BESTCHROM
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Bestdex G-25 Dextran Resin

Instruction for use

Contents

1. Introduction

Bestdex G-25 series resin is made by dextran as raw material and chlorinated propylene oxide as cross-linking agent. Featuring hydrophilicity and multiple pore sizes, the product is widely used for buffer exchange、desalting and small molecule removal of bio-organism samples(protein, polysaccharide and nucleic acid). It can also be used for separation of peptide, oligonucleotides as well as the purification of antibiotics, chemical synthesized medication and natural organism. Besides, application to nanotechnology and sewage disposal is becoming increasingly commonplace.

Functioning as molecular sieves, Bestdes G-25 series separate molecules in various sizes according to the variety in retention time of molecules in the column. Bestdex G-25 is divided into four types based on particle size: Bestdex G-25 C (coarse), Bestdex G-25 M (Medium), Bestdex G-25 F(Fine), and Bestdex G-25 SF (Super fine).

2.Technical characteristics

Product		Bestdex G-25 C	Bestdex G-25 M	Bestdex G-25 F	Bestdex G-25 SF
Appearance		White to yellowish spherical powder			
Matrix		Cross-linking dextran			
Particle size (dry) +		100~300μm	50~150μm	20~80μm	20~50μm
Particle size (wet)++		160~500μm	80~240μm	30~130μm	30~80μm
Average particle size (wet)++		320μm	140μm	90μm	50μm
Max. flow velocity（cm/h）+++		650	430	240	110
Separation range	Linear molecule	100D~5KD			
	Globular molecule	1KD~5KD(10~45 amino acid)			
	nucleotide	<8bp			
Swelling ratio（mL /g dry powder）		2.5			
Volume of swelling glue per gram of dry powder		4~6mL (water) ~4.5mL (0.5M NaCl)			
pH stability		2~13			
Chemical Stability		Stable in common aqueous buffers: 0.2M NaOH，0.2M HCl，1MHAc，6M GuHCl,8M Urea, 1%SDS, 24% ethanol, 30% propanol, 30% acetonitrile			

+Particle size is normally distributed, and particles within this range account for more than 80% of the total.

++ The particle size in different solutions will be different, this data is the particle size in PBS

+++BXK26/70, Column height 60cm, pressure 0.3MPa measured

3. Method of chromatographic

Since the resin is supplied as a dry powder, it is necessary to swell and then fill the chromatography column before use.

3.1 Swelling

- Calculate the required amount of Bestdex G-25 dry powder according to the volume of the chromatography column

  Dry powder(g)=(CV×1.15)÷4.5
- Pour the resin into 0.1M NaCl at 80~100℃ of 5 times the weight of dry powder and stir it slightly, swell for 1h, or swell for 4h at room temperature. (Note that during the swelling process, do not use a magnetic stirrer to stir. Using a magnetic stirrer will cause the resin particles to break), It is best to degas under negative pressure after swelling at room temperature.
- After swelling is completed (the swelling at high temperature needs to be cooled to room temperature), remove part of the supernatant to make the volume of the sedimentation gel account for 50% to 75% of the total volume, and mix well for use.

3.2 Column packing

- The packing buffer has little effect on the packing effect. You can use the buffer for chromatography or water as the packing solution.
- Take a cleaned BXK column (BXK series columns with diameters ranging from 1cm to 30cm can satisfy different scale chromatography applications). Purge the bubbles trapped at the end-piece net by draining some packing solution through the column outlet. Leave about 1cm water at the bottom of the column and close the bottom outlet. Adjust the column so that it is perpendicular to the ground.
- Slowly pour the slurry into the column at one time (use a packing reservoir if necessary). Do not bring any air bubbles into the column.

  Packing reservoir: Empty glasstube with same diameter as the BXK column.
- Fill the remainder of the column with packing solution. Connect the packing reservoir to the chromatography system, open the flow velocity, drain the bubbles in the hose, close the flow velocity, and tighten the top cover of the packing reservoir.

  Note: This operation is only applicable to BXK 50 and below chromatographic columns.

