### Accepted Manuscript

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PII: S0141-8130(18)33428-7

DOI: doi:10.1016/j.ijbiomac.2018.09.141

Reference: BIOMAC 10576

To appear in: International Journal of Biological Macromolecules

Received date: 6 July 2018

Revised date: 10 September 2018 Accepted date: 22 September 2018

Please cite this article as: Wesen Adel Mehdi, Atheer Awad Mehde, Olcay Severgün, Soner Çakar, Mahmut Özacar, Lipase-based on starch material as a development matrix with magnetite cross-linked enzyme aggregates and its application. Biomac (2018), doi:10.1016/j.ijbiomac.2018.09.141

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### **ACCEPTED MANUSCRIPT**

## Lipase -based on starch material as a development matrix with magnetite cross-linked enzyme aggregates and its application

# Wesen Adel Mehdi <sup>a,\*</sup>, Atheer Awad Mehde <sup>a</sup>, Olcay Severgün<sup>a</sup>, Soner Çakar<sup>b</sup>, Mahmut Özacar<sup>a,c</sup>

### **ABSTRACT**

The Fe<sub>3</sub>O<sub>4</sub> magnetic nanoparticles were prepared by precipitating ferrous ion (Fe<sup>2+</sup>) and ferric ion (Fe<sup>3+</sup>)in alkaline solution. The Fe<sub>3</sub>O<sub>4</sub> magnetic nanoparticles were modified by tannic acid. After functionalization process, two methods were used to immobilize Lipase on Fe<sub>3</sub>O<sub>4</sub> magnetic nanoparticles. In the first method, novel tannic acid magnetic cross-linked enzyme aggregates of lipase (TA-MNPs-CLEAs) were synthesized by cross-linking of lipase aggregates and starch as co-feeder with Fe<sub>3</sub>O<sub>4</sub> magnetic nanoparticles improved by tannic acid (TA-MNPs). In the second method, the lipase was successfully immobilized on the surface of TA-MNPs. The properties of Fe<sub>3</sub>O<sub>4</sub> and its modified forms were examined by Fourier transform infrared spectroscopy (FTIR), X-ray diffraction (XRD), vibrating sample magnetometer (VSM), scanning electron microscopy (SEM) and zeta potential measurements. Novel TA-MNPs-lipase and TA-MNPs-CLEAs-starch-lipase were enhanced and provided an effective method to improve the activity and stability of lipase for biodiesel production. Using 1% TA-MNPs-lipase and TA-MNPs-CLEAs-starch (w/w of oil) conversions around 67.87, and 85.88%, respectively, were obtained at 40°C after 2 h of reaction. Furthermore, the immobilized enzyme was easily recovered from the reaction mixture and reused. The obtained results suggest that TA-MNPs-lipase and TA-MNPs-CLEAs-starch-lipase can become a powerful biocatalyst for biodiesel production.

**Keywords:** magnetic cross-linked enzyme aggregates; lipase and enzyme immobilization.

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