

**ISOLASI, PURIFIKASI, DAN KARAKTERISASI *LACCASE*
EKSTRASELULER DARI JAMUR *Aspergillus oryzae* KKB4 YANG
MEMILIKI POTENSI MENDEGRADASI AFLATOKSIN B1**

INTISARI

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Laccase merupakan salah satu enzim ekstraseluler yang mampu mendegradasi aflatoksin B1 (AFB1), salah satu *mycotoxin* paling toksik di rantai pangan, sehingga upaya detoksifikasinya penting. *Aspergillus oryzae* KKB4 diisolasi dari koji dan terbukti mampu mereduksi AFB1. Produksinya dapat ditingkatkan dengan penambahan *inducer* pada media tumbuh, salah satu yang berpotensi yaitu ABTS.

Penelitian ini bertujuan mengetahui adanya enzim *laccase* pada *Aspergillus oryzae* KKB4 serta mengevaluasi karakteristiknya. Dilakukan presipitasi amonium sulfat serta dialisis sebagai upaya pemurnian, dilanjutkan uji ABTS, uji Bradford, SDS PAGE, profil suhu, pH, uji kinetika enzim, kofaktor, serta inhibitor *laccase* pada *Aspergillus oryzae* KKB4.

Hasil menunjukkan aktivitas *laccase* tertinggi pada sampel fraksi 60-80% amonium sulfat (6.44 U/ml), dengan aktivitas spesifik (284.85 U/mg), serta kemurnian 1.47fold. Suhu dan pH optimum *laccase* pada 30°C dan 4,5. Data kinetika *laccase* menunjukkan nilai Km sebesar 0,0194 mM dan Vmaks sebesar 7,4865 U/mg. Kofaktor *copper sulfate* dapat meningkatkan aktivitas *laccase* pada 0,1mM sebesar 64%, serta inhibitor *laccase* terbaik yaitu SDS pada konsentrasi 0,5mM. Hasil penelitian ini menunjukkan adanya potensi *laccase* pada *A. oryzae* KKB4 dalam mendegradasi AFB1.

Kata kunci: *Aspergillus oryzae* KKB4, aktivitas enzim, berat molekul, inhibitor, karakteristik enzim, kofaktor, *laccase*

**ISOLATION, PURIFICATION, AND CHARACTERIZATION OF
EXTRACELLULAR *LACCASE* FROM *Aspergillus oryzae* KKB4 WITH
POTENTIAL TO DEGRADE AFLATOXIN B1**

ABSTRACT

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Laccase is one of the extracellular enzymes that is capable of degrading aflatoxin B1 (AFB1), one of the most toxic mycotoxins in the food chain, therefore the detoxification of it is crucial. *Aspergillus oryzae* KKB4 was isolated from koji and has the capability to reduce AFB1 levels. The fungi production can be enhanced by adding inducers to the growth medium, the most potential one is ABTS.

This study aims to determine the presence of *laccase* enzyme in *Aspergillus oryzae* KKB4 and to characterize its properties. Ammonium sulfate precipitation and dialysis were performed for purification, followed by ABTS assay, Bradford assay, SDS-PAGE, temperature and pH profiling, enzyme kinetics assay, *laccase* cofactor and inhibitor assay.

The results showed the highest *laccase* activity present in the 60-80% ammonium sulfate fraction (6.44 U/ml), with a specific activity of (284.85 U/mg) and a purity of 1.47 fold. The optimum temperature and pH of *laccase* at 30°C and 4.5. The kinetic data for *laccase* showed a K_m value of 0.0194 mM and a V_{max} of 7.4865 U/mg. The cofactor copper sulfate can enhance *laccase* activity by 64% at 0.1 mM, and the best *laccase* inhibitor is SDS at a concentration of 0.5 mM. These research's results indicate the potential of *laccase* from *A. oryzae* KKB4 in degrading AFB1.

Keywords: *Aspergillus oryzae* KKB4, enzyme activity, molecular weight, inhibitors, enzyme characteristics, cofactors, *laccase*