

E309 Purification and characterization of two laccases of the white-rot fungus
Coriolus hirsutus

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Two laccases produced by *Coriolus hirsutus* were purified to electrophoretic homogeneity by acetone precipitation and column chromatographies. The purification of laccases was 14.5- and 35.4-fold with an overall yield of 39.1%. Laccase I (LI) and laccase II (LII) fungus were monomeric glycoproteins with 47 and 16% carbohydrate content, isoelectric points of 7.5 and 4.2, and molecular masses of 74 kDa and 78 kDa, respectively. The N-terminal amino acid sequence of LII showed significant homology to the N-terminal sequences determined for laccases from other fungal laccases. However, the N-terminal amino acid sequence for LI showed low homology to those of laccases of other white-rot basidiomycetes. The highest rate of 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulfonate) (ABTS) oxidation for LI and LII were reached at 45°C, and the pH optima of these enzymes were varied depending on the substrate in the range of 2.0 and 4.5. The enzymes oxidized a variety of usual laccase substrates, including lignin-related phenol with similar affinity and had the highest affinity toward ABTS. Under standard assay conditions, the apparent K_m value of LI and LII toward ABTS was 10.0 and 8.1 μ M, respectively. They were completely inhibited by L-cystein and sodium azide but not by potassium cyanide, SDS, and thiourea.

E310 Characterization of Novel Antifungal Compounds Produced
in *Pseudomonas aurantiaca* KL1326 and its Mutants

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식물병원성 진균류 *Pythium ultimum*, *Botryosphaeri dothidea*와 동물성 병원균 *Candida albicans*에 대하여 강한 항진균 활성을 나타내는 균주 KL1326을 분리하여 동정한 결과, *Pseudomonas aurantica*로 확인되었다. KL1326이 생산하는 항진균 활성물질의 특성을 분석한 결과, *C. albicans*에 활성을 나타내는 20 kDa의 항진균 polypeptide와 식물 병원성 진균류에 대하여 Rf 값이 0.22와 0.29를 나타내는 항진균 활성물질들을 분리 확인할 수 있었다. 또한 20 kDa에 대하여 N-terminal 아미노산 서열을 분석, BlastP 검색을 하였을 때, hemolysin과 매우 높은 상동성을 나타내었다. 한편 KL1326을 대상으로 0.5~30 kGy의 γ -ray(Co^{60}) 조사선량으로 항진균 활성물질 생성능이 증간된 mutant와 항진균 활성이 약하게 나타내는 mutant를 선별한 후 TLC으로 비교한 결과, 두 종류의 항진균물질 중 한 종류가 절실된 것을 확인하였다.

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