✧ After pouring, stir well again with Stirrer, and then wash the resin particles on the inner wall of the column from top to bottom with the packing solution, and let the resin settle naturally until there is about 1cm of clarifying solution on the suspension. Mount the adapter and connect the adapter to the chromatography system or peristaltic pump. Lower the adapter to descend to contact with the clarifying solution and tighten the sealing ring after it is fully immersed in the clarifying solution. With the outlet of the top piece is opened, slowly move the adapter down until all bubbles are drained.

Note: This operation is only applicable to BXK 100 and above columns. Flushing the inner

wall reduces the resin particles sticking between the seal ring and the column wall, avoiding the risk of leakage.

- Set the packing flow velocity. Due to the high rigidity of Bestdex resin, flow velocity and pressure are positively correlated. Since packing pressure lower than 5 bar will hardly damage resin, it is recommended to pack column at highest possible flow. Take BXK50 and column height 30cm for example.

Product	Bestdex G-25 C	Bestdex G-25 M	Bestdex G-25 F	Bestdex G-25 SF
Recommended flow velocity (cm/h)	600	400	200	100

- Open the column bottom plug and start the peristaltic pump or chromatography system at above-set flow velocity. Make sure internal column pressure is lower than the max pressure of column. Thus, appropriately reduce the flow velocity when pressure is too high during column packing.
- When the resin is fully gravity-settled, keep it for more than 30 minutes, mark the resin surface, stop the pump.
- Remove the packing reservoir (if any), mount the adaptor, lower the adaptor to about 0.5cm above the resin surface, and continue to press the column using the above flow velocity until the bed is completely consolidated, mark the consolidated bed height.
- Stop the pump, open top plug, close the bottom plug, loosen the O-ring seal slightly, press the adaptor to about 0.3cm below the marked position, tighten the O-ring seal, close adaptor stop plug, and complete the column packing.

3.3 Evaluation of Packing

- The packing quality of chromatographic column can be confirmed by column efficiency measurement and evaluation. The tests are required after the column packing, during the column working life and when the separation and purification performance weakens. The method usually relies on the height equivalent to a theoretical plate(HETP) and the asymmetry factor(As).
- Acetone or NaCl solution can be used as sample for the testing. Sample solution and mobile phase can be prepared according to the following table.

	Acetone method	NaCl method
Sample	1.0%(v/v)acetone in water	0.8M NaCl in water
Sample volume	1.0%CV	1.0%CV
Mobile phase	Water	0.4M NaCl in water
Flow velocity	30cm/h	30cm/h
Monitor	UV280 nm	Conductivity

- Method for measuring HETP and As:

  Use UV curve or the conductivity curve to calculate the height equivalent of theoretical plate (HETP), number of theoretical plates(N) and the asymmetry (As):

HETP=L/N

$N=5.54(V_R/W_h)^2$

Note: V_R = retention volume

W_h = half-peak width

L = column height

N = the number of theoretical plates

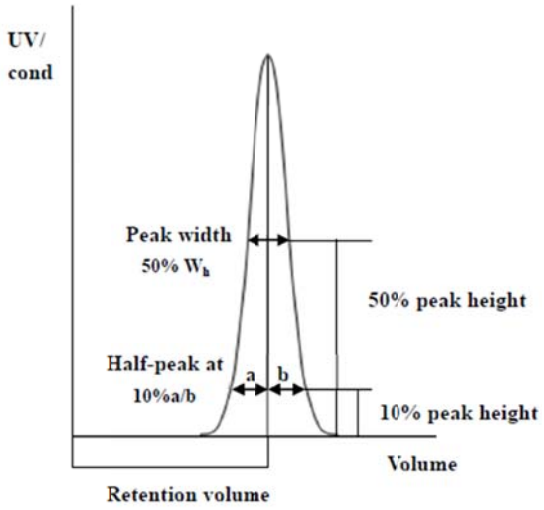
(The units of V_R and W_h should be the same)

As=b/a

Note:

a= 1st half peak width at 10% of peak height

b= 2nd half peak width at 10% of peak height



- Evaluation the column packing

  As a guideline, if the value of HETP is less than 3 times the average particle size(d_{50}) of the resin and the As is between 0.8~1.8, the column is very efficient. The unsatisfactory results should be analyzed and the column should be repacked.

