

Molecular Toolkit Development for Gene Expression and Gene Silencing Technologies in the Homobasidiomycete Fungi *Agaricus bisporus* and *Coprinus cinereus*.

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We have developed a “Molecular Toolkit” comprising interchangeable promoters and marker genes to facilitate transformation of homobasidiomycete mushrooms and subsequent analysis of gene expression. We will describe the testing of a wide range of promoters in both *Agaricus bisporus* and *Coprinus cinereus* when linked to a range of selectable and visual marker genes, along with the parameters required to successfully achieve foreign gene expression within these organisms. It has been previously demonstrated that a prerequisite for GFP expression in *A. bisporus* and *C. cinereus* is an intron. We describe the construction of an expression vector containing a multiple cloning site linked to an intron thus allowing different genes to be easily expressed in *A. bisporus* and *C. cinereus*. We report on the development of gene silencing technologies within *A. bisporus* and *C. cinereus*. In particular the serine protease has been targeted for gene silencing in *A. bisporus*. Serine protease has been implicated in post-harvest and age-related senescence of sporophores. On harvesting, mushrooms degenerate rapidly to give browned caps and loss of texture in the fruit body, and such problems can dramatically reduce saleability of the mushrooms. Suppression of genes involved in these pathways could increase mushroom shelf-life and profitability for mushroom growers, or help to further elucidate the complex biochemical pathways involved in post-harvest degradation. Progress will also be reported on gene silencing in *C. cinereus*.