Bioresource Technology 130 (2013) 801–804

Contents lists available at SciVerse ScienceDirect

Bioresource Technology

journal homepage: www.elsevier.com/locate/biortech

Short Communication

One-pot bioconversion of sucrose to trehalose using enzymatic sequential reactions in combined cross-linked enzyme aggregates

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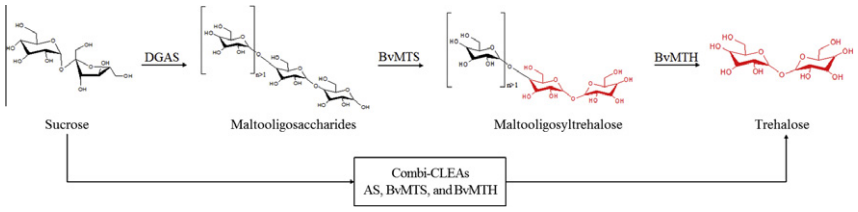
HIGHLIGHTS

- Enzymatic one-pot bioconversion of sucrose to trehalose by combi-CLEAs.
- Development of combi-CLEAs using three enzymes, DGAS, BvMTS, and BvMTH.
- Optimization of various factors for the generation of combi-CLEAs.
- Reusability of DGAS–BvMTS–BvMTH combi-CLEAs was determined.

GRAPHICAL ABSTRACT



ARTICLE INFO

Article history:
Received 6 August 2012
Received in revised form 25 December 2012
Accepted 26 December 2012
Available online 3 January 2013

Keywords:
Amylosucrase
Combined cross-linked enzyme aggregates (combi-CLEAs)
Maltooligosyltrehalose synthase
Maltooligosyltrehalose trehalohydrolase
Trehalose

ABSTRACT

Amylosucrase (AS), maltooligosyltrehalose synthase (MTS), and maltooligosyltrehalose trehalohydrolase (MTH) were used in combined cross-linked enzyme aggregates (combi-CLEAs) to achieve one-step bioconversion of sucrose to trehalose. Combi-CLEAs of three enzymes were successfully established with acetone and glutaraldehyde (GA) as a precipitant and a cross-linker, respectively. The optimum enzyme ratio was 8:0.5:0.5 (AS, 4 mg:MTS, 0.25 mg:MTH, 0.25 mg). To improve trehalose production, bovine serum albumin was co-aggregated with enzymes as a proteic feeder. The trehalose production yield of combi-CLEAs was about 8% in each cycle on the basis of substrate added up to 400 mM. Finally, the combi-CLEAs used in this experiment showed reusability of about five cycles without any activity loss.

1. Introduction

There is an increasing demand for immobilized biocatalysts as useful tools for biotransformation in green and sustainable industries. A significant advantage of immobilized biocatalysts is the increased thermal and operational stability conferred on enzymes, allowing their uses at relatively severe conditions than their soluble counterparts. In addition, the reusability extends the benefit of immobilized biocatalysts. Recently, there has been much interest in cross-linked enzyme aggregates (CLEAs) as an alternative to conventional immobilization methods on solid carriers (Cao et al., 2001; Schoevaart et al., 2004).

In general, CLEAs are prepared by precipitating the enzyme with an addition of a salt or an organic solvent followed by cross-linking with a bifunctional cross-linking agent. CLEAs result in insoluble biocatalysts with highly concentrated enzyme activity, great stability, and low production costs (Cao et al., 2001). Recently, it has been shown that CLEAs can catalyze a sequence of reactions. These forms of CLEAs have been called combi-CLEAs. In combi-CLEAs, heterogeneous populations of enzymes can be simultaneously confined in the same aggregate and they perform a versatile cascade or non-cascade bioconversion (Taboada-Puig et al., 2011).

Trehalose is a non-reducing disaccharide, which serves not only as a carbohydrate reserve but also as an agent that protects against

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0960-8524/$ - see front matter © 2013 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.biortech.2012.12.162