AppliChrom VivoSep SEC Columns

Polymeric Size Exclusion Chromatography Columns for high Resolution and Recovery in Protein Analysis/Purification

supplied and supported by:
mmac
meinhard missbach analytical consulting
Meinhard Missbach GmbH.
1230 Wien, Kanitzgasse 21
Tel: +43 1 889 2351
email: office@missbach.co.at

AppliChrom VivoSep SEC columns for easy going SEC of proteins

- special multistage hydrophilically modified polymeric SEC (size exclusion chromatography). column materials to facilitate/enable many previously difficult protein SEC analyses/separations with high resolution.

- AppliChrom VivoSep SEC materials contain only polymeric material free of silica; means: no silanol activity and no unwanted silica leachables!

- AppliChrom VivoSep SEC columns show high desorption power that is great for SEC of many proteins but – in the past – this was still mostly connected with low pressure stability in case of some in market used cross-linked dextran based media (e.g. p max approx 5bar)

- AppliChrom VivoSep SEC columns show good pressure stability of 50-200 bar (depending on pore size) compared to cross-linked Dextran SEC columns, which are only stable in the range of 5 bar or less.

- AppliChrom VivoSep SEC columns allow (due to their pressure stability) the use of smaller particles, e.g. 7-10µm for for delivering a large separation power extending even 20.000 plates/m for good separation of proteins. These are much more paltecounts/m than you get from 20-50 or larger particles that are cross-linked dextran based.

- AppliChrom VivoSep SEC media show stability in pressurized water up to 121°C.

- AppliChrom VivoSep SEC columns can be used at temperatures from 10-90°C

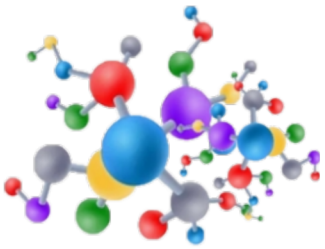
- pH stability 2-11

- AppliChrom VivoSep SEC columns are available with an enormous exclusion limits of 70.000 and > 1.000.000Da*) (many cross-linked dextran based SEC-media for proteins are limited to a maximum molecular size of 100-150kDa or less).

- AppliChrom VivoSep SEC column heads are connected at both sides with 10-32 UNF female thred – making these columns perfect to use with the standard analytical HPLC systems**).

- SEC molecular size measurement/calculation of hydrodynamic size of protein [nm] can be done on base of SEC-system calibration using PEO/PEG reference substances**).

*): Other porosities are needed, please tell us your demand.
**): Questions for column / HPLC-system connection, molecular size [nm] determination, or comments - please tell us what counts for you.



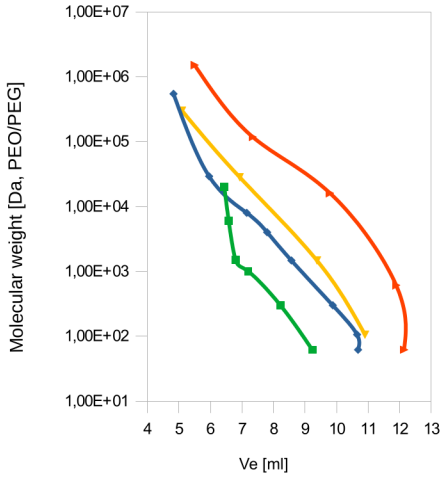
VivoSep SEC Columns therefore offer **three major advantages**, that you will not find on any other system that you can buy.:

1. **Higher separation performance in less time**: By using pressure-stable filling materials, the columns are filled with smaller resin particles, resulting in separation rates of over 20,000 plates/m. This results in up to 4 times higher sample throughput.
2. **Larger Dalton range**The newly developed filling material now allows the separation of proteins over a range of 4 decades (!) from about 100 to greater than 1.00E+06 Da, depending on the column type.
3. **Lower costs** and **better environmental protection**: The separation in VivoSep SEC is carried out in cost-effective, water-based running agents - the same for all size classes and with lower consumption due to the improved separation performance. This not only applies to samples of water-soluble proteins, but also when hydrophobic substructures are present

AppliChrom VivoSep SEC Columns

Polymeric Size Exclusion Chromatography Columns for high Resolution and Recovery in Protein Analysis/Purification

AppliChrom VivoSep SEC calibration curves:
Polymeric high pressure stable high resolution SEC material special designed for demands on SEC of proteins.



