The Divergent Roles of STAYGREEN (SGR) Homologs in Chlorophyll Degradation

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Degradation of chlorophyll (Chl) by Chl catabolic enzymes (CCEs) causes the loss of green color that typically occurs during senescence of leaves. In addition to CCEs, STAYGREEN1 (SGR1) functions as a key regulator of Chl degradation. Although sgr1 mutants in many plant species exhibit a stay-green phenotype, the biochemical function of the SGR1 protein remains elusive. Many recent studies have examined the physiological and molecular roles of SGR1 and its homologs (SGR2 and SGR-LIKE) in Chl metabolism, finding that these proteins have different roles in different species. In this review, we summarize the recent studies on SGR and discuss the most likely functions of SGR homologs.

INTRODUCTION

Mendel’s green cotyledon trait in pea (Pisum sativum) has been widely used to demonstrate the principle of inheritance and the single recessive gene responsible for the non-yellowing trait has intrigued scientists for many years. In 2006, Armstead and colleagues finally identified the gene controlling Mendel’s green cotyledon trait (Armstead et al., 2006), and it is now named STAYGREEN1 (also termed STAY-GREEN1; SGR1) or NONYELLOWING1 (NYE1) in Arabidopsis (Cha et al., 2002; Park et al., 2007; Ren et al., 2007). At about the same time, different groups also independently identified SGR1 homologs in Arabidopsis thaliana (Ren et al., 2007) and rice (Oryza sativa; Jiang et al., 2007; Park et al., 2007), and subsequent work isolated and characterized SGR1 homologs in various plant species, including tomato (Solanum lycopersicum; Barry et al., 2008), bell pepper (Capsicum annuum; Barry et al., 2008), tall fescue (Festuca arundinacea; Wei et al., 2011), Medicago truncatula (Zhou et al., 2011), and soybean (Glycine max; Fang et al., 2014) (Fig. 1A).

Early analysis indicated that SGR1 might function in chlorophyll (Chl) degradation, as: (i) sgr1 knockout mutants exhibit a common stay-green phenotype in several plant species, (ii) sgr1 mutants display a type C nonfunctional (cosmetic) stay-green phenotype, which affects Chl degradation, but not other aspects of senescence (Thomas and Howarth, 2000), and (iii) rice SGR1 physically interacts with light-harvesting complex subunits of photosystem II (LHCII) in vitro and in vivo (Park et al., 2007). Indeed, Park et al. (2007) examined whether SGR1 has enzymatic properties typical of Chl catabolic enzymes (CCEs) in the chloroplast, but found that SGR does not bind to Chl nor does it have chlorophyllase activity. This indicates that SGR1 does not function as a CCE in the chloroplast.

The function of other SGR homologs is another missing piece of the puzzle in the study of SGR1 (Fig. 1B). Previous phylogenetic analysis classified the SGR family of higher plants into two groups (Barry et al., 2008; Hörtensteiner, 2009; Sakuraba et al., 2014b). One group comprises the genuine SGR subfamily, and mutations in these SGR genes cause a type C nonfunctional stay-green phenotype, as described above. In the other group, the SGR-LIKE (SGRL) subfamily, the C-terminal sequence of SGRL proteins differs considerably from that of the SGR subfamily proteins (Hörtensteiner, 2009). To our knowledge, however, all higher plants contain at least one SGRL, indicating that SGR has essential functions in green plants, probably acting in Chl breakdown. Furthermore, some plant species contain two or more SGRs; for example, Arabidopsis has SGR1 and SGR2, in addition to one SGRL (Barry et al., 2008; Sakuraba et al., 2014b). The physiological roles of these SGR homologs had remained unclear. However, after 2010, several studies have addressed the physiological and molecular roles of SGRLs and revealed their functions.

