# ACTA (Anybody Can Titrate Antibody) Report v3.6.11

2026-11-12

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# A. MiFlowCyt metadata

## 1 Experiment Overview

**MiFlowCyt 1.0.** Structured to MiFlowCyt but not self-contained: purpose, experiment variables, organization, conclusions, specimen source and collection, and instrument QC are recorded in the ELN and referenced here. Not a compliant submission document on its own.

| Category | Value |
|---|---|
| Purpose | See ELN EXP1460 (ELN) |
| Keywords | (not supplied) |
| Experiment variables | Stain volume (uL) per antibody; see Reagents. See ELN EXP1460 (ELN) |
| Organization | See ELN EXP1460 (ELN) |
| Primary contact | JD |
| Acquisition date | 2026-10-20 |
| Conclusions | See ELN EXP1460 (ELN) |
| Quality control | 6 automated check(s) per antibody group; see the QC section. |

# 2 Flow Sample/Specimen Details

| Category | Value |
|---|---|
| Test material | HD Human PBMC |
| Cells per well (e6) | 1 |
| Well volume (uL) | 100 |
| Source | See ELN EXP1460 (ELN) |
| Collection | See ELN EXP1460 (ELN) |
| SOP | (not supplied) |
| Fixed | TRUE |
| Permeabilised | TRUE |
| Protocol | See ELN EXP1460 (ELN) |

## 2.4 Fluorescence Reagent Description

**CD19**

| Channel | Markername |
|---|---|
| SSC-H | — |
| SSC-A | — |
| FSC-H | — |
| FSC-A | — |
| SSC-B-H | — |
| SSC-B-A | — |
| BUV496-A | CD4 |
| BUV615-A | CD14_CD16 |
| BUV805-A | CD3 |
| Alexa Fluor 532-A | CD8 |
| Alexa Fluor 660-A | CD19 |
| eFluor 780-A | Viability |
| AF-A | Autofluorescence |

**CD27**

| Channel | Markername |
|---|---|
| SSC-H | — |
| SSC-A | — |
| FSC-H | — |
| FSC-A | — |
| SSC-B-H | — |
| SSC-B-A | — |
| BUV395-A | CD27 |
| BUV496-A | CD4 |
| BUV615-A | CD14_CD16 |
| BUV805-A | CD3 |
| Alexa Fluor 532-A | CD8 |
| eFluor 780-A | Viability |
| AF-A | Autofluorescence |

**CD127**

| Channel | Markername |
|---|---|
| SSC-H | — |
| SSC-A | — |
| FSC-H | — |
| FSC-A | — |
| SSC-B-H | — |
| SSC-B-A | — |
| BUV496-A | CD4 |
| BUV615-A | CD14_CD16 |
| BUV805-A | CD3 |
| Alexa Fluor 532-A | CD8 |
| Alexa Fluor 700-A | CD127 |
| eFluor 780-A | Viability |
| AF-A | Autofluorescence |

**CD45RA**

| Channel | Markername |
|---|---|
| SSC-H | — |
| SSC-A | — |
| FSC-H | — |
| FSC-A | — |
| SSC-B-H | — |
| SSC-B-A | — |
| BUV496-A | CD4 |
| BUV615-A | CD14_CD16 |
| BUV805-A | CD3 |
| BV650-A | CD45RA |
| Alexa Fluor 532-A | CD8 |
| eFluor 780-A | Viability |
| AF-A | Autofluorescence |

**CD45RO**

| Channel | Markername |
|---|---|
| SSC-H | — |
| SSC-A | — |
| FSC-H | — |
| FSC-A | — |
| SSC-B-H | — |
| SSC-B-A | — |
| BUV496-A | CD4 |
| BUV615-A | CD14_CD16 |
| BUV805-A | CD3 |
| BV480-A | CD45RO |
| Alexa Fluor 532-A | CD8 |
| eFluor 780-A | Viability |
| AF-A | Autofluorescence |

**CD62L**

| Channel | Markername |
|---|---|
| SSC-H | — |
| SSC-A | — |
| FSC-H | — |
| FSC-A | — |
| SSC-B-H | — |
| SSC-B-A | — |
| BUV496-A | CD4 |
| BUV615-A | CD14_CD16 |
| BUV805-A | CD3 |
| BV421-A | CD62L |
| Alexa Fluor 532-A | CD8 |
| eFluor 780-A | Viability |
| AF-A | Autofluorescence |

Table 1: Costain panel 'Iteration1' from 'Master_Panel_Inventory.xlsx': 6 of 6 reagents matched.

| Antigen | Fluorochrome | Matched to | Verdict | Catalogue | Vendor | Clone | Type |
|---|---|---|---|---|---|---|---|
| CD14 | BUV615 | BUV615 | matched | 366-0149-42 | Invitrogen | 61D3 | Base |
| CD16 | BUV615 | CD14_CD16 | matched | 751572 | BD OptiBuild | 3G8 | Base |
| CD3 | BUV805 | CD3 | matched | 612893 | BD | SK7 | Base |
| CD4 | BUV496 | CD4 | matched | 612936 | BD | SK3 | Base |
| CD8 | Alexa Fluor 532 | CD8 | matched | 58-0088-42 | Thermo | RPA-T8 | Base |
| Live Dead eFluor 780 (LD) (Viability) | efluor780 | Viability | matched | 65-0865-14 | TS | NA | Base |

# 3 Instrument Details

| Category | Value |
|---|---|
| Manufacturer and model | Aurora |
| Detectors with a recorded gain | 6 |
| Instrument QC | See ELN EXP1460 (ELN) |

### Detector voltages ($PnV)

| detector | marker | voltage |
|---|---|---|
| SSC-H | NA | 125 |
| SSC-A | NA | 125 |
| FSC-H | NA | 125 |
| FSC-A | NA | 125 |
| SSC-B-H | NA | 131 |
| SSC-B-A | NA | 131 |

# 4 Data Analysis Details

Everything below is MiFlowCyt part 4 – what was done to the events between acquisition and the numbers in this report, in the order it happened: events read, compensation, transformation, gating, and the software that did it.

## 4.1 List-mode data files: downsampling

**Info!Downsample_to:** *(blank)*

No downsampling performed.

Events read: 24,354,336 across 66 file(s) (smallest 284,411, largest 473,536).

## 4.2 Compensation Description

**Info!compensation:** *(blank)*

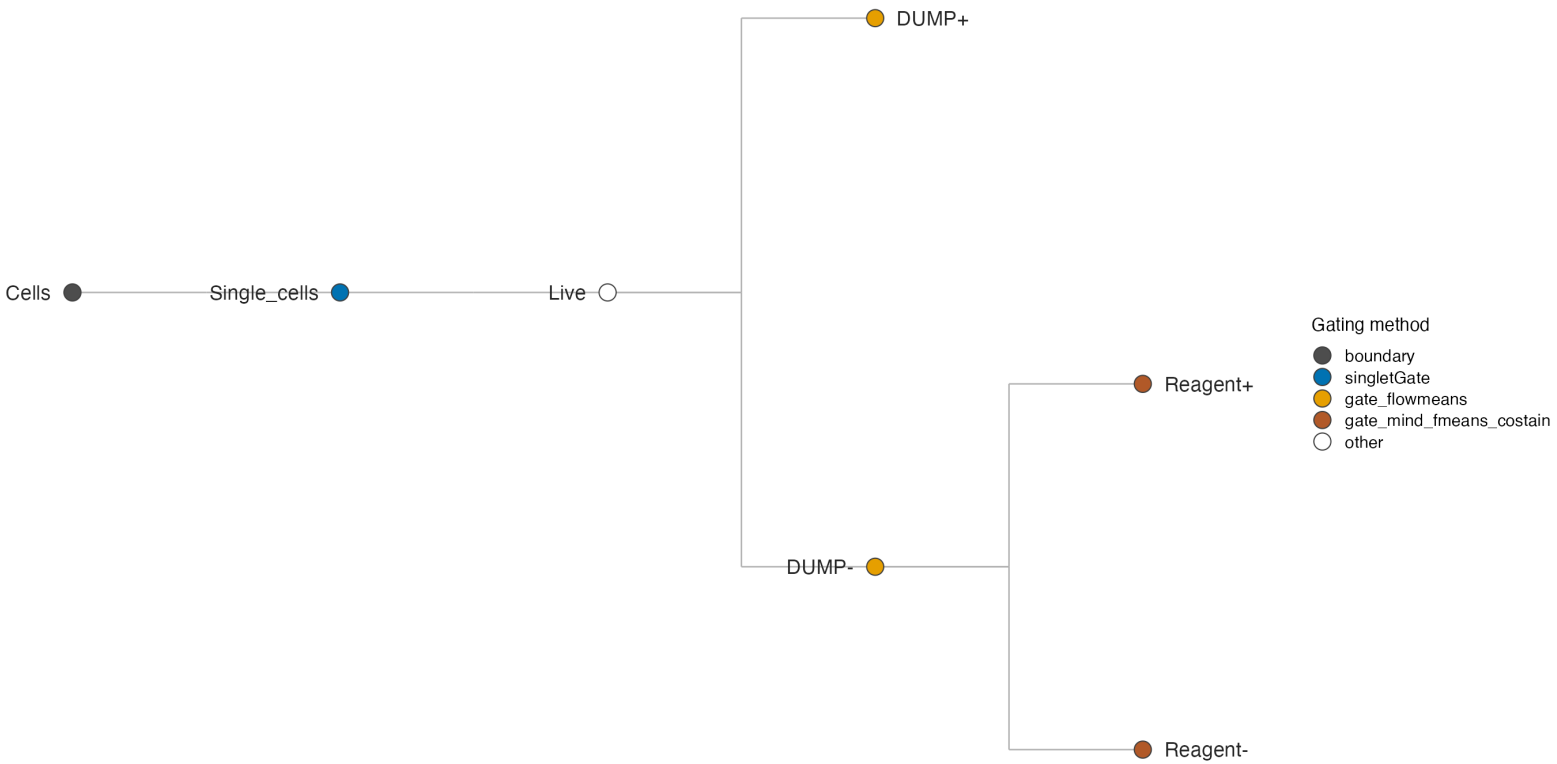
Identity matrix was applied – the data were not compensated.

