Chloroform Methanol Purification of Proteins

Nikolai Slavov

This protocol describes the extraction method used by Slavov *et al* (2014a,b). It is based on the classical method of Wessel and Flügge (1984) that has been used extensively for decades.

To $500\mu l$ protein sample (about $500\mu g$ protein) in a 15 ml falcon tube:

1. Add 2ml methanol and vortex thoroughly.
2. Add 500 μl chloroform and vortex.
3. Add 1500 μl water and vortex; the mixture become cloudy with precipitated protein flakes.
4. Centrifugation for 1 minute at $14,000 \times g$ resulted is three layers: a large aqueous layer on top, a circular flake of protein in the interphase, and a smaller chloroform layer at the bottom.
5. Remove top aqueous layer carefully, trying not to disturb the protein flake.
6. Add $2ml$ methanol and vortex.
7. Centrifuge the resulting mixture for 5 minutes at $20,000 \times g$ until the protein pellets.
8. Remove as much methanol as possible with care since the pellet is delicate.
9. Dry the protein pellet.

References

Slavov N, Budnik B, Schwab D, Airoldi E, van Oudenaarden A (2014a) Constant Growth Rate Can Be Supported by Decreasing Energy Flux and Increasing Aerobic Glycolysis. *Cell Reports* **7**: 705 – 714

Slavov N, Semrau S, Airoldi E, Budnik B, van Oudenaarden A (2014b) Variable stoichiometry among core ribosomal proteins. *arXiv preprint* **1**: arXiv:1406.0399

Wessel D, Flügge UI (1984) A method for the quantitative recovery of protein in dilute solution in the presence of detergents and lipids. *Analytical biochemistry* **138**: 141–143