SGR1, CCEs, and LHCII Interact During Leaf Senescence

Many studies have examined the functions of SGR1 and CCEs in dark-induced senescence (Kusaba et al., 2007; Meguro et al., 2011; Park et al., 2007; Pružinská et al., 2003; Sakuraba et al., 2012; Schelbert et al., 2009). Although SGR1 interacts with the subunits of LHCII in vivo (Park et al., 2007), how SGR activates Chl degradation during leaf senescence has remained unclear. Work in Arabidopsis and rice has identified six plastid-localized CCEs: Non-yellowing coloring1 (NYC1) (Horie et al., 2009; Kusaba et al., 2007) and NYC1-Like (NOL) (Sato et al., 2009) encoding two different Chl b reductases, 7-Hydroxymethyl chlo-
Fig. 1. Phylogenetic and domain structure of SGR homologs in plants. (A) The phylogenetic tree was constructed using MEGA 5.1 (http://www.megasoftware.net/megamac.php). The evolutionary relationship was inferred using the Neighbor-joining method. Numbers at branch points represent bootstrap values of 100 replicate trees. The accession numbers of SGRs and SGRLs in the GenBank, AGI (Arabidopsis Gene Index) or TGI (The Gene Index; TC) are as follows: At SGR1 (Arabidopsis thaliana), At1g44000; Ca SGR (pepper, Capsicum annum), EU414631; AYA98500.1, ACB5686; Os SGR (rice, Oryza sativa), Os09g36200, AY50134; Os SGRL, Os04g59610, AK105982; Gm SGR1 (soybean, Glycine max), AY50141; Gm SGRL2, AY50142; Gm SGRL, TC216309; Mt SGR (Medicago truncatula), BF633258; Mt SGRL, TC182595+GO185310; Ni SGR (tobacco, Nicotiana tabacum), AY850136; Ni SGRL, TC129900; SI SGR (tomato, Solanum lycopersicum), DG100158; SI SGRL, TC118764; Zm SGR1 (maize, Zea mays), AY50136; Zm SGRL2, AY850137; Zm SGRL, TC503941. (B) Domain structure of Arabidopsis and rice SGR homologs. Chloroplast transit peptides (Red line), the conserved SGR domains (Blue line), and the variable C-terminal regions (Green line) are shown.

SGR1 and SGRL function in stress-induced leaf yellowing

Chl breakdown also occurs in response to several abiotic and biotic stresses, in addition to senescence (Lim et al., 2007). Arabidopsis SGR1 and SGRL overexpressors exhibit an early leaf yellowing phenotype, while sgr1/nye1-1 and sgr1 mutants display a stay-green phenotype under abiotic stress conditions, including high salinity, osmotic stress (mannitol), and drought stress conditions (Sakuraba et al., 2014a; 2014b; 2014c, Fig. 3). Furthermore, one of the Arabidopsis sgr1 alleles, termed noc1, develops less severe disease symptoms (leaf chlorosis and/or necrosis) than wild type after infection with the bacterial pathogen Pseudomonas syringae pv. tomato (Pst) DC3000 or the fungal pathogen Alternaria brassicicola (Mceey et al., 2011). Thus, SGR1, SGRL, and CCEs also function in leaf yellowing induced by biotic and abiotic stress.

SGRs and CCEs may also form a multi-protein complex for Chl degradation under biotic and abiotic stress conditions, similar to their interactions during senescence. Interestingly, transcript levels of SGR1, NY1C, PPH, and PAO dramatically increase during senescence, but transcripts of SGRL and NOL are more abundant in pre-senescent leaves during vegetative growth (Sakuraba et al., 2014b). Furthermore, SGRL interacts more strongly with NOL than with other CCEs (Sakuraba et al., 2014c). Thus, the composition of the SGRL-CCE complexes likely differs, with SGRL and NOL as the main components of the SGRL-CCE complex during stress-induced leaf yellowing and SGR1 and NY1C as the main components during senescence (Fig. 2). The transcripts of HCAR and RCCR remain at nearly constant levels throughout development, including during natural senescence (Sakuraba et al., 2013), indicating that HCAR and RCCR participate in the SGR-CCE complex during both stress- and senescence-induced leaf yellowing (Fig. 2).

Distinct relationship of two SGR homologs in soybean and Arabidopsis

As described above, different plant species have different num-
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Fig. 2. Divergent functions of Arabidopsis SGR1, SGR2, and SGRL homologs. In pre-senescence leaves during vegetative growth, SGRL and NOL proteins are much abundant than SGR1 and NYC1 transcripts. Thus, under abiotic/biotic stress conditions, SGRL and NOL are probably the main components of the SGRL-CCEs-LHCII complex. During leaf senescence, SGR1 forms a dynamic, multi-protein complex with CCEs on LHCII to activate Chl degradation. However, SGR2 negatively regulates Chl degradation. Like SGR1 and SGRL, SGR2 can interact with LHCII. However, SGR2 does not interact with most CCEs. Thus, SGR2 likely interrupts the formation of the SGR1-CCE or SGRL-CCE complexes, leading to adjustment of Chl degradation rate in senescing chloroplasts. SGR, STAYGREEN; SGRL, SGR-LIKE; NYC1, NON-YELLOW COLORING1; NOL, NYC1-LIKE; Chl, chlorophyll; CCE, Chl catabolic enzyme; LHCII, light-harvesting complex II.

