Methanol Chloroform Precipitation of Proteins

This is a method to precipitate proteins that are not adequately precipitated by TCA/NaDOC.

1. To 200 µl protein sample, add 480 µl MeOH (filtered), 160 µl CHCl₃ (filtered, add under the hood). Mix sample by vortexing. Add 640 µl ddH₂O, vortex and spin 5 min at 14,000 rpm.

2. Suck off the upper layer (the proteins are between the two liquid phases: top-MeOH/H₂O, bottom-CHCl₃) leaving a small amount of upper phase behind (use a thin gel-loading tip).

3. Add 300 µl MeOH, vortex and spin 30 min, 14,000 rpm, at 4°C.

4. Suck off supernatant completely (carefully) and air dry protein pellet.

5. Resuspend protein pellet in SDS sample buffer (solution A and solution B).