

# Validation data for Anti-mCD3-mIgG2a

<https://www.invivogen.com/anti-mcd3-migg2a-invivoft>

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Version 21A28-NJ

Anti-mCD3-mIgG2a InvivoFit™ is an anti-mCD3ε mAb featuring the variable region of the previously described 145-2C11 armenian hamster IgG1 clone and a murinized IgG2a constant region. The binding capacity of Anti-mCD3-mIgG2a InvivoFit™ to the murine CD3ε (mCD3ε) antigen has been validated by ELISA, using coated mCD3ε protein and a Anti-IgG-HRP secondary antibody (Figure 1). The cell surface staining capacity of Anti-mCD3-mIgG2a InvivoFit™ has been confirmed with Jurkat cells stably expressing mCD3ε (Jurkat-mCD3) (Figure 2).

## Validation of Anti-mCD3-mIgG2a InvivoFit™ by ELISA

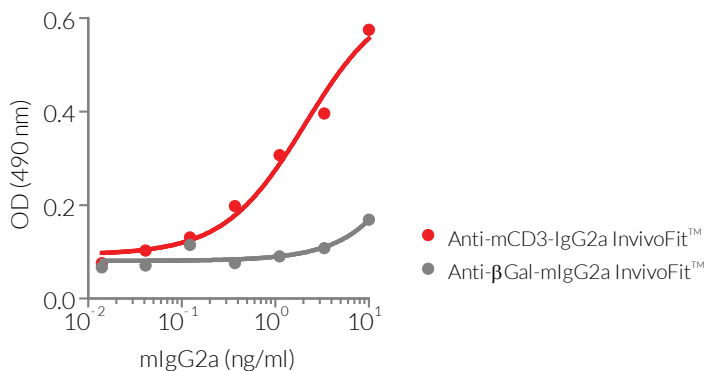


Figure 1: Binding of Anti-mCD3-mIgG2a InvivoFit™ mAb to coated mouse CD3ε protein.

Mouse CD3ε (10 µg/ml) was coated on ELISA plates overnight. A 3-fold serial dilution of the Anti-mCD3-mIgG2a InvivoFit™ mAb (red curve) or of the Anti-βGal mIgG2a control antibody (grey curve) was performed for the capture step. A HRP-labelled anti-IgG antibody (1/1000 dilution) and the HRP substrate OPD (o-phenylenediamine dihydrochloride) were used for the detection step. Absorbance was read at 490 nm.

## Validation of Anti-mCD3-mIgG2a InvivoFit™ binding by FACS

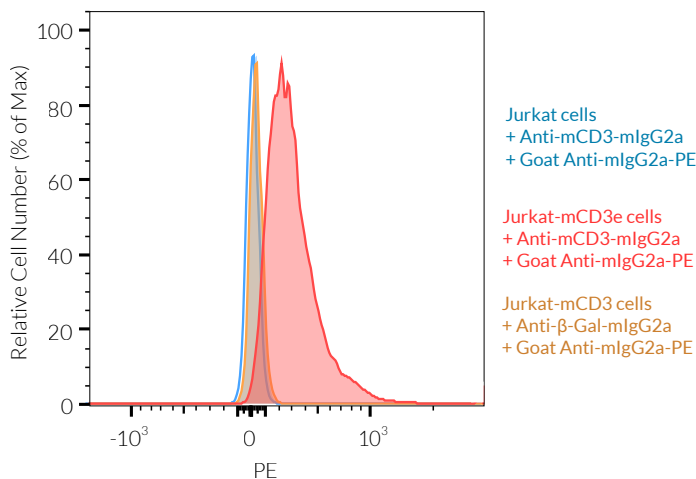


Figure 2: Cell surface staining of murine CD3 using Anti-mCD3-mIgG2a InvivoFit™ mAb.

~5x10<sup>5</sup> Jurkat (parental) or Jurkat-mCD3 cells were incubated with 2 µg of Anti-mCD3-mIgG2a InvivoFit™ mAb or an isotype control for 1h at 4°C. Cells were then washed and incubated with 0.25 µg of goat anti-mIgG2a coupled to PE for 1h at 4°C. Cell surface staining was analyzed by flow cytometry.

### TECHNICAL SUPPORT

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