

Molecular Tools in Filamentous Fungi

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Abstract

Recent progress in DNA-mediated transformation offers the opportunity for rapid development of the molecular genetics of filamentous fungi. Transformation provides a method for isolating specific genes by complementation of mutations using gene bank cloned in plasmid shuttle vector; also, it allows *in vitro* manipulation of cloned sequences and reintroduction of such sequences into the organism.

First transformation requires the presence on the transforming plasmid of a marker allowing selective growth of only transformed colonies. Selection may be by complementation of mutants alleles with cloned genes or by dominant resistance as for *Saccharomyces cerevisiae* and *Escherichia coli*: *Aspergillus nidulans* (*trpC*, *arg B*, *pyrG*, *amdS*), *A. niger* (*A. nidulans-amdS*), *Cephalosporium acremonium* (Tn903-G418 resistance), *Cochiobolus heterostrophus* (*A. nidulans-amdS*), *Magnaporthe grisea* (*A. nidulans-ArgB*), *Mucor racemosus* (*Leu*), *Neurospora crassa* (*qa-2*, *am*, *trp1*, *pyr4*), *Podospora anserina* (*Leu2*, *Ura5*), *Ustilago maydis* (Neomycin resistance).

A second requirement for a useful transformation system is the expression and stable maintenance of transporting genetic material. The rules established for unicellular organisms such as yeast do not necessarily apply to organisms with a syncytial structure in filamentous fungi, it seems that the mere presence of a replication origin (*ars* sequence) is not sufficient to allow stable autonomous replication of the plasmid. Stable maintenance is only achieved by integration into a host chromosome.