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Product no AS09 600

HydA2 | Iron-hydrogenase HydA2

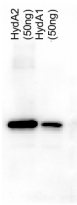
Product information

Immunogen	Recombinant, full length *Chlamydomonas reinhardtii* HydA-2 UniProt: Q8VZZ0
Host	Rabbit
Clonality	Polyclonal
Purity	Serum
Format	Lyophilized
Quantity	100 µl
Reconstitution	For reconstitution add 100 µl of sterile water
Storage	Store lyophilized/reconstituted at -20°C; once reconstituted make aliquots to avoid repeated freeze-thaw cycles. Please remember to spin the tubes briefly prior to opening them to avoid any losses that might occur from material adhering to the cap or sides of the tube.
Additional information	In *Chlamydomonas* **HydA** is present in low levels of **1 µg/liter of culture.** Therefore, an induction of cells by anaerobic adaptation or sulfur depravation (10 x higher amount than with anaerobic adaptation) is necessary for successful detection using this antibody. Methods of HydA induction are described in Hemschemeier et al. 2009. To detect HydA in *Chlamydomonas* extracts amount loaded per well corresponds to 2 µg of chlorophyll for sulfur deprived cells, where relatively much HydA1 is synthesized or corresponds to 2-4 µg of artificially anaerobic induced cultures, where the HydA1 protein level is lower. This antibody is recognizing 1 ng of recombinant HydA protein.

Application information

Recommended dilution	1 : 5000 (WB)
Expected | apparent MW	53,7 | 47,3 kDa (after a signal peptide is cleaved)
Confirmed reactivity	*Chlamydomonas reinhardtii,* recombinant HydA2 expressed in *E.coli*
Predicted reactivity	*Ostreococcus* sp. Species of your interest not listed? Contact us
Not reactive in	No confirmed exceptions from predicted reactivity are currently known
Additional information	HydA2 (505aa) has a calculated MW of 53.7 kDa, but this is including the signal peptide, which gets cleaved off. The protein without TP can only be estimated, since the cleavage site is known only from in silico analysis. It has a calculated MW of 47.3 kDa and should run in the gel also according to its size. HydA1 (497aa) has a calculated MW of 53.1 kDa, but this is including the signal peptide, which gets cleaved off. The protein without TP has a calculated MW of 47.5 kDa and runs according to its size at about 48 kDa

Application example



50 ng of purified protein (HydA1 and HydA2) were separated on 10% SDS-PAGE and blotted 25 min to PVDF membrane. Filters were blocked 1h with 3% low-fat milk powder in PBS-T (0.1% TWEEN 20) and probed with anti-HydA2 (AS09 600, 1:5000, over night at 4°C) and secondary anti-rabbit (1:10 000, 1 h) antibody (HRP conjugated, manufacture Pierce) in PBS-T containing 3% low fat milk powder. Antibody incubations were followed by washings in PBS-T (10, +10min and PBS (+5, +5 min). All washing steps were performed at RT with agitation. Signal was detected with chemiluminescent detection, using CCD camer. Exposure time was 20 min.

Courtesy Dr. Thomas Happe, Ruhr-Universität Bochum, Germany