

# MabSelect PrismA™ X protein A resin

## AFFINITY CHROMATOGRAPHY

MabSelect PrismA™ X protein A resin (Fig 1) is a high-capacity chromatography resin designed to efficiently capture monoclonal antibodies (mAbs). The high dynamic binding capacity (DBC) provides you with a cost-efficient resin for effective capture of high-mAb titers. The resin features the protein A-derived ligand from the well-established MabSelect PrismA resin, offering exceptional alkaline stability for repeated cleaning in place (CIP) to reduce bioburden. It is also well-suited for short bed height applications in rapid cycling chromatography (RCC) to further increase productivity in your capture step. MabSelect PrismA X resin is a part of our mAb resin toolbox providing a range of capture and polishing resins for clinical and commercial-scale mAb production.

### Key features of MabSelect PrismA X resin:

- Enhanced DBC allows high mass throughput of processed mAb per resin volume unit. DBC is higher than 80 g of mAb/L resin at 6 min residence time (RT) in a well-packed column, saving on resin and buffer consumption.
- Excellent alkaline stability enables efficient cleaning and sanitization using 0.5 to 1.0 M NaOH for improved process economy and robustness.
- Designed for use in short bed height applications using RCC to further improve productivity in the protein A capture step.
- Well-established protein A ligand used with several chromatography matrices and supported by the PrismA ELISA kit for accurate and convenient detection of leached protein A ligand.

Production of mAbs is performed across a wide range of scales—from research and process development in the lab—to preclinical and clinical production and all the way to commercial-scale production. The scale of operation, mode of operation, and batch frequency determines the optimal protein A resin for your process. Select from MabSelect PrismA, MabSelect PrismA X, or MabSelect™ SuRe 70 protein A resins—[find out in our guide](#) how to select the protein A resin to fit your regular mAb production.

Table 1 summarizes the characteristics of the MabSelect PrismA X resin.



**Fig 1.** MabSelect PrismA X resin is available in bulk and in prepacked columns.

**Table 1.** Main characteristics of MabSelect PrismA X resin

|  |  |
|--|--|
| Matrix   | Highly cross-linked agarose, spherical                               |
| Ligand   | Alkali-stable protein A-derived ( <i>E. coli</i> )                   |
| Ligand coupling  | Single-point attachment  |
| Coupling chemistry                                       | Epoxy  |
| Particle size $d_{50v}$ <sup>*</sup>                     | ~50 $\mu$ m  |
| DBC QB <sub>10%</sub> <sup>†</sup>                       | > 80 mg mAb/mL resin at 6 min RT<br>> 74 mg mAb/mL resin at 4 min RT |
| Recommended maximum operating flow velocity <sup>‡</sup> | 220 cm/h (20 cm bed height),<br>400 cm/h (10 cm bed height)          |
| Chemical stability                                       | Stable in aqueous buffers commonly used in protein A chromatography  |
| pH stability, operational <sup>§</sup>                   | 3 to 12  |
| pH stability, CIP <sup>¶</sup>                           | 2 to 14  |
| CIP stability <sup>¶</sup>                               | 0.5 to 1.0 M NaOH  |
| Delivery conditions                                      | 20% ethanol  |

<sup>\*</sup> Median particle size of the cumulative volume distribution.

<sup>†</sup> DBC at 10% breakthrough by frontal analysis using trastuzumab at an RT of 6 min (100 cm/h) and 4 min (150 cm/h). Analysis performed in a HiScreen™ column at 10 cm bed height.

<sup>‡</sup> In an AxiChrom™ 300 column with 30 cm i.d. packed according to verified methods, when operated in an aqueous solution with a viscosity of 1 cP at 20°C.

<sup>§</sup> Range where resin can be operated without significant change in function.

<sup>¶</sup> Range where resin can be subjected to cleaning-in-place (CIP) without significant change in function.

