Poster Presentation 2

P2-42

Distribution and shape of organelles visualized with fluorescent proteins in the agaricomycete *Pleurotus ostreatus*

Shivani Shivani, Takehito Nakazawa, Masahiro Sakamoto, Yoichi Honda
Graduate School of Agriculture, Kyoto University, Japan

Purpose: Fluorescent proteins serve as an efficient tool in observing organelle morphology and dynamics. Although changes in mitochondrial morphology during fruiting development has been suggested in agaricomycetes, it has yet to be observed in living cells. Visualizing organelles like endoplasmic reticulum (ER) and Golgi-equivalents (GEs) would be helpful in studying protein secretory mechanisms/ pathways, which has not been well studied in agaricomycetes. The aim of this study is to visualize organelle dynamics using fluorescent proteins in hyphal cells and fruiting bodies of the agaricomycete *Pleurotus ostreatus*.

Methods: Plasmids containing expression cassettes for various EGFP and mCherry fluorescent fusion proteins were introduced into *P. ostreatus* strain PC9. Fluorescence in various parts of hyphal cells and fruiting body was observed via fluorescence microscopy.

Results and Conclusions: EGFP fused with N-terminal 100 and 99 amino acids of Cit1A and Cit1B, respectively, were shown to localize in mitochondria of hyphal cells, gill, stipe, and basidiospores; the morphology was dependant on the cell type. The mCherry attached with peroxisomal targeting signal at C-terminus was observed only in hyphal cells suggesting its absence in fruiting bodies. Sec24 fused with EGFP at C-terminus, Sec24-EGFP, was likely localized at GEs in every observed cell. Sec13-EGFP was likely observed in the ER and GEs of the hyphal cells and at the nuclear pore outer rings, suggesting its association with nucleoporins. It was absent in stipe cells. The fluorescent fusion proteins used in this study would be useful in future cell biological studies on development and protein secretion in *P. ostreatus*.