Enzymatic Hydrolysis of Fish Oil with *Candida cylindracea* Lipase: A Comparison of Reaction Systems

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Omega-3 fatty acids EPA and DHA, plays a significant role in human health. Their main source is marine food, and enzymatic hydrolysis of fish oil constitutes a biotechnological alternative for its obtaining. This study compared three reaction systems for fish oil hydrolysis using *Candida cylindracea* lipase (CCL, LYVEN), which were, I: hydrolysis with soluble lipase (S-CCL), II: hydrolysis with immobilized lipase on octyl-agarose (O-CCL) and III: hydrolysis with O-CCL using solvated reaction medium. In reaction system I and II, the aqueous phase was pH 7 phosphate buffer and the organic phase fish oil. In reaction system III, the aqueous phase was a 40% polyethylene glycol / phosphate buffer pH 7 solution, and the organic phase was 1:4 fish oil / hexane solution. Hydrolysis were carried out for 48 hours at 37°C, pH 7 and 35 U of enzyme per gram of fish oil, with a 1:1 aqueous/organic phase ratio. After reaction, residual enzyme activity was measured with p-nitrophenyl butyrate as substrate. In reaction system I, the fatty acids hydrolysis yield was 65.0% and the concentration of EPA+DHA glycerides in the reaction product was 48.0%. Reaction system II yielded only 61.7% of fatty acids and 35.2% of EPA+DHA glycerides, while in reaction system III fatty acids hydrolysis yield was 94.0% and EPA+DHA glycerides concentration was 53.7%. In reaction system I, residual activity for S-CCL was 3.5 U/mg protein, corresponding to 10% of the initial activity, and 36.8 U/g carrier corresponding to 92% of the initial activity for O-CCL in reaction system II. In reaction system III, O-CCL presents 106 U/g carrier as residual activity, corresponding to 264% of the initial activity, revealing the hyperactivation of the lipase in that environment. Our results demonstrate that the use of solvent / co-solvent in the reaction medium is favourable for obtaining high hydrolysis yield and concentration of EPA+DHA glycerides in the reaction product, from fish oil hydrolysis with CCL lipase immobilized on octyl-agarose. Along with lipase hyperactivation, the elevated residual activity suggests the potential for its reuse in industrial application.