TCA precipitation of cell lysates - April 27, 1987
Margarete Heck

TCA PRECIPITATION OF CELL LYSATES

(It is most convenient to do this in 1.5 ml microfuge tubes.)

1. If in doubt about the protein concentration, add E. coli tRNA (stock at 5 mg/ml in 10 mM Tris-Cl, ph 7.4, 1 mM K-EDTA) to a final concentration of 100 ug/ml.

2. Add 100% TCA to a final concentration of 10%, precipitate on ice at least 30'.

3. 15' spin in the microfuge (cold).

4. Aspirate supe with a drawn pasteur pipette, and resuspend pellet by sonicating in 1 ml of cold 90% Acetone - 10% 0.1N HCl. Extraction (of detergents and acid) can be left on ice or centrifuged immediately.

5. 10' spin in the microfuge (cold).

6. Repeat steps 4. and 5.

7. Aspirate second wash (one wash is usually not sufficient to remove the TCA), and resuspend the pellet by sonicating in the desired volume of SDS-PAGE sample buffer containing DTT or 2-me. Boil 5', spin, and load on gel or freeze at -20.