

Roti[®]-Nanoquant (for protein concentration estimation)

- ◆ Preparation of the work solution
 - ◇ 20mL Roti[®]-Nanoquant (5xconc.)+80mL ddH₂O
 - ◇ Usable for a week at room temperature
- ◆ Preparation of the Eich row:
 - ◇ Prepare X different dilutions of BSA (from 1 µg/mL to 100 µg/mL)
 - ◇ Use the following pipette scheme:

| BSA (µg/mL) | µL out of higher concentrations | µL ddH ₂ O |
|--------------|---------------------------------|-----------------------|
| 0 | - | 200 |
| 1 | 20 µL out of 10 µg/mL | 180 |
| 2.5 | 50 µL out of 10 µg/mL | 150 |
| 5 | 100 µL out of 10 µg/mL | 100 |
| 10 | 40 µL out of 100 µg/mL | 360 |
| 25 | 50 µL out of 100 µg/mL | 150 |
| 50 | 100 µL out of 100 µg/mL | 100 |
| 75 | 150 µL out of 100 µg/mL | 50 |
| 100 | 200 µL out of 400 µg/mL | 600 |

- ◆ Fill the cuvettes:
 - ◇ Zero value: 200 μL ddH₂O + 800 μL working solution
 - ◇ Eich row: each 200 μL of the standard +800 μL working solution
 - ◇ Samples: each 200 μL +800uL working solution
 - ◇ Reference: 1000 μL ddH₂O
 - ◇ Mix all of the cuvettes gently by inverting
- ◆ Measurement:
 - ◇ Measure all OD₅₉₀ values of all cuvettes against the reference
 - ◇ Measure all OD₄₅₀ values of all cuvettes against the reference
- ◆ Evaluation:
 - ◇ Fill in the quotient of OD₅₉₀/OD₄₅₀ into a graph.
 - ◇ Protein concentrations of your samples can easily be read off the eich curve

From: [Roth](#)