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Organic acid derivatization protocol for mass spectrometry analysis

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We use this protocol and it's working

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Abstract

This protocol outlines the derivatization and the detection of organic acids using mass spectrometry analysis. This technique is highly sensitive and allow the detection of low-concentration samples.

Materials

Prepare the following solutions in 66% MS-grade acetronitrile:

- 200 mM 3-NPH HCl in ACN 66%
- 50 mM EDC in ACN 66%
- 7% pyridine in ACN 66%
- 5% formic acid (in MS-grade water)

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1 Prepare the following solutions in 66% MS-grade acetronitrile (ACN):
- 200 mM 3-NPH HCl in ACN 66%
- 50 mM EDC in ACN 66%
- 7% pyridine in ACN 66%
- 5% formic acid (in MS-grade water)

2 Dilute the samples if necessary (This technique is highly sensitive and allows the analysis of samples with low concentration).
- Caution: MOPS (3-(*N*-morpholino)propanesulfonic acid) saturates 3-NPH, preventing proper derivatization of organic acids. A dilution is necessary to achieve a maximum MOPS concentration of 1 mM

3 Prepare the reaction mixture:
- Pipette 50 µL of the sample.
- Add 25 µL of 200 mM 3-NPH HCl.
- Add 25 µL of 50 mM EDC.
- Add 25 µL of 7% pyridine solution.

4 Incubate the samples at 30°C for 5 hours at 500 rpm.

5 Add 125 µL of 5% formic acid to stop the reaction