U-Blot® D-Lactyllysine Specific Antibody Conjugated Agarose Beads (WT9162B)

User Manual

General Information

Product name	U-Blot® D-Lactyllysine Specific Antibody Conjugated Agarose Beads
Product No.	WT9162B
Clonality	Monoclonal
Alternative name	KD-la
Product Description	D-Lactyllysine Specific Antibody Conjugated Agarose Beads is an immunoaffinity enrichment tool for D-lactyl lysine peptides in mass spectrometry-based proteomics. The highly specific antibody selectively captures target modified peptides without cross-reactivity, is applicable to all species, efficiently enriches low-abundance peptides and reduces background interference, delivering stable and reliable results for D-lactyl lysine modification research.

Key Features

Target	D-Lactyllysine
Reactivity	Species Independent
Applications	IAC
Host Species	Rabbit
Modification	Lysine lactylation
Conjugate	Agarose Beads

Storage Information

Storage	Product is stable for several weeks at 4° C. For extended storage, store product at -20° C
Stability	Expiration date is one year from date of shipping if stored properly
Formulation	50/250 µL settled agarose beads supplied as 50% slurry, containing 66% glycerol

Target Information

Background	D-Lactyllysine ($K_{(}$D-la$_{)}$) is the D-stereoisomer of lysine lactylation, a novel type of lysine acylation post-translational modification. By conjugating a D-lactyl group to lysine residues, $K_{(}$D-la$_{)}$ is mainly generated via non-enzymatic pathways from glycolysis byproducts and exists on both histone and non-histone proteins. It participates in chromatin remodeling, gene transcription, and cellular metabolic stress responses, and is closely associated with tumorigenesis, inflammation, metabolic reprogramming, and other pathological processes, serving as a key target in metabolic-epigenetic crosstalk research.

Protocol

Required buffer solutions

1、**IP Buffer:** 50 mM Tris-HCl, 150 mM NaCl, 1 mM EDTA, 0.1% NP-40, pH 7.5

2、**IP Wash Buffer I:** 50 mM Tris-HCl, 100 mM NaCl, 1 mM EDTA, supplemented with 0.5% NP-40 and 0.1% Triton X-100, pH adjusted to 8.0

3、**IP Wash Buffer II:** 50 mM Tris-HCl, 100 mM NaCl, 1 mM EDTA, pH adjusted to 8.0

4、**Elution Buffer:** 0.1% Trifluoroacetic acid (TFA) in water (or 50 mM Ammonium Bicarbonate, pH 8.0)

Immunoaffinity Procedures

1、Gently swirl the bottle of Anti-D-Lactyl Lysine Antibody Conjugated Agarose Beads to obtain an even suspension. Using a wide-bore or cut pipette tip, add 40 µL 50% beads slurry to a 0.6 mL tube.

2、Wash beads with 0.5 mL pre-chilled PBS.

3、Centrifuge at 1000 × g for 1 minute at 4 ℃, discard supernatant and repeat washing twice.

4、Dissolve 2 mg peptides in 400 µL IP buffer.

5、Centrifuge the peptide solution at 12,000 × g for 10 minutes at 4 ℃ to remove possible precipitates.

6、Add the clarified peptide solution to pre-washed beads, incubate overnight with gentle end-over-end rotation at 4 ℃.

Washing Process

1、Centrifuge at 1000 × g for 1 minute at 4 ℃ to harvest beads.

2、Add 0.5 mL Wash Buffer I, invert the tube 15 times for thorough mixing, spin down beads and discard supernatant. Repeat twice.

3、Replace with 0.5 mL Wash Buffer II, invert the tube 15 times for thorough mixing, spin down beads and discard supernatant. Repeat twice.

4、Finally add 0.5 mL deionized water, invert the tube 15 times for thorough mixing, spin down beads and discard supernatant. Repeat twice.

Peptide Elution Operation

1、Ensure beads are fully pelleted after the final wash.

2、Add 100 µL Elution Buffer to the beads.

3、Incubate for 15 minutes with end-over-end rotation at room temperature.

4、Spin down beads and transfer the eluate into a new centrifuge tube.

5、Perform elution twice more and combine all three eluates.

6、(Optional, not included in the original manual: Desalt samples via C18 column, freeze-dry, reconstitute with loading buffer and proceed to mass spectrometry detection.)

Precautions

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.