

## Methanol/Chloroform Protein Precipitation

Modified from Wessel, D., Flugge, U.I., *Anal. Biochem.* 138, 141-144, 1984.

1. To sample of 100uL starting volume add 400uL Methanol.
2. Vortex briefly.
3. Add 100uL Chloroform.
4. Vortex briefly.
5. Add 300uL ddH<sub>2</sub>O. Sample should look cloudy.
6. Vortex briefly.
7. Spin 2 min. @ 14,000 g
8. Pipette off the top aqueous layer. Protein exists between layers and may be visible as a thin wafer. It may be difficult to see so be careful not to disrupt the interface during pipetting.
9. Add 400uL of Methanol.
10. Vortex briefly.
11. Spin 3min. @14,000 g
12. Pipette as much Methanol as possible from the tube without disturbing the pellet.
13. Dry pellet for no more than 1Hr in a fume hood or Speed-Vac to dryness but avoid drying too long as this makes the pellet much harder to re-solubilize.