**TCA-precipitation**

Materials:
55 % TCA
1 % Triton X-100
-20°C acetone
1 M TRIS pH 8
Eppendorf tubes, centrifuge (> 4000g at 4°C) and racks, -20°C freezer, ice, Pipette and tips.

Procedure:
- Mix
  1 vol sample
  + 0.253 vol 55 % TCA
  + 0.14 vol 1 % Triton X-100
- Vortex and store 10 minutes on ice.
- Centrifuge 5 minutes 13000g or 10 minutes 4000g at 4°C.
- remove supernatant
- add 1.2 vol. -20°C acetone
- vortex vigorously until a white protein flake floats freely around
- centrifuge 10 minutes 13000g or 15 minutes 4000g at 4°C
- remove supernatant
- dry 2-6 minutes (not longer, depends on the amount of protein to dry) in the vacuum concentrator
- heat dissolve in SDS or LDS sample buffer (95°C 5 minutes or 70 °C 10 minutes).
- if the sample turns yellow before or during the heating mix in 1 M TRIS pH 8 in 0.5 - 1 µl steps until the color changes to blue again.

Do not store the sample as dry protein pellet, it contains still some TCA and becomes hard to dissolve.
Rather dissolve in SDS-buffer as soon as possible and store this at -20°C.

Solutions:
**55 % TCA**

Wear goggles, lab coat, gloves. Pure TCA is a hygroscopic white solid which appears like old icy snow. It forms a single lump after a while and is quite corrosive, particularly to stainless steel. When you are done, remove thoroughly any stray TCA crystals from balance, spatula and table and clean with enough water.

**Materials:**
TCA
Sturdy spatula suitable to scrape and pinch off from a big lump of TCA
250 ml glass beaker, 100 ml measuring cylinder, 100 ml laboratory bottle, magnetic stirrer and stirring bar.

**Procedure:**
In a 250 ml glass beaker with strirring bar weigh 55 g TCA. Add water to ~ 95 ml and cover the beaker with Parafilm or similar, but not aluminium foil. Stir until everything is dissolved (~ 20 minutes). Transfer to 100 ml measuring cylinder and fill up to 100 ml. Filter through a 0.45 µm or 0.2 µm PVDF syringe filter (sacrifice the first few ml).