Expression and purification of Ulp1

1. Transform *e. coli* BL21 with plasmid pFGET19_Ulp1
2. Pick a colony and grow an overnight culture at 37°C with 40 µgr/ml kanamycin
3. Inoculate 1 L of TB in a 1:1000 ratio with overnight culture
4. Grow to OD600 of 1.5
5. add isopropyl-b-D-thiogalactoside (IPTG) to 1 mM final concentration.
6. Transfer to 25°C and grow overnight
7. Collect cells by centrifugation, 4,000g, 30 min
8. Use immediately or stored frozen at -80°C
9. Resuspend pellet in buffer:
    - 50 mM Tris, pH 8
    - 300 mM NaCl
    - 20 mM imidazole
    - Protease inhibitors
    - 2 mM MgCl2
    - benzonase
10. Dounce cells to homogenize
11. Lyse cells in homogenizer device
12. Remove cell debris centrifugation at 50,000g for 30 min
13. Filter supernatant with a glass filter
14. load (using the sample pump) on a 5 ml Ni-NTA column previously equilibrated with the wash buffer (same as before only w/o PI, MgCl2, benzonase)
15. Wash column with wash buffer until baseline is reached
16. Elute with elution buffer (same as wash buffer only with 250 mM imidazole)
17. Dialyze protein against standard phosphate-buffered saline (PBS) buffer
18. Store at -80°C in storage buffer (50% glycerol v/v, 25 mM DTT)
19. To use mix 0.1% Ulp1 (v/v) plus 1 mM DTT