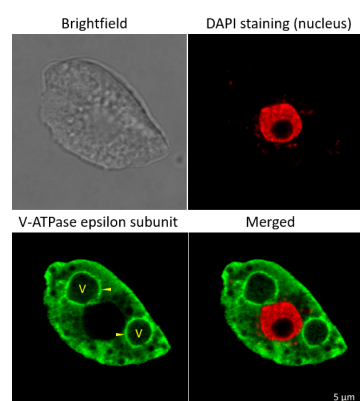


Product no AS10 1116**Donkey anti-Goat IgG (H&L), DyLight® 488 conjugated****Product information****Immunogen** | Purified Goat IgG, whole molecule**Host** | Donkey**Clonality** | Polyclonal**Purity** | Immunogen affinity purified donkey IgG.**Format** | Lyophilized**Quantity** | 1 mg**Reconstitution** | 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 goat IgG, light chains on all goat immunoglobulins

No reactivity is observed to, non-immunoglobulin goat serum proteins

BSA and milk have to be replaced by other blocking reagents, like donkey serum or commercial formulations which are free from bovine IgG.

Application information**Recommended dilution** | 1 : 20-1 : 2000 for most applications

Immunofluorescent localization of V-ATPase epsilon subunit of tonoplast H+ATPase in suspension culture of *Oryza sativa* ssp. japonica cv. 'Unggi 9', using goat anti-V-ATPase, epsilon subunit of tonoplast antibodies (AS09 577A) and donkey anti-Goat IgG (H&L), DyLight® 488 conjugated secondary antibodies (AS10 1116, Agrisera). Vacuolar membrane, tonoplast, is highlighted by yellow arrowheads. DAPI staining of nuclei is pseudocolored red.

MethodMaterial: 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) 0.01% (v/v) Triton-X100 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 min

Hydrophilization: No

Cell wall digestion: Yes Packed cell volume to enzyme ratio: 100 µl : 2 ml 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: 60 min
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: 1:600, 4°C/ON
Secondary antibody: donkey anti-Goat IgG (H&L), DyLight® 488 conjugated secondary antibodies ([AS10 1116](#), Agrisera), 1:600, 1h/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