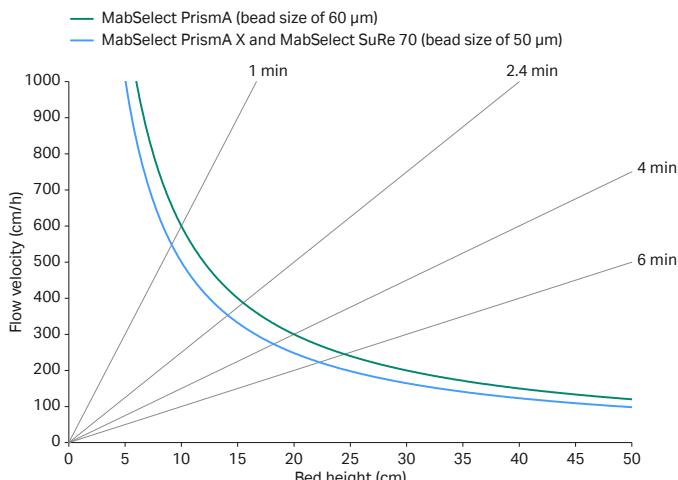
MabSelect PrismA X resin uses the same analytical and qualification methods as MabSelect PrismA resin to save time during implementation in GMP production. MabSelect resins are widely used in regulatory approved processes and build on decades of experience in commercial GMP operations.

## Productivity in the mAb capture step

A productive capture step maximizes the output of your process while minimizing inputs required in the form of raw materials. Critical productivity parameters may be processing time, consumable cost per gram of mAb, or output of your existing facility. Resin-related parameters such as DBC, flow properties, column diameter, alkaline stability, and resin cost, all affect the productivity. We can support you in a discussion about how to make your process more cost efficient. [Contact us for a productivity discussion.](#)

## High flow rates at short bed heights

The working flow velocity for MabSelect PrismA X resin at 20 cm bed height is up to 220 cm/h in an AxiChrom column packed according to verified methods, when operated in an aqueous solution with a viscosity of 1 cP at 20°C. At 10 cm bed height, you can increase flow velocity to approximately 400 cm/h corresponding to 1.5 min RT (Fig 2). The resin is well-suited for high-flow applications at short column bed heights such as those used in RCC.

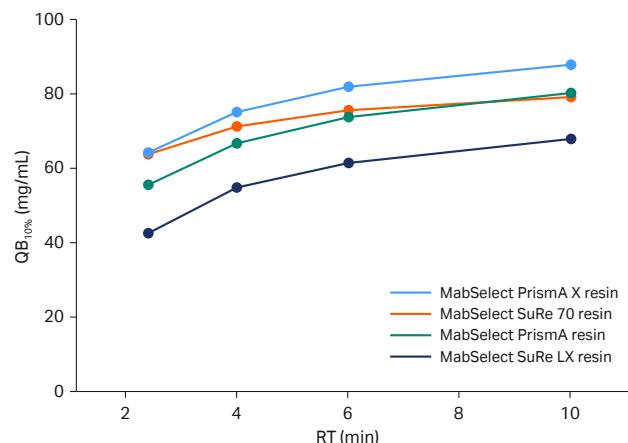


**Fig 2.** The flow velocity that can be used for a given resin increases with decreasing bed height, enabling faster runs at short bed heights. In an AxiChrom column with i.d. > 30 cm packed according to verified methods, when operated in an aqueous solution with a viscosity of 1 cP at 20°C.

## DBC at different residence times

The resin is based on a highly cross-linked agarose base matrix with a median particle (bead) size of 50 µm. The optimized porosity enables a fast uptake of mAb. As shown in Figure 3, the DBC for MabSelect PrismA X resin using trastuzumab is excellent and even higher than that of MabSelect PrismA resin at all RTs tested. At 6 min, the DBC is 82 mg/mL for MabSelect PrismA X resin and 74 mg/mL for MabSelect PrismA resin.

As the high DBC is realized at short RT, the MabSelect PrismA X resin is beneficial for use in RCC to further reduce resin volumes to increase grams of mAb produced per liter of resin and decrease overall resin costs.