3.4 Chromatographic method

- Buffer selection: Aqueous buffer is generally used. The pH and conductivity of the buffer have little effect on the separation. The main consideration is the stability of the sample in the buffer.
- Flow velocity: Operate at the recommended flow velocity, the higher the column height and the slower the flow velocity.
- Sample preparation: In order to prevent blocking of the column, the sample needs to be filtered by microporous membrane of 0.45μm before loading, If the difference between the target and the impurities is relatively small, the sample volume should be 1% to 5% of the column volume. If the target and the impurities to be removed are relatively large (such as the removal of inorganic salts in the sample), the sample volume should 10% to 25% of the sample base on the CV.

4. Influencing factors of separation effect

Bestdex G-25 series resin separate substances with different sizes according to the principle of molecular sieve. The type of buffer and pH generally do not affect the separation. When the separation situation is abnormal, the following aspects can be analyzed:

- Ionic action: The dextran chain contains a small amount of terminal carboxyl groups, and it will interact with the substance to be separated under very low ionic strength, which can be eliminated when the salt concentration in the buffer is greater than 50mM.
- Hydrophobic effect: The cross-linking agent contains a small amount of etheroxy groups. The use of phenol: acetic acid: water = 1: 1: 1 solution and urea or potassium thiocyanate can counteract this effect. When the salt concentration in the buffer exceeds 0.5M and the low pH will cause the hydrophobic effect to increase, organic solvents such as ethanol and acetonitrile can be added to the buffer to eliminate this effect.

5. Cleaning-in-place(CIP)

Bestdex G-25 series resin may decrease the column efficiency after a period of use, and the separation effect becomes poor. The following process can be used for cleaning and regeneration. (CIP).

- Rinse 2CV with distilled water
- Flush 1CV with 1M NaCl
- Rinse 1CV with 0.2M NaOH
- Rinse 4CV with distilled water

6. Sterilization

The swollen Bestdex G-25 can be autoclaved at 121℃ for 30 minutes, or treated with 0.5M NaOH for 30~60 minutes to to reduce the risk of microbial contamination.

7. Storage

The Bestdex G-25 are supplied in a cool and dry place to prevent moisture absorption. The swollen Bestdex G-25 should be stored in 20% ethanol and sealed at 2-30℃, in order to prevent ethanol volatilization and microbial growth, it is recommended to replace the storage solution every 3 months.

8. Disposal and Recycling

Bestdex G-25 are very difficult to degrade in nature, incineration is recommended to protect the environment.

9. Order information

Product	Code No.	Pack size	Product	Code No.	Pack size
Bestdex G-25 C	AG0111	25g	Bestdex G-25 F	AG0131	25g
	AG0112	100g		AG0132	100g
	AG0113	500g		AG0133	500g
	AG0114	1kg		AG0134	1kg
	AG0115	5kg		AG0135	5kg
	AG0116	10kg		AG0136	10kg
	AG331115	25kg		AG333115	25kg
Bestdex G-25 M	AG0121	25g	Bestdex G-25 SF	AG0141	25g
	AG0122	100g		AG0142	100g
	AG0123	500g		AG0143	500g
	AG0124	1kg		AG0144	1kg
	AG0125	5kg		AG0145	5kg
	AG0126	10kg		AG0146	10kg
	AG332115	25kg		AG334115	25kg

Product（wet resin）	Code No.	Pack size
Bestdex G-25 C	AG1402	1L
Bestdex G-25 M	AG0502	1L
	AG0505	5L
	AG0506	10L
	AG0507	20L
Bestdex G-25 F	AG0602	1L
Bestdex G-25 SF	AG0702	1L

Prepacked columns	Code No.	Pack size
BC10 G-25 F	EG002	30
EzFast G-25 F	EG06021	1×1mL
	EG06031	5×1mL
	EG104201	1×5mL
	EG001	5×5mL
EzLoad 16/10 G-25 F	EG025	1 pcs
EzLoad 26/10 G-25 F	EG026	1 pcs
EzFast G-25 M	EG05021	1×1mL
	EG05031	5×1mL
	EG05023	1×5mL
	EG05033	5×5mL
EzLoad 50/20 G-25 M	EG022	1 pcs
EzLoad 16/10 G-25 M	EG104204	1 pcs
EzLoad 26/10 G-25 M	EG008	1 pcs