# Metabolite Extraction Protocol for Cell-Based Experiments

***The Metabolomics Core utilizes a liquid nitrogen ($LN_2$) protocol for the extraction of metabolites from cultured cells. Addition of $LN_2$ directly to the culture dish allows for the immediate quenching of metabolism and efficiently lyses all cellular compartments. The resulting frozen cell "slush" is then delivered to the Core for further processing.***

***On the day of harvest*, the following steps should be performed in a cold room, with all supplies having been allowed to equilibrate to refrigerated temperature:***

1) Transfer culture dish to cold room and aspirate the media. Rinse 1x with PBS.
2) Add $LN_2$ directly to the culture dish and allow it to boil off.
3) Wait for evaporation (dish will look cloudy).
4) When dish starts to thaw, begin scraping frozen material (slush consistency) to center of plate.
5) Use cell lifter and razor blade to quantitatively transfer <u>all</u> material into a 50-mL conical tube submerged in dry ice.
6) Store frozen slush at -80°C and deliver to the Metabolomics Core.

*Important Note: If <u>Acylcarnitines</u> are to be assayed, cell media should be supplemented with 1 mM L-Carnitine (Sigma Catalog Number, C0283) **<u>24 hours prior to harvest</u>**

**<u>Update Jan 2016:</u>** Cell-based experiments require data normalization, usually to protein. The Metabolomics Core offers this service for a nominal fee. If clients wish to measure protein themselves, they will need to contact the Metabolomics Core with their protocol prior to submitting samples.