LET is an important factor affecting several aspects of the irradiation effect, such as cell survival and mutation frequency.1) The effect of LET on deletion induction increases up to an LET value of approximately 100 keV/µm2) while the mutation frequency increases abruptly and peaks at a particular LET value.1) The mechanism between LET and the corresponding mutation frequency is still unknown. In this study, we focused on the effect of LET on gene expression profiles.

Oryza sativa L. cv. ‘Nipponbare’ seeds were imbibed for 3 days. The seeds were irradiated with 22.5 or 50 keV/µm C ions or with 63 or 80 keV/µm Ne ions at a dose of 15 Gy. Embryos were sampled 2 h after irradiation. Gene expression profiles were generated using a rice 4×44K microarray RAP-DB (Agilent Technologies, Tokyo, Japan).

We defined 379 genes showing a significant change in expression level (FC > 2, and p < 0.05) at all four LETs as “LET-independent up-regulated genes” and 628 genes showing a significant change in expression level at an LET of 80 keV/µm, but not at 22.5 keV/µm, as “LET-dependent up-regulated genes”. A singular enrichment analysis (SEA) using a gene ontology (GO) analysis toolkit agriGO revealed that GO terms significantly enriched were different in LET-dependent up-regulated genes and LET-independent up-regulated genes (Fig. 1), which implies that some specific genetic pathway is induced depending on the complexity of DNA damage. Additionally, OsPCNA and OsPARP3 were found to be LET-dependent up-regulated genes (Fig 2). Although molecular functions of rice PARP family genes are largely unknown, some of them are involved in Ku-independent non homologous end joining (A-NHEJ) with Xrcc1 and DNA ligase III in animals3) and plants.4) OsPCNA forms a complex with OsXRCC1 in vivo.5) We assume that the high expression of OsPARP3 and OsPCNA is needed to repair the high-LET induced DNA lesions via Ku-independent NHEJ.

In this study, we found an LET-dependent change of gene expression profile, which is a clue to the clarification of the relationship between LET and mutation frequency.

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Fig 1. SEA analysis. The horizontal axis shows the distribution of the GO term, which is a percentage of genes having each GO term in the analyzed gene set. Blue, red, and white boxes indicate the distribution of LET-independent up-regulated genes, LET-dependent up-regulated genes, and whole genes (rice TIGR locus).

Fig 2. Response of OsPCNA and OsPARP3 gene expressions to heavy-ion beam irradiation. Fold changes in expression level of OsPCNA (A) and OsPARP3 (B) compared to the unirradiated control are shown. *: p < 0.05 (Student’s t-test).

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