

This product is for research use only (not for diagnostic or therapeutic use)

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Product no AS11 1771

Anti-His-tag | 6xHis (clone HIS,H8 / EH158)

Product information

Immunogen <u>KLH</u>-conjugated synthetic peptide 6xHis

Host Mouse

Clonality | Monoclonal

Subclass/isotype | IgG2b

Purity Total IgG fraction. Protein A purified.

Format Liquid

Quantity 50 μg

Storage Store at -20°C.

Additional information Working dilution for ELISA, IL and IP needs to be determined experimentally

Application information

Recommended dilution 1:1000 (WB)

Confirmed reactivity 6xHis

Predicted reactivity 6xHis

Not reactive in No confirmed exceptions from predicted reactivity are currently known

Additional information

Antibody is present in 10 mM PBS, pH 7.2

His-tag (6,8,10xHis) needs to be properly exposed to allow detection. To prevent target protein folding, extraction should be performed with 6 to 8 M urea or using <u>TCA-acetone precipitation method</u>.

Selected references

Morey-Yagi et al. (2025). Expression of spider silk protein in tobacco improves drought tolerance with minimal effects on its mechanotype. Plant J. 2025 Jan;121(2):e17213.doi: 10.1111/tpj.17213.

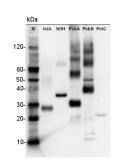
<u>Pinczés</u> et al. (2024). Viral coat proteins decrease the gene silencing activity of cognate and heterologous viral suppressors. Sci Rep. 2024 Dec 28;14(1):31008. doi: 10.1038/s41598-024-81998-4.

<u>De</u> Brasi-Velasco et al. (2021). Autophagy Is Involved in the Viability of Overexpressing Thioredoxin o1 Tobacco BY-2 Cells under Oxidative Conditions. Antioxidants. 2021; 10(12):1884. https://doi.org/10.3390/antiox10121884

<u>Tan</u> et al. (2020). Salicylic Acid Targets Protein Phosphatase 2A to Attenuate Growth in Plants. Curr Biol. 2020 Feb

3;30(3):381-395.e8. doi: 10.1016/j.cub.2019.11.058.

<u>López-Vidal</u> et al. (2020). Is Autophagy Involved in Pepper Fruit Ripening? Cells, 9 (1), DOI: 10.3390/cells9010106 <u>Häggmark-Månberg</u> et al. (2016). Autoantibody targets in vaccine-associated narcolepsy. Autoimmunity. 2016 Sep;49(6):421-433. Epub 2016 May 20.



500 femtomoles of His-tagged proteins IsiA, NifH, PsbA, PsbB and PetC were loaded per gel well in Agrisera PEB extraction buffer. Proteins were separated on 4-12 % NuPAGE PAGE Bis-Tris polycacrylamide gel (Invitrogen) and blotted 1h to PVDF. Blots were blocked with for 1h at room temperature (RT) with agitation. Blot was incubated in the primary antibody at a dilution of 1: 1 000 for 1h at RT with agitation. The antibody solution was decanted and the blot was rinsed briefly twice, then washed once for 15 min and 3 times for 5 min in TBS-T at RT with agitation. Blot was incubated in secondary antibody (goat, anti-mouse IgG horse radish peroxidase conjugated, from Agrisera AS11 1772) diluted to 1:25 000 in 2 % ECL Advance blocking reagent for 1h at RT with agitation. The blot was washed as above and developed for 5 min with ECL according to the manufacturers instructions. Exposure time was 5 seconds.

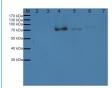


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Apparent molecular weight of recombinant proteins: IsiA - 27 kda, NifH - 34 kDa, PsbA - 30-37 kDa, PsbB - 40 kDa, PetC - 23 kDa.



20 µl of media form *Pichia pastoris* culture overexpressed His-Tegged proteins were separated on 12 % SDS-PAGE and blotted 1h to PVDF. Blots were blocked with 5% skim milk for 1h at room temperature (RT) with agitation. Blot was incubated in the primary antibody at a dilution of 1: 1 000 for 1h at RT with agitation. The antibody solution was decanted and the blot was rinsed briefly twice, then washed once for 15 min and 3 times for 5 min in TBS-T at RT with agitation. Blot was incubated in secondary antibody (goat anti-rabbit IgG horse radish peroxidase conjugated, from Bio-Rad) diluted to 1:15 000 in for 1h at RT with agitation. The blot was washed as above and developed for 5 min with ECL (GE Heltcare) according to the manufacturer's instructions. Exposure time was 15 seconds.

M- protein ladder (Fermentas)

- 2- 20ul of medium before induction (PcGCE protein)
- 3-20ul of medium before induction (PcGCE S217N protein)
- 4-20ul of medium after 48h of induction (PcGCE protein)
- 5-20ul of medium after 48h of induction (PcGCE S217N protein)
- 6-20ul of medium after 96h of induction (PcGCE protein)
- 7-20ul of medium after 96h of induction (PcGCE S217N protein)

Courtesy of Dr. Marta Derba-Maceluch, UPSC, Umeå