# CaptureSelect™ FSH Affinity Matrix

Catalog Numbers 1943180250, 1943180500, 19431801L, 19431805L

Pub. No. MAN0009647 Rev. C.0

**WARNING!** Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves. Safety Data Sheets (SDSs) are available from thermofisher.com/support.

## Product description

The CaptureSelect™ FSH Affinity Matrix purifies human Follicle Stimulating Hormone (FSH) from complex source materials (recombinant sources) in a single step.

The matrix combines selectivity for the intact FSH molecule (not binding to the individual α and β subunits of FSH) with the benefits of a robust and high-quality affinity matrix provided by a 14 kDa single domain [VHH] antibody fragment.

## Product advantages

The CaptureSelect™ FSH Affinity Matrix offers:

- High recovery and purity in a single step
- Binding of only the intact form of FSH
- Neutral pH elution conditions to retain the biological activity of FSH
- Compatibility with FPLC systems

## Specifications

| | |
|---|---|
| Ligand | CaptureSelect™ FSH Affinity Ligand |
| Binding specificity | Human FSH (intact form) from recombinant sources |
| Matrix and particle size | Aldehyde-activated agarose, 65 µm |
| Dynamic binding capacity | ~3 g of FSH/L of matrix |
| Shipping solution | 20% (v/v) ethanol |

## Conditions for use

| Parameter | Conditions for use |
|---|---|
| Equilibration buffer | 20 mM Tris or PBS, pH 7.0–7.5 |
| Elution buffer | • **Neutral:** 20 mM Tris with 2 M $MgCl_2$, pH 7.0–7.5<br>• **Acidic:** 0.1 M glycine, pH 3.0 |
| Strip buffer | Any of the following:<br>• 0.1 M glycine, pH 2.0<br>• 0.5–1.0 M acetic acid<br>• Citric acid |
| Flow rate | 50–200 cm/h |
| Pressure limit | ≤ 2 bar |
| Cleaning solution | Any of the following:<br>• 0.5–1.0 M acetic acid<br>• Citric acid<br>• 10–30 mM NaOH, pH 12 (Higher concentrations affect the functionality of the affinity ligand on the matrix.)<br>• PAB (120 mM phosphoric acid, 167 mM acetic acid, and 2.2% (v/v) benzyl alcohol) (Rogers *et al.*, 2009) Freshly prepare PAB every 4–5 days and store protected from light to minimize radicals that affect the functionality of the matrix. |
| Storage solution | 20% (v/v) ethanol |
| Operating and storage temperatures | • **Operating**: 2–25°C<br>• **Short-term storage**: Room temperature<br>• **Long-term storage**: 2–8°C for up to 2 years |

## Flow characteristics

You can use agarose-based CaptureSelect™ affinity matrices at flow rates of 50–300 cm/h (Figure 1).

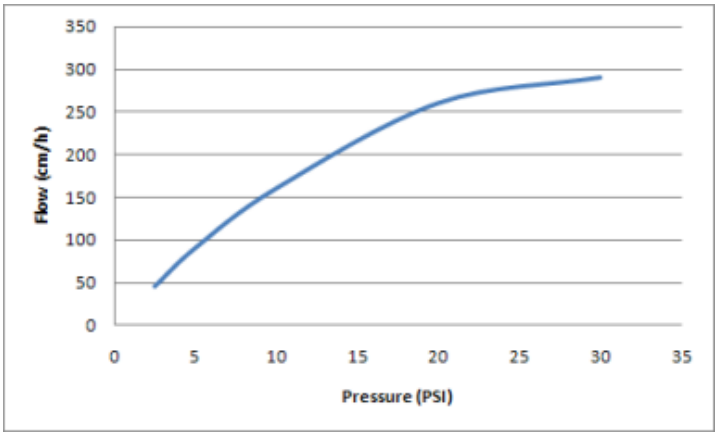


**Figure 1 Pressure-flow properties of an agarose-based CaptureSelect™ matrix tested on a 10-cm diameter column packed to 16-cm bed height. The resin can be operated at flow rates up to 300 cm/h, with a pressure drop that allows use in conventional low-pressure chromatography columns and systems.**

However, for optimal binding capacity and elution efficiency, we recommend flow rates of 50–200 cm/h. A low flow rate results in longer contact time of the load with the affinity matrix and drives the binding capacity. In addition, the elution fraction is more concentrated at a lower flow rate (Figure 2).

