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Degradation of triphenylmethane dyes using a temperature and pH stable spore laccase from a novel strain of *Bacillus vallismortis*

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HIGHLIGHTS

- The strain fmb-103 producing spore laccase is identified as *Bacillus vallismortis*.
- The spore laccase from strain fmb-103 is stable at high temperature.
- The spore laccase from strain fmb-103 is stable both at acidic and alkaline pH.
- The spore laccase can efficiently degrade triphenylmethane dyes.

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ABSTRACT

The characterization of a spore laccase from *Bacillus vallismortis* fmb-103, isolated from textile industry disposal sites, is described. The activity was 6.5 U/g of dry spore with ABTS as the substrate. The enzyme was quite stable at high temperature. It retained more than 90% of its initial activity after 10 h at 70 °C. The enzyme demonstrated broad pH stability in both acidic and alkaline conditions. There was almost no activity loss at pH 3 over an extended period of time, and the relative activity remained at 82% and 38% at pH 7 and pH 9 after 10 days. NaN_3, SDS, L-cysterine, Dithiothreitol, EDTA and NaCl inhibit the enzyme activity. Triphenylmethane dyes, including malachite green, brilliant green and aniline blue were efficiently degraded by the enzyme after 24 h in combination with a mediator with efficiencies of 76.84%, 96.56% and 81.17%, respectively. The reusability of spore laccase for decolorization dyes was also examined.

1. Introduction

Triphenylmethane dyes commonly cause environmental problems (He et al., 2010). Physical and chemical methods can be used for remediation, but these methods are costly and sometimes produce hazardous by-products. Biodegradation is an environment-friendly and economical way to modify chemical processes and substituents (Verma and Madamwar, 2003). For example, many fungi species are known to be capable of decolorizing dyes. These strains can produce different types of enzymes which catalyze the decomposition of the dyes. Laccase is an example of one of the important enzymes involved in the catalytic decomposition of the dyes (Hofrichter, 2002).

Laccases (EC 1.10.3.2) are blue multicopper oxidases that catalyze the oxidation of a variety of aromatic substrates, coupled with the reduction of molecular oxygen to water (Giardina et al., 2010). Laccases are found to be widely distributed among plants and fungi. Many fungal laccases have been isolated and characterized (Baldrian, 2006), and used in biotechnological applications, such as removal of dyes (Rodríguez et al., 2006). However, fungal laccases usually lose their activities rapidly at high temperature and pH (Baldrian, 2006). To date, only a few bacterial laccases have been studied. The most well-known bacterial laccase is CotA from *Bacillus subtilis* spores (Hullo et al., 2001). Compared to fungal laccases, bacterial laccases are much more thermotolerant and pH stable, two characteristics which are advantageous for biodegradation of industrial textile dyes (Singh et al., 2011). Hence, screening and identification of new sources of bacterial laccases should prove useful for industrial applications of enzyme catalyzed bioremediation. In the present study, the *Bacillus vallismortis* strain was identified for the first time to produce laccase. The characterization of a spore laccase from the newly isolated *B. vallismortis* fmb-103 is described. The spore laccase was also tested for its ability to decolorize triphenylmethane dyes.

2. Methods

2.1. Sample collection

Samples were collected from 30 different disposal sites of textile plants in JiangSu, China. Samples including sludge and

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