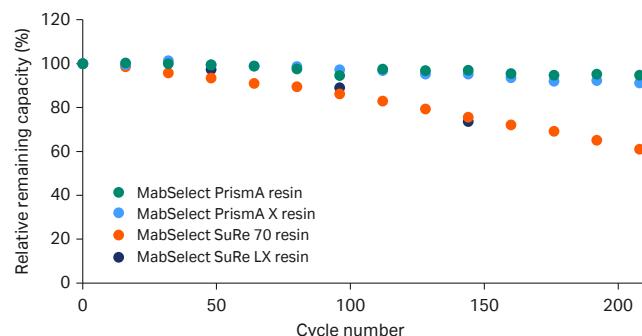
**Fig 3.** Relation between DBC expressed as  $QB_{10\%}$  and RT for MabSelect PrismA X, MabSelect SuRe 70, MabSelect PrismA, and MabSelect SuRe LX resins tested in HiScreen columns with trastuzumab.

## Excellent alkaline stability

We developed the ligand used in MabSelect PrismA X and MabSelect PrismA resins to withstand high concentrations of NaOH. Highly pure ligand is immobilized to the agarose base matrix via a chemically stable thioether linkage. The enhanced alkaline stability lets you perform efficient cleaning of the resin using 0.5 to 1.0 M NaOH over many purification cycles, providing long resin lifetime and minimized risk for bioburden incidents.

Use of 0.5 to 1.0 M NaOH is recommended for CIP. Cleaning with 0.5 to 1 M NaOH is effective, inexpensive, and easy to handle in bulk quantities. Therefore, NaOH is an attractive choice for large-scale commercial manufacturing of monoclonal therapeutic antibodies.

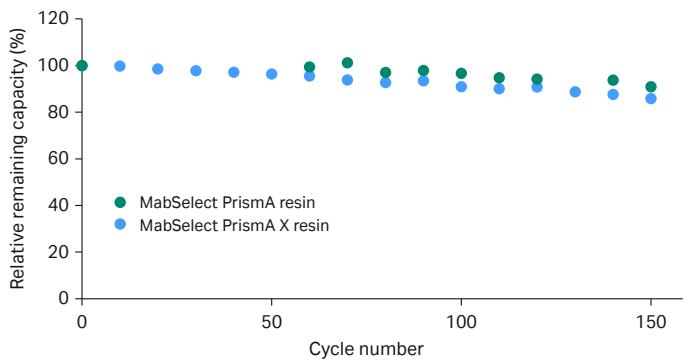
Figure 4 shows the relative remaining DBC of MabSelect PrismA X resin compared with other MabSelect resins over repeated CIP cycles with 0.5 M NaOH in an accelerated cleaning stability study. Accelerated studies provide quick results, whereas lifetime studies, with the column subjected to repeated cycles of loading of cell culture harvest, represent how resins are cleaned in the industry. In the accelerated alkaline stability study, we performed cycling where resins were repeatedly incubated for 4 h (corresponding to 16 cycles of 15 min duration each) in 0.5 M NaOH. As shown, MabSelect PrismA X resin retains more than 90% of its initial DBC after 200 cycles with 0.5 M NaOH and has similar alkaline stability as MabSelect PrismA resin.



**Fig 4.** A study of accelerated alkaline stability using 0.5 M NaOH as cleaning agent showed that MabSelect PrismA X and MabSelect PrismA resins can be used for more than 200 cycles with more than 90% remaining capacity. DBC was determined at 6 min RT with trastuzumab for MabSelect PrismA X, MabSelect SuRe 70, MabSelect PrismA, and MabSelect SuRe LX resins between the incubations.

