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- Diagnostik & molekulare Diagnostik
- Laborgeräte & Service

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Lieferung & Zahlungsart

siehe unsere Liefer- und Versandbedingungen

Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

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Data Sheet

Anti-H4R3me2(sym) polyclonal antibody

Catalog #: 78558

Lot #: 221024	**Host Species:** Rabbit
Conc.: 0.6 µg/µl	**Species Reactivity:** Human
Size: 50 µg	**Immunogen:** Synthetic peptide
Clonality: Polyclonal	**Purification:** Affinity purified

Description: Polyclonal antibody raised in rabbit against the region of histone H4 containing the symmetrically dimethylated arginine 3 (H4R3me2(sym)), using a KLH-conjugated synthetic peptide. The antibody also recognizes H2AR3me2(sym).

Background: Histones are the main constituents of the protein part of chromosomes of eukaryotic cells. They are rich in the amino acids arginine and lysine and have been greatly conserved during evolution. Histones pack the DNA into tight masses of chromatin. Two core histones of each class H2A, H2A, H3 and H4 assemble and are wrapped by 146 base pairs of DNA to form one octameric nucleosome. Histone tails undergo numerous post-translational modifications, which either directly or indirectly alter chromatin structure to facilitate transcriptional activation or repression or other nuclear processes. In addition to the genetic code, combinations of the different histone modifications reveal the so-called “histone code”. Histone methylation and demethylation is dynamically regulated by respectively histone methyl transferases and histone demethylases.

Formulation: PBS containing 0.05% azide and 0.05% ProClin 300

Applications (Suggested dilution amounts): ELISA (1:500), Dot blot (1:5,000), WB (1:1,000), IF (1:500)

Storage/Stability: Store at -80°C for up to 2 years. Centrifuge after first thaw to maximize product recovery. Aliquot to avoid repeated freeze/thaw cycles. Aliquots may be stored at -20°C for at least one month.

Warnings: Avoid freeze/thaw cycles

Notes: The optimal antibody amount per IP should be determined by the end-user. We recommend testing 1-5 µg per IP

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Quality Assurance:

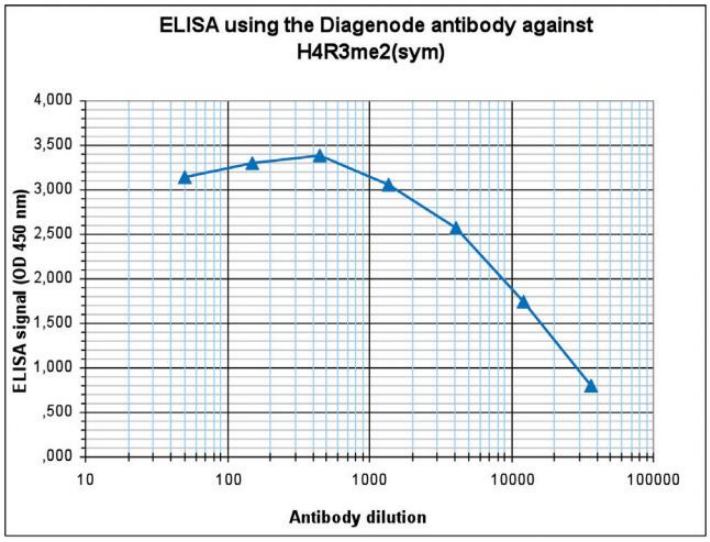

Figure 1. Determination of the antibody titer
To determine the titer of the antibody, an ELISA was performed using a serial dilution of the antibody directed against H4R3me2 (sym) (BPS Bioscience #78558) in antigen coated wells. The antigen used was a peptide containing the histone modification of interest. By plotting the absorbance against the antibody dilution (Figure 1), the titer of the antibody was estimated to be 1:11,750.

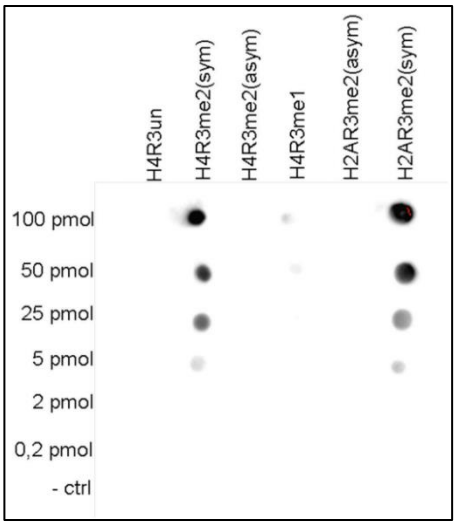

Figure 2. Cross reactivity tests using the antibody directed against H4R3me2(sym)
To test the cross reactivity of the antibody against H4R3me2(sym) (BPS Bioscience #78558), a Dot Blot analysis was performed with peptides containing other histone arginine methylations and the unmodified H4R3. One hundred to 0.2 pmol of the respective peptides were spotted on a membrane. The antibody was used at a dilution of 1:5,000. Figure 2 shows the antibody is specific for the H4R3 symmetric dimethylation and also recognizes the H2AR3 symmetric dimethylation.

