**Protein analysis protocol Degerhamn 2014-11-28**

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Based on Lowry et al 1951, and using a ready-made protein bio assay.

Chemicals;

0.5 M NaOH

Bio-Rad DC Protein Assay

* Bovine serum albumin standard
* Reagent A, alkaline copper tartrate solution
* Reagent B, Folin´s Ciocalteu reagent dilution

Materials;

Freeze dried algae biomass of known quantity.

Glass tubes that can be centrifuged

Aluminium foil

Pipette 100 μl

Pipette 4.0 ml

Water bath set at 90 °C

Spectrophotometer

**Folin´s Ciocalteu reagent contains Phenol. When working with Phenol, work in a fume hood. Inform other people in the lab about what chemicals you are using. Wear neoprene gloves, lab coat and safety goggles**. **Pipet tips, gloves and other contaminated debris should be collected as hazardous waste.**

1. Re-suspend 5 mg biomass samples in 2 ml 0.5 M NaOH.
2. Seal the glass tubes with aluminium foil and parafilm paper
3. Vortex samples gently.
4. Water bath 90 °C 1 h.
5. Let samples cool to room temperature.
6. Prepare a standard curve using the supplied Bovine serum standard. Prepare 5 dilutions ranging from 0.2 to 1.0 mg/ml protein. Prepare a blank.
7. Centrifuge the samples 10 min at 4000 rmp.
8. Transfer 100 μl of the supernatant into clean glass tubes.
9. Add 500 μl Reagent A to each sample.
10. Vortex samples gently.
11. Add 4.0 ml Reagent B and vortex immediately.
12. Wait for ≈15 min.
13. Measure absorbance in a spectrophotometer at 750 nm.
14. Compare your samples to the standard curve.