

Procedure

1. It is very important to use fresh, desiccated ammonium sulfate. This ensures uniform and rapid dissolution.
2. The day before use, place ammonium sulfate over night in ca. 120°C drying oven in a large beaker or drying dish (ammonium sulfate decomposes at 220°C).
3. Clean grinder carefully and grind dry ammonium sulphate to a fine powder. Wear a dust mask (if you don't need a mask, the powder is not dry enough).
4. Use ground powder immediately. For 70% saturation use 43.6 grams/100 mls.
5. Add the powder slowly but steadily with thorough mixing. Do not allow clumps to form.
6. Allow precipitate to form for 30 minutes at 4°C with stirring.
7. Recover precipitate by centrifugation. Solutions highly saturated in ammonium sulfate are quite dense and it can be difficult to pellet the precipitate. For CBF3 prep, spin at 40K in type 45 rotor for 1 hour.
8. Remove supernatant, respin briefly to clear remaining ammonium sulfate.
9. Resuspend pellets in a volume of buffer equal to the volume of the extract.