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Fatty acid extraction and derivatisation

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Protocol status: Working

We use this protocol and it's working

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Keywords: fatty acid extraction, acid extraction, extraction solution, fatty acid standard mix, extraction, ml extraction solution, gc vial with screw cap dilute sample, ml extraction solution to sample, derivatisation c17 internal standard, fatty acid, derivatisation c17, ml hexane, needed amount out of this beaker, screw cap dilute sample, clean glass tube, hcl, gc vial, hexane, hexane phase



Abstract

- C17 internal standard (stock = 1 mg/ml)
- MeOH//3N HCl
- hexane
- 1% NaCl
- clean glass tubes
- always work with glass pipettes or hamilton

Start

samples with 4 oD-units (e.g.: oD = 1, you will need 4 ml culture)
centrifuge in a clean glass tube for 10 min @ maximum speed
discard supernatant
Freeze @ -80 °C until further use

Extraction solution

200 µl C17 internal standard + 9,8 ml MeOH/3N HCl
caution! fill in a beaker and take the needed amount out of this beaker
always work with gloves
pipet C17 internal Std. with hamilton

Extraction

add 1 ml extraction solution to samples and blank
blank: fatty acid standard mix
heat 60 min @ 90°C
let cool down @ RT
add 1 ml hexane
add 1 ml 1% NaCl
vortex for 30 sec
spin down 5 min @ 2000 rpm
transfer hexane phase in to GC vial with screw cap
dilute samples (10 µl sample + 90 µl hexane)
wash hamilton between samples with hexane

Troubleshooting