members of SGR homologs. For example, rice and barley (Hordeum vulgare) have just one SGR homolog, but Arabidopsis and soybean have two SGR homologs, SGR1 and SGR2 (Sakuraba et al., 2014b). Recent discoveries have shed light on the differing physiological functions and relationships of these pairs of SGR homologs. In soybean, the leaves and seed embryos of d1 d2 double mutants show a significant stay-green phenotype (Chao et al., 1995; Guiamet et al., 1991) and recent work showed that D1 and D2 encode GmSGR1 and GmSGR2, respectively (Fang et al., 2014). The GmSGR1 and GmSGR2 homologs resulted from one of the two whole-genome duplication events that occurred in soybean 13 million years ago. Interestingly, d1 leaves showed a much weaker stay-green phenotype than the d1 d2 leaves, and the d1 seeds turned yellow normally, indicating that the two GmSGR homologs function redundantly.

In contrast with the two GmSGR homologs, the two Arabidopsis SGRs have different functions during leaf yellowing. An Arabidopsis SGR2 overexpressor line shows a stay-green phenotype and sgr2 null mutants exhibit a yellow leaf phenotype under dark-induced senescence and abiotic stress conditions (Sakuraba et al., 2014b; Fig. 3). By contrast, SGR1 overexpressor shows early leaf yellowing and sgr1-1/nyc1-1 mutants exhibit a stay-green phenotype, opposite to those of SGR2 overexpressors and sgr2 mutants. SGR2 has a much weaker physical interaction with CCEs compared with the interaction of SGR1 with CCEs. Because SGR2 also can interact with LHCII, an excess of SGR2 may interrupt the formation of the SGR1-CCEs-LHCII complex. Furthermore, SGR1 and SGR2 can interact with each other to form homo- or heterodimers. These two interesting characteristics of SGR2, (i) its lower capacity to interact with CCEs and (ii) its ability to form heterodimers with SGR1, may cause its negative effect on Chl degradation during leaf senescence. Arabidopsis SGR1 and SGR2 likely acquired their antagonistic functions to balance Chl catabolism in senescing chloroplasts, thus preventing inappropriate Chl degradation (Fig. 2). Indeed, rice SGR1 and SGR2 overexpressors showed a premature leaf-yellowing phenotype (Jiang et al., 2011; Rong et al., 2013) and Arabidopsis SGR1 and SGRL overexpressors showed a normal phenotype during vegetative growth (Sakuraba et al., 2012, 2014c). These findings suggest that this difference is partly caused by the existence of SGR2 in Arabidopsis. Thus, SGR homologs are not always positive regulators of Chl degradation, and this depends on the independent evolutionary processes that occurred in each species.

SGRS FUNCTION IN SEED DEGREEING AND MATURATION

Proper seed maturation requires prompt degradation of Chl because any Chl remaining in mature seeds has severe, negative effects on seed storability and longevity (Clerx et al., 2003; Johnson-Flanagan and Spencer, 1994). Although Arabidopsis sgr1 (SALK_007691) and sgr2 (SALK_008330) mutants produce normal yellow seeds, sgr1 sgr2 double mutants produce green seeds, indicating that Arabidopsis SGR1 and SGR2 function redundantly in seed degreening (Delmas et al., 2013). Knockout mutants of the abscisic acid (ABA) signaling-associated transcription factor ABA INSENSITIVE3 (ABI3) also produce green seeds (Clerx et al., 2003; Koornneef et al., 1989) and electrophoretic mobility shift assays revealed that ABI3 directly binds to the promoters of SGR1 and SGR2 (Delmas et al., 2013). Furthermore, ectopic expression of SGR1 and SGR2 in abi3 knockout mutants rescues the phenotype from green to yellow seeds, indicating that SGR1 and SGR2 have pivotal roles in seed degreening during maturation. Arabidopsis nyc1 mutant seeds also contain green pigments (Nakajima et al., 2012), indicating that NYC1 also functions in seed degreening. Zhang et al. (2014) recently reported that
SGR1 and SGR2 also affect the biosynthesis of tocopherol, a major component of Arabidopsis seeds (Sattler et al., 2004), during seed development; for example, the seeds of SGR1-overexpressing plants had significantly lower tocopherol contents. Thus, these results indicate that SGR1 and SGR2 have important roles in balancing Chl degradation and tocopherol biosynthesis during seed maturation.

