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Presence of double stranded RNA in natural isolates of Pycnoporus cinnabarinus

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Presence of double stranded RNA in natural isolates of Pycnoporus cinnabarinus

Abstract

While studying the nucleic acids of different strains of *P. cinnabarinus* (Pc58, Pc470.3, Pc470.6), the presence of dsRNA molecules (1.7 kb to 3.6 kb) was detected in two of them.

Presence of double stranded RNA in natural isolates of *Pycnoporus cinnabarinus*

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While studying the nucleic acids of different strains of *P. cinnabarinus* (Pc58, Pc470.3, Pc470.6), the presence of dsRNA molecules (1.7 kb to 3.6 kb) was detected in two of them.

Pycnoporus cinnabarinus is a basidiomycete capable of degrading lignocellulose and is known as a white rot fungus. *P. cinnabarinus* strains Pc58, Pc470.3 and Pc470.6 were isolated from carpophores collected in the rain forest of southern Chile. In the laboratory these strains were grown in liquid PC medium for 15 days at 30°C without shaking. The mycelia were harvested on Whatman #1 paper and washed with distilled water. Then the mycelia were frozen at -20°C and ground in a mortar. Nucleic acids were prepared by the rapid lithium chloride method (Leach et al. 1987 FGN 34:32-33), except that three phenol:chloroform:isoamyl alcohol (25:24:1) and two chloroform:isoamyl alcohol (24:1) extractions were performed before precipitating with ethanol. Nucleic acids were resuspended in sterile distilled water and later were dialyzed against TE buffer (10 mM Tris-HCl, 1 mM EDTA) for 16 h. The preparations were than analyzed by electrophoresis on a 0.7% agarose gel followed by staining with ethidium bromide.

The electrophoretic analysis showed that strain Pc58 has only chromosomal DNA. Strains Pc470.3 and Pc470.6 have four bands moving faster than chromosomal DNA (Fig. 1). These bands correspond to dsRNA as determined by different tests with nucleases. These dsRNAs, denominated as L-dsRNA, M1-dsRNA, M2-dsRNA and S-dsRNA, have been purified by electrophoresis in low-melting-point agarose followed by CTAB extraction (Langridge et al. 1980 Anal. Biochem. 103:264-271). Their molecular sizes were approximately 3.6 kb, 2.7 kb, 2.3 kb and 1.7 kb respectively.

P. cinnabarinus strains Pc470.3 and Pc470.6 seem to have the same types of dsRNA. Furthermore, the presence of different dsRNA in the same strain has been observed as in other fungi (Myers et al. 1988 Curr. Genet. 13:495-501).

Since dsRNA is an indicator of the presence of viruses in fungi, our observations in *P. cinnabarinus* suggest the presence of a virus-like particle which does not seem to be associated with a visible alteration in phenotype of the fungus.

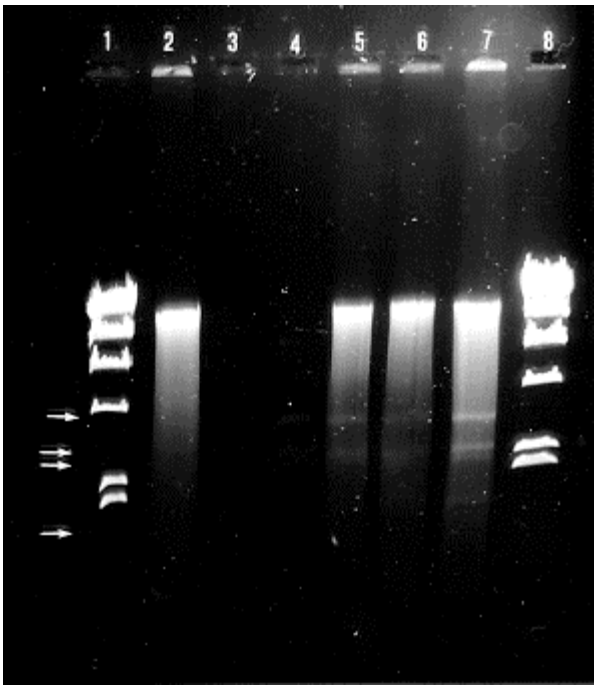

Figure 1. Agarose gel electrophoresis of nucleic acid preparations from natural isolates of *P. cinnabarinus*. Lanes 1 and 8: DNA digested with endonuclease *Hin*dIII; lane 2 strain Pc58; lane 4 strain Pc470.6 digested with DNAse I; lanes 5 and 6 strain Pc470.3; Lane 7 strain Pc470.6

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