

Goat-anti-rabbit Fab fragments

Cat.No. 702-rbCpH; Polyclonal goat antibody, 50 µg Fab1 fragment

Data Sheet

Reconstitution/ Storage	50 µg affinity purified Fab fragments, lyophilized. Rabbit serum albumin was added for stabilization. For reconstitution add 50 µl H ₂ O to get a 1mg/ml solution of antibody in PBS. Either add 1:1 (v/v) glycerol (if this does not interfere with your experiments!), then aliquot and store at -20°C until use, or store aliquots at -80°C without additives. Reconstitute immediately upon receipt! Avoid bright light to minimize photo bleaching of the fluorescent dye.
Applications	ICC: yes (see remarks)
Label	Cypher5E

Remarks

Antigen-purified primary antibodies are recommended. Unpurified rabbit serum or total IgG preparations may contain considerable amounts of unspecific IgGs that will bind plenty of the rbFab-CpH resulting in a lower overall labelling efficiency.

The pH sensitive dye only regaines its fluorescence after the acidification of the targeted cellular compartment.

ICC: Pre-incubate the primary antibody with a 2.5-fold excess of Fab-CpH conjugate (e.g., 2 µg of primary antibody with 5 µg of Fab fragments) in a total volume of 10 µl for 30 min at RT in the dark.
Adjust the antibody to the desired dilution and immediately use it for your internalization assay.

Selected General References

A common origin of synaptic vesicles undergoing evoked and spontaneous fusion.

Hua Y, Sinha R, Martineau M, Kahms M, Klingauf J

Nature neuroscience (2010) 13(12): 1451-3.

Synapse clusters are preferentially formed by synapses with large recycling pool sizes.

Welzel O, Tischbirek CH, Jung J, Kohler EM, Svetlitchny A, Henkel AW, Kornhuber J, Groemer TW

PloS one (2010) 5(10): e13514.

TO BE USED IN VITRO / FOR RESEARCH ONLY

NOT TOXIC, NOT HAZARDOUS, NOT INFECTIOUS, NOT CONTAGIOUS

Cypher5E™ is a pH-sensitive dye, fluorescent only after antibody internalization into an acidic cellular compartment. It can be detected with the filter sets for Cy5™.

Cypher5E-conjugated Fab antibody fragments from goat directed against mouse or rabbit IgGs can be used for the pre-incubation of primary antibodies that are subsequently used for internalization studies into acidic cellular compartments.