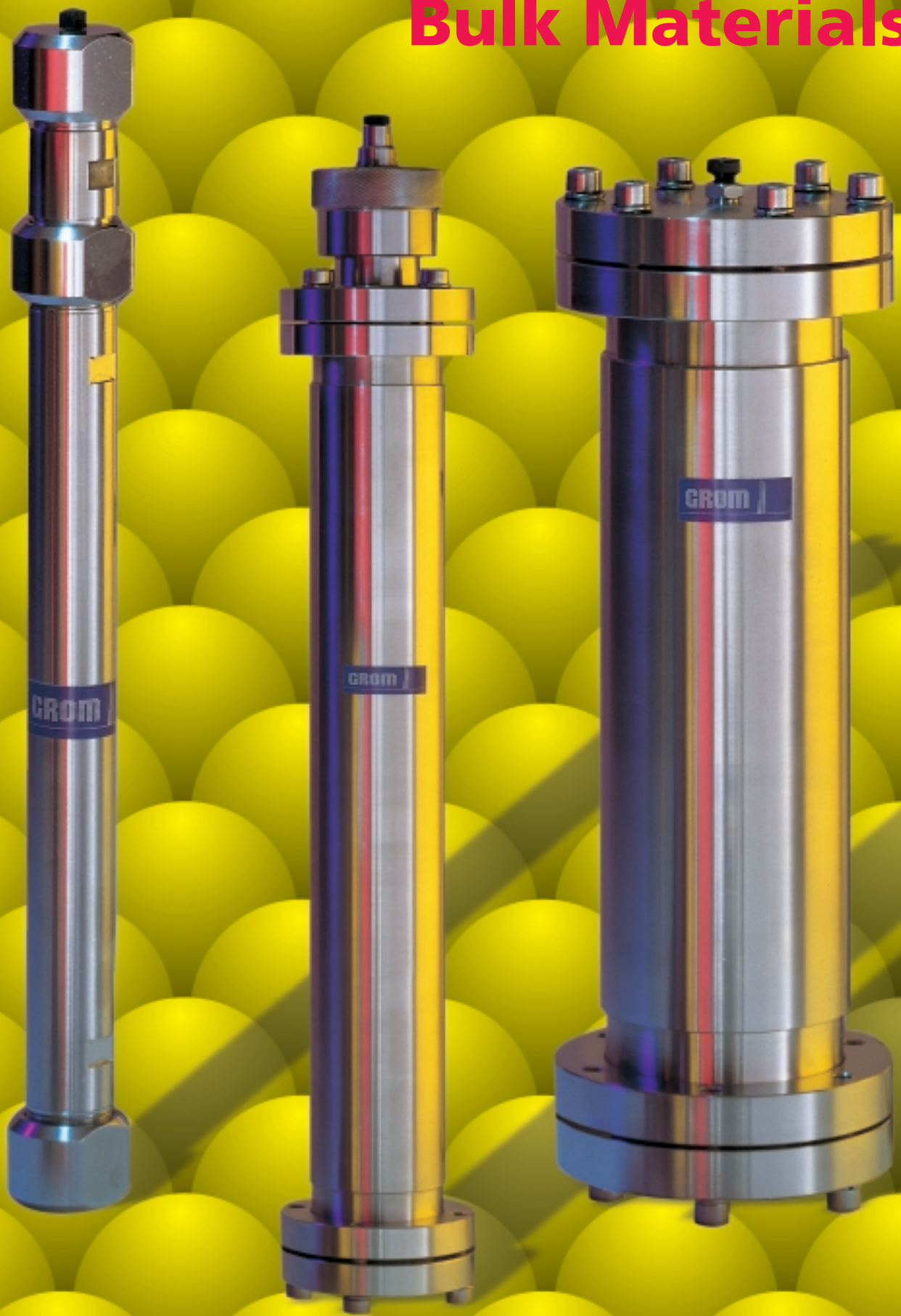
Bulk Materials



20 mm i.d. **60 mm i.d.** **100 mm i.d.**

optimal bulk materials in optimal column hardware for

Preparative HPLC

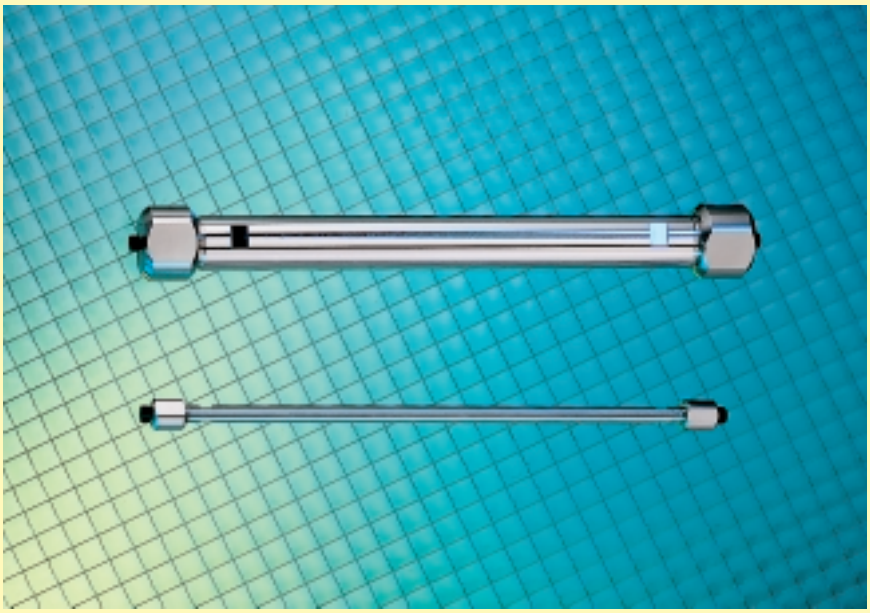
„Up-scaling“- Column Set

For the scale-up of optimized analytical separations without unnecessary waste of expensive samples and solvents, ***GROM*** offers its „Up-scaling“ Sets. These consist of an analytical column (250 x 4 mm) and a preparative column (either 250 x 20 mm or 250 x 40 mm), both of which are filled with completely identical packing (i.e. from the same batch).

This means:
- identical particle diameter,
- identical pore radius,
- identical coating,
- and, most important, the same silica gel as starting material,

guaranteeing identical selectivity of both columns.

A separation optimized on the analytical column (with savings of solvents - both purchase, preparation and disposal) will yield identical resolution when transfered under identical operating parameters (temperature, linear flow rate*, etc.) to the preparative column.
