An Introduction To Lipidomic Workflows

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Course Outline

- Introductory Level

- Goal is to provide information and resources on current lipidomic workflows, from the experimental design stage to data dissemination, so that attendees can learn how to adequately design, perform, and analyze data from lipidomics experiments.

- Course taught with up-to-date guidelines regarding proper lipid measurement and dissemination, as defined by LIPID MAPS and the more recent Lipidomics Standards Initiative

- Basic knowledge of analytical chemistry and mass spectrometry is required
~7000 fatty acyls alone in Lipid Maps
100,000’s of structures – Structure informs Biology
MS Workflows

Shotgun Lipidomics

Chromatography

Ambient Methods

Ion Mobility

https://pubs.rsc.org/nl-be/content/articlelanding/2015/an/c5an00838g
MS Workflows

Targeted Lipidomics (NL, MRM...)

Untargeted Lipidomics (DDA, DIA)

Structural Identification (HRMS, MSn)

Isomer Differentiation (ozID, Paterno-Buchi)

survey scan & precursor selection

fragmentation of selected precursors

survey scan across all isolation windows

fragmentation of all precursors in each window

OzID -110 Da

OzID -110 Da

Normalized intensity

Drift Time / m/z

https://pubs.rsc.org/en/content/articlehtml/2021/mo/d0mo00072h
Stephen Blankspy: “every time you think you have one lipid you actually have 3 or 4”

3 Challenges: sn-position, double bond position, and stereochemistry

<table>
<thead>
<tr>
<th>Structural Resolution</th>
<th>Example</th>
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<tbody>
<tr>
<td>Carbons and Double Bonds</td>
<td>PC(34:2)</td>
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<tr>
<td>Fatty Acid Constituents</td>
<td>PC(16:0_18:2)</td>
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<tr>
<td>Positional Isomers</td>
<td>PC(16:0/18:2)</td>
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<tr>
<td>Double Bond Position</td>
<td>PC(16:0/18:2(9,12))</td>
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<tr>
<td>Double Bond Cis vs Trans</td>
<td>PC(16:0/18:2(9Z, 12Z))</td>
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