The column for gel filtration chromatography

Develosil 100 Diol / 300 Diol

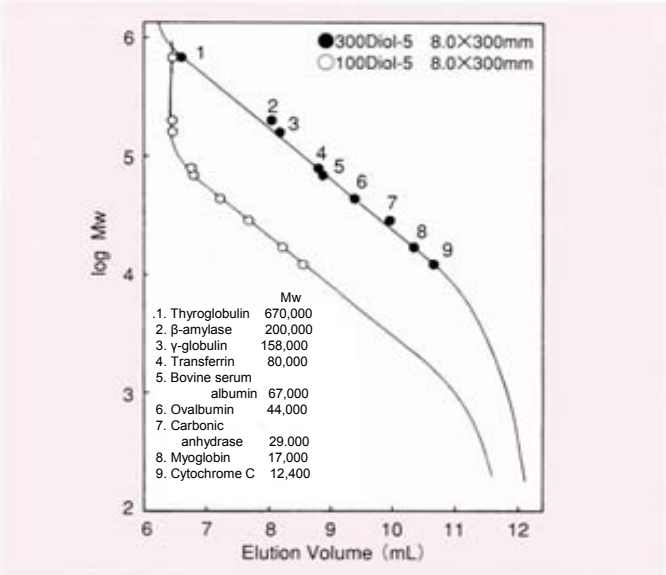
Develosil 100Diol and 300Diol are the columns developed for gel filtration chromatography.
The size exclusion chromatography of water soluble polymers, such as hundreds of thousands of protein, enzymes, etc., is possible from the molecular weight 10,000.
packing material type: For the compounds of low molecular weight ⇒ 100 Diol
For the amounts of macromolecules ⇒ 300 Diol
That is, since there are two kinds, low molecular weight and the amount of macromolecules, a column can be chosen according to the molecular weight of the purpose reagent.

Physical properties of Develosil 100Diol and 300Diol

Column name	Ligand	Carbon	End capping	Surface area	Pore diameter	Pore volume	Range of pH
100 Diol	Glycero propyl radical	12%	NO	350m^2/g	12um	1.05mL/g	pH2-7.5
300 Diol	Glycero propyl radical	9%	NO	180m^2/g	25nm	1.05mL/g	pH2-7.5

Protein and a polyethylene glycol calibration curve

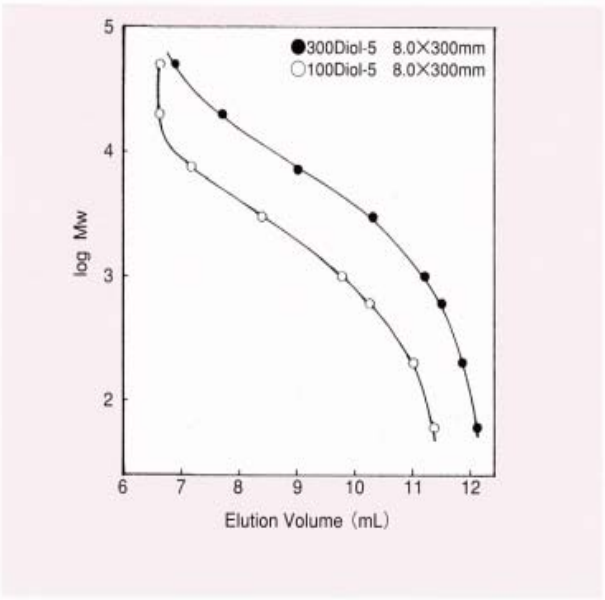
The calibration curve of exclusion limit protein



Conditions:

Column	: Develosil® 100Diol-5 (8.0x300mm)
	Develosil® 300Diol-5 (8.0x300mm)
Mobile phase	: 0.1M Phosphate buffer
	+0.2M NaCl (pH6.8)
Flow rate	: 1.0ml/min
Temperature	: 30℃
Detection	: UV280nm

The calibration curve of polyethylene glycol



Conditions:

Column	: Develosil® 100Diol-5 (8.0x300mm)
	Develosil® 300Diol-5 (8.0x300mm)
Mobile phase	: Water
	: 1.0ml/min
Flow rate	: 30℃
Detection	: RI

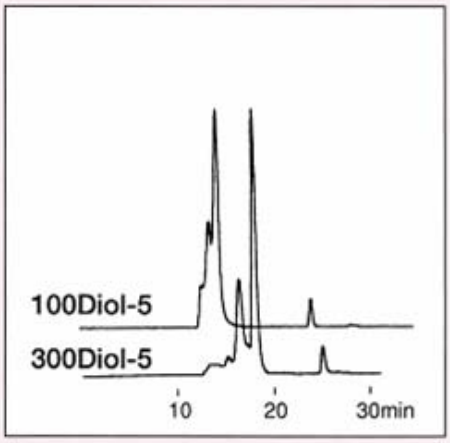
Size exclusion limit molecular weight of Develosil 100Diol and 300Diol

Column name	Proteinic exclusion limit molecular weight	Exclusion limit molecular weight of polyethylene glycol
100 Diol	100,000	10,000
300 Diol	1,000,000	100,000

Please reference this exclusion limit molecular weight as a standard.

