BIDMC Mass Spectrometry Core Facility
Beth Israel Deaconess Medical Center
Center for Life Sciences
3 Blackfan Circle, Room 425
Boston, MA 02115
Ph: 617-735-2651 Fx: 617-735-2655

Metabolomics Sample Submission Guidelines

- Please do not send samples without first discussing the project with John Asara, jasara@bidmc.harvard.edu
- *No radioactive* samples will be accepted
- The submission tube(s) should be a plain clear 1.5mL eppendorf tube
- The metabolite samples should be extracted using 80% methanol and concentrated completely to dryness using either a SpeedVac or lyophilizer
- Store samples dry at -20°C or below until submission
- Be sure to fill out forms completely, 1 form per sample set and send forms with samples. List all sample names on the form(s).
- Please ship samples on dry ice via overnight delivery or drop them off in CLS-425 (posted sample drop-off area)
- *Do not send samples until an account with the core has been set up with John Asara, jasara@bidmc.harvard.edu
- More info and forms to download can be found at www.bidmcmassspec.org

METABOLOMICS Sample Prep Protocol

For collecting adherent cells from 10 cm2 plates

1.) Wash plate with respective medium 1X 5 mL. Add 10 mL respective medium and incubate for 2 hours prior to metabolite collection

2.) Aspirate off medium

- Tilt plate, aspirate of all media
- Keeping plated tilted, wait a few seconds to allow any additional media to collect in corner of plate, and them aspirate to get off as much as possible

3.) Immediately add 4 mL 80% methanol (-80°C)

4.) Immediately transfer to -80°C

- During transfer travel, place plates with 80% methanol on dry ice

5.) Incubate plates at -80°C for 15 minutes

6.) Scrap plates on dry ice with cell scraper

7.) Transfer cell lysate/methanol mixture to 15 mL conical tubes on dry ice

8.) Centrifuge at full speed for 5 minutes in cold room to pellet cell debris and proteins

9.) Transfer supernatant to 50 mL conical tubes on dry ice

- DO NOT THROW AWAY 15 mL TUBES

10.) Add 500 µl 80% methanol (-80°C) to 15 mL tubes and resuspend pellet

- Resuspending the pellet is a little difficult and may require a combination of vortexing and pipetting up and down

11.) Transfer mixture to 1.5 mL Eppendorf tube on dry ice

12.) Spin in microcentrifuge at full speed for 5 minutes in cold room

- KEEP 1.5 ML EPPENDORF TUBE

13.) Transfer supernatant to 50 mL conical tubes on dry ice (from step 9)

14.) Add 500 µl 80% methanol (-80°C) to 15 mL tubes and resuspend pellet

15.) Spin in microcentrifuge at full speed for 5 minutes in cold room

16.) Transfer supernatant to 50 mL conical tubes on dry ice (from step 9)

17.) After pooling the three extractions, the samples are completely dried (speedVac or lyophilizer).

18.) SUBMIT DRIED PELLETS IN 1.5 ML EPPENDORF TUBE – can be stored at -20°C or below.

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Metabolomics Sample Submission Form

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Sample Name:_____ Date Submitted:_____

User Name:_____ Lab/Office Phone:_____

E-mail:_____ Fax:_____

Principal Investigator:_____
Institution:_____
Address:_____
_____
BIDMC Grant #:_____
Non-BIDMC P.O. Number:_____
Billing Contact person/phone/email:_____

Estimated Amount:_____µg/pmol

Number of 10cm^2 cell growth plates used for extraction_____

Volume:_____µL Extraction buffer:_____ Solution color:_____

Organism:_____

*__Project Title__:_____
_____

What is the purpose of the analysis?:
_____
_____
_____

Any special instructions?:
_____
_____