

Data sheet

SepFast™ Butyl (400, 500, 600)
SepFast™ Pentyl (400, 500, 600)
SepFast™ Hexyl (400, 500, 600)
SepFast™ Phenyl (400, 500, 600)
SepFast™ Heptyl (400, 500, 600)
SepFast™ Octyl (400, 500, 600)

1. Introduction

SepFast Butyl, SepFast Pentyl, SepFast Hexyl, SepFast Phenyl, SepFast Heptyl and SepFast Octyl are hydrophobic interaction chromatography (HIC) adsorbents having a carbon chain of C4, C5, C6, C6 ring, C7 and C8, respectively. They are specially designed for the purification of biological molecules based on their hydrophobicity profiles.

HIC is a versatile technique and could show high selectivity to individual molecules according to their exposed hydrophobic zones. It is particularly useful for intermediate and final-stage purifications. A HIC medium normally binds at moderate to high salt concentrations. It is logical to place HIC step after an IEX step where molecules are usually eluted at high salt conditions.

HIC media shows much milder purification conditions than reversed phase chromatography (RPC) media. Better biological activity could be maintained in HIC operations than RPC operations.

SepFast HIC media offers a broad range of choice from C4 to C8. Generally speaking, the longer the carbon chain, the higher the surface hydrophobicity. Researchers have the chance to choose the best HIC medium for their applications. The core advantages are:

- High sample loading capacity
- More choices than other suppliers
- High separation power

The base matrix is made of agarose that has been highly cross-linked. It is very stable to most of the chemical conditions experienced in the bioprocessing industry.

For each type of HIC ligand, there is a choice of 3 different base matrices according to the pore accessibility of target molecules. The feature and selection guide is listed as follows:

400 serial	500 serial	600 serial
SepFast Butyl-400	SepFast Butyl-500	SepFast Butyl-600
SepFast Pentyl-400	SepFast Pentyl-500	SepFast Pentyl-600
SepFast Hexyl-400	SepFast Hexyl-500	SepFast Hexyl-600
SepFast Phenyl-400	SepFast Phenyl-500	SepFast Phenyl-600
SepFast Heptyl-400	SepFast Heptyl-500	SepFast Heptyl-600
SepFast Octyl-400	SepFast Octyl-500	SepFast Octyl-600
The above HIC media is designed to purify peptides or small proteins	The above HIC media is designed to purify most medium to large proteins.	The above HIC media is designed to purify large to very large proteins.

Characteristics of SepFast HIC media:

	Butyl-400	Pentyl-400	Hexyl-400	Phenyl-400	Heptyl-400	Octyl-400
	Butyl-500	Pentyl-500	Hexyl-500	Phenyl-500	Heptyl-500	Octyl-500
	Butyl-600	Pentyl-600	Hexyl-600	Phenyl-600	Heptyl-600	Octyl-600
Matrix	Highly cross-linked agarose					
Functional group	-(CH ₂) ₃ CH ₃	-(CH ₂) ₄ CH ₃	-(CH ₂) ₅ CH ₃	-C ₆ H ₆	-(CH ₂) ₆ CH ₃	-(CH ₂) ₇ CH ₃
Particle size	50 - 150 μm					
Pressure-flow property*	>1000 cm/h for 400 serials; >500 cm/h for 500 serials; >300 cm/h for 600 serials					
Operational pressure	Up to 3 bar					
pH stability	2-14 (short term) and 3-12 (long term)					
Working temperature	+4°C to +30°C					
Chemical stability	All commonly used buffers; 1 M acetic acid, 1 M NaOH, 6M guanidine hydrochloride, 8 M urea, 30% acetonitrile, 30% isopropanol, 70% ethanol, 3 M (NH ₄) ₂ SO ₄					
Storage	20% ethanol					

*Measured in a 32 mm ID column at a bed height of 20 cm.

2. Method optimization

We recommend scouting the parameters among loading capacity, flow velocity, binding pH, binding ionic strength, elution speed and gradient etc. We recommend to pay special attention to optimize elution conditions to achieve the best separation power.

In general, balancing the degree of component separation against process throughput is the major consideration when optimizing a method. Besides, for the purification of instable or shearing-force sensitive molecules, the operational condition needs be optimised to balance the throughput and the possible damage to the target molecule.

3. Maintenance

Depending on the individual applications, the media may be used many times. For the re-use purpose, please see the following instructions.

Cleaning-in-place (CIP)

CIP is a procedure that removes strongly bound materials such as lipids, endotoxins and denatured proteins that remain in the adsorbent surface after regeneration. Regular CIP prevents the build up of contaminants in the packed bed and helps to maintain the column performance.

A specific CIP protocol should be developed for each process according to the type of contaminants present. The frequency of CIP depends on the nature of individual applications.

The following information works as a general guidance.

The contaminants bound by hydrophobic nature can be removed by the following reagents: 1 M NaOH, low percentage non-ionic detergents (e.g. 0.1 – 2%), 30% isopropanol in basic or acidic conditions (e.g. in the presence of acetic acid or phosphoric acid). A combination of the above reagents can be explored as well. In general, the incubation time should be longer (e.g. from 30 minutes to 2 hours) to ensure full dissociation of the contaminants.