# CIP lifetime study with mAb-containing sample and 0.5 M NaOH

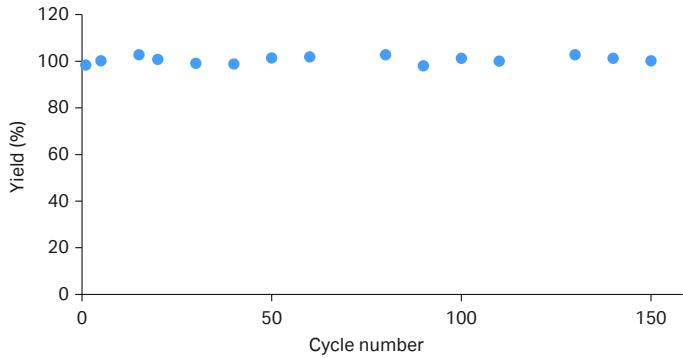
We performed a lifetime study to evaluate the alkaline stability and purification performance of MabSelect PrismA X resin over 150 cycles. We loaded clarified cell culture harvest (CCH) in every cycle, followed by a full purification and cleaning protocol. The mAb load in CCH was set to 80% of  $Q_B_{10\%}$ , based on a  $Q_B_{10\%}$  determination with pure mAb (trastuzumab) at the beginning of the study. The resin was cleaned with 0.5 M NaOH at a contact time of 15 min in each cycle and we found that alkaline stability, defined as relative remaining binding capacity over the cycles, was similar to MabSelect PrismA resin (Fig 5). After 150 cycles, MabSelect PrismA X resin maintains  $\sim 85\%$  of its initial DBC compared to MabSelect PrismA resin, which maintains  $\sim 90\%$  of its DBC after 150 cycles.



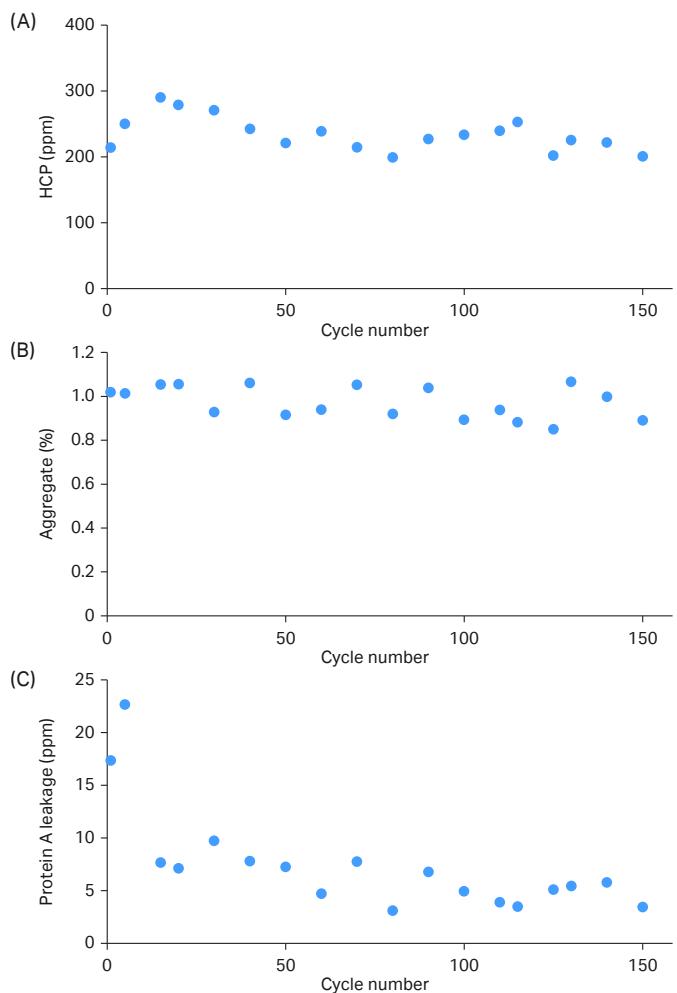
**Fig 5.** Relative remaining DBC of a mAb (trastuzumab) CCH for MabSelect PrismA X resin and MabSelect PrismA resin over 150 cycles. The resins were cleaned with 0.5 M NaOH in each cycle and showed excellent alkaline stability.