Some aspects of seed degreening remain unclear. The seeds of pph mutants do not show a green phenotype, although the pph leaves show a stay-green phenotype (Shelbert et al., 2009). We also confirmed that two other mutants of CCEs, hcar and pao, have a stay-green phenotype but do not produce green seeds (unpublished). Furthermore, some CCE genes, such as HCAR, NOL, and RCCR have lower expression levels during seed maturation than during other developmental stages. Thus, it is not yet clear whether these CCEs function in seed degreening in Arabidopsis. Also, SGR2 acts as a negative regulator of leaf yellowing (Sakuraba et al., 2014b), but it promotes seed degreening. Thus, these results strongly suggest that Chl degradation mechanisms differ during seed degreening and leaf senescence.

OTHER INTERESTING FUNCTIONS OF SGR

Ripening of tomato fruits involves accumulation of carotenoids, including lycopene and β-carotene, along with a concomitant decrease in Chl levels (Fraser et al., 1994). A recent study indicated that tomato SGR1 (Solanum lycopersicum SGR1: SISGR1) balances these two major events of fruit ripening. In SISGR1 fruits, three kinds of carotenoids (β-carotene, lycopene, and phytoene) accumulate to higher levels than in non-transgenic tomato fruits (Luo et al., 2013), indicating that SISGR1 affects the regulation of carotenoid accumulation in ripening tomato fruits. Furthermore, SISGR1 physically interacts with the carotenoid biosynthesis enzyme phytoene synthase 1 (PSY1), which has an important role in tomato fruit nutrition (Fraser et al., 1994; 2004). Based on these results, the authors proposed that the physical interaction of SISGR1 and PSY1 leads to a decrease in PSY1 activity and thus causes reduced lycopene accumulation. This study showed the physical relationship between SGR1 and PSY1 in tomato fruits, prompting the intriguing question of whether SGR1 interacts with PSY1 or other carotenoid biosynthesis enzymes to regulate carotenoid accumulation in other tissues, such as pre-senescent leaves, or in other plant species. During leaf senescence, the level of carotenoids declines as Chl is degraded (Biswal, 1995), indicating the activation of carotenoid degradation pathways and inhibition of carotenoid biosynthesis. Thus, SGR1 may have pivotal functions in the regulation of carotenoid accumulation during leaf senescence.

All SGR family proteins in higher plants are predicted to localize to chloroplasts (Barry et al., 2008), indicating that they likely function in plastids, most likely in Chl degradation. However, in addition to its role in senescing chloroplasts, Medicago truncatula SGR (MISGR) also functions in nodule senescence in roots (Zhou et al., 2011). MISGR expression is higher in senescing nodules than any other organs, including senescent leaves. Furthermore, the nodules of Medicago sgr mutants (termed NF2089) show significant down-regulation of several nodule senescence-associated genes, indicating that MISGR affects nodule senescence in legumes. Nodule cells contain other plastid types, such as leucoplasts, and it would be an intriguing idea (and likely) that SGRs not only occur in chloroplasts, but generally occur in plastids where they execute their diverse functions. Thus, their role in fruit carotenoid biosynthesis likely involves chromoplasts. Functional analyses of MtSGR in nodule senescence of Medicago and SISGR1 in fruit ripening of tomato will expand our understanding of the potential roles of SGRs in physiological processes beyond Chl degradation.

