Proteomics-Facility University of Konstanz	**Colloidal Coomassie staining for proteomic-samples**	SOP Nr.	00
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Protocol is a Modification of: Neuhoff, V., et al., *Improved staining of proteins in polyacrylamide gels including isoelectric focusing gels with clear background at nanogram sensitivity using Coomassie Brilliant Blue G-250 and R-250.* Electrophoresis, 1988. **9**(6): p. 255-62

1. Stock solutions:

Solution A:
10% (w/v) ammonium sulphate, 2% (w/v) phosphoric acid in MilliQ water

Solution B:
5% (w/v) Coomassie Brilliant Blue G-250 in MilliQ water

Washing solution:
25% methanol in MilliQ water

2. Protocol

The gel is incubated for 5 min in water and then stained overnight in staining solution. The detailed protocol is described below:

- The staining solution is prepared by mixing 100 ml of the stock solution A with 2.5 ml stock solution B. Shake strongly this solution for 20 minutes and then add 25 ml pure methanol. Continue shaking the next 20-30 minutes.
- Place the gel in the freshly prepared colloidal Coomassie stain. Stain the gel overnight with gentle shaking.
- Wash the gel with 25 % methanol in MilliQ water for around 1 hour.
- Wash the gel with MilliQ water
- Repeat the last two steps until the protein bands are at the desired contrast against the background of the gel.