A New Chlorophyll Degradation Pathway

Chlorophyll degradation is vital during leaf senescence and fruit ripening, as it allows for recycling of nitrogen and other nutrients and for protection from buildup of phototoxic chlorophyll intermediates (Hörtensteiner, 2006). The first steps in chlorophyll breakdown are the removal of the phytol tail (dehydropheophorbide a) and the central Mg atom. It has been thought that dephosphorylation typically occurs first, catalyzed by the enzyme chlorophyllase, which converts chlorophyll to phytol and chlorophyllide. Removal of Mg subsequently converts chlorophyllide to pheophorbide a. However, Arabidopsis contains just two known chlorophyllases, CHL1 and CHL2, and it has been shown that neither of these isoforms is localized to plastids, and double knockout mutant plants still are able to degrade chlorophyll during leaf senescence (Schenk et al., 2007). Now, Schelbert et al. (pages 5525–5529) report the discovery of a novel plastid-localized enzyme, pheophytinase, that is essential for chlorophyll breakdown during leaf senescence.

Based on the results of Schenck et al. (2007), Schelbert et al. set out to discover the true chlorophyllase that functions during leaf senescence, using a combination of bioinformatics and reverse genetics. They reasoned that the protein should contain an α/β hydrolase fold characteristic of enzymes that catalyze an ester hydrolysis. Of 462 such proteins found in the Arabidopsis genome, the search was narrowed to 30 with predicted chloroplast localization and no previously defined function, and just three of these corresponded to genes that showed a leaf senescence-related pattern of expression. T-DNA insertion mutants of these genes were obtained, and one of these mutants showed a stay-green phenotype, indicating a lesion in chlorophyll breakdown during leaf senescence (see figure). The authors show that the corresponding enzyme is a pheophytinase (PPH), which specifically dephosphorylates the Mg-free chlorophyll pigment pheophytin and does not act on chlorophyll. They also found that putative PPH orthologs are common in eukaryotic photosynthesizing organisms, suggesting that the pathway may be highly conserved.

This work indicates that the previously accepted pathway of chlorophyll degradation must be revised. During leaf senescence, removal of Mg to form pheophytin is likely the first step, followed by removal of the phytol tail, catalyzed by PPH. Chlorophyllide, which is the last precursor of chlorophyll biosynthesis, is most likely not an intermediate of breakdown. Therefore, chlorophyll synthesis and breakdown are metabolically separated during leaf senescence. Based on patterns of expression, chlorophyllase may play a role in chlorophyll breakdown during fruit ripening (Azoulay Shemer et al., 2008) and response to pathogens and wounding (Kariola et al., 2005).

Deficiency of PPH causes a stay-green phenotype. Leaves after 5 d of dark-induced senescence are shown, left to right: wild type (Col-0), pph-1 mutant, complementation of the stay-green phenotype of pph-1 with a 3SS-PPH cDNA construct, and lack of complementation using a construct harboring a mutation of the proposed active-site Ser residue (3SS-PPH5221A). (Figure from Figure 2 of Schelbert et al. [2009].)

REFERENCES
### Permissions


### eTOCs

Sign up for eTOCs at:
[http://www.plantcell.org/cgi/alerts/ctmain](http://www.plantcell.org/cgi/alerts/ctmain)

### CiteTrack Alerts

Sign up for CiteTrack Alerts at:
[http://www.plantcell.org/cgi/alerts/ctmain](http://www.plantcell.org/cgi/alerts/ctmain)

### Subscription Information

Subscription Information for *The Plant Cell* and *Plant Physiology* is available at:
[http://www.aspb.org/publications/subscriptions.cfm](http://www.aspb.org/publications/subscriptions.cfm)