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Integrated process for the extraction and preservation of laccase from Trametes versicolor growth media using aqueous biphasic systems

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PURPOSE OF THE ABSTRACT

Laccases are copper-containing oxidases that are produced by plants, bacteria, insects, and especially by white-rot fungus Trametes versicolor. Since laccases oxidize various aromatic substrates such as polyphenols, aromatic amines, and methoxy-substituted phenols [1], this fungus has been widely used in wood delignification [2], bioremediation [3], decolorization of dyes [3], among other applications. Nevertheless, the purification of laccase from the fermentation medium is still considered an expensive and laborious step, requiring techniques such as precipitation, flocculation, immobilization, gel filtration, chromatography, among others [4]. Some of the applied techniques also require time-consuming sample pretreatment. Therefore, there is a crucial need for the development of cost-effective techniques to recover and purify laccase from fermentation media, as well as to find effective preservation solvents. To attain this aim, aqueous biphasic systems (ABS) can be used. ABS are considered a valid technique for the extraction, concentration, and purification of proteins mainly due to their high-water content [5].

In this work we studied ABS as effective platforms for the extraction of laccase from T. versicolor fermented medium, followed by the use of the ABS coexisting phases as preservation media. Commercial laccase from microorganisms such as Aspergillus oryzae (Novozym ® 51003) is preserved in a mixture containing water (66%), Propylene glycol (PPG) (25%), Sucrose/Glucose (4%) and Glycine (2%). Based on this information, ABS composed of ionic liquids (ILs) or amino acids and polymers, such as polypropylene glycol 400 (PPG 400) and polyethylene glycol (PEG 400 and 600) were investigated. The first step consisted in the screening of the best components that could form ABS in the presence of a commercial laccase solution. These systems were characterized by the determination of the respective phase diagrams at several temperatures, comprising the respective tie-lines, tie-lines lengths and mixture compositions required for given concentration factors. The partition of a commercial laccase was then determined in these ABS, and the best ones were finally tested as new platforms for the extraction of laccase directly from the T. versicolor growth media. The phase enriched in laccase was then evaluated as the preservation medium of laccase. These systems are biocompatible in nature and formed by polymers and amino acids. The gathered results show that designed ABS can act as sustainable separation processes that could be integrated with the preservation step.

FIGURES

FIGURE 1

FIGURE 2

KEYWORDS

ABS | thermoreversible | Waste valorization | Ionic liquids

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