



Exchange Buffers and Concentrate by Spin Filtration

- ☐ Dilute your sample ~10x with your new buffer (e.g., add 54 mL Protein Buffer to 6 mL eluted protein).
- ☐ Load your sample onto a centrifugal filter column.
- ☐ Centrifuge samples at either 4000 rcf (4 mL or 15 mL filters) or 14 000 rcf (0.5 mL filters) at 4 °C until you've reached your target volume. Check on your sample volume in the first 10 min, then again as needed.
- ☐ Continue to dilute and spin your sample, noting your sample volume and dilution volume at each step, until you've reached your target dilution factor (we recommend $\geq 300\times$).
- ☐ Add an equal volume of Protein Buffer (60% glycerol) to your sample to bring the final glycerol concentration to 30%. Freeze samples at $-80\text{ }^{\circ}\text{C}$ for storage.
- ☐ Store columns for later use.
 - ☐ Wash columns by loading with ddH₂O and spinning.
 - ☐ Load columns with EtOH 20% (v/v) and store at room temp.