Procedure 5: To perform a dialysis procedure.

Reagent:
Deionised water
Other reagents as required (e.g. PBS)

Materials & Equipment:

Dialysis membrane (various sizes)
Magnetic stirrer & bar
Plastic pipettes
Scissors
Tissue

Note: Dialysis is used for both the removal of low molecular weight contaminants and buffer exchange of protein containing solutions. Optimal protein concentration range for samples is 0.25 – 5 mg/ml.

Procedure:

1. Cut dialysis membrane to the required length and soak in deionised water for at least 5 minutes prior to use.

2. Clip or tie a double knot in one end of the membrane and dispense sample, of known volume, into the dialysis bag using a plastic pipette.

3. Remove excess air from bag and leave at least 25% extra space for volume expansion. Clip or tie a double knot at the open end to seal the bag.

4. Commence dialysis against at least 50 volumes of buffer at 4^{o}C, twice, for at least 3 hours on each occasion. Ensure that the dialysate is gently stirring throughout the procedure. Maximum dialysis time depends on sample stability and should not exceed 48 hours.

5. Upon completion of dialysis dry the outside of the bag with tissue, cut the bag and remove the sample using a plastic pipette.

6. Measure sample volume post-dialysis.

Note: Removal of unreacted NHS-biotin requires a 48 hour dialysis step with 4 changes of buffer (100 volumes per step) throughout the period. All other conditions are as described in 4 above.