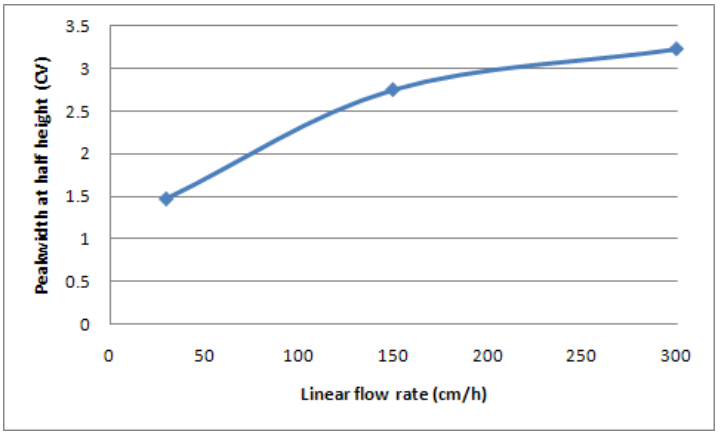


**Figure 2 The peak width at half height (in column volumes) of the elution peak versus the linear flow rate of agarose-based CaptureSelect™ matrices. Lowering the flow rate during elution reduces the volume of the elution peak.**

We recommend that you optimize each of your specific processes to achieve the best conditions for process time, binding capacity, and elution efficiency.

## Guidelines for use with chromatography systems

For optimal matrix performance, optimize the conditions in the following procedure for your application.

1. Pack the column as described in *CaptureSelect™ Affinity Matrices: Guidelines for Packing* (Pub. No. MAN0009645).
2. Attach the packed column to the chromatography system.
3. Equilibrate the matrix with 10 column volumes (CVs) of equilibration buffer.
4. Determine the volume of sample to load based on the dynamic binding capacity, concentration of the target molecule, and the column size. Optimum loading is at physiological pH. Avoid acidic conditions, which decrease binding efficiency.
5. Load the sample on the column.
6. Wash the sample with 5–10 CVs of equilibration buffer. To optimize washing efficiency, you can add NaCl to the equilibration buffer (up to 1.0 M).
7. Elute with 3–5 CVs of elution buffer.
8. Re-equilibrate the column in equilibration buffer.
9. Strip the column with 0.1-M glycine (pH 2.0), citric acid, or acetic acid (0.5–1.0 M).
10. Re-equilibrate the column in equilibration buffer to prepare the column for another purification run.
11. If the column will not be used immediately, store the matrix according to the storage parameters provided in “Conditions for use” on page 1.

## Column cleaning guidelines

Resin lifetime depends on how the resin is used and cleaned. Therefore, we recommend that you specifically evaluate each purification process.

Typical cleaning procedures for CaptureSelect™ resins include combinations of acidic cleaning followed by low concentrations of NaOH, before storing in 20% (v/v) ethanol at neutral pH (Eifler *et al.*, 2014).

To optimize column cleaning, consider these guidelines:

- Pump the cleaning solution through the column for 15–20 minutes in upflow.
- Incorporate a static hold to increase the time that the cleaning solution is in the column while minimizing the volume of cleaning solution required.
- When a combination of acidic and mildly caustic cleaning agents is used, apply the NaOH solution as a final cleaning agent to minimize the risk of irreversibly binding impurities on the column.
- In some purification processes, 20% (v/v) isopropanol (with or without acid) and 6.0 M guanidine-HCl can help remove discoloration.

## Example application - FPLC

In this example, FSH was purified from clarified cell culture harvest. After the resin was loaded, the column was equilibrated, then eluted. Conditions were as follows:

- **Column** – CaptureSelect™ FSH Affinity Matrix
- **Equilibration buffer** – 20 mM Tris, pH 7.4
- **Load** – Feedstock of recombinant FSH production
- **Elution buffer** – 20 mM Tris with 2.0 M $MgCl_2$, pH 7.4
- **Flow** – 200 cm/h

The FSH elutes very well under these conditions. As shown in Figure 3, hardly any protein is detected in the pH strip at pH 2.0.

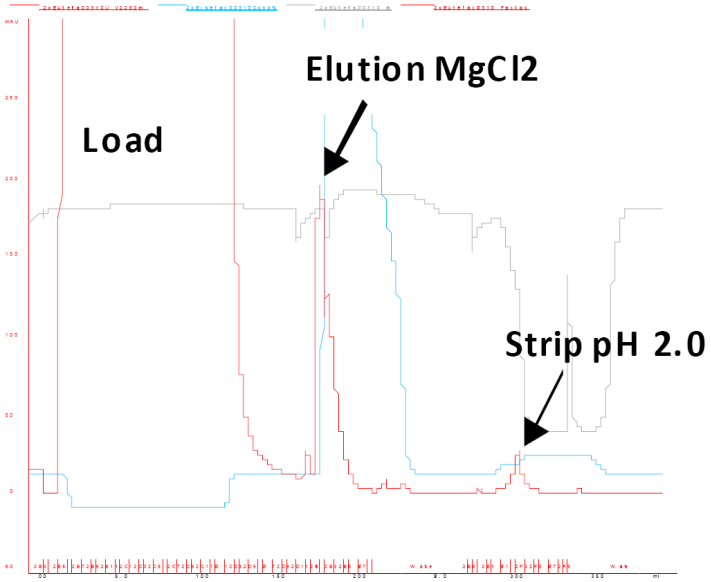


**Figure 3 Chromatogram of FSH purified using the CaptureSelect™ FSH Affinity Matrix.**
Red line: OD280 signal; Blue line: conductivity; Grey line: pH value

The collected fractions were analyzed on a Coomassie non-reduced SDS-PAGE, showing a highly pure recombinant human FSH in the elution fraction (Figure 4).

