

Choosing the Correct MWCO

Once sample volume is determined, the next step is to select the appropriate MWCO (for ultrafiltration) or pore size (for microfiltration). MWCOs are nominal ratings based on the ability to retain > 90% of a solute of a known molecular weight (in Kilodaltons). The table below provides retention characteristics of different MWCO membranes for some solutes. For proteins, it is recommended that an MWCO be selected that is three to six times smaller than the molecular weight of the solute being retained. If flow rate is a consideration, choose a membrane with an MWCO at the lower end of this range (3X); if the main concern is retention, choose a tighter membrane (6X).

It is important to recognize that retention of a molecule by an ultrafiltration membrane is determined by a variety of factors, among which its molecular weight serves only as a general indicator. Therefore, choosing the appropriate MWCO for a specific application requires the consideration of many factors including molecular shape, electrical charge, sample concentration, sample composition, and operating conditions.

Because different manufacturers use different molecules to define the MWCO of their membranes, it is important to perform pilot experiments to verify membrane performance in a particular application.

Common Variables That Increase Molecule Passage:

- ▶ Sample concentration less than 1 mg/mL
- Linear versus globular molecules
- ▶ High transmembrane pressure created by g-force in centrifugal concentrators. (This is especially important in the case of linear molecules, for example DNA fragments. Decreasing the g-force can increase retention of molecules by a membrane.)
- Buffer composition that favors breakup of molecules
- pH and ionic conditions that change the molecule (for example, cause conformational changes or aggregation)

Common Variables That Decrease Molecule Passage:

- ▶ Sample concentration higher than 10 mg/mL
- ▶ Buffer conditions that permit molecules to aggregate
- ▶ Presence of other molecules that increase sample concentration
- Lower transmembrane pressure (in the case of centrifugal concentrators, lower g-force)
- ▶ Adsorption to the membrane or device
- ▶ Low temperature (4 °C versus 24 °C)

MWCO Selection for Protein Applications

MWCO	Pore Size*	Size	Molecular Weight
3K	_	_	10K - 30K
10K		_	30K - 90K
30K		_	90K - 300K
100K	10 nm	30 - 90 nm	300K - 900K
300K	35 nm	90 - 200 nm	900K - 3,000K

MWCO Selection for Virus Applications

MWCO	Membrane Nominal Pore Size*	Virus or Particle Diameter
100K	10 nm	30 - 90 nm
300K	35 nm	90 - 200 nm

MWCO Selection for Nucleic Acid Applications

MWCO	Base Pairs (DS)	Bases (SS)
3K	16 - 50 Bp	32 - 95 Bs
10K	50 - 145 Bp	95 - 285 Bs
30K	145 - 475 Bp	285 - 950 Bs
100K	475 - 1,450 Bp	950 - 2,900 Bs
300K	1,450 - 9,500 Bp	2,900 - 9,500 Bs

^{*}Nominal pore size as measured by electron microscopy



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