



Title	Proteomics of Physcomitrella patens to elucidate the functions of 12-oxo-phytodienoic acid [an abstract of dissertation and a summary of dissertation review]
Author(s)	Luo, WeiFeng
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学位論文内容の要旨

博士の専攻分野名称：博士（農学）

氏 名：Luo Weifeng

学位論文題名

Proteomics of *Physcomitrella patens* to elucidate

the functions of 12-oxo-phytodienoic acid

(プロテオミクスを用いたヒメツリガネゴケにおける

12-オキソファイトジエン酸の機能解析)

Plant hormones regulate a wide variety of physiological events in plants. Many studies have demonstrated that plant hormones, such as auxin, cytokinins, and abscisic acid, also control physiological responses in a model moss, *Physcomitrella patens*. In *P. patens*, the first half of the octadecanoid pathway exists, however, jasmonic acid (JA), a plant hormone, is not synthesized. 12-Oxo-phytodienoic acid (OPDA), an intermediate of JA biosynthesis in flowering plants, inhibits the growth of *P. patens*. Moreover, wounding induces OPDA accumulation in *P. patens*. Thus, OPDA is suggested to be an important oxylipin as a signaling molecule in *P. patens*. Proteomic analysis of *P. patens* gametophores treated with OPDA showed that abundance of proteins is affected by OPDA in gametophores. These results can build a vague vision to understand the functions of OPDA to regulate protein expression. Since OPDA, not JA, was accumulated in response to wounding. OPDA is supposed to play an important role in the expression of genes and proteins, which are related to wounding response in *P. patens*. However the detailed function of OPDA is still unknown in plants, understanding the roles of OPDA in *P. patens* would help to elucidate OPDA functions in plants. In this research, proteomic analysis of *P. patens* protonema treated with OPDA was conducted to compare the OPDA functions in protonema with those in gametophore. Moreover, the comparison of proteomic data between wild-type and OPDA-deficient *P. patens* mutant with disrupted *PpAOS1* and *PpAOS2* genes (*aos* mutant) was done to elucidate the OPDA functions in response to wounding.

1. Proteomic analysis of wild-type *P. patens* protonema treated with OPDA

Proteomic analysis of wild-type *P. patens* protonema treated with OPDA showed that the abundance of 41 proteins was significantly altered by OPDA, with decreased levels for 40 proteins. The proteins for which abundance decreased in response to OPDA at the protonema developmental stage were mainly involved in the metabolism of proteins and carbohydrates. The inhibition for protein abundance is suggested to be a major physiological function of OPDA in *P. patens*. The inhibition of protein synthesis is

likely one of the significant physiological functions of OPDA in *P. patens*. This study also demonstrated that OPDA suppressed to accumulate histones and mRNA of histone genes at the protonema stage. To the best of my knowledge, this is first report that OPDA is involved in gene expression of histones. It is possible that suppression of histone expression at both steps, transcription and translation, is an OPDA-specific function in *P. patens* protonema. In *P. patens*, a subset of the physiological responses caused by OPDA is shown to differ between protonema and gametophore developmental stages.

2. Comparative proteomic analysis of wild-type *P. patens* and OPDA-deficient *P. patens* mutant after wounding

This study showed that the *aos* mutant accumulated proteins involved in protein synthesis in response to wounding, similar to the wild-type. Therefore wound-induced accumulation of proteins involved in protein synthesis is possibly unrelated to AOS-related physiological effects in *P. patens*. In contrast, the fold-changes of the proteins in the wild-type were significantly different from those in the *aos* mutant. The data suggested that wound-induced proteins found in only the wild-type, including proteins involved in amino acid metabolism, protein folding, photosystems, glycolysis, and energy synthesis, were influenced by AOS expression, which caused an increase in OPDA. Moreover it is suggested that *PpAOS* gene expression, which leads to an increase in OPDA, enhances photosynthesis and effective energy utilization in response to wounding in *P. patens*. OPDA was particularly shown to induce gene expression and protein abundance of chaperonins, which are related to correct protein folding. The wounding-responsive proteins in wild-type *P. patens* have also been accumulated in response to wounding in flowering plants, therefore the accumulation of these proteins in response to wounding may be a conserved physiological event in land plants.

Conclusions

OPDA and wounding significantly changed the abundance of proteins in *P. patens*. OPDA-mediated physiological responses are considered to be related to wound-induced responses in *P. patens*. OPDA stimulates various signaling systems. Various signaling triggered by wounding are probably involved in crosstalk with OPDA signaling in *P. patens*. The proteins that are presumably regulated by OPDA in wounded *P. patens* do not completely correspond to the proteins whose abundance is altered in *P. patens* treated with OPDA. The present studies could enhance our understanding of signaling in response to wounding as well as OPDA in *P. patens*.