

# **Product Information**

## **Ultrafiltration Vials**

#### **Catalog Numbers:**

22018 Ultrafiltration Vial, MWCO=3K 22004 Ultrafiltration Vial, MWCO=10K

Storage: Room temperature

Quantity: 5 per pack

### Product Description

Centrifugal ultrafiltration devices contain membranes that allow molecules with molecular weights smaller than the molecular weight cut-off (MWCO) to pass through the membrane into the filtrate collection tube, while molecules larger than the molecular weight cut-off are retained on top of the ultrafiltration membrane. Ultrafiltration can be used to rapidly and easily remove small molecules such as Tris, glycerol and glycine from solutions of antibody or other proteins, or to concentrate antibodies or other proteins prior to conjugation. They also can be used to remove free dye or free biotin from proteins after conjugation.

For ultrafiltration of proteins, it is recommended to use a MWCO at least 3- to 6-times smaller than the protein to be retained. MWCO=10K is recommended for ultrafiltration of antibodies and other proteins larger than 30 kDa, while MWCO=3K is recommended for ultrafiltration of proteins between 10-30 kDa. Ultrafiltration is not effective for fractionating proteins (separating one protein from another protein based on size) unless there is at least a 10-fold difference in the protein molecular weights.

Note: Repeated filtration of large sample volumes (~500 uL) can lead to membrane failure. We therefore recommend keeping sample volumes at or below 350 uL.

### Capacities

Maximum Sample Volume: 500  $\mu$ L (see note above) Final Concentrate Volume: 15  $\mu$ L Filtrate Receiver Volume: 500  $\mu$ L Hold-up Volume (Membrane/Support): < 5  $\mu$ L

#### Protocol for antibody concentration or clean-up

Caution: avoid touching the membrane of the filtration vial with the pipette tip during liquid transfer. Any damage to the membrane may result in loss of sample. You may wish to save the filtrate until sample recovery is verified. Typical protein recovery is 80-90%.

Note: proteins larger than the molecular weight cut-off will remain on the upper surface of the membrane in the sample reservoir. Small molecules (such as Tris, glycerol, glycine, free biotin, and free dyes) will pass through the membrane into the filtrate receiver tube (Figure 1).

- 1. Load the sample solution in the sample reservoir (upper chamber) of a filtration vial and close the cap.
- Centrifuge at 14,000 x g until almost all of the liquid is in the lower filtrate receiver tube. Filtration rate will depend on volume and antibody concentration. Centrifuge for one minute and check the liquid level in the sample reservoir. Continue centrifuging until all of the liquid is in the lower filtrate collection tube.

- Empty the collection tube and add any additional sample solution to the sample reservoir (upper chamber). Repeat steps 1 & 2 until all of the sample solution has been centrifuged.
- 4. For protein concentration, proceed to step 8. For protein clean-up, proceed to step 5.
- Add 300 uL PBS to the sample reservoir (upper chamber) and centrifuge until all of the liquid is in the lower filtrate collection tube.
- 6. Repeat Step 5.
- 7. For clean-up prior to protein conjugation, proceed to step 8. For removal of free dye after protein conjugation, repeat Step 5. By the third ultrafiltration, the fluorescent color of the solution (detected using a UV lamp) in the collection tube should be very light, indicating nearly complete removal of the free dye from the labeled antibody.
- Add the appropriate volume of PBS to the sample reservoir (upper chamber) to resuspend protein at the desired final concentration. Gently pipette up and down to resuspend protein. Transfer the protein solution to a clean tube.



Figure 1. Ultrafiltration vial components.

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