## 4.3 Data Transformation Details

| Argument | CD19 | CD27 | CD127 | CD45RA | CD45RO | CD62L |
|---|---|---|---|---|---|---|
| maxValue (from $PnR) | 4194304 | 4194304 | 4194304 | 4194304 | 4194304 | 4194304 |
| channelRange | 9000 | 9000 | 9000 | 9000 | 9000 | 9000 |
| pos | 4.5 | 4.5 | 4.5 | 4.5 | 4.5 | 4.5 |
| neg | 0 | 0 | 0 | 0 | 0 | 0 |
| widthBasis | -10 | -10 | -10 | -10 | -10 | -10 |

## 4.4 Gating Details

### Gating scheme



### 4.4.2 Gate Statistics

Count, percent of parent, median fluorescence intensity and robust SD were collected for 120 population/well combinations and written to the **Pop_stats** sheet of the titration export beside this report.

In `gating_template`, mark `stats_export` as TRUE to make sure that population's stats are exported.

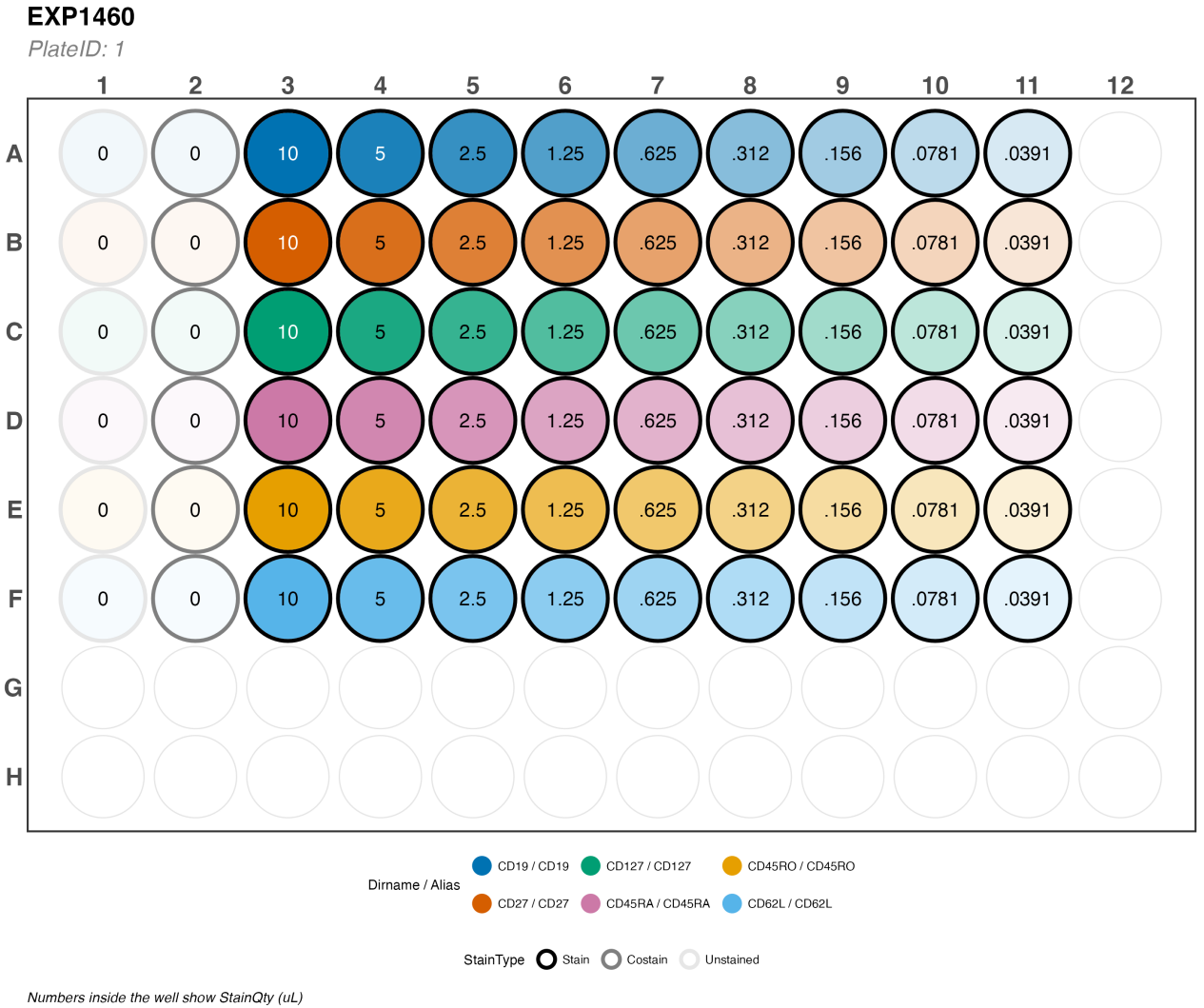
## 4.5 Analysis software

| Category | Value |
|---|---|
| ACTA version | 3.6.11 |
| R version | R version 4.5.1 (2025-06-13) |

This pipeline can be driven from R without the app: see **Automating ACTA** in `README.md` in the script folder.