UP-sets thus help to avoid troublesome, time consuming experiments in transferring an analytical separation to a preparative scale.
This is particularly true when the analytical separation has been performed on a column filled with 3 µm diameter particles whereas the preparative column is packed with cheaper, coarser (e.g., 15 µm) particles, thus resulting in different selectivities of the two columns.

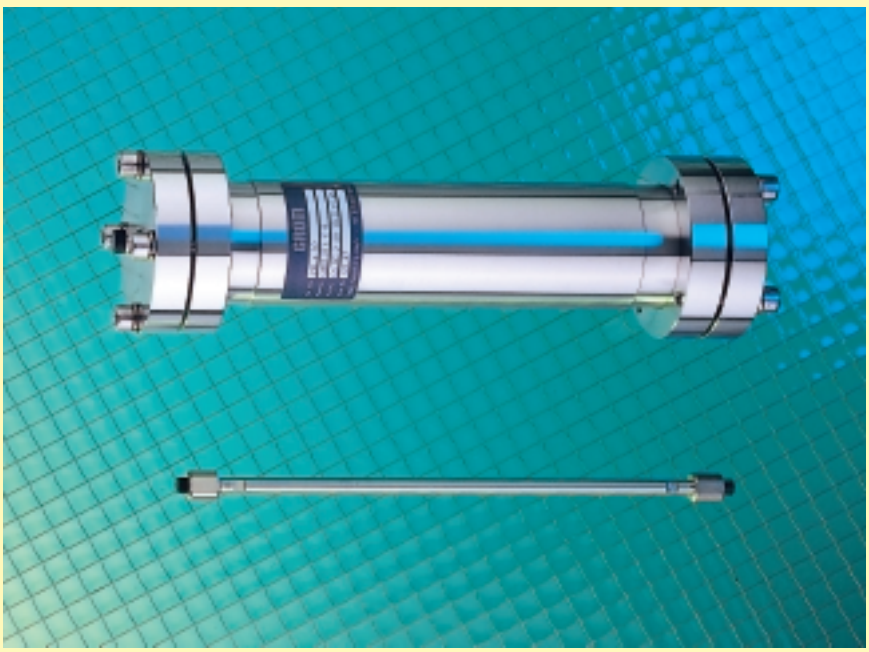


UP-set with 250 x 4 mm analytical column and 250 x 20 mm preparative column

UP- sets, consisting of
one analytical column - 250 x 4 mm
one preparative column - 250 x 20 mm (or 40 mm, respectively)
and with the following packings are available from stock:

GROM-SIL 120 ODS-4 HE, 10 µm	Order No.	GS OD4 1112 U 25 20
	Resp.	GS OD4 1112 U 25 40
GROM Sapphire 110 C8, 10 µm	Order No.	GS OCS 1011 U 25 20
	Resp.	GS OCS 1011 U 25 40
GROM-SIL 120 Si NP-2, 10 µm	Order No.	GS NP2 1112 U 25 20
	Resp.	GS NP2 1112 U 25 40

UP-sets with other stationary phases or with columns of other dimensions upon request.