TRANSCRIPTIONAL REGULATORY NETWORK OF SGR EXPRESSION

Although most studies of SGR have examined its biochemical and physiological functions, recent work in Arabidopsis and rice has identified several transcription factors that directly promote SGR expression. As described above, ABI3 binds to the canonical B3 domain-binding RY motif (CATGCA) in the promoters of SGR1 and SGR2 during seed degreening (Delmas et al., 2013). Recent work showed that Phytochrome Interacting Factor 4 (PIF4) and PIF5 strongly activate SGR1 expression during dark-induced senescence (Sakuraba et al., 2014b). In the PIF4/PIF5-dependent leaf senescence cascade, ABI5 and ENHANCED EM LEVEL (EEL), direct targets downstream of PIF4/PIF5, bind to the ABA-Responsive Element (ABRE) motif (TACGCT) in the SGR1 promoter (Sakuraba et al., 2014d). Notably, ABI3, ABI5, and EEL are ABA signaling-associated transcription factors (Giraudat et al., 1992; Jakoby et al., 2002), and ABRE-Binding Factor 2/3/4 (ABF2/3/4) bZIPs also induced SGR1 expression (Fujita et al., 2009), indicating that ABA signaling has an important role in the activation of SGR1 expression and Chl degradation. Interestingly, NYC1 is also directly activated by ABA-signaling transcription factors, including ABI5 and EEL (Sakuraba et al., 2014d) during dark-induced senescence and ABF4 during seed degreening (Nakajima et al., 2012). Furthermore, the OsNAP transcription factor, which is an ABA signaling-associated gene, directly activates the transcription of both SGR1 and NYC1 in rice (Li et al., 2014). Thus, several ABA-signaling transcription factors likely function to co-
activate SGR1 and NYC1, and this close transcriptional relationship between SGR1 and NYC1 is important for Chl degradation during both leaf senescence and seed degreening.

**FUTURE RESEARCH ON THE FUNCTIONS OF SGRS**

Recent functional studies of SGR1 and its homologs revealed that SGR1 can interact with various chloroplast proteins, including LHCCI proteins, all known CCEs, and PSY1 (Luo et al., 2013; Sakuraba et al., 2012; 2013). However, the amino acids in SGR that affect these physical interactions with other proteins remain to be identified. Park et al. found that V99 is important for OsSGR1 function because the rice sgr mutant has a V99M missense mutation, but this amino acid substitution does not affect the interaction of OsSGR1 with LHCCI (Park et al., 2007). More detailed analyses, such as the combination of site-directed mutagenesis and protein-protein interaction assays, will reveal more about the biochemical function of SGR.

SGR1 could interact with other, unknown proteins. The most intriguing candidate for a potential SGR1-interactor is metal-chelating substance (MCS), which is considered to be required for the removal of the Mg atom of Chl during Chl breakdown, but remains to be identified (Hörtensteiner and Krautler, 2011). Because all of the six known CCEs physically interact with SGR1 (Sakuraba et al., 2012; 2013), MCS likely also interacts with SGR1 and other CCEs as an essential factor in the multi-protein complexes. Another candidate SGR1-interactor is Thylakoid Formation 1 (THF1), a homolog of protein complexes. Another candidate SGR1-interactor is Thy.. Because all of the six known CCEs physically interact with SGR1 (Sakuraba et al., 2012; 2013), MCS likely also interacts with SGR1 and other CCEs as an essential factor in the multi-protein complexes. Another candidate SGR1-interactor is Thylakoid Formation 1 (THF1), a homolog of protein complexes. Another candidate SGR1-interactor is Thy.

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**REFERENCES**

Armstead, I., Donnison, I., Aubry, S., Harper, J., Hörtensteiner, S., James, C., Mani, J., Moffet, M., Ougham, H., Roberts, L., et al. (2007). Cross-species identification of Mendel's locus. Science 315, 73.

Barry, C.S., Miquin, R.P., Chung, M.Y., Besuden, A., and Giovanninoni, J.J. (2008). Amino acid substitutions in homologs of the STAY-GREEN protein are responsible for the green-flesh and chlorophyll retainer mutations of tomato and pepper. Plant Physiol. 147, 179-187.

Biswal, B. (1995). Carotenoid catabolism during leaf senescence and its control by light. J. Photochem. Photobiol. B, Biol. 30, 3-13.

Chen, C.W., Zeng, J., and Liu, D. (2002). Isolation, characterization, and mapping of the stay green mutant in rice. Theor. Appl. Genet. 104, 526-532.

Chao, W.S., Liu, V., Thompson, W.W., Platt, K., and Walling, L.L. (1995). The impact of chlorophyll-retention mutations, d1d2 and cgt1, during embryogenesis in soybean. Plant Physiol. 107, 253-262.

Clerkx, E.J., Vries, S.P., and Koornneef, M. (2003). Characterization of green seed, an enhancer of ab31 in Arabidopsis that affects seed longevity. Plant Physiol. 132, 1077-1084.