Sanitization

Sanitization using 0.5-1.0 M NaOH with a contact time of 1 hour is recommended.

4. Storage

The media should be stored in 20% ethanol or 0.02% sodium azide to prevent microbial growth. Store the media at a temperature of +4°C to +30°C. Before use, equilibrate the media with at least 5 bed volume of running buffer.

5. Ordering information

Product	Quantity	Code no.
SepFast Butyl-400	25 ml	480101
	100 ml	480102
	1 litre	480103
Disposable SepFast Butyl-400 column	5 x 1 ml	480104
	1 x 5 ml	480105
	1 x 10 ml	480106
	1 x 20 ml	480107
SepFast Butyl-500	25 ml	480201
	100 ml	480202
	1 litre	480203
Disposable SepFast Butyl-500 column	5 x 1 ml	480204
	1 x 5 ml	480205
	1 x 10 ml	480206
	1 x 20 ml	480207
SepFast Butyl-600	25 ml	480301
	100 ml	480302
	1 litre	480303
Disposable SepFast Butyl-600 column	5 x 1 ml	480304
	1 x 5 ml	480305
	1 x 10 ml	480306
	1 x 20 ml	480307
SepFast Pentyl-400	25 ml	480401
	100 ml	480402
	1 litre	480403
Disposable SepFast Pentyl-400 column	5 x 1 ml	480404
	1 x 5 ml	480405
	1 x 10 ml	480406
	1 x 20 ml	480407
SepFast Pentyl-500	25 ml	480501
	100 ml	480502
	1 litre	480503
Disposable SepFast Pentyl-500 column	5 x 1 ml	480504
	1 x 5 ml	480505
	1 x 10 ml	480506

	1 x 20 ml	480507
SepFast Pentyl-600	25 ml	480601
	100 ml	480602
	1 litre	480603
Disposable SepFast Pentyl-600 column	5 x 1 ml	480604
	1 x 5 ml	480605
	1 x 10 ml	480606
	1 x 20 ml	480607
SepFast Hexyl-400	25 ml	480701
	100 ml	480702
	1 litre	480703
Disposable SepFast Hexyl-400 column	5 x 1 ml	480704
	1 x 5 ml	480705
	1 x 10 ml	480706
	1 x 20 ml	480707
SepFast Hexyl-500	25 ml	480801
	100 ml	480802
	1 litre	480803
Disposable SepFast Hexyl-500 column	5 x 1 ml	480804
	1 x 5 ml	480805
	1 x 10 ml	480806
	1 x 20 ml	480807
SepFast Hexyl-600	25 ml	480901
	100 ml	480902
	1 litre	480903
Disposable SepFast Hexyl-600 column	5 x 1 ml	480904
	1 x 5 ml	480905
	1 x 10 ml	480906
	1 x 20 ml	480907
SepFast Phenyl-400	25 ml	481001
	100 ml	481002
	1 litre	481003
Disposable SepFast Phenyl-400 column	5 x 1 ml	481004
	1 x 5 ml	481005
	1 x 10 ml	481006
	1 x 20 ml	481007
SepFast Phenyl-500	25 ml	481101
	100 ml	481102
	1 litre	481103
Disposable SepFast Phenyl-500 column	5 x 1 ml	481104
	1 x 5 ml	481105
	1 x 10 ml	481106
	1 x 20 ml	481107
SepFast Phenyl-600	25 ml	481201

	100 ml	481202
	1 litre	481203
Disposable SepFast Phenyl-600 column	5 x 1 ml	481204
	1 x 5 ml	481205
	1 x 10 ml	481206
	1 x 20 ml	481207
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SepFast Heptyl-400	25 ml	481301
	100 ml	481302
	1 litre	481303
Disposable SepFast Heptyl-400 column	5 x 1 ml	481304
	1 x 5 ml	481305
	1 x 10 ml	481306
	1 x 20 ml	481307
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SepFast Heptyl-500	25 ml	481401
	100 ml	481402
	1 litre	481403
Disposable SepFast Heptyl-500 column	5 x 1 ml	481404
	1 x 5 ml	481405
	1 x 10 ml	481406
	1 x 20 ml	481407
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SepFast Heptyl-600	25 ml	481501
	100 ml	481502
	1 litre	481503
Disposable SepFast Heptyl-600 column	5 x 1 ml	481504
	1 x 5 ml	481505
	1 x 10 ml	481506
	1 x 20 ml	481507
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SepFast Octyl-400	25 ml	481601
	100 ml	481602
	1 litre	481603
Disposable SepFast Octyl-400 column	5 x 1 ml	481604
	1 x 5 ml	481605
	1 x 10 ml	481606
	1 x 20 ml	481607
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SepFast Octyl-500	25 ml	481701
	100 ml	481702
	1 litre	481703
Disposable SepFast Octyl-500 column	5 x 1 ml	481704
	1 x 5 ml	481705
	1 x 10 ml	481706
	1 x 20 ml	481707
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SepFast Octyl-600	25 ml	481801
	100 ml	481802
	1 litre	481803

Disposable SepFast Octyl- 600 column	5 x 1 ml	481804
	1 x 5 ml	481805
	1 x 10 ml	481806
	1 x 20 ml	481807



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