LOCASALE LAB PROTOCOLS

Biofluid Sample Preparation

—Plasma, Serum, Urine

1. Thaw serum on ice, gently vortex or pipette up and down several times.
2. Add 20 µL serum to a new Eppendorf tube containing 80 µL ice cold water (HPLC grade). If serum is limited, 10 µL serum added to 90 µL water is also acceptable.
3. Add 400 µL ice cold methanol (HPLC grade) to the tube (final methanol concentration: 80%, v/v), and vortex rigorously.
4. Leave the mix on ice for 10min.
5. Centrifuge at 20 000 rcf for 10 min, 4^{o}C.
6. Transfer the supernatant into two tubes (~200 µL for each), save one tube as backup. Discard the old tube containing the pellet of debris.
7. Dry pellet with a speed vacuum at room temperature, which will take about 1 hr for 200 µL solvent. (The time it takes to dry varies depending on the speed vacuum).
   NOTE: The dried pellet (containing the metabolites) from plasma or serum is usually very small or even not visible, while the pellet from a urine extract tends a white color and usually visible on the bottom of the tube.
8. Store dry pellet in -80 ^{o}C freezer for further LC-MS analysis.

Prepared by Locasale Research Group (September 2016)