*, Subcellular Localization