Delmas, F., Sankaranarayanan, S., Deb, S., Widdop, E., Bouronville, C., Bollier, N., North, J.G.B., McCourt, P., and Samuel, M.A. (2013). ABI3 controls embryo degreening through Mendel’s I locus. Proc. Natl. Acad. Sci. USA 110, e3885-e3894.

Fang, C., Li, C., Li, W., Wang, Z., Zhou, Z., Shen, Y., Wu, M., Wu, Y., Li, G., Kong, L.A., et al. (2014). Concerted evolution of D1 and D2 to regulate chlorophyll degradation in soybean. Plant J. 77, 700-712.

Forsberg, J., Strom, J., Kieselbach, H., Larsson, K., Alexiev, A., Engström, A., and Akerlund, H.E. (2005). Protease activities in the chloroplast capable of cleaving an LHCCI N-terminal peptide. Physiol. Plant. 123, 21-29.

Fraser, P.D., Truesdale, M.R., Bird, C.R., Schuch, W., and Bramley, P.M. (1994). Carotenoid biosynthesis during tomato fruit development (evidence for tissue-specific gene expression). Plant Physiol. 105, 405-413.

Fraser, P.D., and Bramley, P.M. (2004). The biosynthesis and nutritional uses of carotenoids. Prog. Lipid Res. 43, 228-265.

Fujita, Y., Nakashima, K., Yoshida, T., Katagiri, T., Kidokoro, S., Kanamaru, N., Umezawa, T., Fujita, M., Maruyama, K., Ishiyama, K., et al. (2009). Three SnRK2 protein kinases are the main positive regulators of abscisic acid signaling in response to water stress in Arabidopsis. Plant Cell Physiol. 50, 2123-2132.

Giraudat, J., Hauge, B.M., Valon, C., Smalle, J., Parcy, F., and Goodman, H.M. (1992). Isolation of the Arabidopsis ABI3 gene by positional cloning. Plant Cell 4, 1251-1261.

Gray, J., Close, P.S., Briggs, S.P., and Johal, G.S. (1997). A novel suppressor of cell death in plants encoded by the Ls1 gene of maize. Cell 89, 25-31.

Gray, J., Janick-Buckner, D., Buckner, B., Close, P.S., and Johal, G.S. (2002). Light-dependent death of maize lsl1 cells is mediated by mature chloroplasts. Plant Physiol. 130, 1894-1907.

Guimet, J.J., Schwartz, E., Pichersky, E., and Noordon, L.D. (1991). Characterization of cytoplasmic and nuclear mutations affecting chlorophyll and chlorophyll-binding proteins during senescence in soybean. Plant Physiol. 96, 227-231.

Hirashima, M., Tanaka, R., and Tanaka, A. (2009). Light-independent cell death induced by accumulation of phaeophorbide a in Arabidopsis thaliana. Plant Cell Physiol. 50, 719-729.

Horie, Y., Ito, H., Kusaba, M., Tanaka, R., and Tanaka, A. (2009). Participation of chlorophyll b reductase in the initial step of the degradation of light-harvesting chlorophyll a/b-protein complex in Arabidopsis. J. Biol. Chem. 284, 17449-17456.

Hörtensteiner, S. (2009). Stay-green regulates chlorophyll and chlorophyll-binding protein degradation during senescence. Trends Plant Sci. 14, 155-162.

Hörtensteiner, S., and Krautler, B. (2011). Chlorophyll breakdown in higher plants. Biochem. Biophys. Acta 1807, 977-988.

Huang, W., Chen, Q., Zhu, Y., Hu, F., Zhang, L., Ma, Z., He, Z., and Huang, J. (2013). Arabidopsis thylakoid formation 1 is a critical regulator for dynamics of PSII-LHCII complexes in leaf senescence and excess light. Mol. Plant 6, 1673-1691.

Jakoby, M., Weisshaar, B., Droge-Laser, W., Vicente-Carbajosa, J., Tiedemann, J., Kroj, T., and Parcy, F. (2002). bZIP transcription factors in Arabidopsis. Trends Plant Sci. 7, 106-111.

Jiang, H., Li, M., Liang, N., Yan, H., Wei, Y., Xu, X., Liu, J., Xu, Z.,...
Kusaba, M., Ito, H., Morita, R., Iida, S., Sato, Y., Fujimoto, M., Ka-Lim, P.O., Kim, H.J., and Nam, H.G. (2007). Leaf senescence. Annu. Mol. Cells 39, 58-60.

