Manganese Peroxidase Activity and Availability
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Applications of Manganese Peroxidase in Industry

Peroxidase enzymes (EC 1.11.1.7) are heme proteins that contain an iron (III) protoporphyrin IX prosthetic group that is the center of their redox activity. Molecular weight of these enzymes ranges from 30,000 to 150,000 Da (Hamid and Rehman, 2009). They catalyze the reduction of peroxides, such as hydrogen peroxide, followed by the oxidation of a variety of organic and inorganic compounds. Peroxidases are represented by several specific enzymes, such as NADH peroxidase (EC 1.11.1.1), glutathione peroxidase (EC 1.11.1.9), iodide peroxidase (EC 1.11.1.8), as well as manganese peroxidase (MnP; EC 1.11.1.13). The latter, produced by *P. chrysosporium,* a white rot basidiomycete, has a role in degrading lignin in rotting trees in order to liberate the cellulose for fungal nutrition. Because of this activity, it has been tested to determine its ability to catalyze the oxidation of several monoaromatic phenols and aromatic dyes in the presence of both divalent manganese and chelating buffers (Aitken and Irvine, 1989). MnP catalyzes the oxidation of Mn(II) to Mn(III) in the presence of Mn(III)-stabilizing ligands. The resulting Mn(III) complexes can then carry out the oxidation of organic substrates (Aitken et al., 1994).

Synthetic organic azo dyes are used commonly in different industries ranging from food, textile production, printing and pharmaceuticals. The majority of these dyes are recalcitrant, to colorize various raw materials. Certain dyes, their precursors and some aromatic amine metabolites produced through biotransformation of dye compounds have been shown to be carcinogenic. The release of dyes into the environment constitutes water pollution, and the wastewaters represent a serious environmental problem as well as a public health concern. Color removal, especially from textile wastewaters, has been a big challenge over the last few decades. Now enzymatic treatment, including manganese peroxidase, could be an economically attractive treatment that can effectively decolorize textile mill effluent (Chacko and Subramaniam, 2011).

Numerous other applications have been suggested for peroxidases. These include: organic and polymer syntheses, deodorization of swine manure, applications in the paper and pulp industry, as a component of biosensors, for analysis and diagnostic kits, enzyme immunoassays, and potentially for biofuel production processes (Hamid and Rehman, 2009). Analysis and diagnostic kits as well as immunoassays are primarily filled using horse radish peroxidase, produced from horse radish roots. Other applications require too much enzyme to be cost effective.

Limited availability of MnP has restricted use

Many of the described applications will require multiple ton quantities of MnP enzyme, quantities which are not currently available from enzyme suppliers. In fact, only research quantities of MnP are available from a small number of suppliers at quite high cost. Production is problematic because of adverse activity on single celled organisms *in vivo*. In addition, when purification of the enzyme is performed from

fungal cultures, the enzyme is a mixture of multiple isozymes (other forms of the enzyme with similar activity), which lower the overall effectiveness of the isozyme form of choice. Recombinant enzyme produced in a multicellular organism is much cleaner and this organism can mitigate the adverse activity by sequestering activity in a specific location in order to accumulate high levels of enzyme. This is particularly true of the corn production system (Clough et al., 2006) (www.infiniteenzymes.com).

Performance Activity of IE MnP vs. Other Sources

Infinite Enzymes' manganese peroxidase activity was compared with several commercially available sources purified from fungal cultures. The following graphs illustrate resulting activities.

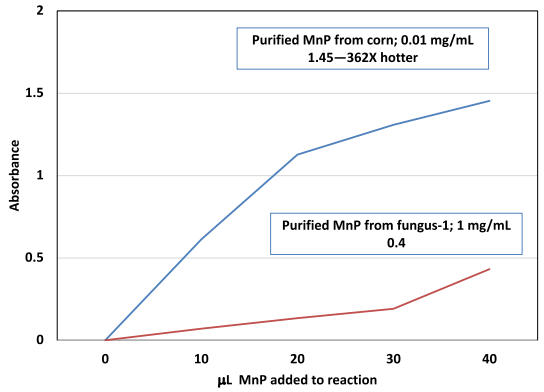

Figure 1: Purified recombinant MnP from corn seed is compared to a purified fungal Mn peroxidase using dimethoxyphenol (DMP) as the substrate. The corn-produced enzyme is >360 times more active on this

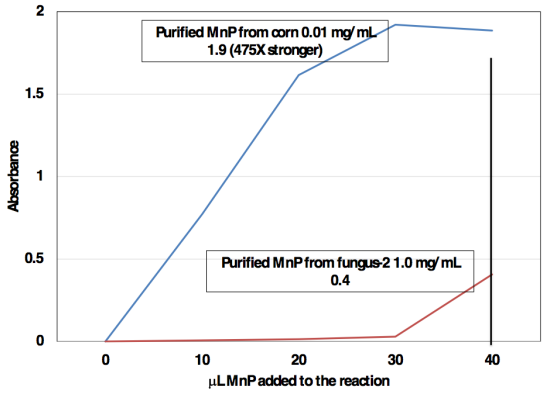

Figure 2: Purified recombinant MnP from corn seed is compared to a second source of purified fungal Mn peroxidase using dimethoxyphenol (DMP) as the substrate. The corn-produced enzyme is 475 times more active on this substrate.

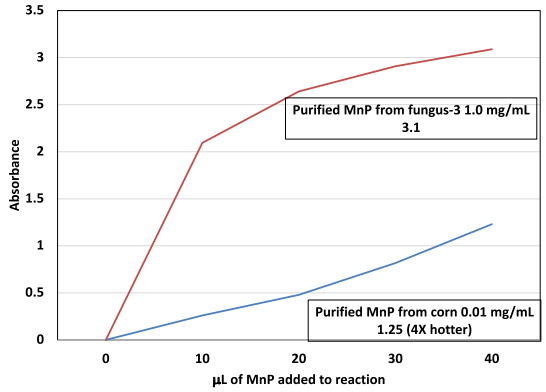

Figure 3: Purified recombinant MnP from corn seed is compared to a third purified Mn peroxidase from a different fungus using dimethoxyphenol (DMP) as the substrate. The corn-produced enzyme is 4 times more active on this

Conclusion:
Of the current sources of MnP, the recombinant protein from corn shows the highest activity. It is a SINGLE activity as well, with no other contaminating enzymes. In addition, one of the advantages of corn production system is the ease of scale-up—just planting more acres. Thus, the Infinite Enzymes produced enzyme can meet the large volume requirements of an industrial enzyme application.

Literature Cited:

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