Red line:	AppliChrom VivoSep SEC Multipore
Yellow Line:	AppliChrom VivoSEP SEC 300
Blue line:	AppliChrom VivoSEP SEC 250XL
Green Line:	AppliChrom VivoSEP SEC 100

Column size 300x8mm e.a.
1.0 ml/min H_2O
molecular weight (PEO/PEG) vs. elution time

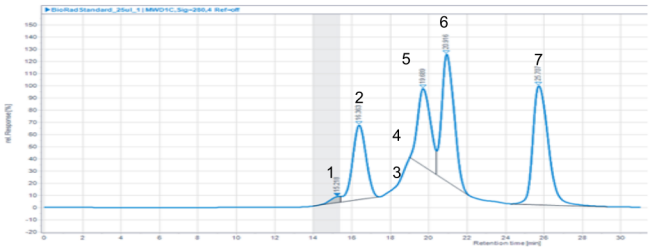
How to use a PEO/PEG SEC calibration data for molecular size determination of an unknown substance?

General: 2 molecules of equal size elute from SEC column in SEC mode at identical elution volume!
E.g.:

PEO 110.000Da in pure water: hydrodynamic size = 12.6nm
=> If IgM elutes at elution volume of PEO 110.000Da, IgM = 12,6nm size.

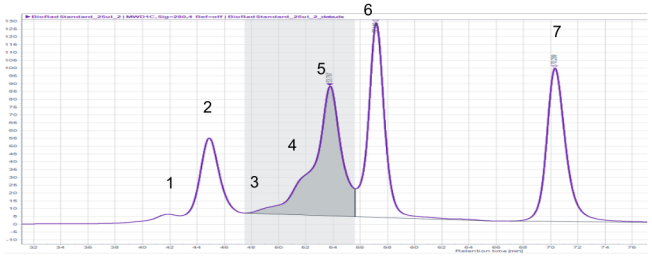
PEO 26.000Da in pure water: hydrodynamic size = 5,3nm
=> If IgG elutes of elution volume of PEO 26.000Da, IgG = 5,3nm

AppliChrom VivoSep SEC for SEC-separation/analysis of 5 proteins + dimers



AppliChrom VivoSep SEC Multipore column
1x 300x8mm

Flow rate:	500µl/min
Back pressure:	40 bar
Detection:	UV 280nm
	PBS buffer
Run time:	30 min



AppliChrom VivoSep SEC Multipore column
2 x 300x8mm

Flow rate:	400µl/min
Back pressure:	40 bar
Detection:	UV 280nm
	PBS buffer
Run time:	120min

Peaks in order of elution:

1 thyroglobulin dimer (1.340.000Da)
2 thyroglobulin (670.000Da)
3 gamma globulin dimer (316.000Da)
4 Gamma globulin (158.000Da)
5 ovalbumin (44.000Da),
6 myoglobulin (17.000Da)
7 vitamin B12 (1350Da)

Chromatograms provided from:

Dr. Max Gilbert
Professor Dr. Pitter Huesgen
Forschungszentrum Jülich
Zentralinstitut für Engineering, Elektronik und Analytik
Institutsbereich Analytik (ZEA-3)
Wilhelm-Johnen-Straße
52428 Jülich

Order information:

AppliChrom VivoSep SEC Multipore
300x8mm
P/N: AVSSECM3008

Precolumn AppliChrom VivoSep SEC Multipore
50x8mm
P/N: AVSSEC508

AppliChrom GmbH | Germendorfer Allee 20 | D-16515 Oranienburg | phone +49 (0)3301 579293 | info@applichrom.com | www.applichrom.com