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Product no AS19 4302 Anti-RPS6A-P240 | Phosphorylated (Ser240) 40S ribosomal protein S6-1

Product information

Immunogen	KLH-conjugated peptide derived from Arabidopsis thaliana RPS6A, UniProt: O48549, TAIR: At4g31700 phosphorylated at Ser240	
Host	Rabbit	
Clonality	Polyclonal	
Purity	Immunogen affinity purified serum in PBS pH 7.4.	
Format	Lyophilized	
Quantity	50 µg	
Reconstitution	For reconstitution add 50 μ l, 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.	

Application information

Recommended dilution	1 : 1000 (WB)
Expected apparent MW	kDa
Confirmed reactivity	Arabidopsis thaliana
Predicted reactivity	Actinidia rufa, Ananas comosus, Beta vulgaris, Cajanus cajan, Capsicum chinense, Citrus clementina, Gossypium australe, Olea europaea subsp. europaea, Oryza sativa, Panicum miliaceum, Populus alba, Sesamum indicum, Senna tora, Solanum lycopersicum, Vigna unguiculata Species of your interest not listed? <u>Contact us</u>
Selected references	Tanigawa et al. (2024). FYVE1/FREE1 is involved in glutamine-responsive TORC1 activation in plants. iScience. 2024 Aug 26;27(9):110814. doi: 10.1016/j.isci.2024.110814. Lando et al. (2024). Nitric oxide participates in sucrose–TOR signaling during meristem activation in Arabidopsis thaliana. Planta, Short Communication, October 2024, Volume 260.
	2 3 4 5 6

kDa 1 2 3 4 5 6 250-	kDa 1 2 3 4 5 6 250-
100-	100-
75-	75-
50-	50-
37-	37-
RP56	RPS6
25-	25-
20-	20-
15-	15-

Left panel is developed with anti-RPS6-p240, while right panel anti-RPS6, which serves as a loading control

Lanes:

1 and 4 - 0.5 µg of Arabidopsis thaliana whole leaf extract from untreated leaf discs

2 and 5 - 0.5 µg of Arabidopsis thaliana whole leaf extract from leaf discs treated with 10 mM glutamine (TOR activator) for 8 hours

3 and 6 - 0.5 µg of Arabidopsis thaliana whole leaf extract from leaf discs treated with 2 µM AZD-8055 (TOR inhibitor) for 8 hours

0.5 µg/well of total protein extracted freshly from mature *Arabidopsis thaliana* leaves with 50 mM HEPES pH 7.5, 5mM NaF, 2.5 mM NaPPi, 25 mM B-phosphoglycerol, Roche Complete inhibitor tablet (1x), 2% PVPP, 2 mM PMSF) and denatured with 2x SDS sample buffer (80 mM tris pH 6.8, 2% SDS, 10% glycerol, 100 mM DTT, bromophenol blue) at 99 °C for 3 min. were separated on AnyKd (BioRAD) gradient % SDS-PAGE and blotted 1h to PVDF (pore size of 0.45 um) using semi-dry transfer. Blot was blocked with 2% milk for 1h/RT with agitation. Blot was incubated in the primary antibody at a dilution of 1: 1 000 in TBS-T + 2% milk powder ON/4 °C with agitation. The antibody solution was decanted and the blot was rinsed briefly twice, then washed 3 times for 5 min in TBS-T at RT with agitation. Blot was incubated in Agrisera matching secondary antibody



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(anti-rabbit IgG horse radish peroxidase conjugated, <u>AS09 602</u>) diluted to 1:25 000 in TBS-T + 2 % milk powder for 1h/RT with agitation. The blot was washed as above and developed for 5 min with Agrisera ECLBright with GE Amersham Imager 600. Exposure time was ~60.seconds.

Courtesy of Dr. Brendan O'Leary, University of Western Australia, School of Molecular Sciences, ARC Centre for Plant Energy Biology, Australia