

Product no **AS08 295****GLN1 GLN2 | GS1 GS2 glutamine synthetase global antibody****Product information**

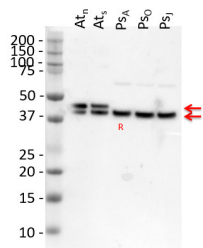
<b>Immunogen</b>	KLH-conjugated synthetic peptide derived from a wide range of available sequences including all isoforms of <i>Arabidopsis thaliana</i> <u>GLN1-1,1-2,1-3</u> and <u>1-4</u> , ( <u>At5g37600</u> , <u>At1g66200</u> , <u>At3g17820</u> , <u>At5g16570</u> )
<b>Host</b>	Rabbit
<b>Clonality</b>	Polyclonal
<b>Purity</b>	Serum
<b>Format</b>	Lyophilized
<b>Quantity</b>	50 µl
<b>Reconstitution</b>	For reconstitution add 50 µl of sterile water
<b>Storage</b>	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** | The antibody will recognize both, cytoplasmic and chloroplastic forms of the GS enzyme

**Application information**

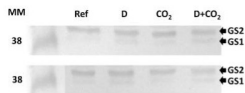
<b>Recommended dilution</b>	1 : 10 000 (WB)
<b>Expected   apparent MW</b>	39-40 kDa (GLN1, cytoplasmic form), 44-45 kDa (GLN2, chloroplastic form)
<b>Confirmed reactivity</b>	<i>Arabidopsis thaliana</i> , <i>Eragrostis tef</i> , <i>Gracilaria gracilis</i> (red algae), <i>Gracilaria lemaneiformis</i> , <i>Leptodictyum riparium</i> (Hedw.) Warnst (moss), <i>Medicago truncatula</i> , <i>Physcomitrium patens</i> , <i>Pinus strobus</i> , <i>Spinacia oleracea</i> , <i>Solanum lycopersicum</i> , <i>Triticum aestivum</i> , <i>Zea mays</i>
<b>Predicted reactivity</b>	<i>Brachypodium distachyon</i> , <i>Brassica napus</i> , <i>Camellia sinensis</i> , <i>Citrus clementina</i> , <i>Cucumis melo</i> , <i>Daphnia magna</i> , <i>Datisca glomerata</i> , <i>Emiliania huxleyi</i> , <i>Eucalyptus grandis</i> , <i>Gazania splendens</i> , <i>Genlisea aurea</i> , <i>Glycine max</i> , <i>Helianthus annuus</i> , <i>Hordeum vulgare</i> , <i>Oryza sativa</i> , <i>Panax quinquefolius</i> , <i>Phaseolus angularis</i> , <i>Phytophthora cinnamomi</i> , <i>Populus trichocarpa</i> , <i>Saccharum officinarum</i> , <i>Securigera parviflora</i> , <i>Solanum lycopersicum</i> , <i>Solanum tuberosum</i> , <i>Stevia rebaudiana</i> , <i>Theobroma cacao</i> , <i>Zea mays</i> , <i>Vitis labrusca</i>
	GLN1 dicots including: <i>Brassica napus</i> , <i>Phaseolus vulgaris</i> , monocots including: <i>Hordeum vulgare</i> , <i>Oryza sativa</i> , trees: <i>Pinus sylvestris</i> , <i>Populus</i> sp., <i>Zosteria marina</i>
	GLN2 dicots including: <i>Brassica napus</i> , <i>Glycine max</i> , <i>Phaseolus vulgaris</i> , monocots including: <i>Triticum aestivum</i> , <i>Oryza sativa</i>
	GLN3: <i>Zea mays</i> ,
	GLN1 in algae: <i>Chlamydomonas reinhardtii</i>
	Species of your interest not listed? <a href="#">Contact us</a>
<b>Not reactive in</b>	No confirmed exceptions from predicted reactivity are currently known
<b>Selected references</b>	<a href="#">Maresca</a> et al. (2021) Biological responses to heavy metal stress in the moss <i>Leptodictyum riparium</i> (Hedw.) Warnst. <i>Ecotoxicol Environ Saf.</i> 2022 Jan 1;229:113078. doi: 10.1016/j.ecoenv.2021.113078. Epub 2021 Dec 17. PMID: 34929502. <a href="#">Silva</a> et al. (2019). Characterization of plant glutamine synthetase S-nitrosation. <i>Nitric Oxide.</i> 2019 Apr 23;88:73-86. doi: 10.1016/j.niox.2019.04.006. <a href="#">Wang</a> et al. (2018). Response of <i>Gracilaria lemaneiformis</i> to nitrogen deprivation. <i>Algal Research</i> Volume 34, September 2018, Pages 82-96. <a href="#">Witzel</a> et al. (2017). Temporal impact of the vascular wilt pathogen <i>Verticillium dahliae</i> on tomato root proteome. <i>J Proteomics.</i> 2017 Oct 3;169:215-224. doi: 10.1016/j.jprot.2017.04.008. <a href="#">Silva</a> et al. (2015). Possible role of glutamine synthetase of the prokaryotic type (GS1-like) in nitrogen signaling in <i>Medicago truncatula</i> . Volume 240, November 2015, Pages 98–108. <a href="#">Lang</a> et al. (2011). Simultaneous isolation of pure and intact chloroplasts and mitochondria from moss as the basis for sub-cellular proteomics. <i>Plant Cell Rep.</i> 2011 Feb;30(2):205-15. doi: 10.1007/s00299-010-0935-4.

