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NBP1-19015 Protocol

Immunocytochemistry/Immunofluorescence Protocol for GLO1 Antibody (NBP1-19015)

Immunocytochemistry Protocol

1. Cells are cultured one day prior to the experiment.
2. After washing twice with PBS and they are fixed with 4% paraformaldehyde in PBS at ?20C for 15 min.
3. Followed by two washes with PBS, they are permeabilized with 0.1%Triton X-100 in PBS at ?20C for 5 min.
4. To remove the detergent the cells are washed5 times with PBS and then blocked with 2.5 % goat serum in PBS for 2 hr at RT.
5. Cells are then incubated with GLOI mAb (10 μg/ml) in PBS for 1 hr at RT and washed twice for 5 min with PBS.
6. Cells are incubated with secondary antibody (anti-mouseIgG) conjugated with Texas Red (1: 400 dilution in PBS) (Molecular Probes) for 1 hr at RT.

Images of lenses were acquired on a Leica DMI 6000 B inverted microscope using a 20x objective connected to a Retiga EXI camera (Q-imaging Vancouver British Columbia). Secondary Ab contribution to immune reaction was verified by staining without the primary Ab.