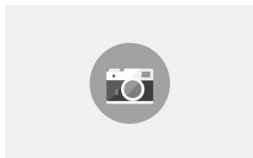


Anti-PIP2;1, PIP2;2, PIP2;3 | Plasma membrane intrinsic protein 2-1,2-2,2-3

Qty: AS09 491



AS09 491 | Clonality: **Polyclonal** | Host: **Rabbit** | Reactivity: ***A. thaliana*, *G. coquereliana*, *R. sativus***

Replaced by AS22 4810

Price:

[Agrisera Western Blot protocol and video tutorials](#)

[Protocols to work with plant and algal protein extracts](#)

[Agrisera Educational Posters Collection](#)

Method for isolation of plant plasma membranes

Isolation of Plasma Membranes from Plant Tissues

Stock solutions

- (1) 100 mM K-phosphate buffer, pH 7.8 [200 mL]
 (2) 0.25 M sucrose, 10 mM K-phosphate buffer, pH 7.8, 1 mM DTT [sucrose/KP/DTT] [500 mL]
 (3) 300 mM NaCl in sucrose/KP/DTT [100 mL]

Phase separation solution (four tubes, 50 mL Falcon tube)

PEG 3640 (Sigma P-3640)	3.0 g
Dextran T500 (Pharmacia)	3.0 g
Sucrose/KP/DTT (2)	28.0 mL
300 mM NaCl/sucrose/KP/DTT (3)	4.8 mL

Store at 0°C
(very important!)



Tissue homogenate

Filtration 10,000xg, 15 min (6,000 – 10,000 rpm)

Sup

150,000xg, 30 min (35,000 – 42,000 rpm)

Ppt (ER, Golgi, Plasma membrane, vacuolar membrane)

10 mL of Sucrose/KP/DTT (2)

Suspension

Phase separation solution (0°C)

Mix strongly 4,500xg, 4 min

upper phase

Plasma membrane

lower phase

Mix 4,500xg, 4 min

upper phase

3 vol. of 0.25 M sucrose, 20 mM Tris-acetate, pH 7.2, 1 mM DTT

45,000xg, 30 min

Ppt. suspended in sucrose/KP/DTT

Plasma membranes

Courtesy of Dr. Masayoshi Maeshima, Laboratory of Cell Dynamics, Graduate School of Bioagricultural Sciences Nagoya University Nagoya, Japan

- Product Info
- Immunogen:

KLH-conjugated synthetic peptide derived from *Arabidopsis thaliana* PIP2 proteins: AtPIP2-1, At3g53420, AtPIP2-2 P43287, At2g37170, AtPIP2-3 P30302, At2g37180

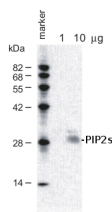
Host:	Rabbit
Clonality:	Polyclonal
Purity:	Serum
Format:	Lyophilized
Quantity:	100 µl
Reconstitution:	For reconstitution add 100 µl of sterile water
Storage:	Store lyophilized/reconstituted at -20 °C; make aliquots to avoid repeated freeze-thaw cycles. Please remember to spin the tubes briefly prior to opening them to avoid any losses that might occur from material adhering to the cap or sides of the tube.
Tested applications:	ELISA (ELISA), Western blot (WB)
Recommended dilution:	1 : 8000 (ELISA), 1 : 1000 (WB)
Expected apparent MW:	30.4 28 (PIP2-1, PIP2-2, PIP2-3) kDa
• Reactivity	
• Confirmed reactivity:	<i>Arabidopsis thaliana</i> , <i>Camelina sativa</i> , <i>Gromphadorhina coquereliana</i> , <i>Raphanus sativus</i> <i>Brassica napus</i> , <i>Cucumis sativus</i> , <i>Glycine hispida</i> , <i>Gossypium hirsutum</i> , <i>Hedychium coronarium</i> , <i>Mimosa saman</i> , <i>Nicotiana glauca</i> , <i>Petunia hybrida</i> , <i>Pisum sativum</i> , <i>Ricinus communis</i> , <i>Populus tremula</i> <i>x Populus tremloides</i> , <i>Physcomitrium patens</i>
Predicted reactivity:	

Species of your interest not listed? [Contact us](#)

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

• Application Examples

• Application example



1 µg and 10 µg of crude membrane fraction/lane from *Arabidopsis thaliana* were separated on 12 % **SDS-PAGE** and blotted 1h to **PVDF membrane** (40 min. at 10 V using BioRad semidry transfer). Filters were blocked 1h with 5 % low-fat **milk powder** in TBS-T (0.05% Triton X.100). Membranes were washed 5 times with TBS-T, each time in a fresh polystyrene box and probed with anti-PIP2s antibodies (AS09 491, **1:1000**, 1h) and secondary anti-rabbit (**1:2000**, 1 h). All steps were performed in RT with agitation.

• Additional Information

- 0.1% sodium azide is added as preservative. For antibody re-suspending information check the tube label.

Additional information:

Antibodies will detect target protein in a few µg of a crude preparation loaded per well. If purified preparations of vacuolar and plasma membranes are used, one µg load per well should be sufficient. Protein or membrane sample should be treated at 70 °C for 10 min before loading on the gel.

Diluted antibody solution can be used 2 to 3 times within one month if it contains 0.1% sodium azide as preservative and is stored at -20°C to -80°C.

Additional information (application):

Triton X-100 should not be included in the protein extraction buffer, when cell organelles or membrane proteins must be separated from soluble proteins. Because, Triton X breaks membrane structure and solubilizes most membranes proteins. Furthermore, it should be noted that Triton X at high concentrations binds SDS and mask the detergent effect of SDS for SDS-PAGE. Also, micelles of Triton X behave as a large complex with molecular mass of 90 kDa at high concentrations in SDS-PAGE.

• Background

- **PIP2;2** is a plasma membrane aquaporin.

Background:

Alternative names of isoforms: aquaporin PIP2-1, plasma membrane intrinsic protein 2a, PIP2a, aquaporin PIP2-2, plasma membrane intrinsic protein 2b, PIP2b, TMP2b, Aquaporin PIP2-3, plasma membrane intrinsic protein 2c, PIP2c, TMP2C, RD28-PIP, water stress-induced tonoplast intrinsic protein, (WSII-TIP)

- Product Citations

- Carmona-Salazar et al. (2021). Plasma and Vacuolar Membrane Sphingolipidomes: Composition and Insights on The Role of Main Molecular Species. *Plant Physiol.* 2021 Feb 11:kiab064. doi: 10.1093/plphys/kiab064. Epub ahead of print. PMID: 33570616.
- Cano-Ramirez et al. (2021) M. Plasma Membrane Fluidity: An Environment Thermal Detector in Plants. *Cells.* 2021 Oct 17;10(10):2778. doi: 10.3390/cells10102778. PMID: 34685758; PMCID: PMC8535034.
- Hyun-Sung et al. (2019). NaCl-induced CsRCI2E and CsRCI2F interact with aquaporin CsPIP2;1 to reduce water transport in *Camelina sativa* L. *Biochemical and Biophysical Research Communications*, Available online 4 April 2019.
- Chowanski et al. (2015). Cold induced changes in lipid, protein and carbohydrate levels in the tropical insect *Gromphadorhina coquereliana*. *Comp Biochem Physiol A Mol Integr Physiol.* 2015 May;183:57-63. doi: 10.1016/j.cbpa.2015.01.007. Epub 2015 Jan 23.

Selected
references: