ESRF

ESRF	**Experiment title:** STRUCTURE OF *Rigidoporus lignosus* LACCASE	**Experiment number**: MX129
Beamline: ID14-EH2	**Date of experiment**: from:10/07/03 to:11/07/03	**Date of report**: 22th June 2004
Shifts: 3	**Local contact(s)**: Stephanie Monaco	*Received at ESRF:*

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Report:

Background – Laccase is a multicopper blue oxidase that couple the four electron reduction of oxygen with the oxidation of a broad range of organic substrates, including phenols and arylamines. The enzyme is the object of intense biotechnological research, due to its employment in bioremediation of soils and water as well as in other biotechnological applications. We report here the cDNA and protein sequences, the post-translational modifications, the crystallization and X-ray structure determination of a laccase from the white-rot fungus *Rigidoporus lignosus*. The amino acids sequence deduced from cDNA clearly identified a pre-sequence of 21 residues representing the signal for extra-cellular localization. Mass spectrometry analysis performed on the salvage enzyme, confirmed the deduced sequence and precisely mapped two glycosylation sites at Asn337 and Asn435, determining the nature of the bound glycosidic moieties. The crystal structure was determined at 1.7 Å resolution from perfectly hemihedrally twinned crystals, by molecular replacement technique. While the overall structure closely resembled those reported for other fungal laccases, the analysis of the T2/T3 trinuclear cluster revealed an unprecedented coordination sphere for the T3 copper pair. No bridging oxygen ligand was present between the two T3 copper ions, which were no longer symmetrical coordinated.

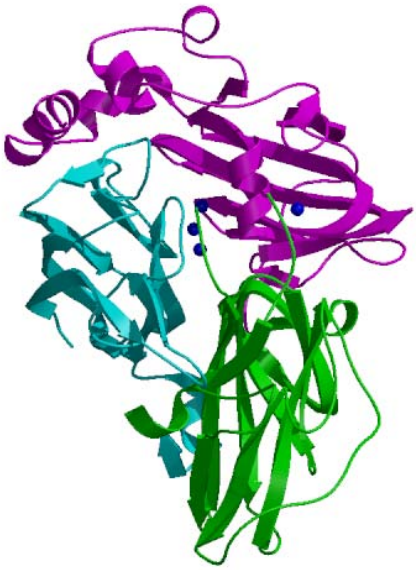

Figure 1: Ribbon representation of the laccase from Rigidoporus lignosus

1. Silvia Garavaglia, Maria Teresa Cambria, Marco Miglio, Santa Ragusa, Vito Iacobazzi, Ferdinando Palmieri, Chiara D'Ambrosio, Andrea Scaloni, and Menico Rizzi (2004). The structure of *Rigidoporus lignosus* laccase containing a full complement of copper ions, reveals an asymmetrical arrangement for the T3 copper pair. *Submitted*