

Protein Dialysis Protocol

Target Concentration: PBS 0.01M

Original Concentration: 250mM Imidazole + 20mM Tris(pH 7.6) + 300mM NaCl

1. Cut a proper length of dialysis bag (about 8cm for 3ml of solution).
2. Rinse the inside and outside of dialysis bag with PBS 0.01M.
3. Clip the bottom of dialysis bag with metal clip.
4. Add the eluted proteins (3~4ml) into the dialysis bag
5. Clip the other end of dialysis bag with plastic clip, and tie an Eppendorf on it to keep it suspended in the dialysis buffer.
6. Put a stir bar in the dialysis buffer and stir at 4°C

The stir bar would attract the metal clip by magnetic force, hence the dialysis bag would rotate with the stir bar

7. Change dialysis buffer according to the chart below:

Dialysis Concentration		Time
Imidazole 0.15M 、 PBS 0.01M	5M Imidazole 30ml, add 0.01M PBS to 1L	2hr
Imidazole 0.075M 、 PBS 0.01M	5M Imidazole 15ml, add 0.01M PBS to 1L	2hr
Imidazole 0.05M 、 PBS 0.01M	5M Imidazole 10ml, add 0.01M PBS to 1L	3hr
Imidazole 0M 、 PBS 0.01M	0.01M PBS 1L	O/N
Imidazole 0M 、 PBS 0.01M	0.01M PBS 1L	2hr

8. Collect samples from the dialysis bag.
9. Centrifuge at 13,000 rpm, 20mins, 4°C, and obtain the centrifuge. Collect the supernatant of T as S(solution), and dissolve the palate with 50ul ddH20 as P(palate).
10. Store at -20°C or quantify the product via protein assay.