

Anti-ASN (asparagine synthetase, plant) antibody, rabbit polyclona 81-031 200 µg

Storage: Shipped at 4°C and store at -20°C. Do not freeze.

Validation: The specificity of the antibody has been validated by western blotting with mutant plants.

Immunogen: Purified recombinant Arabidopsis asparagine synthetase isoprotein 2 (ASN2), full-size, no-tag attached.

Reactivity: Plant ASN2 and ASN1 isoproteins

Applications:

- 1. Western blotting (1/1,000-1/2,000 dilution)
- 2. Immunohistochemistry, paraffin section (1/100-1/500)
- 3. ELISA (assay dependent)

Other applications have not been tested.

Purity: IgG purified with protein A/G mix

Form: 4 mg/ml in PBS, 50% glycerol. Filter sterilized. No preservative or carrier protein added.

Background: Asparagine synthetase 2 (ASN2) is essential for nitrogen assimilation, distribution and remobilization within the plant via the phloem. ASN2 is expressed in leaf and ASN1 is expressed in floral organs.

The amino acid sequences of Arabidopsis ASN1 and ASN2 are 76% identical.

The amino acid sequences of Arabidopsis and Maize ASN2 are 73.6% identical. **Data Link**: UniProtKB: <u>Q9LV77</u> (A. thaliana), <u>B5U8J7</u> (Z. mays)

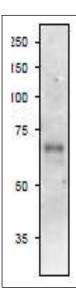


Fig.1 Western Blot of ASN2 in arabidopsis leaf extract.

Anti-ASN2 antibody was used at 1/1,000 dilution. Secondary antibody (goat anti-rabbit IgG antibody HRP-conjugated, ab97051) was used at 1/10,000 dilution.

1. Arabidopsis leaf extract, 10 μg

Molecular mass of Arabidopsis ASN2 is 65 kDa.





Fig.2 Absence of ASN2 protein in leaf extracts of asn-2-1 and asn2-2 mutants.

AS is the position of ASN2 protein migrated at 65 kDa in SDS-PAGE and 70 kda is the position of pre-stained protein size marker. Col0 is wild-type Arabidopsis plant. *asn2-1* and *asn2-2 are* T1 insertion mutants.

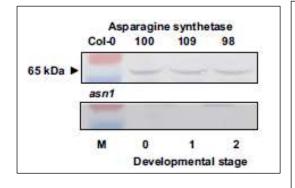


Fig.2 Absence of ASN1 protein in extracts of floral organ of *asn1* insertion mutant.

Wild type (Col-0) samples are analyzed in the upper window and *asn1* mutant samples are in the lower window. The upper numerals are the protein levels measured by densitometric tracing of western blot. Developmental stages are indicated below. Marker proteins are 72 and 55 kDa.

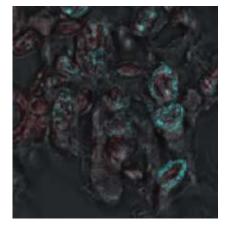


Fig.3 Immunofluorescence analysis of ASN2 in plant leaf section. Arabidopsis thin leaf section was subjected to indirect

immunofluorescence analysis using the Anti-ASN antibody as the primary antibody. Goat anti-rabbit IgG labelled with Alexa 405 (Molecular Probes) was

Goat anti-rabbit IgG labelled with Alexa 405 (Molecular Probes) was used as a secondary antibody.

Blue: ASN2 protein probed with anti-ASN antibody Red: Autofluorescence

Reference: This product has been used in the following publications.

1. Gaufichon L, Masclaux-Daubresse C, Tcherkez G, Reisdorf-Cren M, Sakakibara Y,



Hase T, Clément G, Avice JC, Grandjean O, Marmagne A, Boutet-Mercey S, Azzopardi M, Soulay F, Suzuki A. "Arabidopsis thaliana ASN2 encoding asparagine synthetase is involved in the control of nitrogen assimilation and export during vegetative growth." Plant Cell Environ. 2013 Feb;36(2):328-42. PMID: <u>22789031</u> WB, IHC ;Arabidopsis

2. Gaufichon L, et al. ASN1-encoded asparagine synthetase in floral organs contributes to nitrogen filling in Arabidopsis seeds. <u>Plant J.</u> 2017 Aug;91(3):371-393. PMID:28390103 WB, IHC; Arabidopsis