# B. Titration results

## 1 Plate map



# 2 QC

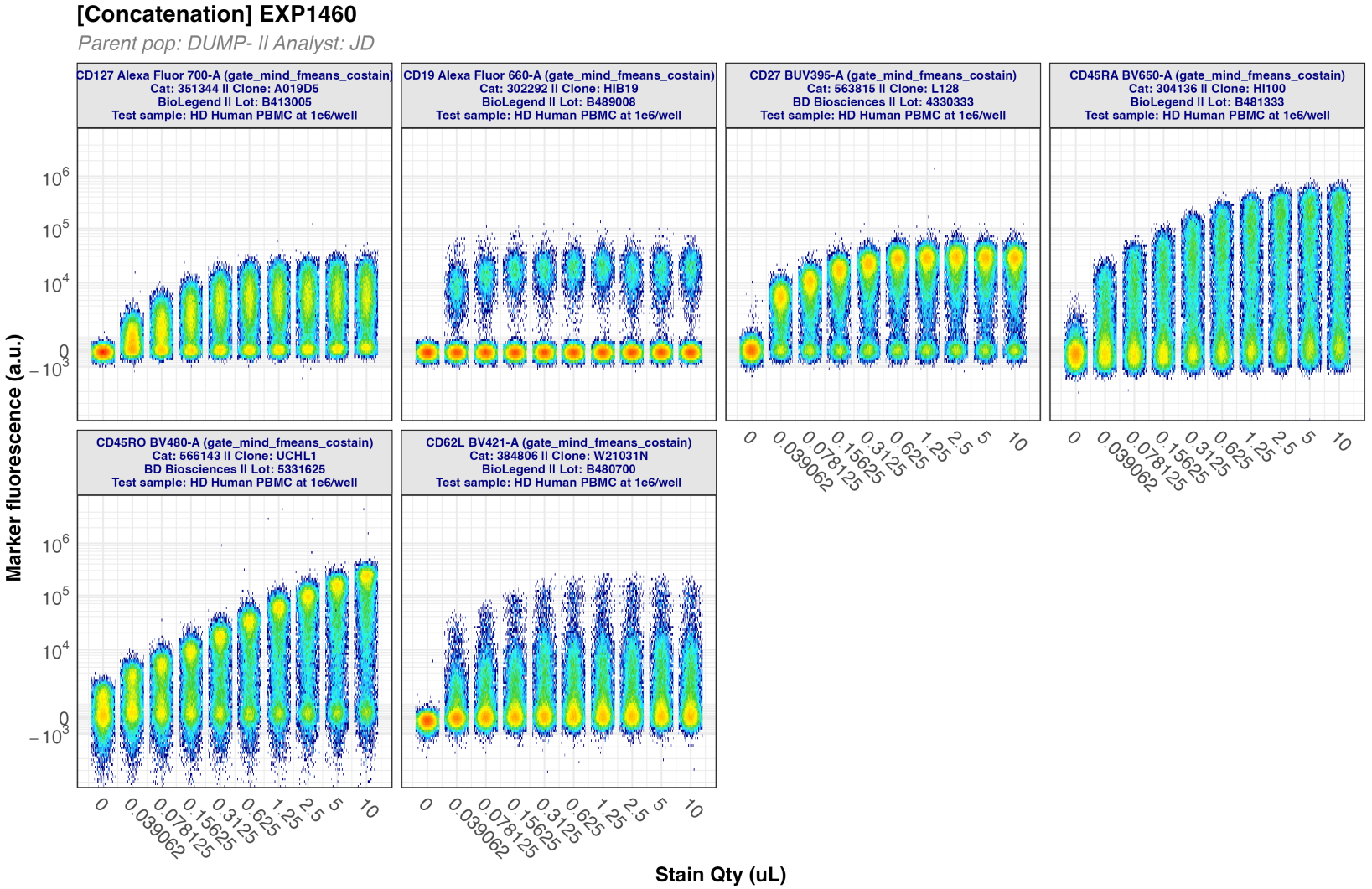
*Groups 1-4 of 6*

| Check | CD19 | CD27 | CD127 | CD45RA |
|---|---|---|---|---|
| Median FSC-A of Cells is at least 50% of the median of all events | **PASS** | **PASS** | **PASS** | **PASS** |
| More than 50% Live cells | **PASS** | **PASS** | **PASS** | **PASS** |
| More than 80% single cells | **PASS** | **PASS** | **PASS** | **PASS** |
| Costain SI is the lowest in the group | **PASS** | **PASS** | **PASS** | **PASS** |
| Co-stain is less than 5% positive | **PASS** | **PASS** | **PASS** | **PASS** |
| Every population has at least 100 events in every well (except costain) | **PASS** | **PASS** | **PASS** | **PASS** |
| **Overall** | **PASS** | **PASS** | **PASS** | **PASS** |

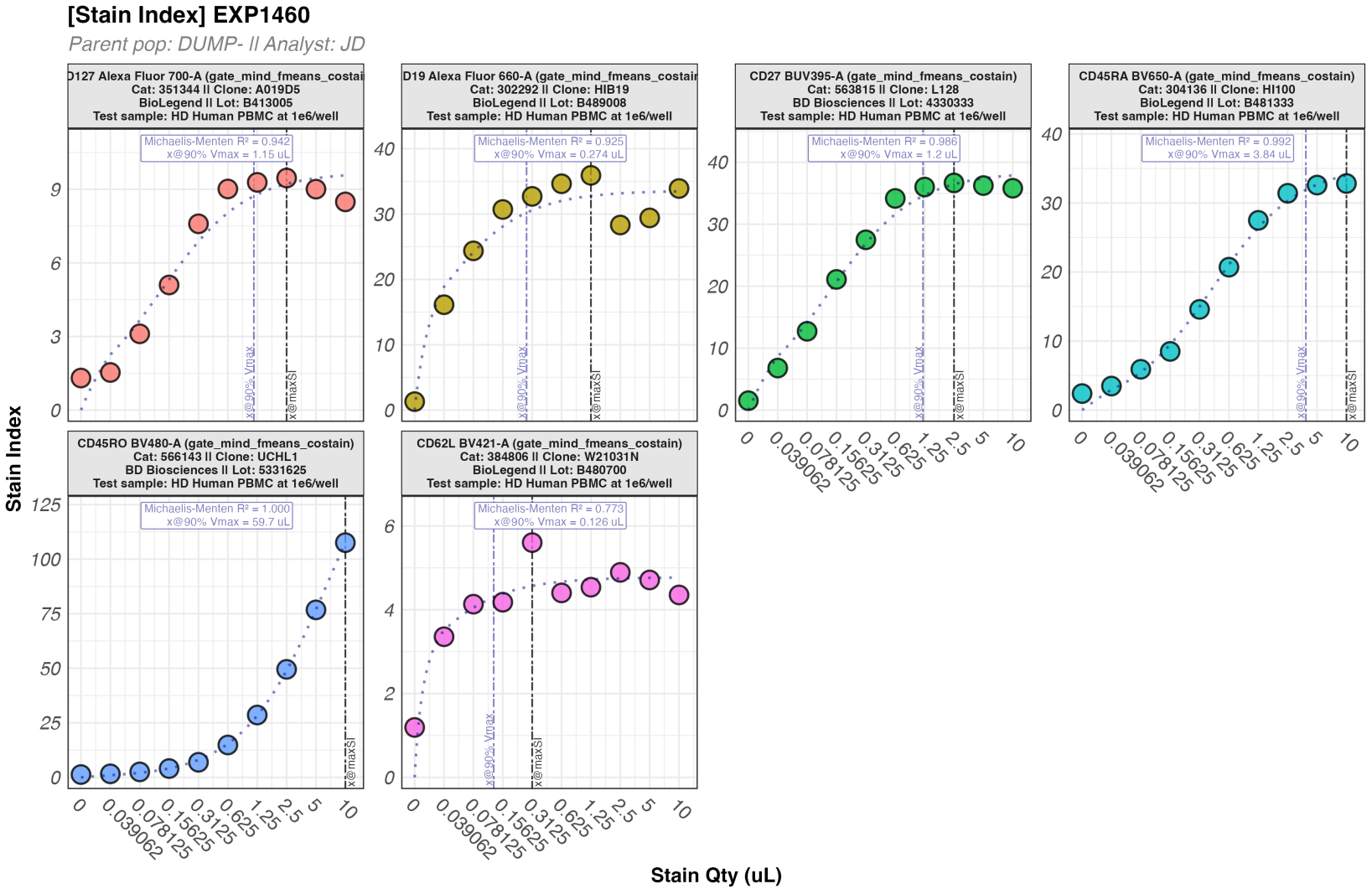
*Groups 5-6 of 6*

| Check | CD45RO | CD62L |
|---|---|---|
| Median FSC-A of Cells is at least 50% of the median of all events | **PASS** | **PASS** |
| More than 50% Live cells | **PASS** | **PASS** |
| More than 80% single cells | **PASS** | **PASS** |
| Costain SI is the lowest in the group | **PASS** | **PASS** |
| Co-stain is less than 5% positive | **PASS** | **PASS** |
| Every population has at least 100 events in every well (except costain) | **PASS** | **PASS** |
| **Overall** | **PASS** | **PASS** |

