

SDR HyperD[®]

Solvent-detergent removal
chromatography resin

Benefits

- Binds solvent and detergent molecules used in viral inactivation processes (TnBP and Triton* X-100)
- High recovery for proteins (exclusion limit 10 kDa)
- High binding capacity for small hydrophobic molecules
- Stable in acid, polar organic and oxidizing solutions



Product Information

Plasma preparations may contain viruses that are effectively removed and inactivated by combining nanofiltration (for size exclusion removal of non-lipid enveloped viruses) and treatment with non-ionic solvents and detergents (effective for lipid-coated viruses).

The elimination of solvent and detergent from biological fractions is necessary, and can be achieved by various methods including resin partitioning, size exclusion, affinity or batch extraction with vegetable oils combined to reverse phase on C18.

SDR HyperD[®] is a unique resin designed to eliminate solvent and detergent from biological fluids.

Product description and mechanism of solvent detergent adsorption

SDR HyperD[®] is a composite resin that combines a silica-bead moiety filled with a three-dimensional cross-linked hydrophobic polymer.

The particle size distribution (40–100 μm), the small pore size of the silica beads and the nature of the polymer have been optimized for a retention of solvents and detergents typically used in viral inactivation processes (i.e. Tri-n-Butyl Phosphate – TnBP – and Triton[®] X-100).

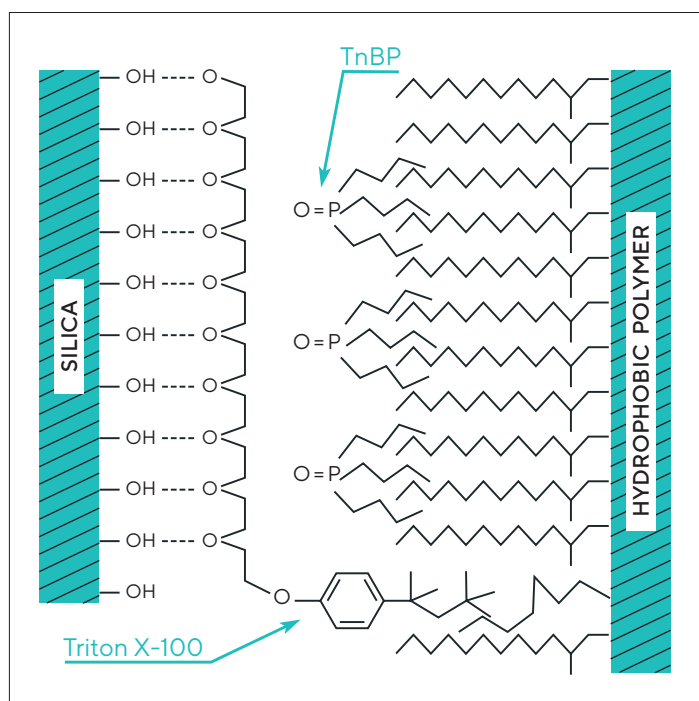


Figure 1: Schematic interaction mechanism of Triton[®] X-100 and TnBP on SDR HyperD[®] resin.

Triton[®] X-100 interacts both with the silica surface (formation of hydrogen bonds between the silanols and the polyoxyethylene chain) and with the hydrophobic polymer moiety. TnBP interacts only with the hydrophobic polymer of the resin.

The adsorption mechanism involves both the silica moiety and the hydrophobic polymer. The adsorption of Triton[®] X-100 is proportional to the silica surface area, whereas the adsorption of TnBP is linked to binding to the organic polymer moiety. The main features of SDR HyperD[®] resin are summarized in Table 1 and the adsorption mechanism represented in Figure 1.

Table 1: Main Properties of SDR HyperD[®] resin

| | |
|--|---|
| Structure | Spherical silica beads filled with a three-dimensional hydrophobic polymer. |
| Average particle size | 40–100 μm |
| Nature of polymer | Hydrophobic, long aliphatic chains bind solvents. 10 kDa limit prevents proteins from being retained. |
| Typical sample load | 2–3 times the column volume with residence times of 5 min using IgG or ATIII treated solutions. |
| Recommended residence time | 5–15 min |
| Binding capacity for Triton [®] X-100 | ≥90 mg/ml ⁽¹⁾ |
| Adsorption buffer | PBS |
| Solvent detergent elution buffer | PBS Ethanol (50 50) and EtOH or and isopropanol |
| Operating pH range | 2–12 |
| Pressure resistance | 70 bar (1000 psi) |

⁽¹⁾ Determined using 5 mg/ml Triton[®] X-100 in PBS, pH 7.4, 10% breakthrough, 300 cm/h.

