Quick check if His-tagged protein is present

1. Add 500 µl Ni-NTA slurry to a 1.5 ml tube
    a. Add 1 ml H_2O & invert the tube 10x
    b. Centrifuge at 2000 rpm for 1 min and carefully take off supernatant
    c. Repeat this wash step
    d. Add 1 ml medium & invert the tube 10x
    e. Centrifuge at 2000 rpm for 1 min and carefully take off supernatant
    f. Repeat this equilibration step

2. Add 1 ml supernatant & invert the tube 10x
    a. Incubate tube on rotator at 4 °C for 30 min
    b. Centrifuge at 2000 rpm for 1 min and carefully take off supernatant (keep as flow through sample)

3. Add 1 ml wash buffer & invert the tube 10x
    a. Centrifuge at 2000 rpm and carefully take off supernatant
    b. Repeat washing step 3 times in total (keep the last wash supernatant)

4. Add 100 µl wash buffer again, mix and take a sample from beads for the gel

5. Add 1 ml elution buffer & invert the tube 10x
    a. Centrifuge at 2000 rpm for 1 min
    b. Take off supernatant and transfer to a new tube → this is the protein!!!!

6. Add 100 µl elution buffer again, mix and take a sample from beads for the gel

Wash buffer (5ml)
100 µl 1M Tris
300 µl 5M NaCl
1 ml 50 % Glycerol
5 µl 0.1 M PMSF
50 µl 2 M Imidazole

Elution buffer (5ml)
100 µl 1M Tris
300 µl 5M NaCl
1 ml 50 % Glycerol
5 µl 0.1 M PMSF
1.25 ml 2M Imidazole