# 3 Resolution of populations

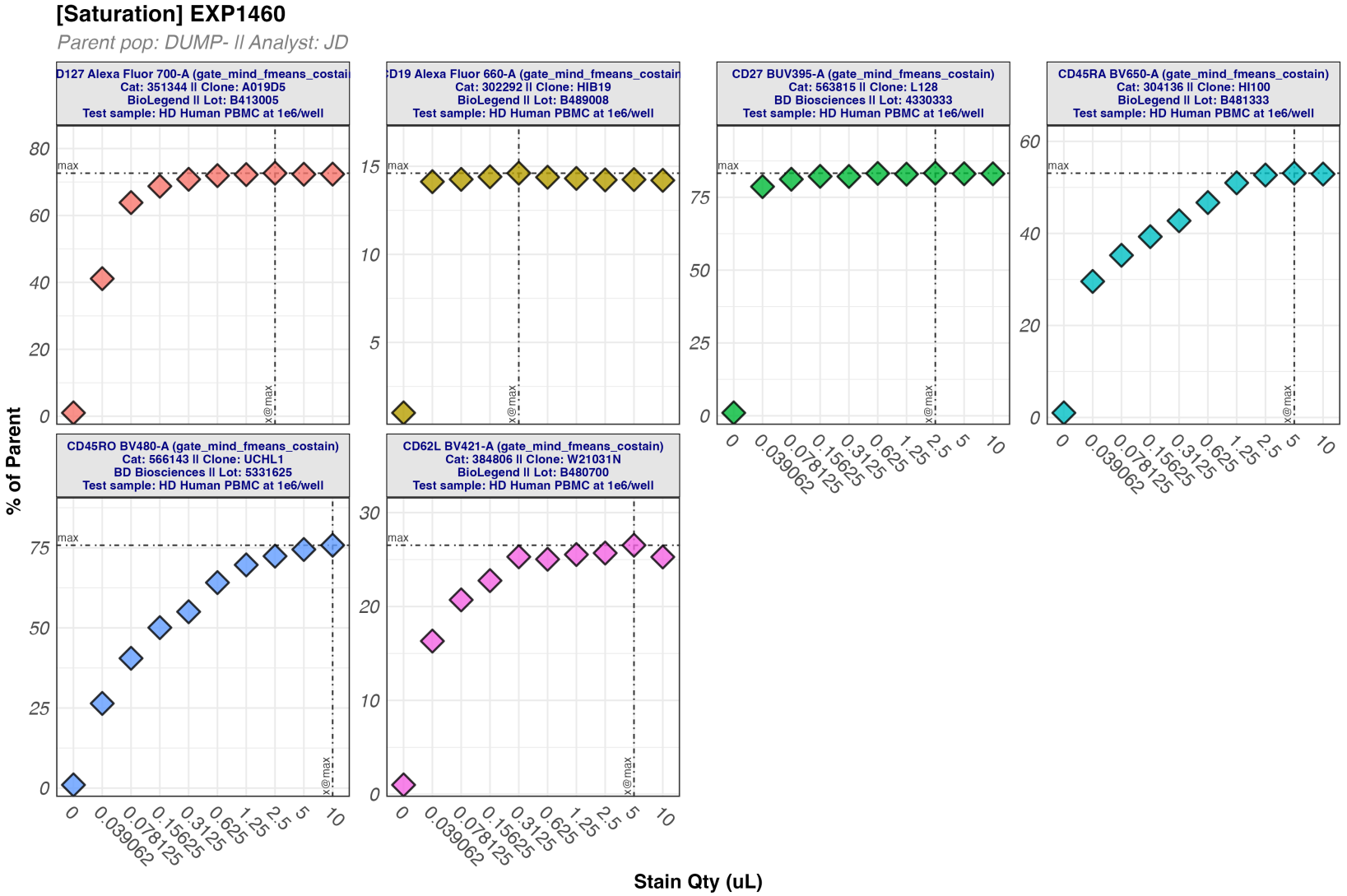



*Concatenated events, up to 20,000 per FCS.*



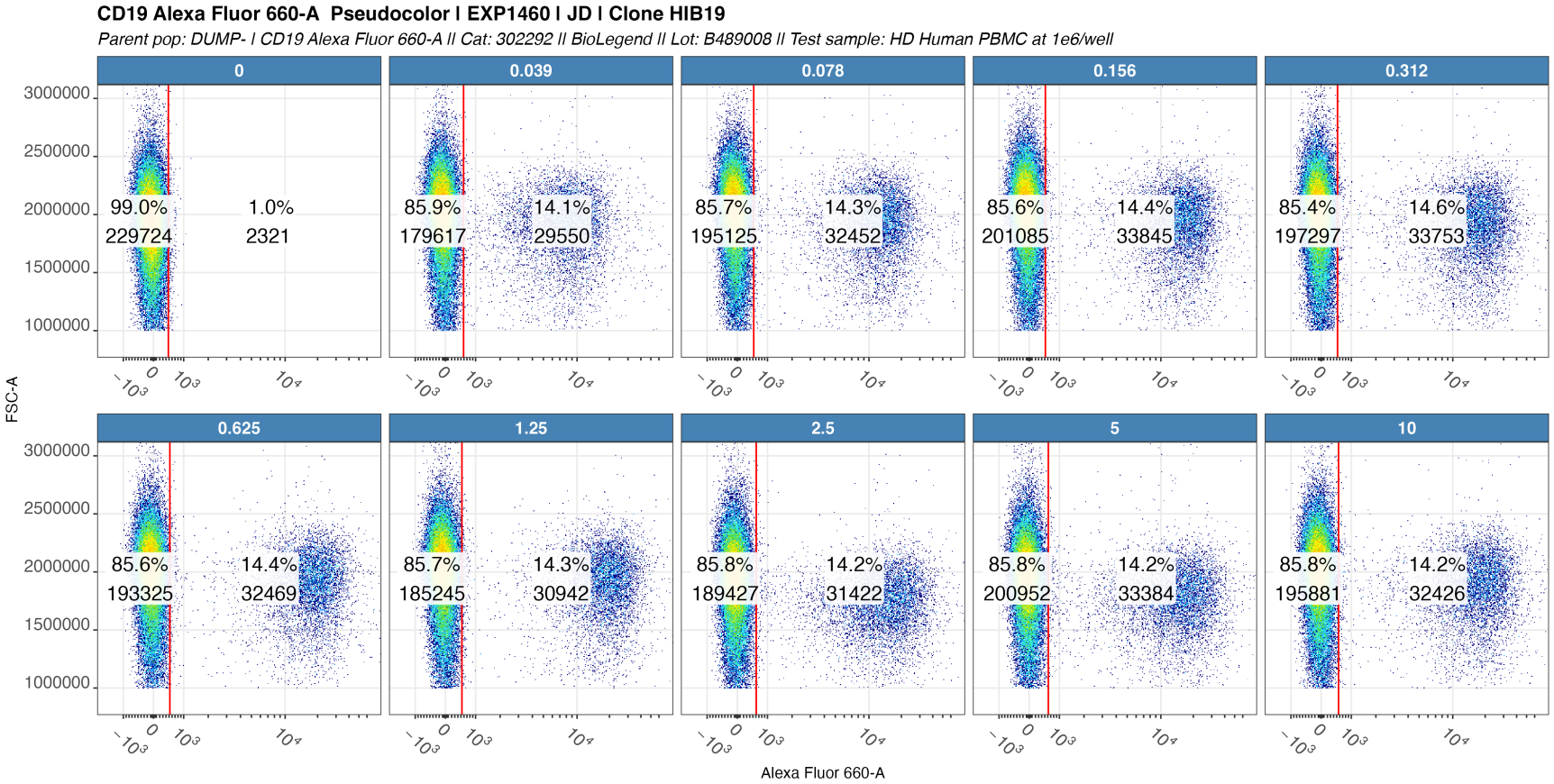

*MFI calculated on untransformed (linear) values. // Compensation: —*

# 4 Saturation of positive population proportion



# 5 Layouts for each antibody

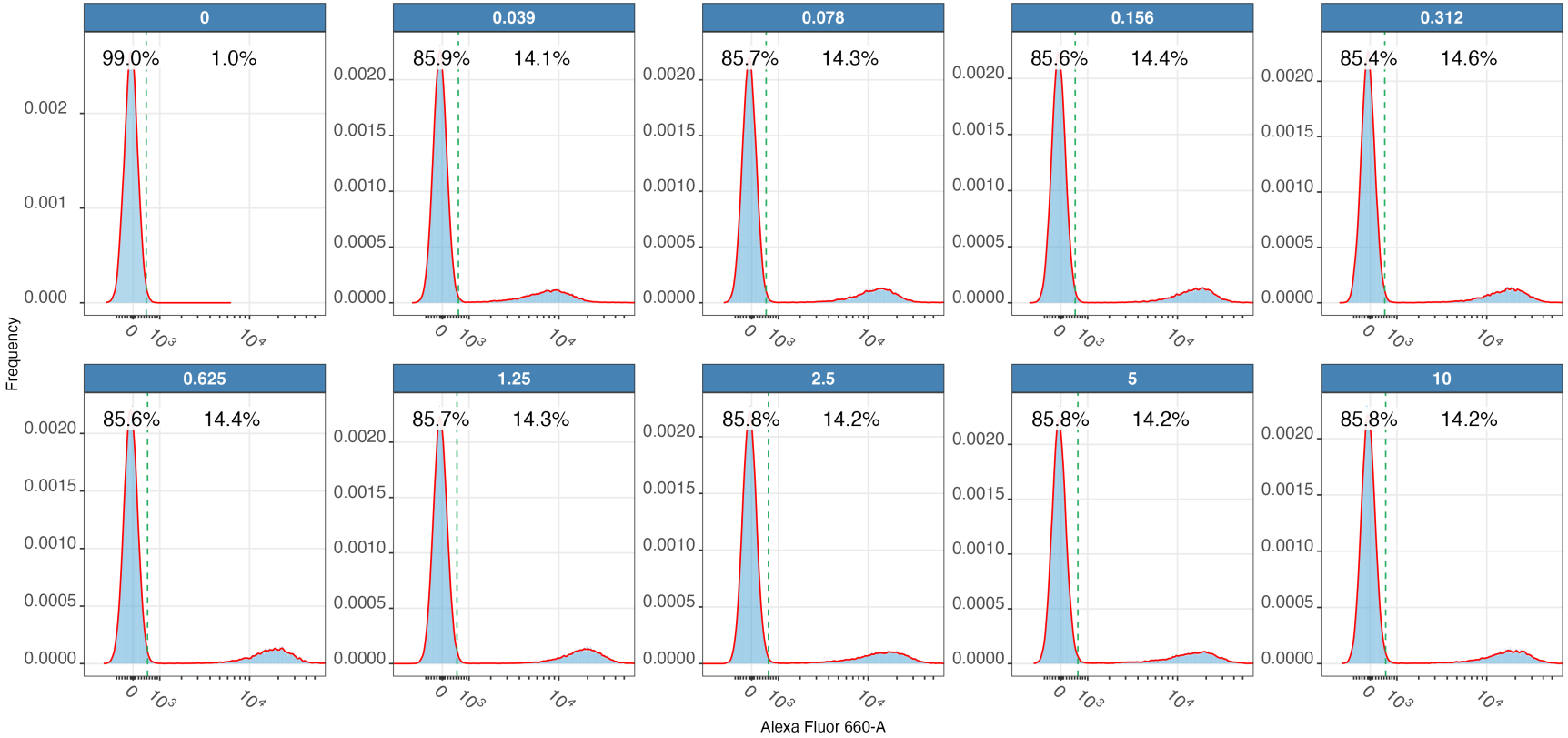
### 5.1 CD19




*Gating method: gate_mind_fmeans_costain || args: K=2, cluster='neg', quantile=0.99, gate_range=NULL, adjust=2 || collapse: FALSE || groupBy: Dirname || pp: pp_costain_floor || pp args: costain_quantile=0.99*
*Compensation: —*

**CD19 Alexa Fluor 660-A Histogram | EXP1460 | JD | Clone HIB19**

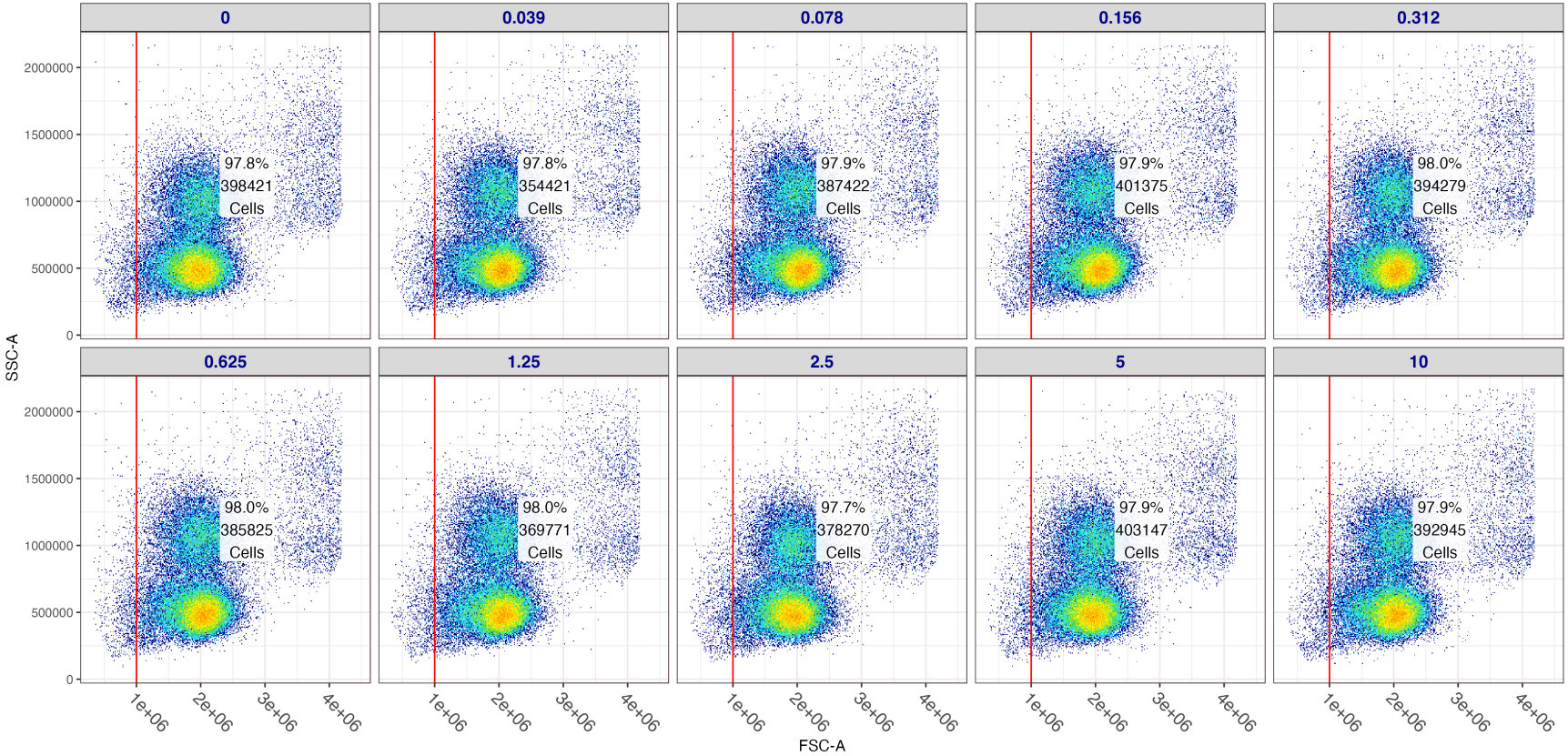
