

Dialysis Protocol

By Gail Shirley, August 1994

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1. Put on gloves.
2. Cut off a length of dialysis tubing that is longer than you think you will need.
3. Wet the dialysis tubing under the distilled water faucet. **Once the tubing gets wet, keep it wet.** Crinkle up the tubing under the running water. Rinse out the inside - there is glycerol inside and this needs to be washed away. Continue this washing for 5 - 10 minutes.
4. Tie a knot near one end of the tubing. Tie another. Check for leakage. If any solution leaks through these knots, tie another.
5. Immerse the knotted tubing in dialysis buffer (keep a tub near side of sink). This will be whatever buffer your protocol calls for. Keep the tubing in the buffer until you are ready to load your sample.
6. Load your sample into tubing using a pasteur pipette. Clip an orange dialysis closure just above the sample, leaving a little room for bubbles. Check for leakage. If necessary (and if you have room) clip on another closure. Tie one or two knots above the closure(s).
7. Fill a 2L flask almost to the top with dialysis buffer. Put a stir bar at the bottom, and put your dialysis bag containing sample on top. Place flask on a magnetic stirring plate in the cold room. Turn stirrer to a setting that will allow dialysis bag to slowly float around the top of the solution.
8. Check the dialysis situation a few times during the first half hour of stirring to make sure that the bag is not moving too quickly. Sometimes as the stirring plate warms up, the stirrer will start stirring faster. If this is the case, turn it down.
9. After the dialysis has proceeded for 2 hours, replace the buffer with fresh dialysis buffer. Allow this to stir overnight.
10. Remove the dialysis bag from the flask, and carefully remove the closure(s). Cut the bag right beneath the upper knot, and carefully remove your sample from the bag using a pasteur pipette. Work over a funnel and beaker just in case bag slips from your hand.