Protein Purification Gel, 45-135 µm-Streptavidin XT
PRODUCT DATA SHEET

Protein Purification Gel, 45-135 µm-Streptavidin XT

Cat No: AG-4500-Streptavidin XT

Description

Strep-Tactin XT (Strep-Tag II) gel, is composed of a highly cross-linked 4% agarose gel as a matrix covalently combined with a large amount of the latest Strep-Tactin ligand - Streptactin XT protein. Strep-tag II is a widely used affinity tag in protein purification systems. It includes two types Strep-tag II and Twin Strep-tag II. Strep-tag II is a short peptide tag composed of eight amino acids (WSHPQFEK) that can be fused to proteins as an N-terminal or C-terminal tag with little effect on recombinant proteins. The further improved Twin Strep-tag II is a sequence of two Strep-tag II sequences (linked by internal amino acids), which enables gentle and rapid purification like Strep-tag II.

Strep-Tactin XT (Strep-Tag II) gel is primarily used to purify Strep-tag II and Twin Strep-tag II proteins from any expression system, including baculoviruses, mammalian cells, yeast, and bacteria.

Characteristics

Matrix	Highly crosslinked 4% agarose gel (4FF)
Ligand	Recombinant Streptactin XT protein
Particle size	45~135 µm
Concentration	The gel volume accounts for 50% of the suspension volume
Pressure flow rate	80 ~ 150 cm/h (0.3 MPa, 3 bar)
Binding ability	10 mg Twin Strep-tag II protein/mL gel
Scope of application	Purification of Strep-tag II or Twin-Strep-tag proteins
Shelf life	Stable storage at 2 ~ 8°C for two years

Strep-Tactin XT (Strep-Tag II) gel tolerance

Reagent	**Concentration**
Guanidine hydrochloride	6 M
Urea	8 M
NaCl	5 M
DTT	50 mM

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β-mercaptoethanol	50 mM
EDTA	50 mM
Tween-20	2%
Imidazole	0.25 M

Operation process

(1) Biotinylation molecular purification process

1. Prepare before use

1.1 Buffer:

Buffer	Formula
Balance/washing solution	0.15 M NaCl, 20 mM Na_2HPO_4, pH 7.2
Eluate	Balance/washing solution with 1 ~ 5 mM D-biotin
Regenerated solution	10 mM NaOH

1.2 Vortex oscillator, rotary mixer, pipette, gun head and centrifugal tube

2. Sample preparation

It is recommended that the samples be centrifuged and filtered with 0.22 μm or 0.45 μm filter membrane before loading to reduce impurities and improve the efficiency of protein purification.

3. Gel filling

1) Take the appropriate size of the affinity chromatographic column, put it into the gasket, add an appropriate amount of deionized water washing column tube and gasket, and close the lower outlet.

2) Mix **Strep-Tactin XT (Strep-Tag II) gel** evenly, use a pipette to absorb an appropriate amount of slurry and add it to the chromatograph column (the actual volume of the gel accounts for half of the suspension), and open the lower outlet to flow out the protective liquid.

3) Add appropriate amount of deionized water to rinse the gel, and close the lower outlet when the liquid in the column flows out.

4) The loaded chromatographic column is added to **Balance/washing solution** for balance, and the protective liquid is added when not used for the time being, and stored at 2 ~ 8°C.

4. Sample purification

4.1 Purification by incubation

1) According to the purified sample, add an appropriate amount of **Strep-Tactin XT (Strep-Tag II) gel** to a centrifuge tube, centrifuge it at 1000 rpm for 1 min, and remove the supernatant;

2) Add 5 times the gel volume of **Balance/washing solution** into the centrifuge tube to clean the gel, centrifuge at 1000 rpm for 1 min, remove the supernatant, repeat more than 2 times.

3) The samples were added and incubated at 2~8°C for 2~ 4 h or overnight.

4) After incubation, centrifuge at 1000 rpm for 1 min, absorb the supernatant to the new centrifuge tube, and retain the supernatant as a flow for electrophoresis identification.

5) Clean the gel with a **Balance/washing solution** of 5 times the gel volume, centrifuge at 1000 rpm for 1 min, remove the supernatant (be careful not to inhale the gel), repeat 3 to 5 times, and it is recommended to replace a new centrifuge tube in the middle.

6) Add 3 ~ 5 times the gel volume of **eluent** for elution, incubate at room temperature for 5~15 min, centrifuge at 1000 rpm for 1 min to collect eluent, can be repeated 2 ~ 3 times.

4.2 Purification by chromatographic column

The amount of solution is calculated by the column volume (for example, 5 times the column volume, 1 mL specification corresponds to 5 mL solution, 10 mL specification corresponds to 50 mL solution).

1) The loaded **Strep-Tactin XT (Strep-Tag II) gel** chromatography column is balanced with 5 times the column volume **Balance/washing solution** so that the gel is under the same buffer liquid system as the target protein, and repeat 2 ~ 3 times.

2) The sample was added to the balanced chromatographic column and incubated in the rotary mixing apparatus for 30 ~ 60 min, then the effluent was collected, and the sample could be repeated to increase the binding efficiency.

3) Use a **Balance/washing solution** of 10 ~ 15 times the column volume for washing, remove the impurity protein unless specifically adsorbed, and collect the wash solution.

4) Use **eluent** of 5 ~ 10 times column volume to eluate, collect in sections, collect one tube for each column volume, and test separately, which can ensure that all the bound target proteins are elated, and can obtain high purity and high concentration of proteins.

4.3 SDS-PAGE detection

Samples obtained from purified products (including flow through components, wash components, and elution components) and original samples were tested using SDS-PAGE.

5. Gel regeneration and cleaning

Regeneration: Strep-Tactin XT (Strep-Tag II) gel should be regenerated after each application, removing the D-biotin attached to the gel to ensure consistent results by washing with 5 times the

column volume of deionized water and reproducing with 10 times the column volume of 10 mM NaOH. Then clean it with 5 times the volume of column deionized water.

Preservation: The gel is stored in the same volume of protective liquid after regeneration and cleaning, and stored at 2~8°C to prevent the gel from being contaminated by bacteria.

Notes

1. Before performing experimental operation, please read this operation manual carefully.
2. In the experiment, the adhesion of Strep-Tactin XT (Strep-Tag II) tactin to Strep-tag II and Twin Strep-tag II proteins is different, and the adhesion is also affected by the Buffer. Therefore, **it is possible to optimize the operation details or screen and prepare buffers for experiments**.
3. The gel should be fully oscillated and evenly before use. The gel should be kept in storage solution to prevent drying.
4. This product is for scientific research only.

Ordering Information

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