ABSTRACT

Laccase is an extensively studied enzyme because of its apparent use in several areas such as food, textile, paper and pulp industries. Laccase is the model enzymes for multi-copper oxidases can be used in bioremediation, beverage processing, ascorbic acid determination, baking, as a biosensor and to improve food sensory factors. Laccases have become significant industrial enzymes that can be used for a number of diverse applications. Laccase is broadly dispersed in the wide range of fungi as well as in bacteria. Laccase is more important because it oxidizes both toxic and nontoxic substrates.

Laccase producing microorganism, *Bacillus subtilis* (MTCC 2057), *Streptomyces lavendulae* (MTCC 6821), *Ganoderma lucidum* (MTCC 1039) and *Pleurotus ostreatus* (MTCC 1802), were subjected to laccase production. The laccase production was optimized in the presence 2% carbon, 3% nitrogen, temperature 30°C, incubation time 60 hr and pH 5. The laccases were partially purified using ammonium salt precipitation method at an of 80% saturation. The 3% nitrogen sources, peptone and beef extract and 2% carbon sources, glucose and sucrose in the medium considerably increased the laccase production. Purified laccase showed highest laccase activity by ammonium sulfate precipitation at a saturation of 80%. The molecular weights of partially purified laccase were ~ 80kDa. The dialysis of bacterial fraction showed 1.43U/ml and 1.59U/ml enzyme activity, total protein 0.034mg/ml and 0.036mg/ml. The dialyzed fungal fraction showed 1.86U/ml and 2.03U/ml enzyme activities, total protein 0.031mg/ml and 0.032mg/ml. Subsequently, laccase was purified by DEAE-cellulose anion exchange ion-exchange chromatography. After each step of purification of the bacterial and fungal enzyme, the specific activity was increased.

In this study, it was also identified that the purified laccase showed comparatively higher activity at pH 7 and high concentrations of metal $CuSO_4$ and $FeSO_4$ and organic solvent methanol. Enzymes were able to decolorize several dyes with over 80% and 90% reduction in color in case of methyl orange and trypan blue. This investigation thus conveys the higher yields of laccase produced from *B. subtilis* (MTCC 2057), *S. lavendulae* (MTCC 6821), *G. lucidum* (MTCC 1039) and *P. ostreatus* (MTCC 1802) strains with good potential for several industrial applications.