UP- set with 250 x 4 mm analytical column and 250 x 40 mm preparative column.

* For constant linear flowrate: volumetric flowrate of the prep. column = (vol. flow rate of the analytical column / cross-section of the anal. column) x cross-section of the prep. column.

GROM-SIL - optimal bulk materials in optimal column hardware for preparative HPLC

For preparative HPLC, a wide range of ***GROM-SIL*** phases (see overview table pages 12/13) is available as bulk material in quantities of 100g, 500g, and 1kg. (Larger quantities are available on request).
GROM-SIL phases are characterised not only by high chemical selectivity, but also by their excellent physical properties: they are absolutely spherical, fully porous and are of high specific surface area. As a consequence, GROM-SIL-packed columns show superior flow characteristics (e.g., low back pressure) and high efficiency (i.e., high plate number) and remarkable peak symmetry.

Phase type	Pore diameter	Particle size						
		10 µm	12 µm	15 µm	20 µm	40 µm	50 µm	150 µm
C 18	65 Å	Sapphire		Sapphire	Sapphire	Sapphire		
	100 Å	ODS-2 FE	ODS-2 FE	ODS-2 FE				
	110 Å	Sapphire		Sapphire	Sapphire	Sapphire		
	120 Å	ODS-3 CP		ODS-3 CP	ODS-3 CP			
		ODS-4 HE		ODS-4 HE	ODS-4 HE	ODS-4 HE	ODS-4 HE	ODS-4 HE*
		ODS-5 ST		ODS-5 ST	ODS-5 ST	ODS-5 ST	ODS-5 ST	ODS-5 ST*
	200 Å	ODS-4 HE		ODS-4 HE	ODS-4 HE	ODS-4 HE*	ODS-4 HE	
		ODS-5 ST		ODS-5 ST	ODS-5 ST	ODS-5 ST	ODS-5 ST	ODS-5 ST*
	300 Å	ODS-2 FE	ODS-2 FE	ODS-2 FE				
		ODS-5 ST		ODS-5 ST	ODS-5 ST	ODS-5 ST	ODS-5 ST	ODS-5 ST*
	500 Å		ODS-2 FE					
C8	65 Å	Sapphire		Sapphire	Sapphire	Sapphire		
	100 Å	Octyl-4 FE	Octyl -4 FE	Octyl-4 FE				
	110 Å	Sapphire		Sapphire	Sapphire	Sapphire		
	120 Å	Octyl-6 MB		Octyl-6 MB	Octyl-6 MB	Octyl-6 MB	Octyl-6 MB	
	200 Å	Octyl-6 MB		Octyl-6 MB	Octyl-6 MB	Octyl-6 MB	Octyl-6 MB	
	300 Å			Octyl-4 FE*				
		Octyl-6 MB		Octyl-6 MB	Octyl-6 MB	Octyl-6 MB	Octyl-6 MB	
	500 Å		Octyl-4 FE*					
Phenyl	120 Å	Phenyl-1 FE		Phenyl-1 FE			Phenyl-1 FE	
	200 Å			Phenyl-1 FE*				
	300 Å	Phenyl-1 FE*		Phenyl-1 FE				
C4	65 Å	Sapphire		Sapphire	Sapphire	Sapphire		
	110 Å	Sapphire		Sapphire	Sapphire	Sapphire		
	120 Å	Butyl-1 ST		Butyl-1 ST	Butyl-1 ST	Butyl-1 ST	Butyl-1 ST	
	200 Å	Butyl-1 ST		Butyl-1 ST	Butyl-1 ST	Butyl-1 ST	Butyl-1 ST	
	300 Å	Butyl-1 ST		Butyl-1 ST	Butyl-1 ST	Butyl-1 ST	Butyl-1 ST	
	500 Å		Butyl 2 FE*					
C1	120 Å	TMS-1 ST		TMS-1 ST	TMS-1 ST	TMS-1 ST	TMS-1 ST	
	200 Å	TMS-1 ST*		TMS-1 ST*			TMS-1 ST*	
	300 Å	TMS-1 ST*		TMS-1 ST			TMS-1 ST	
Si	65 Å	Sapphire		Sapphire	Sapphire	Sapphire		
	100 Å	Norm Ph-1 ST	NormPh-1 ST	Norm Ph-1 ST				
	110 Å	Sapphire		Sapphire	Sapphire	Sapphire		
	120 Å	Norm Ph-2 SP		Norm Ph-2 SP	Norm Ph-2 SP	Norm Ph-2 SP	Norm Ph-2 SP	Norm Ph-2 SP
	200 Å	Norm Ph-2 SP		Norm Ph-2 SP	Norm Ph-2 SP	Norm Ph-2 SP	Norm Ph-2 SP	
	300 Å	Norm Ph-2 SP		Norm Ph-2 SP	Norm Ph-2 SP	Norm Ph-2 SP	Norm Ph-2 SP	
	500 Å		Norm Ph-1 ST					
	1000 Å	Norm Ph-2 SP			Norm Ph-2 SP			

