



Title	Molecular biological studies on Arabidopsis DEAD-box RNA helicase 7 [an abstract of entire text]
Author(s)	刘, 跃林
Citation	北海道大学. 博士(農学) 甲第12250号
Issue Date	2016-03-24
Doc URL	http://hdl.handle.net/2115/61983
Type	theses (doctoral - abstract of entire text)
Note	この博士論文全文の閲覧方法については、以下のサイトをご参照ください。
Note(URL)	https://www.lib.hokudai.ac.jp/dissertations/copy-guides/
File Information	Liu_Yuelin_summary.pdf



[Instructions for use](#)

Molecular biological studies on *Arabidopsis* DEAD-box RNA helicase 7
(シロイヌナズナ DEAD-box RNA helicase 7 の機能に関する分子生物学的研究)

Liu Yuelin

Abstract

DEAD-box proteins comprise the largest family of RNA helicases, and exist in most organisms. They participate in a wide range of RNA processing events from transcription to RNA decay. To date, several DEAD-box proteins are known to regulate plant development through ribosome biogenesis. In this dissertation, I isolated AtRH7 as a common interactor of AtCSP2 and AtCSP3, and characterized the physiological and molecular functions of *AtRH7* in plant morphogenesis and abiotic stress tolerance.

1. AtCSP2 is a negative regulator of salt tolerance

Cold shock domain (CSD) proteins are RNA chaperones that destabilized RNA secondary structures. In previous study, one of the *Arabidopsis* CSD proteins, AtCSP2, was found to be induced during cold stress but negatively regulates freezing tolerance. Here, I found AtCSP2 negatively regulates salt tolerance in addition to cold stress. Interactome analysis showed that AtCSP2 interacts with proteins involved in variety RNA-associated events, suggesting the multi-functions of AtCSP2 in RNA metabolisms. Among the interactors, AtRH7 was selected for further functional characterization.

2. AtRH7 regulates plant growth and development and is involved in pre-rRNA processing.

In this study, the biochemical and genetic functions of AtRH7 were analyzed. The recombinant AtRH7 showed ATPase activity that was stimulated by addition nucleic acid, indicating AtRH7 is a functional RNA helicase. Phylogenetic analysis showed AtRH7 belongs to a subfamily that includes all *E.coli* DEAD-box RNA helicases. Heterologous expression of *AtRH7* complemented the cold-sensitive phenotype of *csdA* mutant. Promoter:GUS activity showed that AtRH7 was expressed ubiquitously in plants, with higher levels in rapidly developing tissues. Two knockout mutant lines displayed several morphological alterations such as short roots and pointed first true leaves, which were similar to several ribosome related mutants. In addition, aberrant floral development was also observed in *rh7* mutants. At low temperature (12°C), germination and growth of the *rh7* mutants were severely delayed; after prolonged exposure to

cold, the mutants developed extremely small, miss-shaped leaves. I examined the pre-rRNA processing by RNA blots and circular RT-PCR, and found that several rRNA precursors accumulated to higher levels in the mutants than in WT under both optimal and cold conditions. Taken together, the results suggest that AtRH7 affects pre-rRNA processing and plays an important role in plant growth under low temperatures.

3. AtRH7 is involved in plant abiotic stress tolerance

As AtCSP2 regulates stress tolerance, AtRH7 may also be involved in response to abiotic stress. Interestingly, the *rh7* mutants were more tolerant to freezing stress than WT only after cold acclimation, suggesting AtRH7 negatively regulates plant cold acclimation. The qRT-PCR revealed that the CBF pathway is not the main factor involved in AtRH7-mediated cold acclimation. Soluble sugar accumulated to higher levels in *rh7* mutant than in WT after cold acclimation, which may partially explain the increased freezing tolerance of *rh7* mutants. In addition to freezing stress, *rh7* mutants displayed enhanced tolerance against salt and drought. The microarray data revealed that several stress responsive genes were up-regulated in *rh7* mutant. Together, our results demonstrated that AtRH7 negatively regulates multiple abiotic stress tolerance in *Arabidopsis*.

Taken together, I concluded that AtCSP2 is a negative regulator of salt tolerance and physically interacts AtRH7. AtRH7 is a ribosome biogenesis factor that regulates plant growth and development under both optimal and cold conditions. AtRH7 negatively regulate plant freezing, salt and drought tolerance.