*Parent pop: DUMP- || CD19 Alexa Fluor 660-A || Cat: 302292 || BioLegend || Lot: B489008 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: gate_mind_fmeans_costain || args: K=2, cluster='neg', quantile=0.99, gate_range=NULL, adjust=2 || collapse: FALSE || groupBy: Dirname || pp: pp_costain_floor || pp args: costain_quantile=0.99*
*Compensation: —*

**CD19 Alexa Fluor 660-A : Cells | EXP1460 | JD | Clone HIB19**

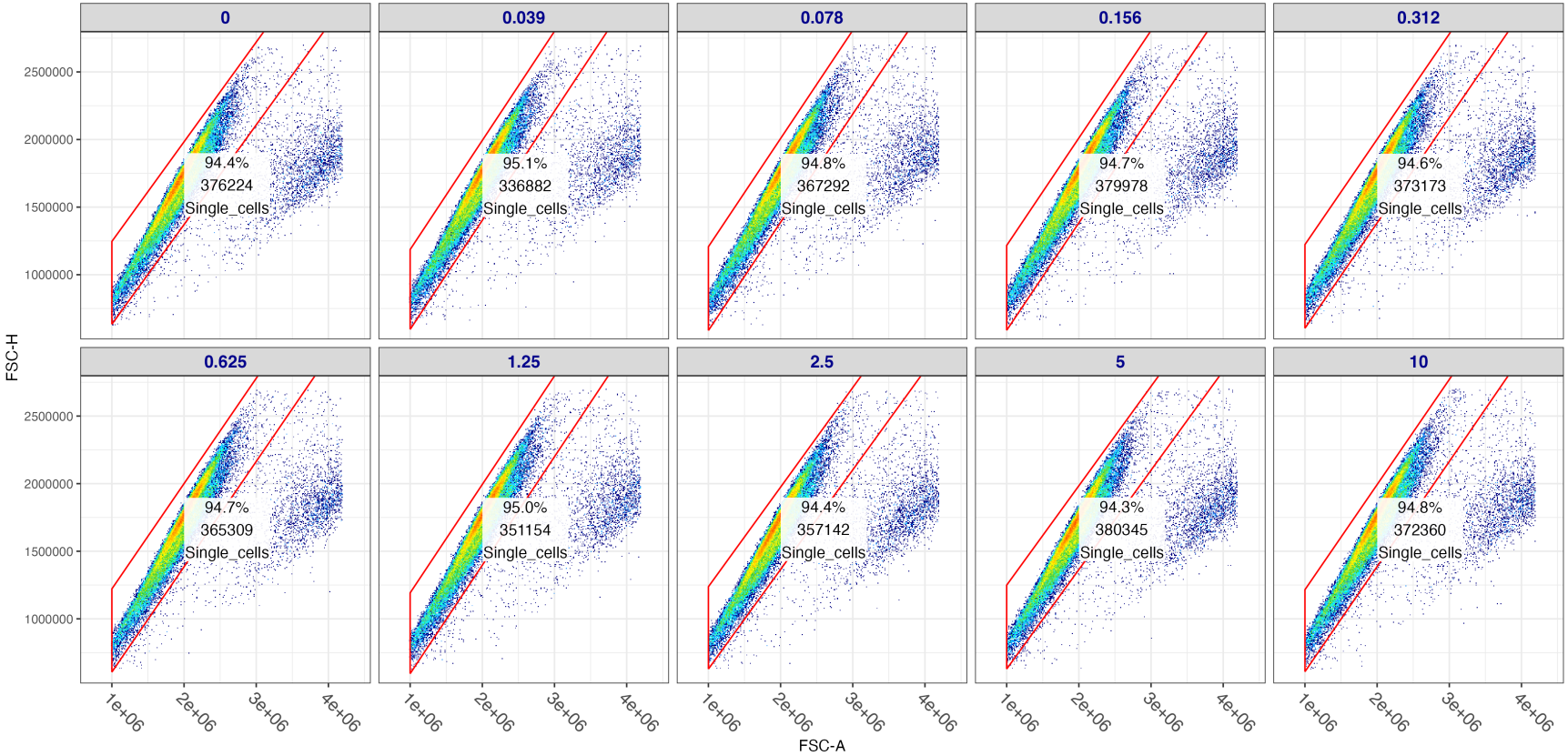
*Parent pop: root | CD19 Alexa Fluor 660-A || Cat: 302292 || BioLegend || Lot: B489008 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: boundary || args: min=c(1e6,-Inf), max=c(Inf,Inf) || collapse: TRUE || groupBy: Dirname || pp: — || pp args: —*
*Compensation: —*