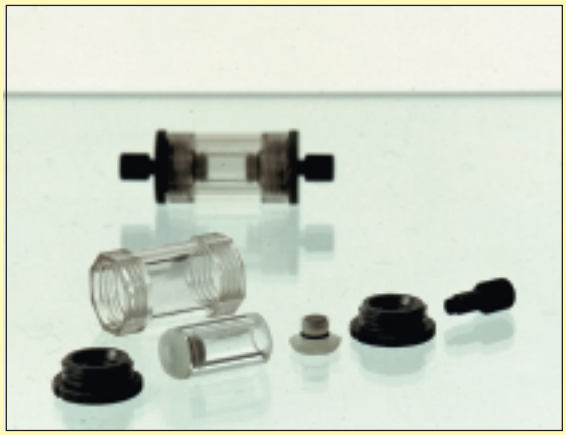
* These phases are available only in amounts of ≥ 1kg.
Upon request, further stationary phases (see overview-table pages 12/13 and 144) are also available (minimum quantity 1kg).

Activated Agarose Gels for Affinity Chromatography

Novarose™ Act^{High} and Novarose™ Act$_{Low}$

The ***Novarose™*** series of activated gels are based on cross-linked agarose beads activated according to a new method [1]. ***Novarose™ Act^{High}*** and ***Novarose™ Act$_{Low}$*** are the perfect choice for easy and reliable immobilization of ligands.

- **Ready for immediate use**
- **Simple coupling procedures at room temperature**
- **Stable at room temperature in aqueous solution and at neutral pH**
- **High flexibility; three pore sizes, each with two degrees of substitution**
- **High flow rate**
- **Coupling of ligands containing sulfhydryl, amino or hydroxyl groups**



Novarose Act Gels

The ***Novarose*** cross-linked agarose beads are activated according to the bromohydrine method. This activation method is based upon well-known chemistry wich allows the coupling to be performed in aqueous solutions.

Novarose Act is supplied as an aqueous suspension. After washing, the gel is immediately ready for use. Since no toxic chemicals are involved and the Novarose products are stable at room temperature, the coupling procedure can, as long as the application allows, easily be performed at the bench at ambient temperature.

The cross-linked, spherical ***Novarose Size Exclusion*** media are used as starting material for the activated gels. They are available in different particle and pore sizes. Agarose beads are normally too soft for modern high performance methods, but, due to a new, patented cross-linking method, ***Novarose SE*** beads are rigid and therefore ideal for all modern chromatography techniques. A spacer of 4 to 16 C-atoms separates the ligand from the Novarose bead.

Being a cross-linked agarose matrix, ***Novarose Act*** possesses most of the characterisics of an extraordinary, ***ideal matrix*** for ***affinity chromatography*** (2). To make it easy to find the right gel for each application, ***Novarose Act*** is available with ***three different pore sizes***, each with ***two degrees of substitution***.

Novarose Act is a simple matrix for convenient immobilization of ligands. It is very easy to use and offers high flexibility and high performance. The gels can be stored at room temperature for one year in aqueous solution at pH 7 (containing ~20 % methanol as preservative) without any significant decrease in coupling activity.

Application Areas

Novarose™ Act High and ***Novarose™ Act Low*** are tailor-made for purification of biomolecules using different chromatographic techniques – ***IEX, HIC, IMAC*** or ***affinity chromatography*** – after coupling with a suitable ligand. As mentioned, they are the perfect gels for easy and reliable immobilization of ligands containing sulfydryl, amino or hydroxyl groups.

Product Specifications

Novarose Product	Exclusion [kD]	Bead Size [µm]	Max. Flow [cm/min]	Degree of Substitution [mol/mol]	Group to be coupled	Field of Applications
Nov.Act$_{Low}$ 100/40	ca. 200	32–60	15	0.10–0.20	-SH, -NH_2, -OH	peptides
Nov.Act^{High} 100/40	ca. 200	32–60	15	0.65–0.75	-SH, -NH_2, -OH	peptides
Nov.Act$_{Low}$ 1 000/40	ca. 1 500	32–60	15	0.10–0.20	-SH, -NH_2, -OH	peptides/proteins
Nov.Act^{High} 1 000/40	ca. 1 500	32–60	15	0.60–0.70	-SH, -NH_2, -OH	peptides/proteins
Nov.Act$_{Low}$ 10 000/40	> 10 000	32–60	15	0.10–0.20	-SH, -NH_2, -OH	proteins
Nov.Act^{High} 10 000/40	> 10 000	32–60	15	0.45–0.55	-SH, -NH_2, -OH	proteins

Chelating High Performance Agarose for Immobilised Affinity Chromatography

Novarose™ IDA^{High}, Novarose™ IDA$_{Low}$, Novarose™ TREN^{High}, Novarose™ TREN$_{Low}$, Novarose™ DPA^{High} and Novarose™ DPA$_{Low}$

The **Novarose™** series of immobilized affinity chromatography (**IMAC**) gels are cross-linked agarose beads activated and coupled according to the **bromohydrine** method [1]. This method gives rise to a spacer arm of 4 to 16 atoms length between the agarose backbone and the attached chelator.

