DAISEP SPIN XL
PROTOCOL

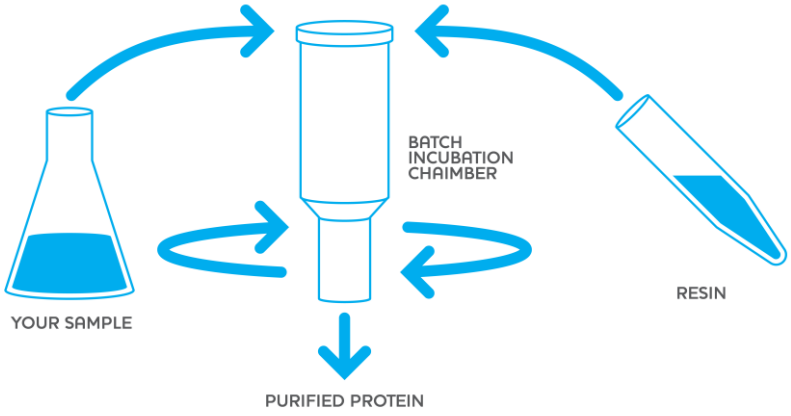
MATERIALS SUPPLIED IN THE KIT:

- DAISEP Spin XL column (22 mL capacity in a swing bucket rotor)
- Two caps:
  1 clear spin push cap for all centrifugation steps
  1 yellow batch incubation cap for the incubation step only
- 50 mL centrifuge tubes
  80 units

ADDITIONAL MATERIALS REQUIRED:

- 0.2 µm syringe filters for clarification
- 50 mL centrifuge tubes
- A bench-top centrifuge with swing bucket rotor capable of handling 50 mL centrifuge tubes (the preferred rotor is a swing bucket rotor)
- Quartz cuvettes for UV absorbance measurements
- UV/VIS spectrophotometer

PROTEIN PURIFICATION PROTOCOL:



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RECOMMENDED PROTOCOL

The following spin speeds and times are appropriate for a 0.25 – 1 mL resin bed volume. Spin times for each of the following steps may increase with larger bed volumes.

PRE-EQUILIBRATION

1. Pipette the appropriate resin slurry into the batch incubation chamber of the spin column barrel. Wash the resin by centrifuging at 750 x g for 5 min (with the clear spin push cap). This step is critical to ensure that storage solution is removed from the resin.

NOTE: Ethanol does interfere with sealing properties of the Self Seal™ membrane technology.

2. Pre-equilibrate the media with 15 mL equilibration buffer by centrifuging the spin column (with the clear spin push cap) at 750 x g for 5 min. It is **critical** that you repeat this step one more time with a further 15 mL fresh equilibration buffer.

NOTE: If using one spin column, ensure that the spin column is counterbalanced with a unit of equal weight (adjusted with distilled water).

CLARIFICATION OF SAMPLE

3. Pre-filter the sample through a single 0.2 µm filter (e.g. syringe filter).

NOTE: It is critical that the sample is filtered through a final 0.2 µm syringe filter **immediately** before loading it on the spin column. Optimal performance of these devices will depend on these instructions being rigorously followed.

SAMPLE LOADING

4. Transfer the spin column barrel to a fresh 50 mL centrifuge tube and load up to 22 mL of your filtered sample into the batch incubation chamber to reach the appropriate resin/sample mix ratio (maximum sample volume 22 mL). Tightly screw on the yellow batch incubation cap and invert 2-3 times to mix the sample and the resin. Place the column on a standard tube roller and mix during appropriate contact times.

5. After batch incubation, replace the yellow cap with the clear spin push cap. Centrifuge the column at 750 x g for up to 10 min and collect the flowthrough fraction.

NOTE: If using one spin column, ensure that the spin column is counterbalanced with a unit of equal weight (adjusted with distilled water).

WASHING STEPS

6. If you need to perform washing, transfer the spin column barrel to a fresh 50 mL centrifuge tube and load wash buffer. Add the clear spin push cap to the end and wash off the unbound protein at 750 x g for 5 min. Remove the filtrate and repeat this step.