



## BV421 Mouse Anti-Human CD3 (SP34-2)

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### Goal

To test the reactivity of BV421 Mouse Anti-Human CD3 (Catalog# 562877) from BD Biosciences on *Callithrix jacchus* (Common marmoset) peripheral blood mononuclear cells (PBMCs).

### Antibodies

	Primary Antibody	Isotype control
<b>Antibody name</b>	BV421 Mouse Anti-Human CD3	BV421 Mouse IgG1, k Isotype Control
<b>Alternate name</b>	CD3E; CD3-epsilon; T3E; TCRE	IgG1, kappa Isotype Control (Anti-KLH)
<b>Clone name</b>	SP34-2	X40
<b>Immunogen</b>	Purified Human CD3-epsilon Protein	Keyhole limpet hemocyanin (KLH)
<b>Isotype</b>	Mouse BALB/c IgG1	Mouse BALB/c IgG1
<b>Vendor</b>	BD Biosciences	BD Biosciences
<b>Catalog number</b>	562877	562438
<b>Lot number</b>	1025796	136398

### Experiment Information

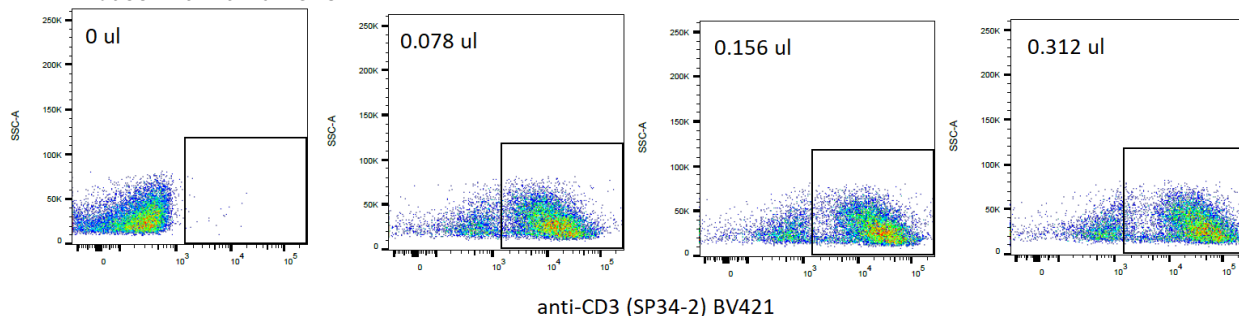
<b>Species</b>	<i>Callithrix jacchus</i>
<b>Common name</b>	Common marmoset
<b>Animal ID</b>	cj2060
<b>Animal facility</b>	MD Anderson
<b>Sample type</b>	PBMC
<b>Experiment type</b>	Flow cytometry/ Titration
<b>Number of cells/experiment</b>	100,000
<b>Volume/ experiment</b>	100 ul
<b>Date of Experiment</b>	May 06, 2021
<b>Experiment by</b>	Shubham Dutta

## Protocol

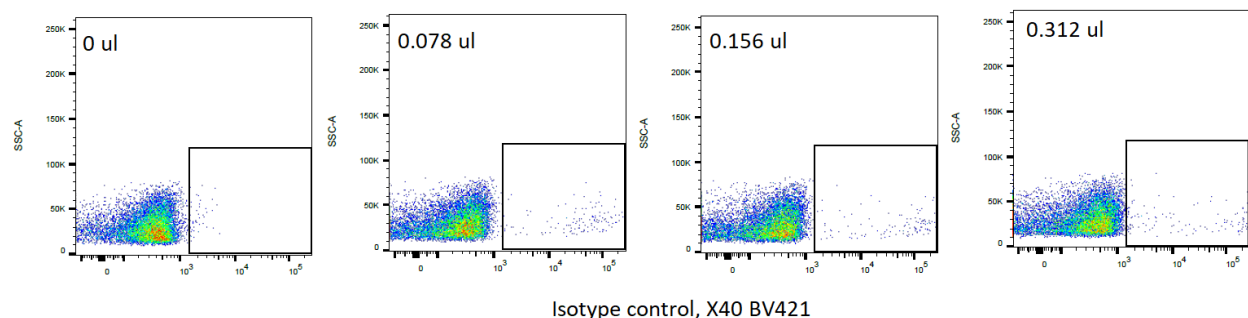
- Thaw PBMCs on ice, centrifuge at 500xg for 5 minutes.
- Add 1000 ul of ice cold Flow staining media (RPMI1640 w/ L-glutamine+2% FBS+1mM EDTA) and centrifuge at 500xg for 5 minutes.
- Add 1000 ul of ice cold Flow staining media and pass through nylon mesh.
- Count cells using AOPI and adjust volume such that there are 100,000 cells/ 100 ul of ice cold Flow staining media.
- Block with BioRad human blocking buffer for 10 minutes on ice according to manufacturer's protocol (5 ul per 100 ul of cells).
- Add primary antibodies and incubate for 30 minutes on ice in the dark.
- Centrifuge at 500g for 5 minutes to remove primary antibody and wash cells with Flow staining media.
- Add 100 ul of 1% PFA in PBS for 30 minutes on ice to fix the cells.
- Centrifuge at 500g for 5 minutes to remove 1% PFA solution.
- Add 100 ul of PBS and store in dark at 4C until acquisition.

## Results

### BV421 Mouse Anti-Human CD3



### BV421 Mouse IgG1, k Isotype Control



## Conclusion

- BV421 Mouse Anti-Human CD3 (SP34-2) stains on Marmoset (cj2060) PBMCs.