Figure 6 shows high and consistent relative mAb yields over the 150 cycles. Figure 7 demonstrates robust removal of host cell protein (HCP) with consistent and low levels of mAb aggregates and protein A ligand leakage.

We evaluated aggregate level and monomer purity by size exclusion chromatography (SEC) using a Superdex™ 200 Increase 10/300 GL column. We used a Gyrolab CHO-HCP E3G kit to measure the levels of HCP in samples. The analysis was performed using a Gyrolab xPand instrument. The PrismA ELISA kit was used to determine protein A ligand leakage.

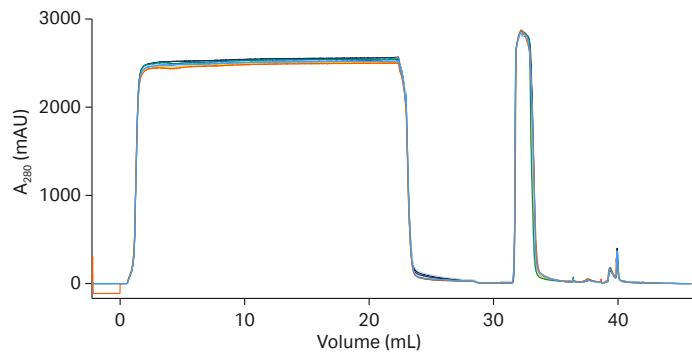


**Fig 6.** MabSelect PrismA X resin demonstrates high relative yield of mAb (trastuzumab) over 150 cycles.



**Fig 7.** Purification performance over the lifetime study of 150 cycles is high and consistent as shown by (A) HCP removal, (B) mAb aggregates levels, and (C) protein A ligand leakage.

During the lifetime study of MabSelect PrismA X resin, we see robust alkaline stability as no observed breakthrough of mAb occurred over the 150 cycles in the CCH loading phase (Fig 8).



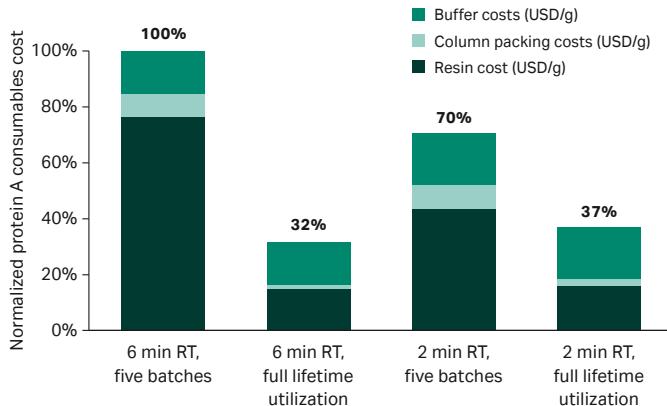
**Fig 8.** Chromatogram overlay of cycle 0 to 150 shows no breakthrough in the lifetime study and therefore no indication of target mAb (trastuzumab) breakthrough.

The alkaline stability for MabSelect PrismA X resin at 1 M NaOH is estimated to be similar to for MabSelect PrismA resin, see data in the [MabSelect PrismA data file](#).

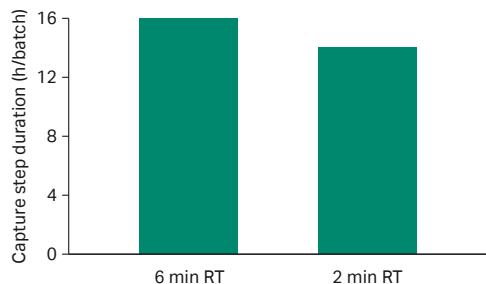
## Different protein A productivity scenarios

Protein A resin costs are relatively low in commercial manufacturing due to the extensive reuse of resin. However, in clinical scenarios where only a part of the resin lifetime is used, the protein A resin contribution to the overall batch cost is large. RCC can help reduce costs in processes in this situation.

