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PurKine™ MBP-Tag Dextrin Resin

Cat #: BMR2020 Size: 5 mL/25 mL

[<u>=</u> Q	MBP-Tag Dextrin Resin, crosslinked 4% agarose		
REF	Cat #: BMR2020	LOT	Lot #: Refer to product label
	Capacity: >20 mg MBP-tagged protein/mL		Bead size: 45-165 μm
	Tolerance: 0.1 MPa, 1 bar		Buffer: PBS containing 20% ethanol
Ŷ	Storage: Stable for 12 months at 4°C from date of shipment		

Assay Principle

Maltose binding Protein (MBP) is a member of the maltose/maltodextrin system of E.coli which is accountable for the uptake and efficient catabolism of maltodextrins. MBP-Tag is very effective in improving the expression level and solubility of many proteins as a fusion protein. PurKine™ MBP-Tag Dextrin Resin provides a rapid and efficient purification of MBP-Tag proteins.

Reagent Preparation

Water and chemicals used for buffer preparation should be of high purity. It is recommended to filter all buffers by passing through a $0.22 \mu m$ or $0.45 \mu m$ filter before use. For most proteins, the following buffer are recommended:

Binding/Wash Buffer: 20 mM Tris-HCl, 200 mM NaCl, 1 mM EDTA, pH 7.4

Elution Buffer: 20 mM Tris-HCl, 1 mM EDTA, 10 mM Maltose, pH 7.4

Note:1 mM DTT or 10 mM β -mercaptoethanol can be included in the Binding and Elution Buffer.

Sample Preparation

The sample should be centrifuged and/or filtered through a $0.22~\mu m$ or $0.45~\mu m$ filter before it is applied to the medium to prevent clogging the column. If the sample is too viscous, dilute it with binding buffer to prevent clogging the column. Be careful not to exceed the resin's binding capacity.

Procedure for Sample Purification

- 1. Pack column with an appropriate amount of MBP-Tag Dextrin Resin. Allow storage buffer to drain from resin by gravity flow.
- 2. Add 2 resin-bed volume Binding buffer to the column. Equilibrate the column, and drain away the Binding buffer. Repeat this step for three times.
- 3. Add the prepared sample (prepare sample by mixing protein extract with equal Binding buffer) to the column, collect the effluent liquid which can be analyzed by SDS-PAGE.

Note: For maximal binding, the sample can be incubated for 30 min at room temperature or 4°C. Be careful not to exceed the resin's binding capacity.

- 4. Add 2 resin-bed volume Wash buffer to the column to remove the non-specific adsorption protein. Collect the wash liquid which can be analyzed by SDS-PAGE. Repeat this step for six times.
- 5. Add 5-10 resin-bed volume Elution buffer to the column to wash the target protein, or until the absorbance of the effluent at 280 nm is stable. Collect the wash liquid, and analyzed the content in each tube respectively.



6. Examine and identify the fractions containing the target protein. Use UV absorbance, SDS-PAGE, or Western blotting.

Storage of the Column

Use 2 resin-bed volume Binding buffer and 2 resin-bed volume deionized water to equilibrate the column in turn, repeat twice. Then add 2 resin-bed volume 20% ethanol, repeat once. Add equal volume PBS containing 20% ethanol as storage buffer, store the column in 4°C to keep bacteria away.

Cleaning-in-Place (CIP)

In general, resin may be used at least five times. When a column used to purify protein from cell exact usually has buildup of insoluble substances, that is cell debris, which are non-specifically absorbed onto the matrix support and cannot be completely removed during washing steps. If the column is to be reused, these contaminants should be cleaned from the column. Cleaning-in-Place helps eliminating materials and preventing progressive buildup of contaminants.

- 1. Add 3 column volumes of deionized water.
- 2. Add 3 column volumes 0.1% SDS or 0.5 M NaOH solution.
- 3. Add 3 column volumes of deionized water. Add equal volume PBS containing 20% ethanol as storage buffer, store the column in 4°C.

Trouble Shooting

Problem	Cause	Solution
Back pressure exceeds	Column is clogged	Cleaning-in-place. Increase the centrifugation speed or f iltering the sample.
Sample contamination	Expression of target protein in extract is very low	Check expression level of protein by estimating the amount in the extract, flow through, elute fraction and pellet upon centrifugation. Or apply larger sample volume.
Low protein/sample	Target protein is degraded	Perform purification at 4°C in the presence of protease of protease inhibitors.
recovery	Lots of amylase exist in sample or buffer	Add glucose in culture medium to inhibit amylase expre ssion.

Recommended Products

Catalog No.	Product Name
KTP2020	PurKine™ MBP-Tag Protein Purification Kit (Dextrin)
A02070	Anti-MBP Tag Mouse Monoclonal Antibody (9Y5)
KTP2001	PurKine™ His-Tag Protein Purification Kit (Ni-NTA)
KTP2010	PurKine™ GST-Tag Protein Purification Kit (Glutathione)
KTP2030	PurKine™ Biotin-Tag Protein Purification Kit (Streptavidin)
KTP2070	PurKine™ Antibody Purification Kit (Protein A/G)
KTP2140	PurKine™ Endotoxin Removal Kit (Polymyxin B)

Disclaimer

The reagent is only used in the field of scientific research, not suitable for clinical diagnosis or other purposes.