**CD19 Alexa Fluor 660-A : Single_cells | EXP1460 | JD | Clone HIB19**

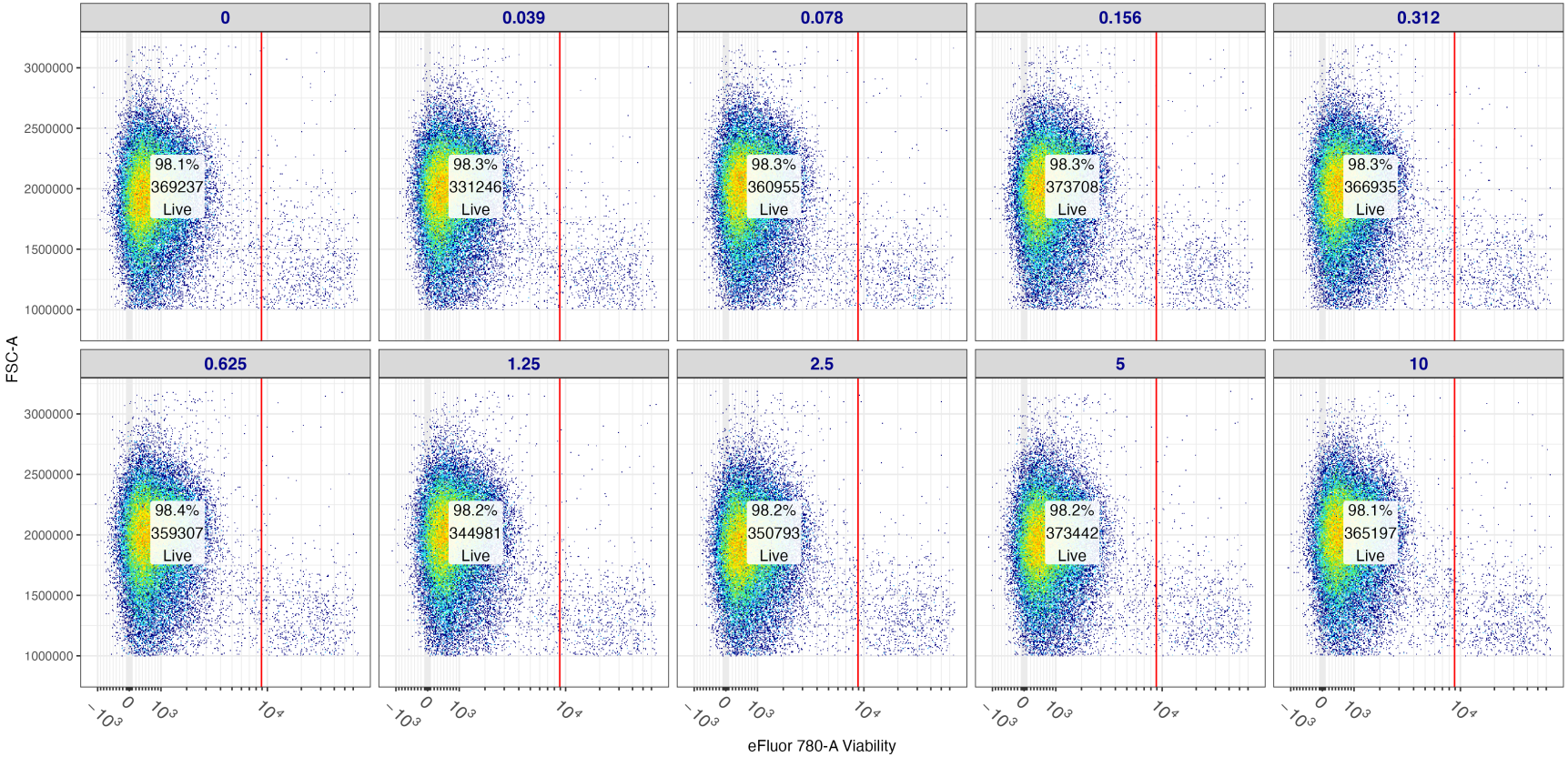
*Parent pop: Cells | CD19 Alexa Fluor 660-A || Cat: 302292 || BioLegend || Lot: B489008 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: singletGate || args: prediction_level=0.99, wider_gate=FALSE || collapse: FALSE || groupBy: Dirname || pp: — || pp args: —*
*Compensation: —*

**CD19 Alexa Fluor 660-A : Live | EXP1460 | JD | Clone HIB19**

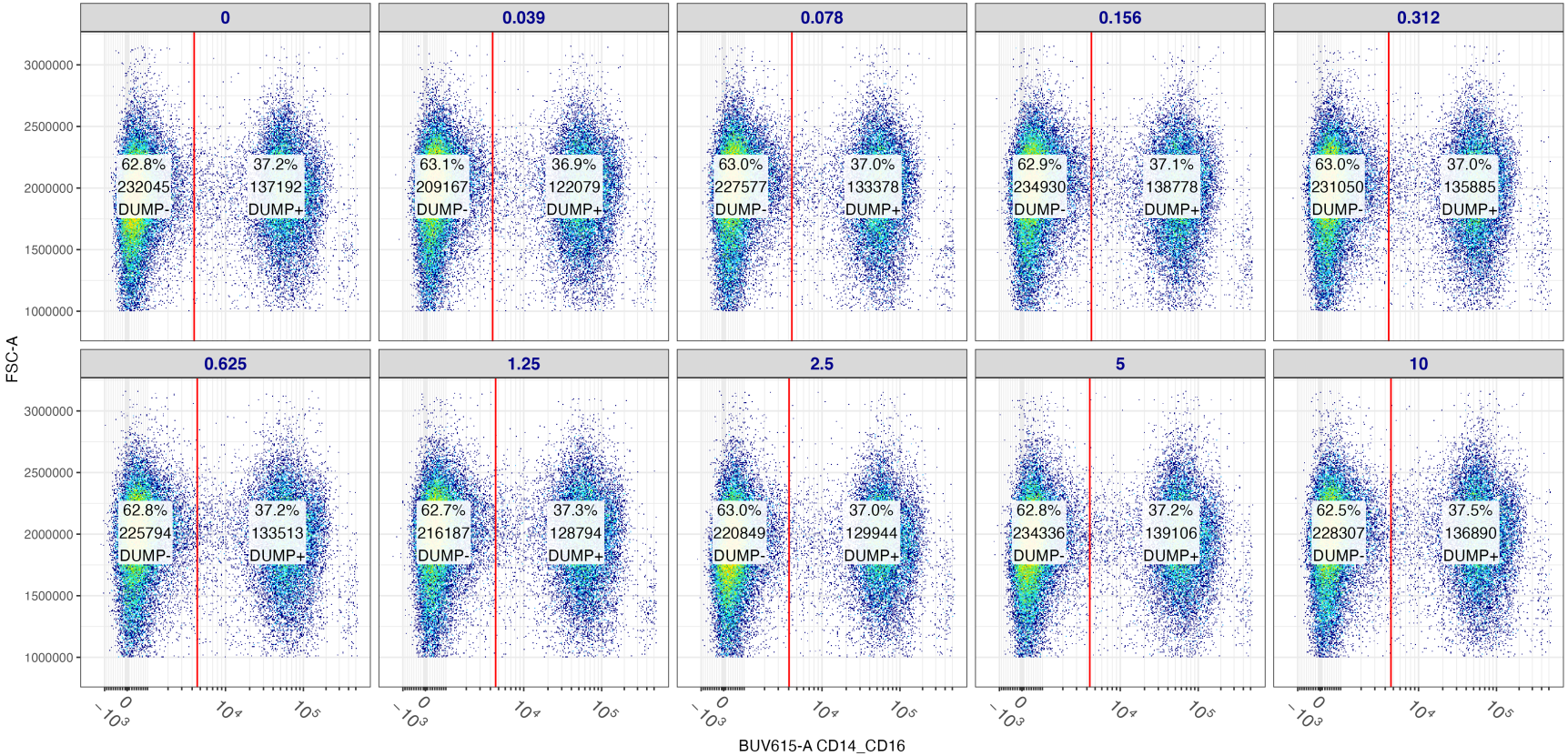
*Parent pop: Single_cells | CD19 Alexa Fluor 660-A || Cat: 302292 || BioLegend || Lot: B489008 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: gate_mindensity || args: gate_range=NULL, adjust=1, min=0 || collapse: TRUE || groupBy: Dirname || pp: — || pp args: — Compensation: —*