- **Ready for immediate use**
- **High flexibility; three pore sizes, each with two reactive ligand densities**
- **High flow rates**
- **Easy to pack in specially designed columns**

Novarose gels are supplied in aqueous suspensions with ethanol as a preservative and are immediately ready for use after washing.
The ligand density has been shown to have an great impact on the separation (3, 4). In some cases, a high capacity gel is needed, while in other cases a low capacity gel may solve the problem. ***Novarose IMAC*** gels are available in three pore sizes and two degrees of substitution, low and high, to allow maximum flexibility when selecting the optimal IMAC conditions. Furthermore, if there is a need for other capacities, the ***Novarose IMAC*** series may be mixed to obtain any capacity between the specified limits in a simple and reproducible way.

Chelating Groups

There are three different chelators attached to **Novarose** gels: the well known **iminodiacetic acid (IDA)** and two new chelators, namely **tris-(2-ethylaminoethyl) amine (TREN) and dipicolyl amine (DPA)**. These chelators exhibit different interactions with metal ions and consequently show different adsorption/desorption properties towards the solute molecule.

Application Areas

Novarose IMAC gels are designed for purification of biomolecules using immobilized metal ions. This technique is

Product Specifications

Novarose Product*	Exclusion Limit [kD]	Bead Size [µm]	Max. Flow [cm/min]	Mequ. Capacity [µeqv. Cu^{2+}/ml gel]
Novarose IDA$_{Low}$ 100 / 40	ca. 200	32–60	15	10–20
Novarose IDA^{High} 100 / 40	ca. 200	32–60	15	50–60
Novarose IDA$_{Low}$ 1 000 / 40	ca. 1 500	32–60	10	10–20
Novarose IDA^{High} 1 000 / 40	ca. 1 500	32–60	10	50–60
Novarose IDA$_{Low}$ 10 000 / 40	> 10 000	32–60	10	10–20
Novarose IDA^{High} 10 000 / 40	> 10 000	32–60	10	40–50
Novarose TREN$_{Low}$ 100 / 40	ca. 200	32–60	15	10–20
Novarose TREN ^{High} 100 / 40	ca. 200	32–60	15	50–60
Novarose TREN$_{Low}$ 1 000 / 40	ca. 1 500	32–60	10	10–20
Novarose TREN^{High} 1 000 / 40	ca. 1 500	32–60	10	50–60
Novarose TREN$_{Low}$ 10 000 / 40	> 10 000	32–60	10	10–20
Novarose TREN^{High} 10 000 / 40	> 10 000	32–60	10	40–50
Novarose DPA$_{Low}$ 100 / 40	ca. 200	32–60	15	10–20
Novarose DPA^{High} 100 / 40	ca. 200	32–60	15	50–60
Novarose DPA$_{Low}$ 1 000 / 40	ca. 1 500	32–60	10	10–20
Novarose DPA^{High} 1 000 / 40	ca. 1 500	32–60	10	40–50
Novarose DPA$_{Low}$ 10 000 / 40	> 10 000	32–60	10	10–20
Novarose DPA^{High} 10 000 / 40	> 10 000	32–60	10	35–45

* pack size 25 ml, 150 ml and 500 ml

well-known, having been established in 1975 and the interest in its applications has grown rapidly during the last years. Typically, biomolecules (proteins, polypeptides and peptides) containing one or several exposed **histidines** interact strongly with certain metal ions. **Tryptophan** and **cysteine** have also been shown to interact under some conditiones. IMAC gels have also been used quite successfully to fractionate nucleic acids and to separate cells.

Metal Ions and Loading

Most frequently, the following metal ions have been used for IMAC: Zn^{2+}, Cu^{2+}, Ni^{2+}, Fe^{3+}, Co^{2+}, Ca^{2+} and Al^{3+}. But, in principle, any metal ion which interacts with the protein of interest may be used. It should be noted that the total binding capacity for the different gels is always specified only for Cu^{2+} (see table page 139) and will vary slightly for other metal ions.

