Anti-Succinyl Lysine Antibody Conjugated Agarose Beads

Catalog # PTM-402

Pack Size: 100/500 µL

Formulation: 50/250 µL settled agarose beads supplied as 50% slurry, containing 66% glycerol

Species Reactivity: All species expected

Application: Peptide immunoaffinity enrichment followed by Mass Spectrometry-based proteomics

Storage & Stability: Store at -20°C and avoid freezing. Stable for 12 months from date of receipt

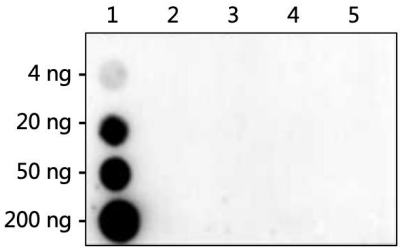
Usage Recommendation:

1. The usage amount for each sample (2 mg peptides) is 40 µL resin (containing 20 µL dry resin).
2. The resin product has stable performance and is not recommended to be aliquoted. Aliquoting will lead to beads loss.
3. It is recommended to use the beads with specialized buffer kit for optimal performance.
4. It is recommended to use sterile low-adsorption pipette tips for aspiration.

Specificity:

With the immobilization of highly specific anti-succinyl lysine antibodies, the anti-succinyl lysine antibody conjugated agarose beads selectively capture peptides bearing succinyl lysine residues, but does not cross-react with the peptides bearing other structurally similar modified residues.

Dot Blot



1. Succinyl lysine peptide library
2. Acetyl lysine peptide library
3. Propionyl lysine peptide library
4. Butyryl lysine peptide library
5. Control lysine peptide library

Scientific Background:

Succinylation modification refers to the addition of succinyl groups to the lysine residues of proteins. Compared with methylation and acetylation, succinylation modification can cause more changes in protein structure and function. The substrate for succinylation modification is succinyl coenzyme A. Since succinyl coenzyme A is mainly produced in the TCA cycle in mitochondria, succinylation was initially considered as a non-enzymatic process occurring in mitochondria and is regulated by factors such as succinyl coenzyme A concentration. Succinylation modification plays a crucial role in tumor progression, liver diseases, neurological diseases, metabolic diseases, and plant stress. In the brain of Alzheimer's disease patients, significant changes in protein succinylation occur. In particular, there is a reduction in mitochondrial protein succinylation and an increase in some extramitochondrial protein succinylation. This change may be related to the pathological process of the disease. Succinylation can change enzyme activity and metabolic pathways and affect processes such as insulin signal transduction, glucose metabolism, and fat metabolism, thus playing a role in the pathogenesis of metabolic diseases. In tumor cells, succinylation modification can regulate the expression of oncogenes and tumor suppressor genes and affect biological behaviors such as tumor cell proliferation, apoptosis, invasion, and metastasis.

Please note: This product is "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES".

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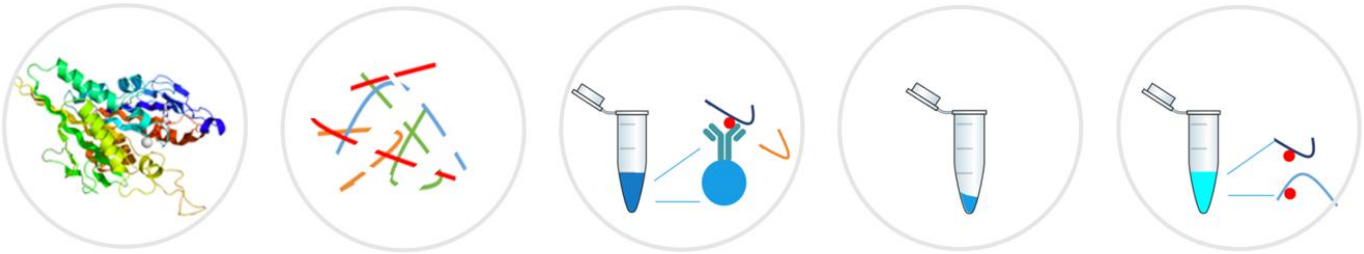
Outline of Peptide Immunoaffinity Enrichment



① **Protein extraction** ② **Protein digestion** ③ **Peptide enrichment** ④ **Beads washing** ⑤ **Peptide elution**

Product Manual

1. Gently swirl the bottle of Anti-Succinyl Lysine Antibody Conjugated Agarose Beads to obtain an even suspension. Using a wide-bore or cut pipette tip, add 40μL 50% beads slurry to 0.6 mL tube.
2. Wash beads with 0.5 mL pre-chilled **PBS**. Spin down beads at 1000 x g for 1 min at 4°C, and remove the supernatants. Repeat twice.
3. Dissolve 2 mg peptides in 400 μL **IP buffer**.
4. Remove any possible precipitates in peptide solution by centrifuging at 12,000 x g for 10 min at 4°C.
5. Mix peptide solution with pre-washed beads. Incubate at 4°C overnight with gentle end-over-end rotation.
6. Harvest beads by centrifuging at 1000 x g for 1 min at 4°C.
7. Wash beads with 0.5 mL **Wash Buffer I** by inverting tube 15 times. Spin down beads and discard supernatants. Repeat twice.
8. Wash beads with 0.5 mL **Wash Buffer II** by inverting tube 15 times. Spin down beads and discard supernatants. Repeat twice.
9. Wash beads with 0.5 mL deionized water by inverting tube 15 times. Spin down beads and discard supernatants. Repeat twice.
10. Elute bound peptides with 100 μL **Elution Buffer**. Incubate for 15 min by end-over-end rotating at room temperature. Spin down beads and transfer eluates into a new tube.
11. Repeat twice and combine all three eluates.

Additional Reagents Required

1. Phosphate-buffered saline (PBS, pH=7.2): 10 mM Na_2HPO_4, 1.8 mM KH_2PO_4, 2.7 mM KCl, 137 mM NaCl.
2. IP buffer, Wash Buffer I, Wash Buffer II and Elution Buffer are from PTM BIO buffer kit and should be purchased seperately.

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