

# PurKine<sup>™</sup> Biotin-Tag Protein Purification Kit (Streptavidin)

Cat #: KTP2030

Size: 1 mL/1 mL×5

[ <u>;</u> ]	Biotin-Tag Protein Purification Kit (Streptavidin), crosslinked 6% agarose			
REF	Catalog Number: KTP2030	LOT	Lot Number: Refer to product label	
	Capacity: >6 mg Biotin-tagged protein/mL		<b>Bead size:</b> 45-165 μm	
	Tolerance: 0.3 MPa, 3 bar		Buffer: PBS containing 20% ethanol	
Ĵ,	Storage: Stored at 4°C for 12 months	$\land$	<b>Note:</b> Storing according to the recommended storage conditions after the package is opened	

#### **Assay Principle**

Biotin is also called B complex vitamin. It is a dicyclic compound with a molecular weight of 244.31 Da. Biotin is an important cofactor of mammalian carboxylase. It is present in cells in the form of free and binding mixtures. Essential vitamin with important roles in amino acid, energy metabolism and fatty acid synthesis. It is a cogroup in the enzyme carboxylase of four mammalian species that promotes the binding and transport of carbon dioxide. Based on the interaction between streptavidin and Biotin, PurKine<sup>™</sup> Biotin-Tag Protein Purification Kit provides a simple, rapid, and efficient purification of Biotin-Tag proteins.

## **Materials Supplied and Storage Conditions**

	Size		Storege conditions
Kit components	1 mL	1 mL×5	Storage conditions
Biotin-Tag Streptavidin Packed Column 6FF	1 mL	1 mL×5	4°C
Binding/Wash Buffer (10×)	30 mL	100 mL+50 mL	4℃
Elution Buffer (1×)	100 mL	250 mL×2	4°C

## **Materials Required but Not Supplied**

- 0.22 μm or 0.45 μm filter
- Precision pipettes, disposable pipette tips
- Deionized water
- Various glassware for preparing reagents and buffer solutions

## **Sample Preparation**

The sample should be centrifuged and/or filtered through a 0.22 µm or 0.45 µm 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. Avoid

1/2



using protease inhibitors or other additives that contain chelators, such as EDTA, or strong reducing agents, such as DTT or  $\beta$ -mercaptoethanol, which will disrupt the function of the resin. Be careful not to exceed the resin's binding capacity.

### **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.

1×Binding/Wash Buffer: Before use, Binding/Wash Buffer (10×) was diluted with deionized water to 1×Binding/Wash Buffer, and mixed well for use. Store at 4°C.

## **Procedure for Sample Purification**

1. Fix Column. Move the top and bottom stopper, and let the storage buffer drain away.

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  $4^{\circ}$ C to keep bacteria away.

## **Recommended Products**

Catalog No.	Product Name	
BMR2030	PurKine™ Biotin-Tag Streptavidin Resin 6FF	
KTP2001	PurKine™ His-Tag Protein Purification Kit (Ni-NTA)	
KTP2020	PurKine™ MBP-Tag Protein Purification Kit (Dextrin)	

## Disclaimer

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



212