Adsorption and Desorption

Chromatography conditions vary, of course, with the separation problem. While adsorption solvents are usually aqueous, organic solvents in low concentrations can also be used. Depending on the nature of the chelator, both electrostatic and hydrohobic interaction may be involved and care has to be exercised with respect to the ionic strength of the buffer. Buffers possessing moieties with appreciable affinities for the metal ions are generaly avoided in IMAC.
Protein desorption is done either by altering of the pH or by competition for the metal binding sites between the protein and an another compound like ammonium salts or imidazole buffer.

Storage and Removal of the Metal Ions

Novarose IMAC gels are supplied as aqueous suspensions containing 22 % ethanol as a preservative. They can be stored at room temperature. However, to prevent bacterial growth, it is recommended to add ethanol or sodium azide if stored in buffer.
Many metal ions undergo redox reactions and this may cause deviations in the properties of the gel during long-term storage. If the gel is not to be used for a long time, it is recommended that the metal ions be removed. This can easily be done with a solution of ethylenediamine tetraacetic acid (EDTA).

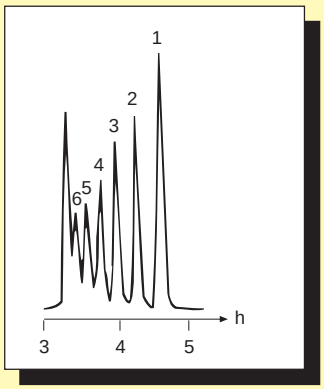
References

1. Patent SE 91 02 211 - 1, Inovata AB
2. G. T. Hermansson, A. K. Mallia and P. K. Smith, „Immobilized Affinity Ligand Techniques“, Academic Press Inc., 1992
3. Inovata Application, No. 305 *(will be supplied upon request by GROM GmbH)*
4. J. J. Winzerling et al., „How to use immobilized metal ion affinity chromatography“, A Companion to Methods in Enzymology **4**, 4–13, 1992

Gel Permeation, Size Exclusion Chromatography (SEC)

Employing Toyopearl Resins packed in the unique *NovoGROM* column hardware

… as described on pages 120 and 121



Toyopearl HW size exclusion chromatography (SEC) resins are macroporous packings dedicated to the separation of large biopolymers such as proteins, nucleic acids, etc. They are also appropriate for molecular weight determination. These resins are semi-rigid, spherical beads with a hydrophilic surface. They are synthezised by co-polymerisation of ethylene glycol and methacrylate-type polymers. There are five types of Toyopearl HW resins available, i.e., HW-40, HW-50, HW-55, HW-65 and HW-75. As easily can be concluded from figure and table, each of these is defined by a given molecular size separation range. They span peptide and protein molecular weights, from 100 to 50,000,000 Daltons depending on resin type.

Separation of ß-cyclodextrin hydrolysate

Experimental conditions: **column:** Toyopearl HW-40S, 600 x 22 mm (2x in series); **sample:** 500 µl 5% (wt) hydrolyzed ß-cyclodextrin: 1) glucose, 2) maltose, 3) maltotriose, 4) maltotetraose, 5) maltopentaose, 6) maltohexaose, **eluent:** distilled water; **flow rate**: 1.0 ml / min; **temperature:** 40°C; **detection**: RI

Properties and Molecular Weight Separation Ranges

Toyopearl Resin	Particle Size [µm]	Pore Size [Å]	Polyethylene Glycols/Oxides [Dalton]	Dextrans [Dalton]	Globular Proteins [Dalton]
HW-40S HW-40F HW-40C	20–40 30–60 50–100	50	100 – 3 000	100 – 7 000	100 – 10 000
HW-50S HW-50F	20–40 30–60	125	100 – 18 000	500 – 20 000	500 – 80 000
HW-55S HW-55F	20–40 30–60	300	100 – 150 000	1 000 – 200 000	1 000 – 700 000
HW-65S HW-65F	20–40 30–60	1 000	500 – 1 000 000	10 000 – 1 000 000	40 000 – 5 000 000
HW-75F	30–60	>1 000	4 000 – 5 000 000	100 000 – 10 000 000	500 000 – 50 000 000

S, superfine; **F,** fine; **C,** coarse

Hydrophobic Interaction Chromatography (HIC)

… a powerful tool for biochemical applications

Toyopearl HW-65 resins, rather well-accepted for size exclusion chromatography, are an excellent base matrix for the Toyopearl 650 series of hydrophobic interaction resins. The hydrophobicity of these packings increases through the series: Ether-, Butyl-, Phenyl. They provide outstanding performance when packed in the unique *NovoGROM* columns …