Capacity and solventdetergent removal efficiency

The structure of SDR HyperD[®] resin has been engineered to optimize the solvent | detergent retention. Due to the specific degree of three-dimensional polymer cross-linking, a low exclusion limit of 10 kDa means target proteins are “excluded” from the resin, and are found unretained in the column void volume.

On the other hand, the high specific surface area (200 m²/g) of the porous silica allows a high capacity for Triton[®] X-100 and TnBP. The dynamic binding capacities currently obtained are 60–80 mg/ml for Triton[®] X-100 and 40–50 mg/ml for TnBP at 100 cm/h (initial concentration of Triton[®] X-100 and TnBP in bovine plasma are respectively 10 and 5 mg/ml).

Examples of removal efficiencies from various feedstreams are summarized in Table 2.

Table 2: Solvent-Detergent Depletion Example

| | | Before depletion | After depletion | Removal efficiency |
|--------------|---------------|------------------|-----------------|--------------------|
| IgG | TnBP | 5,000 ppm | < 0.4 ppm | 99.9% |
| | Triton* X-100 | 10,000 ppm | < 10 ppm | 99.9% |
| ATIII | TnBP | 5,000 ppm | < 0.4 ppm | 99.9% |
| | Triton* X-100 | 10,000 ppm | < 10 ppm | 99.9% |
| Bovine serum | TnBP | 5,000 ppm | < 0.4 ppm | 99.9% |
| | Triton* X-100 | 10,000 ppm | < 10 ppm | 95.5% |

Sample volume: 3.6 CV, Flow rate: 150 cm/h;
Column length: 10 cm; Residence time: 4 min.

Note: The removal efficiency is also dependent on flow rate and column loading: i.e. when using a 10 cm column at 150 cm/h (2 CV load of bovine serum supplemented with Triton* X-100 or TnBP), a removal of 95.5% was observed for Triton* X-100; this removal efficiency was decreased to 80% when 8 CV loads were used.

A comparative study shows SDR HyperD® has a capacity twice that of the used C18 silica for the removal of 1%TnBP and 1% Triton -X100 maintaining a good recovery. SDR HyperD® has also the ability to remove Triton X-80, Triton X-45 and Tween.

SDR HyperD® resin can be also used for the removal of TWEEN, Triton X80 from biological solution as feedstock.

Guidelines for binding capacity optimization

As for any resin, the dynamic binding capacity (DBC) of SDR HyperD® resin is sensitive to the linear flow rate, to the residence time on the column and to the nature of the sample. Therefore, it is recommended to start trials by loading not more than 5 CV of sample on a column with a minimum height of 15 cm, and a working flow rate not exceeding 150 cm/h. Adsorption buffer sample load and flow rate can be increased according to performance. Note that the rigid nature of SDR HyperD® resin allows to use higher flow rates (i.e. >600 cm/h) for washing and cleaning, with moderate backpressure (< 2 bar).

Desorption of retained solvent | detergent

It is typically achieved by injecting 1-10 CV of PBS | Ethanol 95° (50 | 50), followed by 3-10 CV of ethanol 95°. If necessary, 2-isopropanol can also be used (10 CV) in the washing sequence.

Cleaning in place and chemical stability

SDR HyperD® resin is insoluble in water and in organic solvents. It is also very stable to strong denaturing agents and chaotropic agents. The resin can be treated with 0.01 to 0.1 M hydrochloric acid or water-miscible organic solvent. SDR HyperD® resin can also be treated with oxidizing agents such as peracetic acid (1500 ppm in sodium acetate, pH 5.0), which is a well-known bactericide and sporicide.

Recommendations for sanitization are shown in Table 3.

Table 3: Recommended clean-in-place method.

Alcohol | acid treatment

Wash with at least 3 CV of a solution of 20% (v | v) ethanol containing 1 M acetic acid.

This solution should be injected after removal of dissolved gas at a flow rate of 10 - 20 cm/h (1 hour contact time). After treatment, reequilibrate with normal sterile pyrogen-free buffer.

SDR HyperD® resin is supplied in a storage solution containing 20% ethanol.

For industrial applications, bulk quantities are available on request.

Ordering Information

| Product | Cat. No. | Size |
|-------------|-----------|--------|
| SDR HyperD® | 20033-031 | 25 mL |
| | 20033-023 | 100 mL |
| | 20033-015 | 1 L |
| | 20033-056 | 5 L |
| | 20033-049 | 10 L |

Reference


- Specific resin to remove solvent-detergent mixtures from virus-inactivated biological fluids. Guerrier, L., et al., J. Chromatogr. B, 664 (1995) 119.
- Comparative removal of solvent and detergent viral inactivating agents from human intravenous immunoglobulin G preparations using SDR HyperD® and C18 sorbents. Burnouf, T, and al, Analytical Biochemistry 389 (2009) 69-73.

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