Figures 9 and 10 show different scenarios (simplified here for clarity) in clinical (five batches) and commercial (full resin use) manufacturing using MabSelect PrismA X resin for short bed heights and high titers (see Table 2 for the process parameters). This example shows that for a process run in five batches, the resin cost was reduced by > 40% and buffer cost was increased by 20% when running at 2 min RT in RCC mode compared to 6 min RT in batch mode.



**Fig 9.** Normalized protein A consumable cost using MabSelect PrismA X protein A resin for five batches typical in clinical production and utilizing the full resin lifetime, respectively, at 6 min and 2 min RT. Resin and column packing cost per gram of mAb produced is calculated based on the number of cycles used. Using short RT reduces resin cost at low resin lifetime utilization and increases consumable costs at high lifetime utilization.



**Fig 10.** Duration of the MabSelect PrismA X protein A chromatography step decreases with shorter RT despite more cycles being run with smaller column volumes. Resin utilization does not impact the time required to run a batch.

**Table 2.** Process parameters

| Parameter                          | 6 min RT     |               | 2 min RT     |               |
|------------------------------------|--------------|---------------|--------------|---------------|
| Resin utilization                  | Five batches | Full lifetime | Five batches | Full lifetime |
| Volume clarified cell culture feed |              |               |              | 2000 L        |
| Titer                              |              |               |              | 4.25 g/L      |
| Column diameter                    |              |               |              | 45 cm         |
| Column bed height                  | 20 cm        |               | 10 cm        |               |

## Resin and column formats

MabSelect PrismA X resin is available as bulk resin in 25 mL, 200 mL, 1 L, and 5 L containers. The resin is also available in prepacked column formats such as HiTrap™ columns (1 and 5 mL), HiScreen columns, and on request as production-scale ReadyToProcess™ columns. These formats enable use from lab to commercial-scale manufacturing.

## Determining ligand leakage

You can easily measure protein A leakage using the [PrismA ELISA kit](#), which is specifically designed for use with MabSelect PrismA and MabSelect PrismA X resins. The PrismA ELISA kit contains all reagents required for you to perform the assay. This includes the PrismA ligand optimized for use in ELISA and polyclonal antibodies raised specifically against the PrismA ligand. The PrismA ELISA kit was developed with a focus on usability, robustness, and environmental sustainability.

## Recommended protocol

Table 3 outlines a typical mAb capture process using MabSelect PrismA X resin.

**Table 3.** Typical protein A capture protocol

| Step                            | CV*  | RT (min)/flow velocity (cm/h) at 20 cm bed height | Buffer  |
|---------------------------------|--|---|---|
| Equilibration                   | 3  | 6/200   | 20 mM sodium phosphate, pH 7.4 + 150 mM NaCl <sup>†</sup> |
| Load                            | 70% to 80% of QB <sub>10%</sub>                        | 6/200   | As required   |
| Wash 1                          | 5  | 6/200   | 20 mM sodium phosphate, pH 7.0 + 500 mM NaCl              |
| Wash 2                          | 1  | 6/200   | 50 mM sodium acetate, pH 6.0                              |
| Elution step                    | 3  | 6/200   | 50 mM sodium acetate, pH 3.5                              |
| Strip                           | 2  | 6/200   | 100 mM acetic acid, pH 2.9                                |
| CIP*                            | 3 ( $\geq$ 15 min contact time in upflow) <sup>‡</sup> | 5.5/220   | 0.5 M NaOH to 1 M NaOH                                    |
| Re-equilibration                | 3 to 5 (in upflow)                                     | 5.5/220   | 20 mM sodium phosphate, pH 7.4 + 150 mM NaCl <sup>†</sup> |
| Only after last run/for storage | 4  | 7.5/160   | 20% ethanol   |

\* CV is column volume; CIP is cleaning in place.

<sup>†</sup> For lab-scale use, 20 mM sodium phosphate, pH 7.4 + 500 mM NaCl can be used also for equilibration and reequilibration to reduce the number of different buffers.

<sup>‡</sup> MabSelect PrismA X resin cannot be cleaned using 3 CV of CIP solution with a contact time of 15 min (5 min residence time) when the column bed height exceeds 18.7 cm due to flow limitations of the resin. For columns with greater bed heights, increase the contact time as needed.

