

**NBP1-85740 Lot A40214**

**Type of samples: Human aortic valve endothelial cells.**

- Cells should be grown, treated, fixed and stained directly in multi-well plates, chamber slides or on coverslips.
- Aspirate liquid, then cover cells to a depth of 2–3 mm with 4% formaldehyde diluted in warm PBS.
- Block coverslips in 5% BSA for 30 min.
- While blocking, prepare primary antibody by diluting 1/100 in 5% BSA.
- Aspirate blocking solution, apply diluted primary antibody.
- Incubate 1h in room temperature.
- Rinse three times in 1x PBST for 5 min each.
- Incubate coverslips in fluorochrome-conjugates secondary antibody diluted in 5% BSA for 1,5 h at room temperature in the dark.
- Rinse three times in 1xPBS for 5 min each.
- Incubate coverslips with DAPI reagent for 15 min.
- For best results, allow mountant to cure overnight at room temperature. For long term storage, store slides flat in 4°C protected from light.