**Immobilization of *Rhizopus oryzae* lipase in corn cob powder for application in dietetic triglicerydes synthesis**

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**Highlights**

* ID of physical adsorption immobilized derivative was 28.55%.
* ID of covalent binding immobilized derivative was 34.47%.
* Corn cob powder is a successful immobilization carrier for lipase.

**1. Introduction**

Lipases are glycerol ester hydrolases (EC 3.1.1.3) and are widely employed as biocatalysts in Enzymatic Biotechnology. These enzymes are commonly used for oils and fats modification in the immobilized form. When immobilized, they are easily recovered from the reaction medium. A great variety of methods are used to immobilize enzymes, of which physical adsorption and covalent binding are the most important [1]. Therefore, the objective of this work was to immobilize *Rhizopus oryzae* lipase in corn cob powder in order to apply the immobilized derivative to the synthesis of dietary triglycerides by enzymatic acidolysis of grape seed oil.

**2. Methods**

*2.1 Immobilization of* Rhizopus oryzae *lipase*

Immobilization by physical adsorption (PA) was carried out according to Costa (2015) [2]. Immobilization by covalent binding (CB) was carried out according to Bassan et al. (2016) [3].

*2.2 Enzymatic acidolysis of grape seed oil*

The immobilized derivatives (10% w/w) were used in acidolysis of grape seed oil with capric acid (C10:0) at molar ratio 1:3 in batch reactor (6 x 3 cm) for 24 h (45 °C, 800 rpm). The product was analysed by CG – FID. The incorporation degree (ID, %) was calculated according to equation 1:

$ID(\%)=(\frac{MFA}{TFA})\*100$ (Equation 1)

MFA: moles of medium fatty acids (C10:0); TFA: moles of total fatty acids.

**3. Results and discussion**

The results of ID (%) are presented in Table 1. The ID obtained for the immobilized derivative by PA was 28.55%. Costa et al. (2018) [4] obtained an ID of 34.8%, using a different immobilization carrier (Eupergit) for olive oil and capric acid acidolysis. The lower ID obtained in this work can be justified by the difference in the carrier material and the substrate used.

**Table 1.** ID (%) of the fatty acid (C10:0) from *Rhizopus oryzae* lipase immobilized derivative

|  |  |  |
| --- | --- | --- |
| **Fatty acid** | **Fatty acids (%)** | **Moles** |
| **PA** | **CB** | **PA** | **CB** |
| C10:0  | 19.83 | 24.52 | 0.12 | 0.14 |
| C16:0  | 8.03 | 5.68 | 0.03 | 0.02 |
| C18:0  | 2.75 | 1.95 | 0.01 | 0.01 |
| C18:1n9C  | 20.45 | 19.95 | 0.07 | 0.07 |
| C18:2n6C  | 45.64 | 44.63 | 0.16 | 0.16 |
| C18:3n3  | 3.31 | 3.28 | 0.01 | 0.01 |
| **Sum**  | 100.00 | 100.00 | 0.40 | 0.41 |
| **ID (%)** |  |  | **28.55** | **34.47** |

For the CB immobilized biocatalyst, the ID was 34.47%. Considering that the maximum ID for 1,3 specific lipase dietary triglyceride synthesis is 66.66%, the obtained ID was satisfactory. According to Bassan et al. (2018) [5], who performed the enzymatic acidolysis of grape seed oil using a commercially immobilized lipase (Lipozyme RM IM), the ID was of 34.53%. Our results are similar to that obtained by Bassan et al., which shows the potential of the corn cob powder as a carrier for lipase immobilization.

**4. Conclusions**

The use of corn cob powder as a carrier for immobilization of lipases presents great potential for the application in dietary triglycerides synthesis.

**5. Acknowledgements**

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**References**

[1] SHELDON, R. A.; PELT, S. Enzyme immobilization in biocatalysis: why, what and how. Chemical Society Reviews, v. 42, n. 15, Mar. 2013.

[2] COSTA, D. M. Sabugo de milho como suporte para imobilização de lipase. 2015.157 f. Tese (Doutorado em Engenharia de Processos) – Universidade Tiradentes, Aracajú/SE, 2015.

[3] BASSAN, C. J.; SOUZA, B. M. T.; PEIXOTO, G.; CRUZ, P. Z. C.; GALÁN, M. P, J.; VAZ, S. B .A.; GARRIDO, S.S.; FILICE, M.; MONTI, R. Immobilization of trypsin in lignocellulosic waste material to produce peptides with bioactive potencial from whey protein. Materials, v. 9, n. 5, 2016.

[4] COSTA, C. M.; OSÓRIO, N. M.; CANET, A.; RIVERA,I.; SANDOVAL, G.; VALERO,F.; FERREIRA-DIAS, S. Prodution of MLM Type Structured Lipids From Grapeseed Oil Catalyzed by Non-Commercial Lipases. European Journal of Lipd Science and Technology, v. 120, 2018.

[5] BASSAN, N. RODRIGUES, R. H.; TECELÃO, C.; MONTI,R.; FERREIRA-DIAS, S.; PAULA, A. V. Enzymatic modification of grape (Vitis vinifera L.) seed oil aimig to obtain dietary triacylglycerols in a batch reactor. LWT - Food Science and Technology, 2018.