

Immunofluorescence Staining

PSG5 Γ Kappa	50 μ g	FITC & R
PSG5 Flag Γ Kappa	50 μ g	FITC & M
HACAN Γ Kappa	50 μ g	FITC & M

- 1 million cells
- (a) - wash in PBS Do No vortex pipette up and down to mix
- aspirate leaving approx. 20 μ l of 1x PBS
- mix cells pipetally uniformly
- (b) approx 1 μ l cells to spread to evenly distribute cells
- let cells air dry
- (c) Fix 1:1 Acetone / Methanol -20° C for 10 mins.
- (d) Air dry
- (e) Block incubate 20% goat serum 30 mins in humid chamber at R. temp.
- (f) Wash with 1x PBS Dilute in 20% Goat serum.
- (g) Incubate (humid chamber) with antibody 1-2 hours at R. temp
- (h) Wash well with 1x PBS 4-5 times 5 minute washes 10% goat serum
- (i) Incubate to goat α Mouse Ig - FITC 1/1000 dilute in 20% Goat serum 1 hour
- (j) Wash well with 1x PBS $\times 2$. Keep moist
- (k) Add antifade 100 μ l and coverslip. Flip slide and press firmly to remove any trapped bubbles