

LABORATORY OF BIOLOGICAL DIVERSITY

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Active DNA transposons are important tools for gene functional analysis. The endogenous non-autonomous transposon, *nDart1-0*, in rice (*Oryza sativa* L.) is expected to generate various transposon-insertion mutants because *nDart1-0* elements tend to insert into genic regions under natural growth conditions. The transpositions of *nDart1-0* were promoted by an active autonomous element, *aDart1-27*, on chromosome 6. By using the endogenous *nDart1/aDart1-27* system in rice, a large-scale *nDart*-inserted mutant population could be easily generated under normal field conditions, and the resulting tagging lines were free of somaclonal variation.

I. A mutable albino allele in rice reveals that formation of thylakoid membranes requires SNOW-WHITE LEAF1 gene Activation and Epigenetic Regulation of DNA Transposon *nDart1* in Rice

To understand chloroplast biogenesis and development, various chloroplast-defective mutants have been analyzed, yet they remain to be discovered. Although analyses of albino plants provide important information about mechanisms of plastid development, albino mutants are seedling lethal under natural growth conditions, owing to the complete loss of photosynthetic apparatus. Variegated mutants are excellent models for exploring the mechanism of chloroplast biogenesis because green and white sectors in the leaves allow an increased chance of survival. The variegation caused by somatic excision of DNA transposon is known as a mutable allele. The endogenous *nDart1/aDart1* tagging system is a powerful tool for investigating various unidentified and/or uncharacterized albino alleles. We report a novel variegated albino mutant, *snow-white leaf1-variegated* (*swl1-v*), caused by insertion and excision of *nDart1-0* in the *SNOW-WHITE LEAF1* (*SWL1*) gene (Figure 1). We have developed a specific method (*nDart1-0*-iPCR)

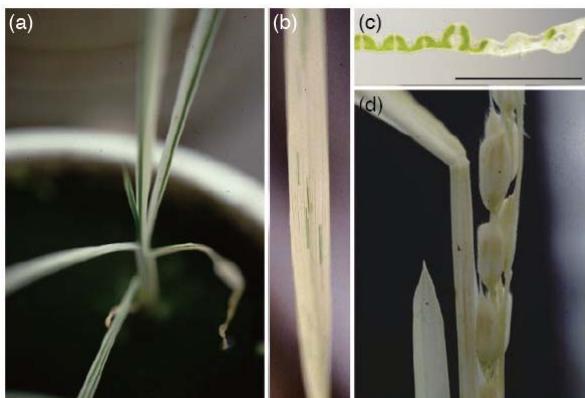


Figure 1. Phenotypes of *swl1-v* plants. Mutable albino plants with large green sectors (a) and with fine sectors (b). (c) Transverse section of the leaf blade. Scale bar = 1.0 mm. (d) Panicles.

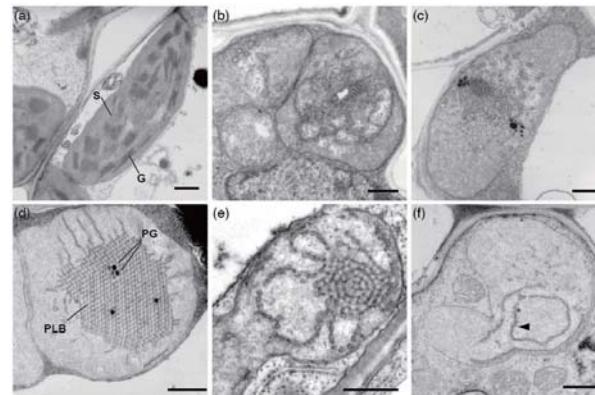


Figure 2. Plastid ultrastructures of WT and *swl1-stb* plants. Chloroplasts from tertiary leaf blades of WT (a) and *swl1-stb* (b and c) seedlings grown under light. Etioplasts from the tertiary leaf blades of WT (d) and *swl1-stb* (e and f) seedlings germinated in the dark. G, grana thylakoid; S, stroma thylakoid; PG, plastoglobule; PLB, prolamellar body. The arrowhead indicates a ring-shaped structure formed from vesicles. Scale bars = 500 nm.

for efficient detection of *nDart1-0* insertions and successfully identified the *snow-white leaf1* (*swl1*) gene in a variegated albino (*swl1-v*) mutant obtained from the *nDart1*-promoted rice tagging line. The variegated albino phenotype was caused by insertion and excision of *nDart1-0* in the 5'-untranslated region of the *SWL1* gene predicted to encode an unknown protein with the N-terminal chloroplast transit peptide. *SWL1* expression was detected in various rice tissues at different developmental stages. However, immunoblot analysis indicated that *SWL1* protein accumulation was strictly regulated in a tissue-specific manner. In the *swl1* mutant, formations of grana and stroma thylakoids and prolamellar bodies were inhibited (Figure 2). This study revealed that *SWL1* is essential for the beginning of thylakoid membrane organization during chloroplast development. Furthermore, we provide a developmental perspective on the *nDart1*-promoted tagging line to characterize unidentified gene functions in rice.

Publication List

[Original paper (E-publication ahead of print)]

- Hayashi-Tsugane, M., Takahara, H., Ahmed, N., Himi, E., Takagi, K., Iida, S., Tsugane, K., and Maekawa, M. A mutable albino allele in rice reveals that formation of thylakoid membranes requires SNOW-WHITE LEAF1 gene. *Plant Cell Physiol.* 2013 Oct. 21.