Luo, Z., Zhang, J., Li, J., Yang, C., Wang, T., Ouyang, B., Li, H., Mach, J.M., Castillo, A.R., Hoogstraten, R., and Greenberg, J.T. (2009). Defect in non-yellow coloring 3, an alpha/beta hydro-lase of the chlorophyll cycle in Arabidopsis. Plant Cell 21, 1362-1375.

Morita, R., Sato, Y., Masuda, Y., Nishimura, M., and Kusaba, M. (2009). Two short-chain dehydrogenase/reductases, AYEY1 and AYEY2, regulate chlorophyll degradation during leaf senescence in rice. Mol. Cells 27, 106-115.

Pružinská, A., Tanner, G., Anders, I., Roca, M., and Hörtensteiner, S. (2003). Chlorophyll breakdown: pheophorbide a is a product of a cDNA encoding a beta-hydro-lase. Plant Physiol. 131, 1699-1706.

Rong, H., Tang, Y., Xiong, H., Wu, P., Chen, Y., Li, M., Wu, G., and Jiang, H. (2013). The Stay-Green rice (SGRL) gene regulates chlorophyll degradation in rice. J. Plant Physiol. 170, 1367-1373.

Sakuraba, Y., Schelbert, S., Park, S.Y., Han, S.H., Lee, B.D., Andres, C.B., Kessler, F., Hörtensteiner, S., and Paek, N.C. (2012). STAY-GREEN and chlorophyll catabolic enzymes interact at light-harvesting complex II for chlorophyll detoxification during leaf senescence in Arabidopsis. Plant Cell 24, 507-518.

Sakuraba, Y., Kim, Y.S., Yoo, S.C., Hörtensteiner, S., and Paek, N.C. (2013). A role for chlorophyll a catabolite reductase in the metabolism of chlorophyll breakdown intermediates during leaf senescence. Biochem. Biophys. Res. Commun. 430, 32-37.

Sun, X., Chu, J., et al. (2014). OsNAP connects abscisic acid signaling and directly targeting senescence-associated genes in rice. Proc. Natl. Acad. Sci. U.S.A. 111, 1266-1274.

Zhang, W., Liu, T., Ren, G., Huang, Z., Zhou, Y., Cahoon, E.B., and Zhang, C. (2014). Chlorophyll degradation: the tocopherol biosynthesis-related phytohormone 1 gene product in Arabidopsis leads to deficient tocopherol accumulation and variegated leaves. Plant Physiol. 163, 3594-3604.

Wei, Q., Guo, Y., and Kuai, B. (2011). Isolation and characterization of a chlorophyll degradation regulatory gene from tall fescue. Plant Cell Rep. 30, 1201-1207.

Yamatani, H., Sato, Y., Masuda, Y., Kato, Y., Morita, R., Fukunaga, K., Nagamura, Y., Nishimura, M., Sakamoto, W., Tanaka, A., et al. (2013). NYC4, the rice ortholog of Arabidopsis THF1, is involved in chlorophyll degradation during leaf senescence in Arabidopsis. Plant Cell 25, 767-785.

Zhang, W., Liu, T., Ren, G., Hörtensteiner, S., Zhou, Y., Cahoon, E.B., and Zhang, C. (2014). Chlorophyll degradation: the tocopherol biosynthesis-related phytohormone 1 gene product in Arabidopsis leads to deficient tocopherol accumulation and variegated leaves. Plant Physiol. 163, 3594-3604.

Wei, Q., Guo, Y., and Kuai, B. (2011). Isolation and characterization of a chlorophyll degradation regulatory gene from tall fescue. Plant Cell Rep. 30, 1201-1207.

Yamatani, H., Sato, Y., Masuda, Y., Kato, Y., Morita, R., Fukunaga, K., Nagamura, Y., Nishimura, M., Sakamoto, W., Tanaka, A., et al. (2013). NYC4, the rice ortholog of Arabidopsis THF1, is involved in chlorophyll degradation during leaf senescence in Arabidopsis. Plant Cell 25, 767-785.

Chen, F., and Wu, G. (2007). Molecular cloning and function analysis of the stay green gene in rice. Plant J. 52, 197-209.