## Resin storage

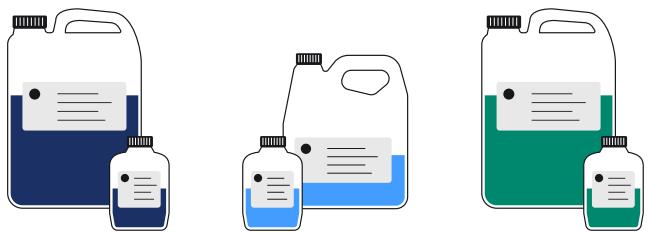
MabSelect PrismA X resin is delivered in 20% ethanol. Store unused resin in its container between 2°C and 8°C. Ensure that the screw top is fully tightened. Equilibrate packed columns in buffer containing 20% ethanol or 2% benzyl alcohol to prevent microbial growth. After storage, equilibrate with starting buffer and perform a blank run, including CIP, before use.

## mAb toolbox by Cytiva

Process volumes, number of batches, target molecule, and cleaning protocols influence which protein A affinity chromatography resin will be most cost-efficient and productive in your application. Figure 11 shows different protein A resins and what to consider to develop a cost-efficient protein A step for regular mAbs. All Cytiva MabSelect protein A resins are suitable for large-scale manufacturing and our ligands and base matrices are widely used in commercial manufacturing.

The protein A affinity capture step in antibody purification is followed by one or two polishing steps to remove impurities, see our website for the full range of **Capto™ resins** for chromatography polishing.

For mAb variants such as bispecific antibodies and fragments, we provide additional [affinity capture resins](#).



### MabSelect PrismA resin

Excellent productivity at commercial scale. Wide window of operation and well-suited for purification platforms.

### MabSelect PrismA X resin

Highest DBC of the MabSelect resins for cost-efficient mAb capture. Excellent durability.

### MabSelect SuRe 70 resin

Cost-efficient resin at low utilization with Cytiva quality and support.

**Fig 11.** Protein A resins for different process scenarios.

## Supply chain stability

We recognize the critical importance of reliable and secure supply to you. We are rapidly expanding our robust manufacturing capacity and maintaining open communication with you to support supply resilience.

We recommend that you work closely with your Cytiva representative to forecast demand to support our production planning and manufacturing operations.

Find out more about how we support [secure supply to the biopharma industry](#).

## Support and training

MabSelect PrismA X resin belongs to the BioProcess™ family of products developed and supported for large-scale manufacture of biopharmaceuticals. This support includes validated manufacturing methods, secure long-term resin supply, and regulatory support files (RSF) to assist process validation and submission to regulatory authorities. In addition, Fast Trak™ training and education provide high-level, hands-on training in key aspects of process development and manufacturing.

You can also access [Cytiva online learning](#) to build your skills and continue your education.

## Ordering information

| Product                                   | Size                        | Product code |
|---|-----------------------------|--------------|
| HiTrap MabSelect PrismA X column          | 1 x 1 mL                    | 17550151     |
|   | 5 x 1 mL                    | 17550152     |
|   | 1 x 5 mL                    | 17550153     |
|   | 5 x 5 mL                    | 17550154     |
| HiScreen MabSelect PrismA X column        | 1 x 4.7 mL                  | 17550115     |
| PreDictor™ RoboColumn                     | 8 x 200 µL                  | 17550133     |
| MabSelect PrismA X column                 | 8 x 600 µL                  | 17550134     |
| MabSelect PrismA X resin                  | 25 mL                       | 17550101     |
|   | 200 mL                      | 17550102     |
|   | 1 L                         | 17550103     |
|   | 5 L                         | 17550104     |
| ReadyToProcess MabSelect PrismA X columns | See sizes and product codes |              |
| PrismA ELISA kit                          | 1 kit                       | 29707299     |



Please [contact us](#) to request samples of MabSelect PrismA X resins.



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