

## Enzymatic Synthesis of Palm Based Fatty Amides

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### Introduction

Fatty monoethanolamides, a class of non-ionic surfactants derived from a reaction between triglycerides or fatty acid methyl esters (FAMES) or free fatty acids and various primary amine alcohols can be synthesised enzymatically (3,5), under milder conditions than the energy intensive chemical reaction (1,2). Lipase catalysed alkanolamide synthesis is studied, owing to the importance of alkanolamides as detergents, emulsifiers and intermediates to produce oleochemicals. The main objective of the study is to obtain, optimised and controlled reaction system of fatty monoethanolamide production of palm kernel oil fraction as substrate.

### Materials and Methods

**Screening:** Few types of immobilized enzymes and native enzyme were screened in the amidation reaction between free fatty acid and monoethanolamine in organic solvent (hexane). **Enzymatic synthesis:** Reactions were set up at equimolar ratio of reagents, palm kernel olein (PKL) and monoethanolamine (MEA) and in the presence of enzyme at 37°C for 72 hours in shaker bath with 150rpm shaking rate. **Analysis of products:** The purified of various individual fatty monoethanolamides were obtained after isolation and purification process. The various fatty monoethanolamides standards and PKL monoethanolamide mixture were characterised by infrared spectroscopy (IR), thin-layer chromatography (TLC), nuclear magnetic resonance (NMR) and gas chromatography (GC) (4,5). Gas chromatograph was also used for the quantitative analysis to calculate percent yield by using internal standard method. **Comparative study:** Five different enzymes were used in this study at 37°C with 1:1 and

1:3 ratio of PKL: MEA to investigate the selectivity of enzyme towards the various fatty monoethanolamides in product mixture. **Kinetics study:** This was done to determine the  $V_{max}$  and  $K_m$  values for different enzymes and different substrates to further confirm the selectivity of enzyme. **Optimisation studies:** Five reaction parameters were studied (temperature, time, mole ratio, organic solvent and water removal) to obtain the best condition for the optimum yield in the amidation reaction system.

### Results and Discussion

In the screening study, immobilised enzymes such as Lipozyme IM, Novozyme 435 and Amano PSC-lipase were found produced better yield compared to the native enzyme, *Candida rugosa* (4). Mixture of palm based fatty monoethanolamide was successfully obtained from the reaction between palm kernel olein (PKL) and monoethanolamine (MEA) in the presence of enzyme at 37°C and 150rpm shaking rate with better yield than the control experiment (without enzyme). The purified of various individual fatty monoethanolamides were also obtained from the reaction between free fatty acids ( $C_{10}$ - $C_{18}$ ) and MEA after the isolation and purification processes. In the comparative study, Amano PSC-lipase was observed to have good selectivity towards  $C_{18}$  fatty acid (4). This was further confirmed by kinetics study whereby the  $K_m$  value for  $C_{18}$  fatty acids was lower when PSC-lipase was used than the other enzyme (Novozyme 435). The optimal yield (90%) was achieved at 60°C reaction temperature after 24 hours incubation time in the presence of excess of amine (1: 5 PKL: MEA). Lipase worked better in hydrophobic solvents, which have higher log P compared to hydrophilic solvents. Hexane was found to be the best sol-

vent for amidation reaction in the system. The addition of 0.1 gram desiccant (molecular sieve) could increase the yield.

### Conclusions

Fatty monoethanolamides were produced by using lipases from the amidation reaction between palm kernel olein and primary amine alcohol, MEA in organic solvent system. Products were qualitatively and quantitatively detected by GC analysis to determine the optimum yield at optimum condition.

### Benefits from the study

Environmental friendly, enzymatic synthesis of biodegradable surfactant from the cheap raw material, palm oil fraction can be used as a potential route to replace the energy intensive chemical procedures in the biotechnology industrial.

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### Graduate Research

None.