Toyopearl Hydrophobic Interaction Chromatography Resins, typical properties

Toyopearl Resin	Particle Size [µm]	Exclusion Limit [Daltons]	Mass Recovery Lysozyme [%]	Mass Recovery Ovalbumin [%]	Adsorption Capacity [mg/ml]
Ether-650S	20–50	1×10^6	94	83	10–30
Ether-650M	40–90	1×10^6	94	94	10–30
Butyl-650S	20–50	1×10^6	85	73	30–50
Butyl-650M	40–90	1×10^6	85	73	30–50
Butyl-650C	50–150	6×10^6	85	73	30–50
Phenyl-650S	20–50	1×10^6	92	88	30–50
Phenyl-650M	40–90	1×10^6	92	88	30–50
Phenyl-650C	50–150	6×10^6	92	88	30–50

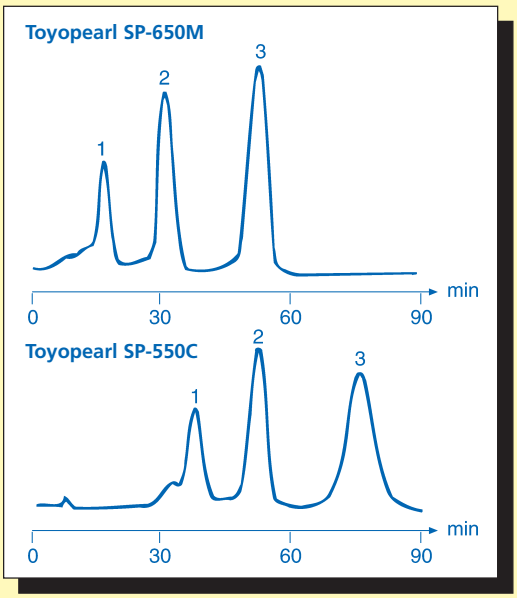
S, superfine; **M,** medium; **C,** coarse

Toyopearl Ion Exchange Chromatography (IEC)

Packed in unique *NovoGROM* column hardware

… as described on pages 120 and 121

The numerous surface hydroxyl groups of the **Toyopearl HW-55**, and **HW-65** resins, which have been well-proven in size exclusion chromatography, provide excellent attachment points for other functional groups and ligands. Thus, these resins are used as the principal base matrix for the **Toyopearl 550** and **650** series of ion exchange resins. The Toyopearl 550 series of IEC resins is designed to enable high volume sample throughput, whereas Toyopearl 650 series provides the high resolution required during the mid to final stages of a purification process. They have an estimated globular protein exclusion limit of 700000 and 5000000 Daltons respectively. All Toyopearl ion exchangers are chemically and thermally stable due to their polymeric backbone. They can be used from pH 3.0 to pH 10.5. Caustic or acidic solutions can also be applied for sanitation (e.g., 1 % sodium hypochlorite) and depyrogenation. Furthermore, in some cases, overnight cleaning or sterilisation procedures with strong acid or base are acceptable, for instance with **Toyopearl DEAE-650**.



Comparison of resolution by strong cation exchangers

Experimental conditiones: **column:** 100 x 15 mm; **sample:** 1) ribonuclease A, 2) cytochrome C, 3) lysozyme; **elution:** linear gradient (within 60 min) 0 → 0.5 M NaCl in 0.02 M Na-phosphate pH 7.0; **flow rate:** 1.1 ml/min; **UV detection:** 280 nm

Toyopearl Ion Exchange Resins, typical properties

Toyopearl Resin		Functional Group	Particle Size [µm]	Ion Exchange Capacity [meq/ml]	Adsorption Capacity [mg/ml]	Exclusion Limit [Daltons]	Pressure Drop [bar]
CM-650S	HW-65	-O-CH_2-COOH	20–50	0.08–0.12	40–60	1×10^6	max. 0.4
CM-650M			40–90	0.08–0.12	40–60	1×10^6	max. 0.2
CM-650C		weak cation exchanger	50–150	0.05–0.11	35–55	6×10^5	max. 0.15
SP-650S	HW-65	-O-CH_2-CH_2-CH_2-SO_3	20–50	0.13–0.17	40–60	1×10^6	max. 0.5
SP-650M			40–90	0.13–0.17	40–60	1×10^6	max. 0.2
SP-650C		strong cation exchanger	50–150	0.12–0.18	35–55	6×10^5	max. 0.15
DEAE-650S	HW-65	-O-CH_2-CH_2-N-$(C_2H_5)_2$	20–50	0.08–0.12	25–35	1×10^6	max. 0.4
DEAE-650M			40–90	0.08–0.12	25–35	1×10^6	max. 0.2
DEAE-650C		anion exchanger	50–150	0.05–0.11	25–35	6×10^5	max. 0.2
QAE-550C	HW-55	-O-R´-N^+-$(R)_3$ strong anion exchanger	50–150	0.28–0.38	60–80	5×10^5	max. 0.15
Super Q -650S	HW-65	-O-R´-N^+-$(R)_3$	20–50	0.20–0.30	105–155	–	max. 0.4
Super Q -650M			40–90	0.20–0.30	105–155	–	max. 0.2
Super Q -650C		strong anion exchanger	50–150	0.20–0.30	105–155	–	max. 0.15
SP-550C	HW-55	-O-CH_2-CH_2-CH_2-SO_3 strong cation exchanger	50–150	0.14–0.18	80–120	5×10^5	max. 0.15

