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学位論文題目	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

学位論文の内容の要約

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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.

A putative SnRK2 substrate has been selected and confirmed by the protein-protein interaction and in vitro phosphorylation assay, so far. Even the relationship between SnRK2 and this putative substrate has been studied, but the effect of phosphorylation remains 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.