E355 Cloning and Sequencing of the Genomic DNA Encoding Putative Mitogen-activated Protein Kinase from a Phytopathogenic Fungus *Botrytis cinerea*

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Mitogen-activated protein kinase (MAPK) which is activated by a wide variety of extracellular signals is an ubiquitous kinase conserved from fungi to mammals. In order to clone a MAP kinase gene from a phytopathogenic fungus *Botrytis cinerea*, a cell growth inhibition test has been used. A *Botrytis cinerea* genomic DNA library was prepared using pRS316 CEN shuttle vector and transformed into a MAP kinase defective yeast mutant EY1095 (*Δfus*3-8). When a yeast muant containing any fungal DNA fragment was treated with extracellular signal such as α -mating factor and thereby showed inhibited cell growth, it would suggest that transformed fungal DNA complements yeast MAPK gene. The 2087 bp fungal genomic DNA has been isolated with this strategy and fully sequenced. Comparison with the sequence databases using BLAST search program indicated that it is very much homologous to the yeast uracil DNA glycosylase gene. The reason why this gene is cloned with the pheromone sensitivity assay is currently being examined.

E356 Regulation of chitin synthase 3 of *Saccharomyces cerevisiae* in the vegetative cell cycle

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In *Saccharomyces cerevisiae*, the three chitin synthases participate in septum and cell wall formation of vegetative cells. Of these, the chitin synthase 3 (Chs3) is responsible for the synthesis of most chitin and chitin ring and is essential for cell division. The message of *CHS3* is preferentially expressed at G1 phase of the cell cycle. In this study, we examined the enzymatic activity of *CHS3* and the transcription of two positive regulator, *CHS4* and *CHS5* whereas the former corresponded well to its message level, the latter remained constant throughout the cell cycle. These results suggest that *CHS3* may be regulated temporally at transcriptional level and spacially at post-translational level. As its regulator, *CHS4* and *CHS5* involved in protein interaction with Chs3 but their expression pattern might intimate the existence of another protein or Chs4 and Chs5 might mediate third interaction of Chs3 with another regulator.