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Liquid chromatography-mass spectrometry based approach for rapid comparison of lysophosphatidic acid acyltransferase activity on multiple substrates

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HIGHLIGHT

- Non-radio active LPAAT assay with native and non-native LPA substrates
- Robust LC-MS approach for LPAAT assay with multiple acyl-CoA substrates in a single reaction
- Adaptable for other lipid enzymatic assays

ABSTRACT

Lysophosphatidic acid acyltransferases (LPAAT) play an essential role in generating phosphatidic acid (PA), a key intermediate for phospholipids and triacylglycerol synthesis. The individual members have a diversity of localisation, and a strong fatty acid substrate preference. *In vitro* LPAAT enzymatic activity assays are necessary for understanding the physiological function of these enzymes. In this work, we have developed a liquid chromatography-mass spectrometry (LC-MS) based rapid enzymatic assay without using radioactive labelling. We show that this approach is comparable to radioactive labelling assays, using either native or non-native lysophosphatidic acid receiver molecules. Most importantly, this approach can be applied to the comparison of multiple substrates in a single assay. The approach is also adaptable for other lipid enzymatic assays.

Abbreviations

CoA Coenzyme A

DAG Diacylglycerol