

### Discovery of genes related to leaf senescence during long-term dry stress

Extreme weather events frequently occur around the world, causing crop damage. The damage caused by droughts is extensive and has become a serious global problem. Leaf senescence occurs as the late stage of leaf development and in response to environmental stress. Ethylene is a major plant hormone inducing leaf senescence. Recent studies have shown that abscisic acid (ABA) also induces leaf senescence. The elucidation of the mechanism of leaf senescence during long-term dry stress is important to enable stable food production under drought via development of crops. However, the detailed mechanisms of ABA-induced leaf senescence remain unclear. We focused on the A subfamily of stress-responsive NAC (SNAC-A) transcription factors, the expression of which is induced by abiotic stresses, particularly ABA.

Gene expression analysis revealed that seven SNAC-A genes (ANAC055, ANAC019, ANAC072/RD26, ANAC002/ATAF1, ANAC081/ATAF2, ANAC102 and ANAC032) were induced by long-term treatment with ABA and/or during age-dependent senescence. The SNAC-A septuple mutant clearly showed retardation of ABA-inducible leaf senescence. Microarray analysis indicated that SNAC-As induce ABA- and senescence-inducible genes. In addition, comparison of the expression profiles of the downstream genes of SNAC-As and ABA-responsive element (ABRE)-binding protein (AREB)/ABRE-binding factor (ABF) (AREB/ABFs) indicates that SNAC-As induce a different set of ABA-inducible genes from those mediated by AREB/ABFs. These results suggest that SNAC-As play crucial roles in ABA-induced leaf senescence signaling.

Our results indicate that the SNAC-A subfamily genes are mainly involved in ABA-induced leaf senescence. Controlling gene expression will enable researchers to regulate leaf senescence during stress responses, and thus contribute toward improving the yield of stress-tolerant crops.

1) Nakashima K et al. (2012) *Biochimica et Biophysica Acta* 1819: 97-103

2) Takasaki H et al. (2015) *Plant J.* 84: 1114-1123

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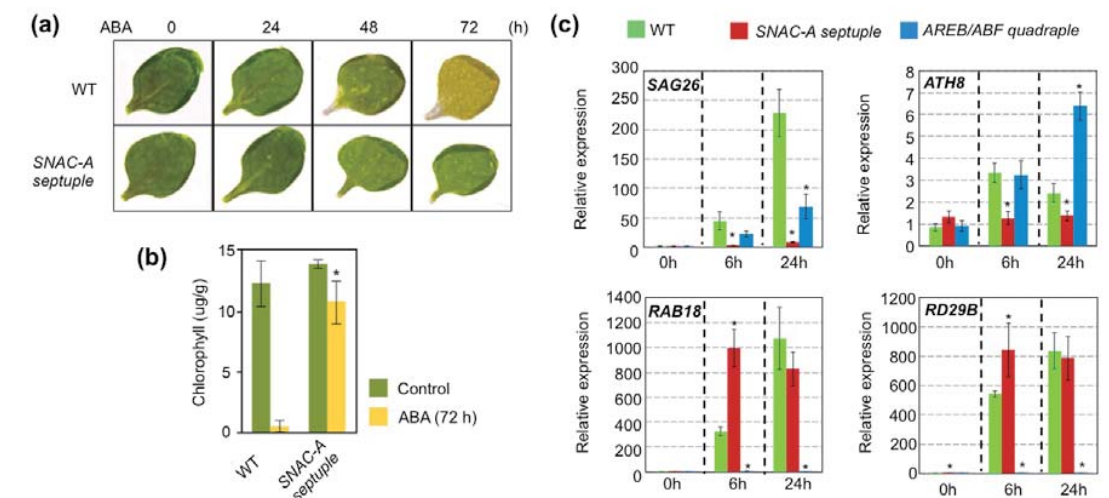


Fig. 1. Chlorophyll degradation and gene expression in ABA-treated wild-type and ABA-treated mutant leaves.

(a) Reduced sensitivity to ABA in chlorophyll degradation in ABA-treated wild-type (WT) and mutant (*SNAC-A septuple*) leaves. Each number indicates the time after 100  $\mu$ M ABA treatment.

(b) The change in chlorophyll content in ABA-induced senescence. First rosette leaves of wild-type and mutant plants were grown on GM agar plates with or without treatment with 100  $\mu$ M ABA for 72 h. Student's t-test: \* $P < 0.05$ . Each value represents mean  $\pm$  SD ( $n = 4$ ).

(c) ABA-responsive gene expression in the SNAC-A septuple and AREB/ABF quadruple mutants. Expression profiles of downstream genes of AREB/ABF and SNAC-A transcription factors. Two-week-old seedlings grown on GM agar plates were transferred to GM agar plated with 100  $\mu$ M ABA for 6 and 24 h. Total RNA was isolated from seedlings.

The figures were adapted from Takasaki *et al.* (2015).

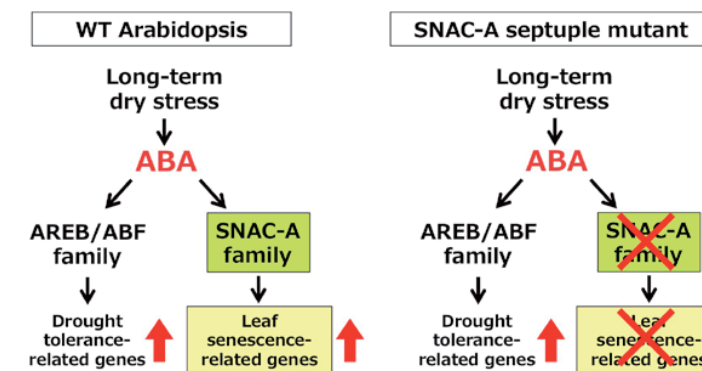


Fig. 2. Molecular mechanism of leaf senescence during long-term dry stress.

SNAC-As induce ABA-inducible genes related to senescence during stress, and SNAC-As induce a different set of ABA-inducible genes from those mediated by AREB/ABFs.