## Application example



10 µg of total protein extracted freshly from *Arabidopsis thaliana* wt leaf tissue ( $At_1$ , non-senescent leaves), *Arabidopsis thaliana* wt leaf tissue ( $At_5$ , senescent leaves), *Pinus strobus* needle tissue ( $PS_{A-J}$ ) with 1 M Tris-HCl, pH 6.8, 10 % SDS, 15 % sucrose, 0.5 DTT and denatured at 75 °C for 5 min. were separated on 10 % Bis-Tris Nupage Novex gel (120 V/45 min. using MES buffer system) and blotted 30 min. to PVDF. Blot was blocked with 5 % non-fat milk 45 min./RT with agitation. Blot was incubated in the primary antibody at a dilution of 1: 10 000 for 1h/RT with agitation in TBS with 2 % non-fat milk or ON/4 °C with agitation. The antibody solution was decanted and the blot was rinsed briefly twice for 10 min. in TBS at RT with agitation. Blot was incubated in Agrisera matching secondary antibody (anti-rabbit IgG horse radish peroxidase conjugated, [AS09 602](#)) diluted to 1:75 000 in for 1h/RT with agitation. The blot was washed as above and developed using chemiluminescent detection. Exposure time was 26.5 seconds.

Courtesy of Dr. Christine Yao-Yun Chang and the Ensminger lab, University of Toronto, Canada



The detection of GS1 and GS2 proteins was performed using the crude extract of soluble proteins from *Oryza sativa* plants: Ref: Reference (control); D: Drought;  $CO_2$ : High  $CO_2$ ; D+ $CO_2$ : Drought + High  $CO_2$ . Fresh leaves samples were ground until obtaining a fine powder in presence of liquid  $N_2$ , ice-cold 100 mM K-phosphate buffer (pH 7.0) containing 1 mM EDTA and 2 mM ascorbic acid. After centrifugation at 14,000 x g for 30 min, the supernatant was collected and used as protein extract. All extraction stages were carried out at 4 °C. The total soluble protein was measured according to the Bradford's method. Leaf protein extracts were first separated by SDS-PAGE (Laemmli 1970). Equal amounts of protein (20 µg) were electrophoretically transferred to a nitrocellulose membrane (Towbin et al. 1979). Polypeptide detection was performed using specific polyclonal antibodies against GS1 and GS2 (AS08 295, Agrisera, Sweden). Membranes were blocked for 3 hours with 5% non-fat milk in saline Tris-HCl buffer (100 mM Tris-HCl, pH 7.6, 150 mM NaCl), incubated with GS antibody overnight and after with alkaline phosphatase-conjugated secondary antibody by 6 hours. The protein detection was developed using NBT/BCIP (Sigma-Aldrich®, USA) by adding 1 tablet to 10 mL  $dH_2O$ , until bands were visualized.

Courtesy of Dr. Ana Karla Lobo, Laboratory of Plant Metabolism, Federal University of Ceara, Brazil