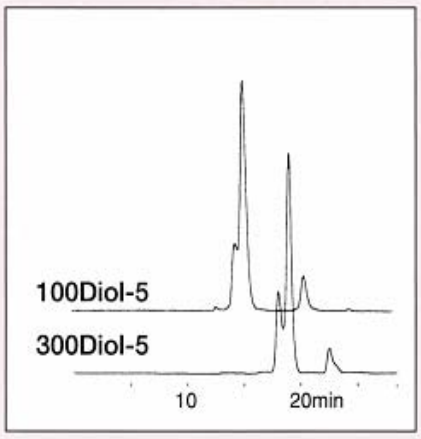
Application

Analysis of a person blood serum



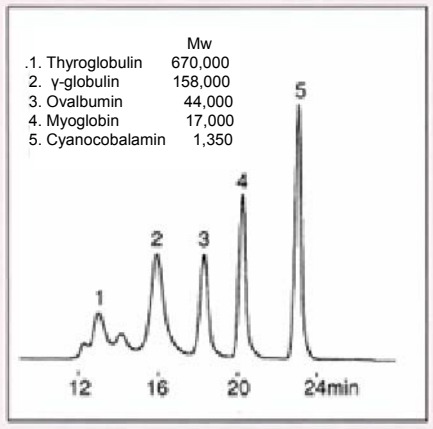
Column: Develosil 300 Diol-5 8.0x300mm
Develosil 100 Diol-5 8.0x300mm
Eluate: 0.1M phosphate buffer + 0.2M NaCl (pH6.8)
Flow reate: 0.5mL/min
Detection: UV280nm

Analysis of albumen



Column: Develosil 300 Diol-5 8.0x300mm
Develosil 100 Diol-5 8.0x300mm
Eluate: 0.1M phosphate buffer + 0.2M NaCl (pH6.8)
Flow reate: 0.5mL/min
Detection: UV280nm

Analysis of standard protein



Column: Develosil 300 Diol-5 8.0x300mm
Eluate: 0.1M phosphate buffer + 0.2M NaCl (pH6.8)
Flow reate: 0.5mL/min
Detection: UV280nm

Bibliography

【Measurement of the uncombined type concentration of drug in the blood by the HPFA method using Develosil 100Diol-5 column】

Determination of Unbound Concentration of Hydrophobic Drugs in Albumin Solutions by High-Performance Frontal Analysis Using a Diol-Silica Column.

Akimasa Shibukawa, Chikako Nakao, Takeshi Sawada, Akira Terakita, Noritsugu Morokoshi, Terumichi Nakagawa.

J.Pharm.Sci.,83〔1994〕868-873.

Table 1—HPLC Conditions for the Determination of the Unbound Diclofenac Concentration

	HPFA	Off-Line Reversed-Phase HPLC
Column	Develosil® 100Diol-5 (30cm×8.0mm i.d.,Nomura Chemical)	LiChrosphere 100 RP-18 (12.5cm×4mm i.d.,Merck)
Mobile phase	Sodium phosphate buffer (pH7.4, I =0.17)	Sodium phosphate buffer (ph6.0) /MeOH=1/1 (v/v)
Flow rate	1.0mL/min	0.8mL/min
Detection	UV,280nm	UV,280nm
Inj vol	167μL	100μL
Temperature	25℃	25℃

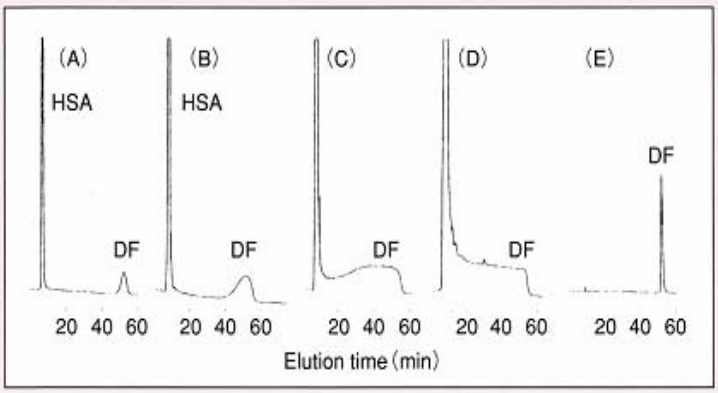


Figure 2 — Elution profile of 200μM diclofenac (DF) -550μM human serum albumin (HSA) mixed solution (A-D) and 200μM DF solution (E). Injection volume: (A) 2 μL, (B) 5 μL, (C) 20 μL, (D) 167 μL, (E) 20 μL. The AUFS of chromatogram E is 8 times larger than that of A-D. See Table 1 for the HPLC conditions.

Table 3 — Unbound Diclofenac (DF) Concentration in Human Serum Albumin (HSA) Solution

	Unbound Concentration (nM) [a]	
	HPFA/HPLC	Ultrafiltration/HPLC
100μM DF-550μM HSA	48.7±2.09 (99.95%)	53.1±3.74 (99.95%)
200μM DF-550μM HSA	112±2.24 (99.94%)	154±14.8 (99.92%)

[a]Mean ±SD (n=5 for HPFA/HPLC, n=4 for ultrafiltration/HPLC). The values in parentheses are the bound DF fraction.