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学 位 論 文 内 容 の 要 旨
Abstract of Doctoral Thesis

博士の専攻分野の名称　博士（生命科学）　　氏　名　孫 惠慧
Degree requested　Doctor of Life Science　Applicant name

学 位 論 文 題 名
Title of Doctoral Thesis

Studies on proteasome function in response to environmental stress in Arabidopsis
（シロイヌナズナを用いた環境ストレス応答時のプロテアソーム機能に関する研究）

26S proteasome is a multicatalytic complex to selectively degrade damaged proteins, nonfunctional proteins and short life proteins to modulate organism's homeostasis and regulate activities of growth, development and adaption to various stresses. Regulation of proteasome proteolytic capacity is essential to adapt to different circumstances. Refer to publications proteasome activity can be regulated by self's subunit composition, complex conformational structure, proteasome interactor proteins (PiPs), post-translational modification and other mechanisms in response to different environmental stress. In this doctoral dissertation, I investigated the proteasome function in response to abiotic sugar stress and biotic pathogen-derived MAMP (microbe-associated molecular pattern) Flg22 stress. The results revealed that appropriate proteasome subunit composition and putative post-translational modification were involved in the regulation of proteasome function in response to environmental stress in Arabidopsis.

1. Studies on proteasome RPT2a subunit function and sugar stress in Arabidopsis.

Proteasome is a multiply-subunits protease; each subunit plays a crucial role in plants although most proteasome subunits function has not been well understood. On the base of RPT2a research background, which *RPT2a* gene transcription level was significantly increased under high sugar stress (microarray data). In my study, further function of RPT2a participats in sugar stress was elucidated. The results showed *rpt2a* knock out

mutant was hypersensitivity to high sugar stress compared with wide type (WT) and *rpt2b*；*rpt2a* mutant growth was completely arrested by high sugar threats; *RPT2a* gene was markedly transcriptionally promoted by sugar application; poly-ubiquitinated proteins was slightly accumulated in *rpt2a* mutant under 6% sucrose conditions compared with WT; transcription level of *RPT2a* gene in *gin2 (glucose insensitive 2)*, was not up-regulated by sugar application, indicating that RPT2a is involved in the hexokinase-dependent sugar response. Taken together, the above findings indicated that proteasome RPT2a subunit plays an essential in response to sugar stress.

2. Studies on proteasome function and immune response in Arabidopsis.

Different from mammal and yeast, most subunits of Arabidopsis proteasome are coded by duplicated genes, suggesting the highly heterogenous composition of proteasome in plant. In this study, a high-throughput method was established to identify detailed proteasome composition of paralogous and modified subunits with affinity-purification and combination of 2-dimentional electrophoresis and LC-MS/MS analysis. Moreover, the functional regulation of proteasome activity in response to biotic Flg22 stress signal was examined, which demonstrated that specific subunit composition and proteasome peptidase activity was affected by Flg22 treatment. Interestingly, mRNA expression of all peptidase genes, *PBA*, *PBB1/2*, *PBE1/2* and total proteasome amount were not changed by Flg22. These results proposed the fine-tune mechanism regulating proteasome function via putative post-translational modification in response to Flg22 treatment.

Taken together, proteasome function and activity were regulated by subunit composition and putative post-translational modification response to high sugar abiotic stress and Flg22 biotic stress.