

TCA precipitation of proteins

Please first perform a protein assay (Bradford or micro BCA) before doing the precipitation protocol if you have no clue on the amount of proteins in your samples. Also, make sure that your sample buffer is compatible with the protein assay protocol.

Use cleaned tubes that are washed with all solvents used in the following protocol.

1. Starting solution of proteins: 100 ul (can be scaled up to 500 ul)
2. Add in the following order:
 - 100 ul of **10X** TE (standard Tris-EDTA buffer)
 - 100 ul of 0.3% NaDoc (sodium deoxycholate)
 - 100 ul of 72% TCA
3. Incubate on ice for 1 hour
4. Spin 14Krpm, 20 min, 4°C
5. Aspirate supernatant
6. Resuspend pellet in 90% room temperature acetone or methanol (volume of step 1)
7. Incubate in the freezer o/n
8. Spin 14Krpm 20 min, 4°C
9. Aspirate supernatant being careful to not dislodge the protein pellet, allow protein pellet to air-dry at room temperature for 15 minutes (cover the tubes with a clean Kimwipes to avoid dust deposition in tubes)
10. Leave samples dry and send to proteomic core facility or freeze them at -80°C until ready to submit.

N.B. This protocol is not suited for samples containing SDS, Triton or NP-40. They precipitate along with the proteins. Use the acetone precipitation protocol instead.