Enzymatic synthesis of sugar fatty acid esters in monophasic solvent

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Abstract

Enzymatic synthesis of sugar fatty acid esters was investigated in organic solvent using *Candida rugosa* lipase. To overcome sugars insolubility in organic solvent, sugar absorption procedure was done on a silica gel. The product yield was determined by using Ion Chromatography, with various factors such as reaction time, enzyme fatty acid molar ratio, number of carbon in fatty acid.

Introduction

Lipase has proved to be of great interest in the industrial flavour ester synthesis. Ester of sugar and fatty acid has been employed widely as surfactants in food, detergent, and pharmaceutical application. The synthesis of sugar esters using lipase has been studied extensively in recent years and enzymatic synthesis has provided regio- and stereoselective products, as compared with chemicals synthesis. In this work, various parameters affecting enzyme activity such as temperature, pH, organic solvent, along with the influence of sugar/acyl donor molar ratio, enzyme concentration, number of carbon in fatty acid, and reaction time on the production of glucose laurate were studied. In recent years, much research has centered on the conduct of enzyme reactions in organic solvent. Biocatalysis in organic media has been the object of intensive basic and application such as biphasic liquid systems, reverse micelles and monophasic liquid systems. During recent years, the biotechnological use of lipase in nonoaqueous media has become a common approach in industrial application. It is well known that lipase exhibit a high catalytic activity in water-restricted environments.

Material and Methods

Analysis of sugar fatty acid esters by Ion Chromatography

IC analysis was carried out using the PAD II which use the highest value for E 2 (+0.75V) and the lowest value for E 3(-0.25V). A CarboPac PA 1 (4 X 50mm) Guard Column was used with the CarboPac PA 1 (4 X 250mm) Analytical Column. The guard column was to prevent sample contamination from eluting onto the analytical column. The products were eluted with 200mM NaOH buffer at 0.5l/min flow rate, 80 kgf/cm2 pressure. All samples were filtered through membrane filter (0.45m) and degassed under vacuum.

Synthesis of sugar fatty acid ester by lipase

20 mg silica gel was added to a solution of the glucose (50 mg) in 4ml methanol was evaporated. The reaction mixture consisted of 50 mg fatty acid, 5 mg lipase from *Candida rugosa* preparation which were aced to 4 ml hexane. The reaction time was 5 hours at room temperature under constant stirring. The products were determined by ion chromatography.

Effect of sugar to fatty acid molar ratio on synthesis of sugar fatty acid ester

20 mg silica gel was added to a solution of the glucose (50 mg) methanol was evaporated. A typical esterification reaction was carried out in solution containing lipase from *Candida rugosa* (5 mg), hexane (4 ml), lauric acid and glucose

Effect of fatty acid chain length on synthesis of sugar lauric acid ester by lipase

A typical esterification reaction was carried out in solution containing lipase from *Candida rugosa* (5 mg), hexane (4 ml), fatty acid(50mg) and glucose. The rate of fatty acid/sugar was 1/3. The sample was taken by IC methods.

Results and Discussions

In order to study the influence of sugar to acyl donor molar ratio on the glucose

laurate production, different glucose concentration to lauric acid molar ratio were used. While lipase from *Candida rugosa* seems to be more efficient in this type of reaction. The best initial molar ratio was 1/5. The production was increased at the higher degree of fatty acid concentration. If fatty acid ratio is not different, the production was enhanced by decreasing sugar concentration. As already reported, it can be seen that in all cases the reaction were dependent in the chain length of acyl donor. For short chain length of fatty acid the reaction rates gradually increase to capric acid (C_{10}). For short chain length of fatty acid, the reaction rates gradually increasedto capric acid. The highest conversion rate obtained using a lignoceric acid, 22.6%. With capric acid and oleic acid, the product yield were similar to the product yield about 17%.

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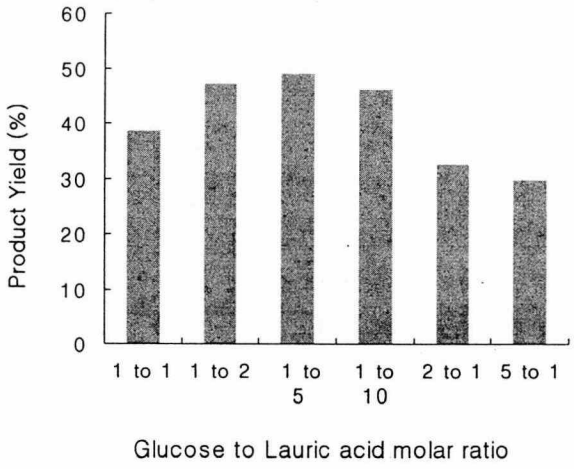

Fig. 1. Effect of glucose to lauric acid molar ratio on the synthesis of glucose laurate. 20 mg silica gel was added to a solution of the glucose (50 mg) methanol was evaporated. A typical esterification reaction was carried out in solution containing lipase (5 mg), hexane (4 ml), lauric acid and glucose.

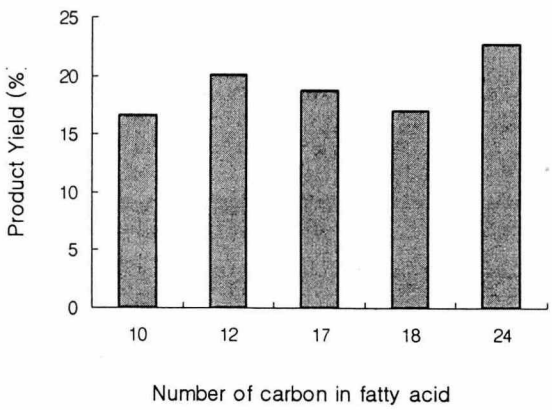

Fig. 2. Effect of the number of carbon in fatty acid on activity of lipase. Reaction mixture contained fatty acid and glucose at 1:3 ratio. Fatty acid such as capric acid (C_{10}), lauric acid (C_{12}), heptadecanoic acid (C_{17}), oleic acid (C_{18}), and lignoceric acid (C_{24}) was used.