**CD19 Alexa Fluor 660-A : DUMP | EXP1460 | JD | Clone HIB19**

*Parent pop: Live | CD19 Alexa Fluor 660-A || Cat: 302292 || BioLegend || Lot: B489008 || Test sample: HD Human PBMC at 1e6/well*

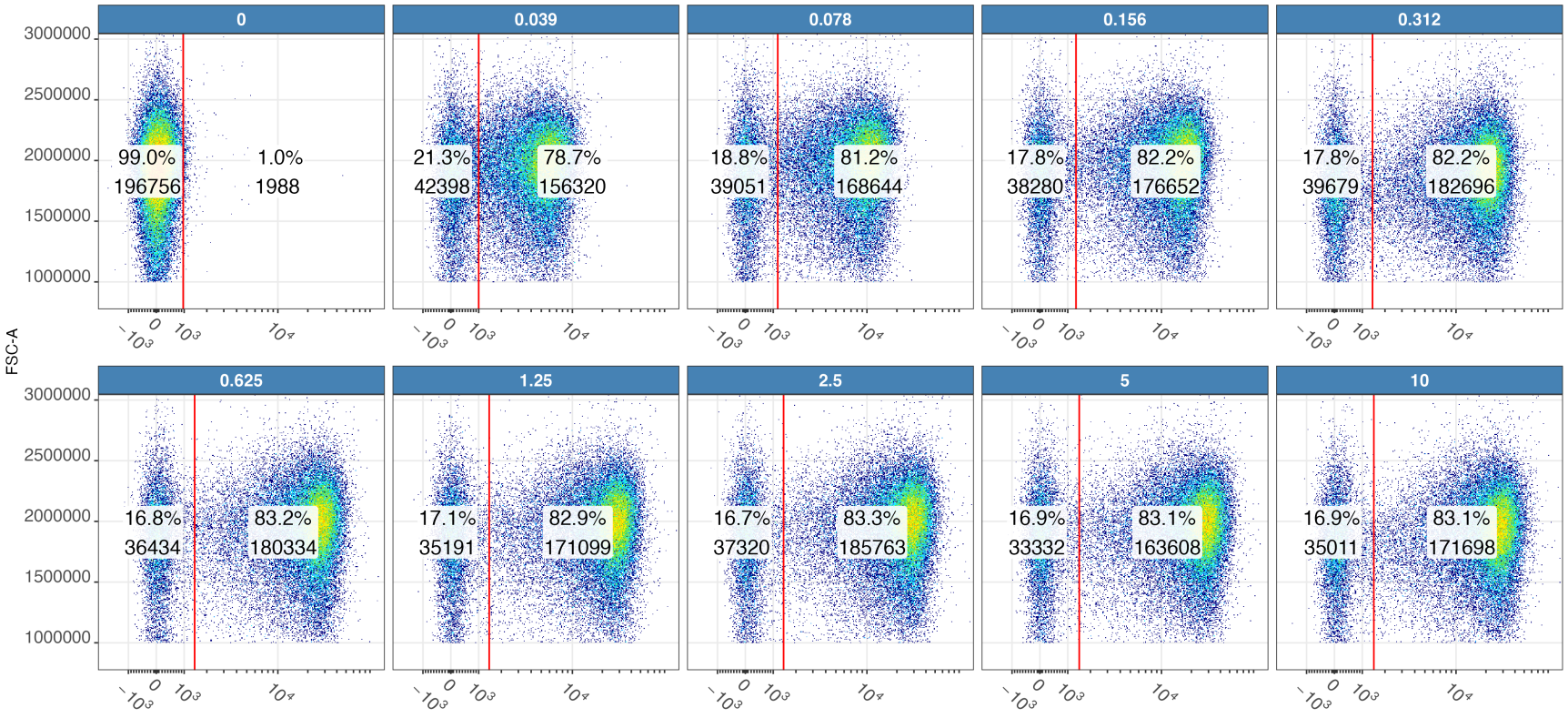



*Gating method: gate_flowmeans || args: K=2, cluster='neg', quantile=0.99 || collapse: FALSE || groupBy: Dirname || pp: — || pp args: —*
*Compensation: —*

### 5.2 CD27

**CD27 BUV395-A Pseudocolor | EXP1460 | JD | Clone L128**

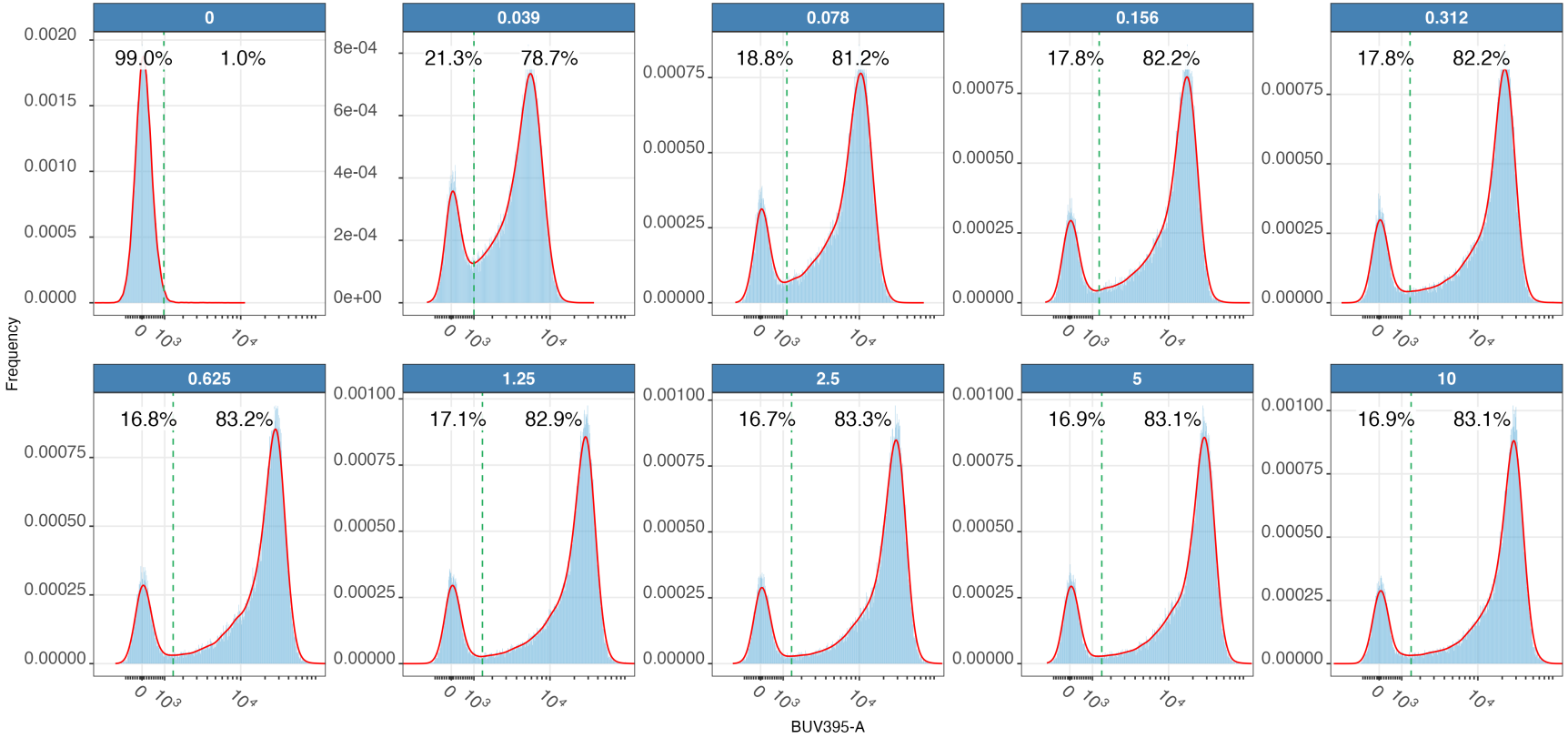
*Parent pop: DUMP- | CD27 BUV395-A || Cat: 563815 || BD Biosciences || Lot: 4330333 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: gate_mind_fmeans_costain || args: K=2, cluster='neg', quantile=0.99, gate_range=NULL, adjust=2 || collapse: FALSE || groupBy: Dirname || pp: pp_costain_floor || pp args: costain_quantile=0.99*
*Compensation: —*