Johnson-Flanagan, A.M., and Spencer, M.S. (1994). Ethylene production during development of mustard (Brassica juncea) and radish (Raphanus sativus) seeds. Plant Physiol. 106, 601-606.

Kato, Y., and Sakamoto W. (2009). Protein quality control in chlo-roplast: a current model of D1 protein degradation in the photo-system II repair cycle. J. Biochem. 146, 463-469.

Koornneef, M., Hanhart, C.J., Hillhorst, H.W., and Karssen, C.M. (1989). In vivo inhibition of seed development and reserve pro-tein accumulation in recombinants of abscisic acid biosynthesis and responsiveness mutants in Arabidopsis thaliana. Plant Physiol. 90, 463-469.

Kusaba, M., Ito, H., Morita, R., Iida, S., Sato, Y., Fujimoto, M., Ka-Lim, P.O., Kim, H.J., and Nam, H.G. (2007). Leaf senescence. Annu. Mol. Cells 39, 58-60.

Luo, Z., Zhang, J., Li, J., Yang, C., Wang, T., Ouyang, B., Li, H., Giovanni, J., and Ye, Z. (2013). A STAY-GREEN protein SIGSR1 regulates lycopene and beta-carotene accumulation by interfering directly with SPSY1 during the ripening processes in tomato. New Phytol. 198, 442-452.

Mach, J.M., Castillo, A.R., Hoogstraten, R., and Greenberg, J.T. (1990). The Arabidopsis-accelerated cell death gene ACD2 encodes red chlorophyll carotolite reductase and suppresses the spread of disease symptoms. Proc. Natl. Acad. Sci. U.S.A. 98, 771-776.

Mecsey, C., Hauck, P., Trapp, M., Pumplin, N., Plovanich, A., Yoo, J., and He, S.Y. (2011). A critical role of STAYGREEN/Mendel’s loci in controlling disease symptom development during Pseudomonas syringae pv tomato infection of Arabidopsis. Plant Physiol. 157, 1965-1974.

Meguro, M., Ito, H., Takabayashi, A., Tanaka, R., and Tanaka, A. (2011). Identification of the 7-hydroxymethyl chlorophyll a reductase of the chlorophyll cycle in Arabidopsis. Plant Cell 23, 3442-3453.

Morita, R., Sato, Y., Masuda, Y., Nishimura, M., and Kusaba, M. (2009). Defect in non-yellow coloring 3, an alpha/beta hydro-lase-fold family protein, causes a stay-green phenotype during leaf senescence in rice. Plant J. 59, 947-952.

Nakagawara, E., Sakuraba, Y., Yamashita, T., Tanaka, R., and Naka-taka, A. (2007). Clp protease controls chlorophyll b synthesis by regulating the level of chlorophyllide a oxygenase. Plant J. 49, 800-809.

Nakajima, S., Ito, H., Tanaka, R., and Tanaka, A. (2012). Chlorophyll b reductase plays an essential role in maturation and storability of Arabidopsis seeds. Plant Physiol. 160, 261-273.

Ollinares, P.D., Kim, J., and van Wijk, K.J. (2011). The Clp protease system: a central component of the chloroplast protease network. Biochem. Biophys. Acta 1807, 999-1011.

Park, S.Y., Yu, J.W., Park, J.S., Li, J., Yoo, S.C., Lee, N.Y., Lee, S.K., Jeong, S.W., Seo, H.S., Koh, H.J., et al. (2007). The senescence-induced staygreen protein regulates chlorophyll degrada-tion. Plant Cell 19, 1649-1664.

Průžinská, A., Tanner, G., Anders, I., Roca, M., and Hörtensteiner, S. (2003). Chlorophyll breakdown: phaeophorid b is an Rieske-type iron-sulfur protein, encoded by the accelerated cell death 1 gene. Proc. Natl. Acad. Sci. U.S.A. 100, 15259-15264.

Průžinská, A., Anders, I., Aubry, S., Schenk, N., Tapernoux-Luthi, M., Euller, T., Krautler, B., and Hörtensteiner, S. (2007). In vivo particip-ation of red chlorophyll carotolite reductase in chlorophyll breakdown. Plant Cell 19, 369-387.

Ren, G., An, K., Liao, Y., Zhou, X., Cao, Y., Zhao, H., Ge, X., and Kuai, B. (2007). Identification of a novel chloroplast protein