

## Isolation, Molecular Characterization and Location of Telomeric Sequences of Basidiomycete *Pleurotus ostreatus* var. *florida*

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The white rot fungus *Pleurotus ostreatus* is an edible basidiomycete of increasing biotechnological interest due to its ability to degrade both wood and chemicals related to lignin degradation products.

Telomeres are specialized structures at the end of all eukaryotic chromosomes. Ensure chromosome stability and protect the ends from degradation and from fusing with other chromosomes. Telomeres sequences are extraordinary highly conserved in evolution. The loss of telomeric repeats triggers replicative senescence in cells.

For identification of restriction telomeric fragments in a previously described linkage map of *Pleurotus ostreatus* var. *florida* (Larraya et al., 2000), dikaryotic and eighty monokaryotic genomic DNAs were digested with different restriction enzymes (*Bam*HI, *Bgl*II, *Hind*III, *Eco*RI, *Pst*I, *Sal*I, *Xba*I and *Xho*I) electrophoresed and transferred to nylon membranes. Numerous polymorphic bands were observed when membranes were hybridized with human telomeric probe (TTAGGG)<sub>132</sub> (heterologous probe).

Telomeric restriction fragments were genetically mapped to a previously described linkage map of *Pleurotus ostreatus* var. *florida*, using RFLPs identified by a human telomeric probe (tandemly repeating TTAGGG hexanucleotide).

Segregation of each telomeric restriction fragment was recorded as the presence vs. absence of a hybridizing band. Segregation data for seventy three telomeric restriction fragments was used as an input table to be analysed as described by Ritter et al. (1990) and by Ritter and Salamini (1996) by using the MAPRF program software. Seventeen out of twenty two telomeres were identified.

Telomere and telomere-associated (TA) DNA sequences of the basidiomycete *Pleurotus ostreatus* were isolated by using a modified version of single-specific-primer polymerase chain reaction (SSP-PCR) technique (Sohal et al., 2000). Telomeres of *Pleurotus ostreatus* contain at least twenty five copies of non-coding tandemly repeated sequence (TTAGGG).