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令和１年　１２月　１６日
Year　Month　Day

学 位 （ 博 士 ） 論 文 要 旨
(Doctoral thesis abstract)

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論文題目 Title	オオムギ種子胚のアブシジン酸応答におけるリン酸化シグナル伝達経路の大規模解析				

論文要旨 (和文要旨(2000 字程度)または英文要旨(500words))
※欧文・和文どちらでもよい。但し、和文の場合は英訳を付すこと。
Write a summary in Japanese (2000 characters) or in English (500words).
If the abstract is written in Japanese, needed to translate into English.

Plants developed dormancy and germination mechanism to germinate under favorable conditions. Seed dormancy mechanism has been impaired by breeding for uniform and synchronized germination. In some instances, such as cereal crops, the loss of the dormancy at grain maturity has created a widespread problem known as preharvest sprouting (PHS), which is the premature germination of grain in the spike following moist conditions. Therefore, the control of seed dormancy is essential for stable food production.

Abscisic acid (ABA) is a phytohormone and a major determinant of seed dormancy in plants. Seed dormancy is gradually lost during dry storage, a process known as 'after-ripening', and this dormancy decay is related to a decline in ABA content and sensitivity in seeds after imbibition. This study aimed at investigating the effect of after-ripening on ABA signaling and imbibiton in barley, cereal model species. Phosphosignaling networks in barley grains were investigated by a large-scale analysis of phosphopeptides to examine potential changes in response pathways to after-ripening. This study used freshly harvested (FH) and after-ripened (AR) barley grains which showed different germination ability and ABA sensitivity.

In ABA treatment, a total of 1,730 phosphopeptides were identified in barley embryos isolated from half-cut grains. A comparative analysis showed that 329 and 235 phosphopeptides were upregulated or downregulated, respectively after ABA treatment, and phosphopeptides profiles were quite different between FH and AR embryos. These results were supported by peptide motif analysis which suggested that different sets of protein kinases are active in FH and AR grains. Furthermore, in vitro phosphorylation assays confirmed that some phosphopeptides were phosphorylated by SnRK2s, which are major protein kinases involved in ABA signaling.
On the other hand, the LC-MS/MS analysis identified 2346 phosphopeptides in barley embryos, with 269 and 97 of them being up- or downregulated during imbibition, respectively. A number of

phosphopeptides were differentially regulated between FH and AR samples, suggesting that phosphoproteomic profiles were quite different between FH and AR grains. Motif analysis suggested multiple protein kinases including SnRK2 and MAPK could be involved in such a difference between FH and AR samples.

The different phospho-signalings between FH and AR embryo of barley were identified by two phosphoproteomic analysis. Although phospho-profiles were investigated on a large scale, the individual mechanism causing the difference between FH and AR embryos remained unknown. To understand the molecular mechanism of seed dormancy, I selected phosphoprotein for further analysis based on previous results. AFP2 is known as a negative regulator of ABA signaling. AFP2 degrades ABI5, one of the major dormancy regulators in ABA signaling. I hypothesized SnRK2 phosphorylates and regulates AFP2 to promote seed dormancy.

To clarify the function of ARP2, protein-protein interaction assay and phosphorylation assay between SnRK2 and AFP2 were performed, so far. Even the relationship between SnRK2 and AFP2 have been studied, but the effect of phosphorylation of AFP2 remain unclear. This should be revealed by further experiments.

Taken together, this study revealed very distinctive phosphosignaling networks in FH and AR embryos of barley, and suggested that the after-ripening of barley grains is associated with differential regulation of phosphosignaling pathways leading to a decay of ABA signaling.

(英訳) ※和文要旨の場合(300 words)

If the abstract is written in Japanese, needed to translate into English.(300 words)