IF only

Fixation:

1. apirate media, wash briefly w/PBS.
2. Fix for 10 minutes @ RT using 4% PFA in PBS.
3. Permeabilize on ice for 10 minutes with cold PBS/0.5% Triton X-100/1:200 NEB VR inhibitor complex
4. Store cells in 70% EtOH at -20 C.

IF

1. 2x 5 min PBS
2. block 30 min RT in PBS 0.2% Tween-20 0.2% IgG-free BSA (0.2 g /100ml)
3. 1 hr RT antibody in block
4. wash 3x 4 min in PBS 0.2% Tween-20
5. biotinylated Goat anti-rabbit (or whatever) secondary, 30 min
6. wash 3x 4 min in PBS 0.2% Tween-20
7. Fix coverslip to slide with prolong gold w/dapi