

Phosphorylase to produce glycosides

Ghent Bio-Energy Valley, a consortium of research laboratories of Ghent University, is seeking partners interested in the enzymatic production of glycosides such as glycolipids and cellobiose derivatives.

Introduction

Cellobiose derivatives have applications as chromogenic substrates or inhibitors of cellulases. Glycolipids, in turn, are high value products which are used as clean and biodegradable biosurfactants in detergent formulations. To date, these products are mainly synthesized via chemical or enzymatic glycosylation reactions. However, chemical synthesis is laborious, has low yields and generates toxic waste. The general industrial feasibility of using enzymes for glycosylation reactions is usually very low.

Technology

Researchers at Ghent University have identified a cellodextrin phosphorylase derived from the bacterium *Clostridium stercorarium* that can efficiently glycosylate glucose derivatives, using either glucose-1-phosphate or galactose-1-phosphate as donor substrate. The enzyme can be applied for the synthesis of glycosides such as cellobiosides and lactosides in an industrial context.

Applications

Enzymatic and/or recombinant methods to produce high value glycosides such as, among others, alkyl/aryl cellobiosides and lactosides, cellobiose and lactose lipids,.

Advantages

- industrially feasible, high yield, enzymatic/recombinant production method of high value compounds;
- less laborious and less toxic waste compared to chemical procedures to produce said compounds.

Status of development

Heterologous expression of the cellodextrin phosphorylase in *E. coli* and purification has been achieved. Activity assays were performed with 5% purified enzyme in the presence of glucose-1-phosphate or galactose-1-phosphate as donor and several acceptor molecules. The amount of inorganic phosphate released from the donor was measured and HPLC analysis revealed the existence of high value products. The degree of glycosylation was confirmed by MS-analysis and the high value products were purified.

Partnership

UGent is seeking a partner for further validation and upscaling of the present methodology.

Intellectual property

A PCT application covering a method to produce glycosides using the cellodextrin phosphorylase from *Clostridium stercorarium* and specified acceptors was filed on May 19, 2011 (priority date May 21, 2010)

Glycolipid production at 45 °C and pH 6.5*

Donor	Acceptor	Activity (%) [#]	Product	Solubility	Yield (%) [§]
Glc1P	Cellobiose	100	Cello-oligosaccharide	+	50
Glc1P	Glucolipid	126	Cellobiolipid	-	80
			Cellotriolipid	-	55
Glc1P	Sophorolipid	36	Glucosophorolipid	-	75
			Cellobiosophorolipid	-	50
Gal1P	Glucolipid	11	Lactolipid	+	50

* using 20 mM of acceptor and either 30 mM Glc1P or 100 mM Gal1P as donor

[#] relative to the activity on cellobiose, [§] relative to the acceptor concentration

The Inventors

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References

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Keywords

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