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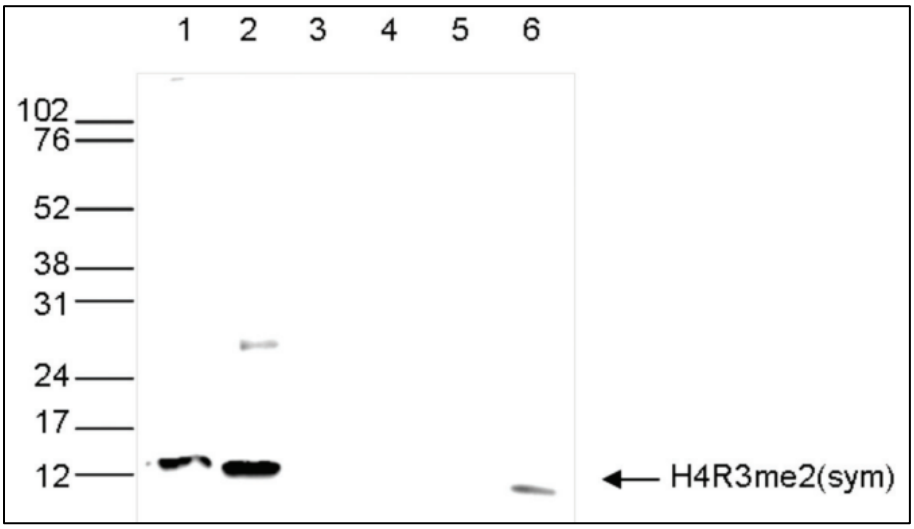

Figure 3. Western blot analysis using the antibody directed against H4R3me2(sym)
Western blot was performed on whole cell (25 µg, lane 1) and histone extracts (15 µg, lane 2) from HeLa cells, and on 1 µg of recombinant histone H2A, H2B, H3 and H4 (lane 3, 4, 5 and 6, respectively) using the antibody against H4R3me2(sym) (BPS Bioscience #78558). The antibody was diluted 1:1,000 in TBS-Tween containing 5% skimmed milk. The position of the protein of interest is indicated on the right; the marker (in kDa) is shown on the left.

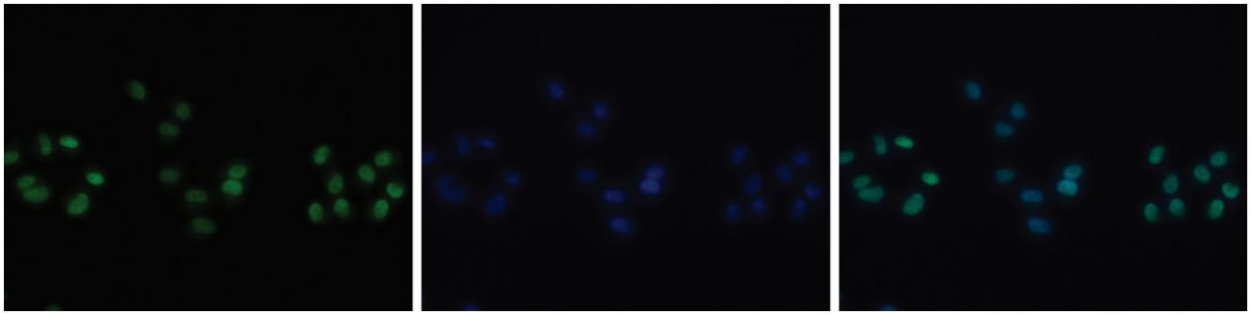

Figure 4. Immunofluorescence using the antibody directed against H4R3me2(sym)
HeLa cells were stained with the antibody against H4R3me2(sym) (BPS Bioscience #78558) and with DAPI. Cells were fixed with 4% formaldehyde for 10' and blocked with PBS/TX-100 containing 5% normal goat serum and 1% BSA. The cells were immunofluorescently labeled with the H4R3me2(sym) antibody (left) diluted 1:500 in blocking solution followed by an anti-rabbit antibody conjugated to Alexa488. The middle panel shows staining of the nuclei with DAPI. A merge of the two stainings is shown on the right.