



Identifying Lipid Mixtures for Optimal Tethered Bilayer Lipid Membrane Sensing of Lipase Activity

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Lipases are enzymes that have various industrial applications in the food industry, as detergents and as pharmaceuticals [1]. Examples include *Candida rugosa* (CR) lipase used in dairy production; the *Aspergillus niger* (AN) lipase used in the transesterification production of biodiesel; wheat germ (WG) lipase is used in food production and is an efficient chemical catalyst; and *Rhizopus oryzae* (RO) lipase which is a commonly used food additive and has applications in the pharmaceutical industry [1]. However, the availability of these enzymes is limited by high production costs [1]. The development of cost-effective protocols for monitoring enzyme production would therefore be of keen interest to the enzyme production industry. Tethered lipid bilayer membranes (tBLMs) are model lipid substrates that enable an analysis of the enzymatic processes occurring at the lipid-water interface using electrical impedance spectroscopy techniques [2]. We have recently demonstrated that it is possible to produce tBLMs using *triolein*, a naturally occurring, symmetrical 18-carbon triglyceride. These triolein tBLMs can undergo hydrolysis by lipases, making them a potential real-time biosensor of lipase activity [3]. In order to improve the sensitivity of this biosensor, this study aims to identify lipid mixtures that can be used to create triolein-fatty acid tBLMs that potentially have an improved membrane structure in terms of their molecular packing, which is better suited to lipase hydrolysis. Here, we present data showing the effects of incorporating fatty acids such as 18:1 Lyso PC and 17:1 lyso PC into triolein tBLMs and their subsequent hydrolysis by CR, AN, WG and RO lipases. The lipase activity was measured in terms of a change in the tBLM normalised conductance. With this data, it is hoped that an optimal triglyceride tBLM architecture can then be manufactured and used *in-line* to monitor enzyme production at an industrial scale.

[1] Houde, A., Kademi, A. and Leblanc, D., 2004. Lipases and Their Industrial Applications: An Overview. *Applied Biochemistry and Biotechnology*, 118(1-3), pp.155-170.

[2] Garcia, A., Deplazes, E., Aili, S., Padula, M., Touchard, A., Murphy, C., Mirissa Lankage, U., Nicholson, G., Cornell, B. and Cranfield, C., 2020. Label-Free, Real-Time Phospholipase-A Isoform Assay. *ACS Biomaterials Science & Engineering*, 6(8), pp.4714-4721.

[3] Rebaud, S., Maniti, O. and Girard-Egrot, A., 2014. Tethered bilayer lipid membranes (tBLMs): Interest and applications for biological membrane investigations. *Biochimie*, 107, pp.135-142.