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This product is **for research use only** (not for diagnostic or therapeutic use) **contact: support@agrisera.com**

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Product no AS10 1261

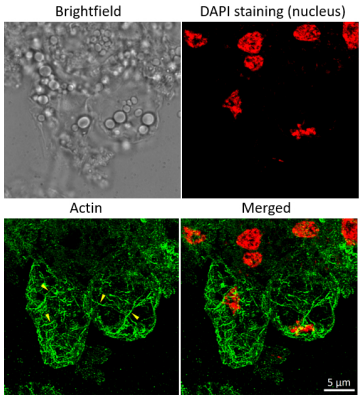
Donkey anti-Mouse IgG (H&L), DyLight® 488 conjugated

Product information

Immunogen	Purified Mouse IgG, whole molecule
Host	Donkey
Clonality	Polyclonal
Purity	Immunogen affinity purified donkey IgG.
Format	Lyophilized
Quantity	1 mg
Reconstitution	For reconstitution add 1,1 ml of sterile water, Let it stand 30 minutes at room temperature to dissolve, Prepare fresh working dilutions daily
Storage	Store lyophilized material at 2-8°C. Product is stable for 4 weeks at 2-8°C after rehydration. For long time storage after reconstitution, dilute the antibody solution with glycerol to a final concentration of 50% glycerol and store as liquid at -20°C, to prevent loss of enzymatic activity. For example, if you have reconstituted 1 mg of antibody in 1,1 ml of sterile water add 1,1 ml of glycerol. Such solution will not freeze in -20°C, If you are using a 1:5000 dilution prior to diluting with glycerol, then you would need to use a 1:2500 dilution after adding glycerol. Prepare working dilution prior to use and then discard. Be sure to mix well but without foaming.
Additional information	Conjugate is present in 10 mM Sodium Phosphate, 0.15 M Sodium Chloride, pH 7.2, 1 % (w/v) BSA, Protease/IgG free. 0.05 % (w/v) sodium azide is added as preservative. Based on immunoelectrophoresis, this antibody reacts with: heavy ( ) chains on mouse IgG, light chains on all mouse immunoglobulins No reactivity is observed to: non-immunoglobulin mouse serum proteins

Application information

Recommended dilution 1 : 20-1 : 2000 for most applications



Immunofluorescent localization of actin on suspension culture of *Oryza sativa* ssp. japonica cv. 'Unggi 9', using anti actin (AS21 4615) and anti-mouse IgG DyLight® conjugated secondary antibodies (AS10 1261). Few representative actin filaments are highlighted by yellow arrowheads. DAPI staining of nuclei is pseudocolored red.

Material: Suspension cultures of *Oryza sativa* ssp. japonica cv. 'Unggi 9'
Fixation: Packed cell volume to fixer ratio: 250 µl : 5ml Fixer composition and buffer: 4% (w/v) paraformaldehyde (freshly prepared as 8% stock and 0.2 µm filtered) in Phosphate Buffered Saline (PBS), pH 7.4 (2x stock, 0.2 µm filtered)
Container and method: in 6 cm Petri dish, gentle shaking at room temperature (RT)
Duration: 40 minutes. Triton X100 is not used in fixer. Cells were not shaken during the first 5 mins of fixation to allowed to partially recover from osmotic shock induced by formaldehyde.
Hydrophilization: no
Cell wall digestion: Yes Packed cell volume to enzyme ratio: 100ul : 2ml Enzyme composition: 1% (A) 1.2% (R) Cellulase (chromatically purified, powder, Worthington) 1% (A) 1.2% (R) Pectinase (protease free, liquid, Sigma) Buffer: 0.5% (w/v) MES buffer, pH 5.6
Container and method: in 2 ml microfuge tube by rolling at room temperature (RT)
Duration: 30 mins (A) or 60 mins (R)

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Membrane permeabilization: Triton-X100 (0.35%), 7 min/RT
Antigen retrieval: no
Blocking buffer: Fish gelatin (5% v/v)
Washing buffer: PBS
Primary antibody dilution and incubation time: 1:500, 1hr/RT
Secondary antibody dilution and incubation time and supplier: DyLight® 488 (AS10 1261) 1:600, 45 min/RT
Co-staining of the nucleus (DAPI): Yes
Cell wall and nucleus staining: 100 ng/ml DAPI

Courtesy of Dr. Ferhan Ayaydin, Hungarian Centre of Excellence for Molecular Medicine (HCEMM), Szeged, Hungary.