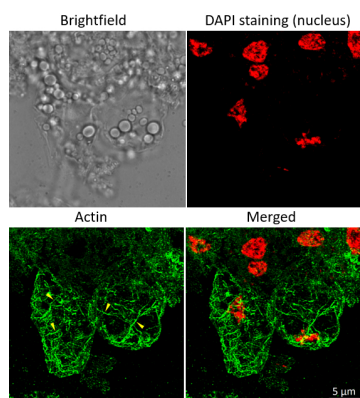


**Product no** **AS10 1261****Donkey anti-Mouse IgG (H&L), DyLight® 488 conjugated****Product information****Immunogen** Purified Mouse IgG, whole molecule**Host** Donkey**Clonality** Polyclonal**Purity** Immunogen affinity purified donkey IgG.**Format** Lyophilized**Quantity** 1 mg**Reconstitution** 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)

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.