S, superfine; **M,** medium; **C,** coarse

Conditions: Exclusion Limits are +/- 30 % and were determined using polyethylene glycol, polyethylene oxide or dextran standards, as appropriate. Pressure drop has been determined in a 600 x 22 mm column at 1.0 ml/min using 0.1 M NaCl as eluent. Samples used to determine adsorption capacity were: CM-650, hemoglobin; SP-650, lysozyme; DEAE-650, bovine serum albumin.

TSK-GEL 5PW Preparative LC Resins packed in *NovoGROM* columns

The ultimate alternative to preparative HPLC

TSK-GEL 5PW Preparative LC Resins are most often used for up-scaling in biopharmaceutical applications. The TSKgel DEAE-5PW and TSK-gel SP-5PW resins permit high resolution in ion exchange chromatography, whereas the outstanding, TSKgel Phenyl-5PW and TSKgel Ether-5PW resins are used for up-scaling in hydrophobic interaction chromatography. They have a particle size of 30 µm. All are made from the same hydrophilic G5000 PW- polymer and guarantee excellent preparative separations, especially when packed in the unique ***NovoGROM*** columns (s. pages 120 and 121).

Table 10 **TSK-GEL Preparative PW Resins for Ion Exchange Chromatography**

TSK-GEL Resin	Particle Size [µm]	Exclusion Limit [Daltons]	Adsorption Capacity [mg/ml]
SP-5PW	20–40	5 000 000 (globular proteins)	20–40 (lysozyme)
DEAE-5PW	20–40	5 000 000 (globular proteins)	20–40 (bovine serum albumin)

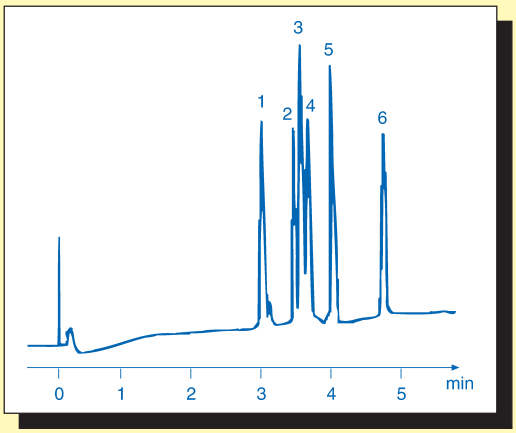
TSK-GEL Preparative PW Resins for Hydrophobic Interaction Chromatography

TSK-GEL Resin	Particle Size [µm]	Exclusion Limit [Daltons]	Adsorption Capacity [mg/ml]
Ether-5PW	20–40	9×10^5 (polyethylene glycol)	20–40 (lysozyme)
Phenyl-5PW	20–40	9×10^5 (polyethylene glycol)	20–40 (lysozyme)

The huge variety of ***NovoGROM*** HPLC columns is offered packed with

Resin-based TSK-GEL reversed phases

They provide maximum selectivity and resolution for a wide range of peptides and proteins. The hydrophilic backbone of these TSKgel reversed phase packings guarantees unsurpassed chemical and physical stability for longer column life. Several problems inherent to silica based packings, such as a lack of pH stability or secondary interactions due to exposed silanol groups, are eliminated when columns are packed with TSKgel reversed phase resins.



Rapid peptide separation on TSK-gel Octadecyl-NPR
Experimental conditions: **column:** TSK-gel Octadecyl-NPR, 4.6 x 35 mm; **eluent / graduent:** 10 min linear from 0 % to 80 % ACN in 0.2 % TFA; **flow rate:** 1.5 ml / min; **temperature:** ambient; **detection:** UV, 220 nm; **sample:** 1) α-endorphin, 2) bombesin, 3) γ–endorphin, 4) angiotensin, 5) somatostatin, 6) calcitonin

Resin-based TSK-gel reversed phase packings, typical properties

Toyopearl Resin	Functional Group	Particle Size [µm]	Pore Size [Å]	Ligand Density [meq/ml]	Linear Flow [cm/min]	Max. Pressure [MPa]
Phenyl-5PW RP	phenyl, monomeric	10 13 20	1 000 1 000 1 000	1 1 1	3.0–6.0 1.6–2.2 —	3.0 3.0 —
Octadecyl-4PW	C18 alkyl, monomeric	7 13	500 500	1 1	3.0–6.0 0.8–1.6	1.2 3.5
Octadecyl-NPR	C18 alkyl, monomeric	2.5	nonporous	1	6.0–9.0	20.0