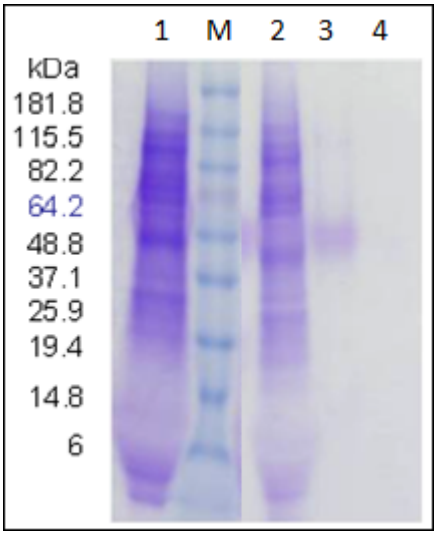


**Figure 4 SDS PAGE analysis of the fractions: Non-reduced Coomassie-stained starting material, flow through, and elution fractions of FSH purification using the CaptureSelect™ FSH Affinity Matrix.**
Lane 1: starting material; Lane M: molecular weight marker; Lane 2: flow through; Lane 3: elution; Lane 4: strip

## Ordering information

| Product | Size | Cat. no. |
|---|---|---|
| CaptureSelect™ FSH Affinity Matrix | 250 mL | 1943180250 |
| | 500 mL | 1943180500 |
| | 1 L | 19431801L |
| | 5 L | 19431805L |

### Regulatory Support File

A Regulatory Support File (RSF) is available for the resin. It contains detailed information about the resin and the manufacturing process. Contact your local sales representative to obtain access.

### Supporting products

Pre-packed affinity HPLC columns are available for determining titers and analyzing in-process samples during the production and purification of FSH. The pre-packed columns contain the same affinity ligand as the CaptureSelect™ FSH Affinity Matrix, but the ligand is immobilized on POROS™ 20-µm beads that are suitable for HPLC applications.

A biotinylated anti-FSH conjugate is also available. Applications for the CaptureSelect™ Biotin Anti-FSH Conjugate include:

- ELISA
- Western blot
- Gyros™ Gyrolab™-based immunoassays
- Label-free detection platforms, such as those based on surface plasmon resonance (Biacore™ and IBIX-MX96 systems) and bio-layer interferometry (ForteBio™ Octet™ systems)

In addition, a ligand leakage ELISA is available for detecting possible leached ligand in the elution fractions of the CaptureSelect™ FSH Affinity Matrix.

| Product | Size | Cat. no. |
|---|---|---|
| POROS™ CaptureSelect™ FSH Affinity Column | 2.1 × 30 mm | 4481822 |
| | 4.6 × 50 mm | 4481824 |
| | 4.6 × 100 mm | 4481826 |
| | 10 × 100 mm | 4481828 |
| CaptureSelect™ Biotin Anti-FSH Conjugate | 100 µg | 7103180100 |
| | 500 µg | 7103180500 |
| CaptureSelect™ FSH Ligand Leakage ELISA Kit | 1 assay | 810318001 |
| | 10 assays | 810318010 |

## For more information

For more information on CaptureSelect™ products and ligand leakage ELISA products, go to **www.thermofisher.com/captureselect**.

## Customer and technical support

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- Worldwide contact telephone numbers
- Product support information
  - Product FAQs
  - Software, patches, and updates
  - Training for many applications and instruments
- Order and web support

- Product documentation
  - User guides, manuals, and protocols
  - Certificates of Analysis
  - Safety Data Sheets (SDSs; also known as MSDSs)

    **Note:** For SDSs for reagents and chemicals from other manufacturers, contact the manufacturer.

## Limited product warranty

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## References


Rogers, M. *et al.* 2009. Development of a rapid sanitization solution for silica-based protein A affinity adsorbents. *Journal of Chromatography A.* 1216:4589–4596.

Eifler, N. *et al.* 2014. Development of a novel affinity chromatography resin for platform purification of lambda fabs. *Biotechnology Progress* DOI:10.1002/btpr.1958.



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**Revision history:** Pub. No. MAN0009647

| Revision | Date | Description |
|---|---|---|
| C.0 | 25 January 2022 | Document branding was updated. |
| B.0 | 19 February 2015 | Baseline for this revision. |