Data sheet

MabPolish®

(Type I, Type II, Type III)

MabPolish is a novel group of mixed mode chromatography media designed for the efficient removal of impurities (e.g. host cell proteins, aggregates, DNAs and viruses etc) in monoclonal antibody products.

1. Properties

MabPolish is special mixed-mode chromatography media having strong binding to a broad range of host cell proteins but little binding to antibodies (typically >90% yield).

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

MabPolish Type I has anion-exchange functionality and very mild hydrophobicity. It is ued in flow-through mode to bind a wide range of impurities at lower pH (< 5.0) and at mild salt conditions (up to 0.15 M).

MabPolish Type II is more hydrophobic than MabPolish Type I under the similar working conditions. It is very good for removing antibody aggregates from monomers.

MabPolish Type III has anion-exchange group with moderate hydrophocity. It can be used in bind-elute mode or flow-through mode to remove impurities from monoclonal antibodies or other antibody materials.

Table 1: Characteristics of MabPolish:

Matrix	Spherical beads of cross-linked agarose
Particle size	50 – 150 µm
Functional group	Mixed mode ligands involving multi interaction mechanisms
Operational pressure	Up to 3 bar
pH stability	2-14 (short term) and 3-12 (long term)
Working temperature	+4^{O}C to +30^{O}C
Chemical stability	All commonly used buffers
Avoid	Oxidizing agents, detergents

2. Applications

MabPolish can be used in flow-through mode to remove impurities from monoclonal antibodies or other antibody materials, at various stages of the whole antibody purification process. Examples of the possible application scenario could be:

- After an antibody is first purified by affinity chromatography media (e.g. Protein A, Protein G or Protein L etc);
- After an antibody is first purified by ion-exchangers;
- After an antibody is first processed through precipitation;
- Direct removal of impurities from crude clarified antibody materials (for MabPolish Type I) before next purification step

The typical working condition:

- pH 4 to 5.
- Typical buffers include sodium acetate buffer, sodium phosphate buffer etc
- Typical ionic strength up to 0.15 M salt

Please see Section 4 for process optimisation.

3. Operations

The loose media is stored in 20% ethanol on delivery. It can be easily packed to any commercially available chromatography columns.

Column packing can be done in deionised water or low salt buffers using all the common methods. For flow packing, particular attention should be given to the maximum packing pressure. The typical packing pressure is 0.2 – 0.3 MPa. Increase or decrease the packing pressure if the peak asymmetry becomes >1.5 or <0.7. Operate the column at a pressure lower than the maximum packing pressure.

Packing Efficiency Assessment

To check the quality of the packing and to monitor this during the working life of the column, column efficiency should be tested directly after packing, prior to re-use and if there is an observed deterioration in separation performance. The efficiency of a packed column is expressed in terms of the height equivalent to a theoretical plate (HETP) and the asymmetry factor (As). These values are easily determined by applying a sample such as 1% acetone solution to the column and using water as eluent. Sodium chloride can also be used as a test substance. Use a concentration of 1 M NaCl in water with 0.15 M NaCl in water as eluent. It is important that conditions and equipment are kept constant so that results are comparable. Changes in solute, solvent, eluent, sample volume, flow rate, liquid pathway, temperature, etc., will influence the results. A sample volume of less than 2.5% of the column volume and the flow velocity between 15 and 30 cm/h will give the most optimal results.

4. Method optimization

We recommend scouting for optimal binding pH, ionic strength and flow velocity (i.e. residence time). We recommend special attention be paid to optimising the flow velocity to balance product yield and product purity.

The typical working pH is 4 to 5. The typical salt concentration is 0.15 M. Type of buffers can be screened to identify the best suitable buffer for a given antibody.

Tips: If unexpected high loss of product is noticed, consider to reduce the pH, decrease the ionic strength, or run at increased flow rates.

In general, balancing product recovery against process throughput is the major consideration when optimizing a method. However, for the purification of shearing-force sensitive molecules, the operational flow velocity needs to be optimised to balance the throughput and minimise the possible damage to the target molecule.

5. Process scaling up

MabPolish range of media is designed for bioprocessing use with regulatory support documents. Please contact us for further information.

6. Maintenance

Depending on the individual applications, please see the following recommendations.

Note: when sodium hydroxide solution or organic solvent (e.g. 20% ethanol or 30% IPA etc) is used, the flowrate must be less than 50% of the normal operational flowrate, because the column pressure will increase under these chemical conditions.

Regeneration
After each run, elute any reversibly bound material with 0.5 M NaOH. For MabPolish Type I, following wash with 2 M NaCl, 1M acetic acid is recommended before introducing equilibration buffer.

Cleaning-in-place (CIP)
CIP is a procedure that removes strongly bound materials such as lipids, endotoxins and denatured proteins that remain in the column 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.

Reverse the flow to introduce the following liquid from the bottom of the column.

- 1 M NaOH (+ 30% isopropanol if necessary) followed by
- 2 M NaCl followed by
- 1M acetic acid (or 0.5 M HCl for MabPolish Type I) followed by
- 2 M NaCl followed by
- Equilibration buffer

Longer contact time in the acetic acid step and NaOH step may improve the CIP performance.

Sanitization
Sanitization using 0.5-1.0 M NaOH with a contact time of 30 mins is recommended.

7. Storage

The loose media or column should be stored in 20% ethanol to prevent microbial growth. Store the column at a temperature of +2^{o}C to +8^{o}C. After storage, equilibrate the column with at least 5 bed volumes of running buffer before use.

8. Order information

Product	Quantity	Code no.
MabPolish Type I	25 ml	270201-25ML
	100 ml	270201-100ML
	1 litre	270201-1L
Pre-packed column	5 x 1 ml	270201-5x1ML
	1 x 10 ml	270201-11x100
MabPolish Type II	25 ml	270202-25ML
	100 ml	270202-100ML
	1 litre	270202-1L
Pre-packed column	5 x 1 ml	270202-5x1ML
	1 x 10 ml	270202-11x100
MabPolish Type III	25 ml	270203-25ML
	100 ml	270203-100ML
	1 litre	270203-1L
Pre-packed column	5 x 1 ml	270203-5x1ML
	1 x 10 ml	270203-11x100
MabPolish Selection Kit	5 x 1 ml	270000-5x1ML
	(1 ml each of MabPolish Type I, Type II, Type III, MabPolish DUO 150A, 150C)	

Note: other volumes available on request

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