**CD27 BUV395-A Histogram | EXP1460 | JD | Clone L128**

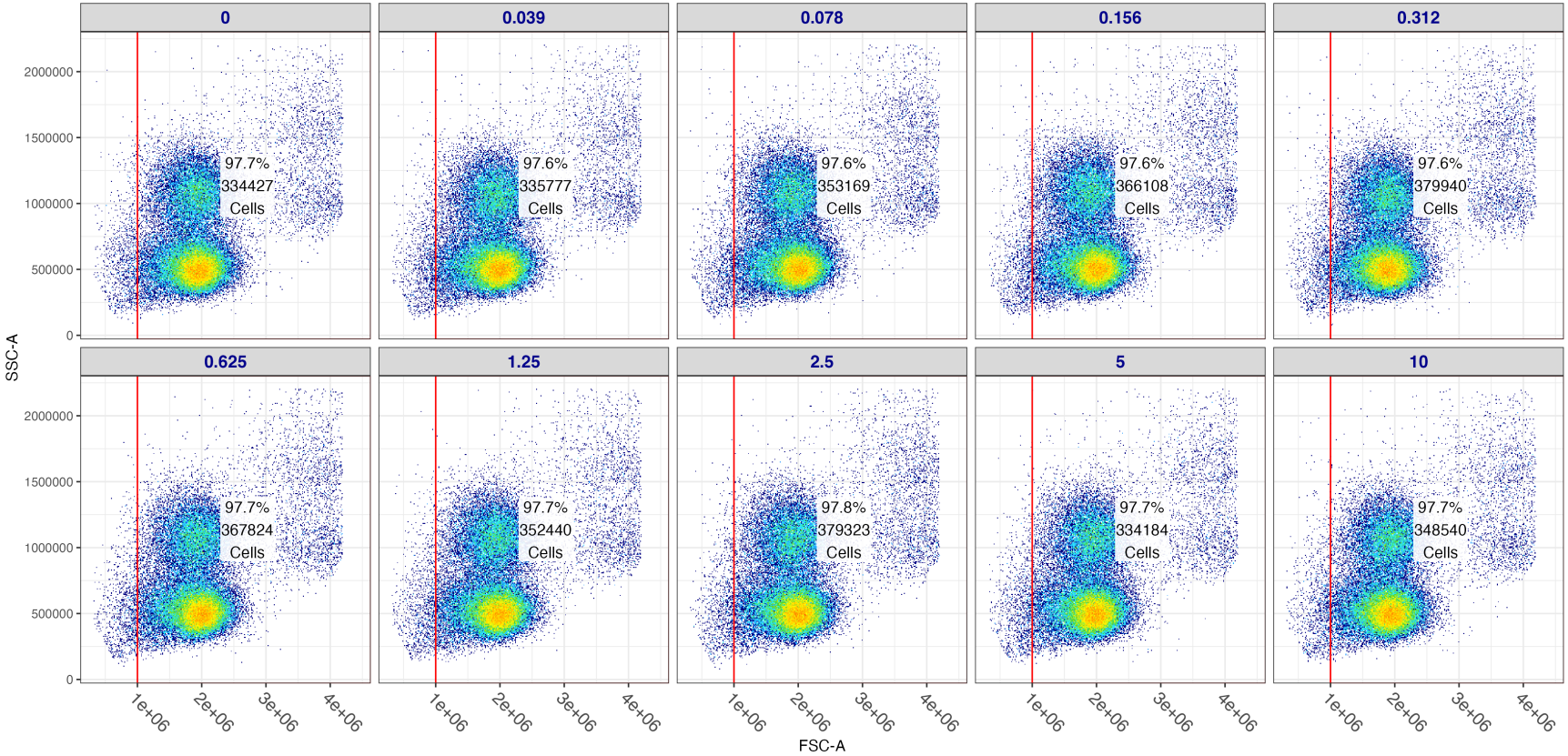
*Parent pop: DUMP- || CD27 BUV395-A || Cat: 563815 || BD Biosciences || Lot: 4330333 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: gate_mind_fmeans_costain || args: K=2, cluster='neg', quantile=0.99, gate_range=NULL, adjust=2 || collapse: FALSE || groupBy: Dirname || pp: pp_costain_floor || pp args: costain_quantile=0.99*
*Compensation: —*

**CD27 BUV395-A : Cells | EXP1460 | JD | Clone L128**

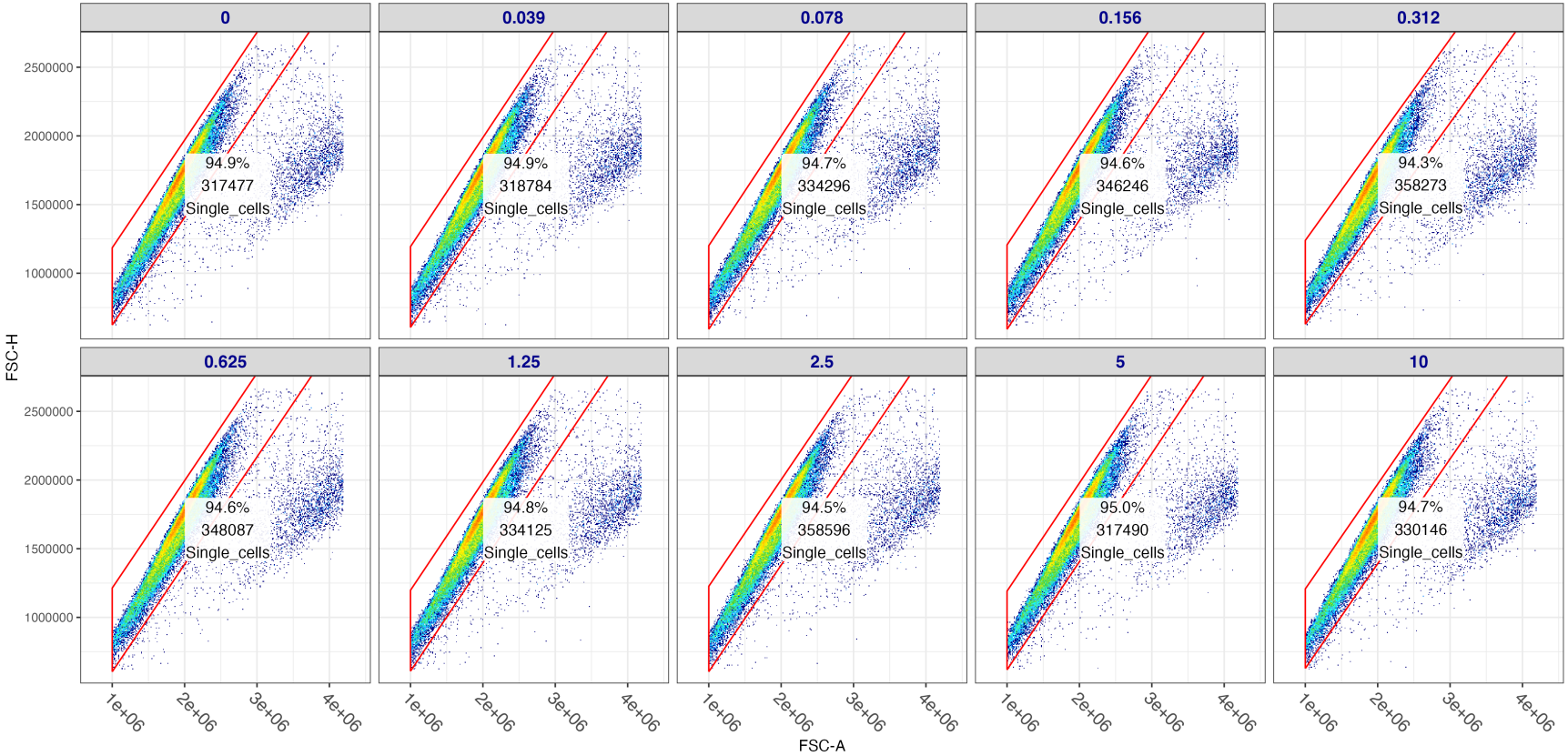
*Parent pop: root | CD27 BUV395-A || Cat: 563815 || BD Biosciences || Lot: 4330333 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: boundary || args: min=c(1e6,-Inf), max=c(Inf,Inf) || collapse: TRUE || groupBy: Dirname || pp: — || pp args: —*
*Compensation: —*

**CD27 BUV395-A : Single_cells | EXP1460 | JD | Clone L128**

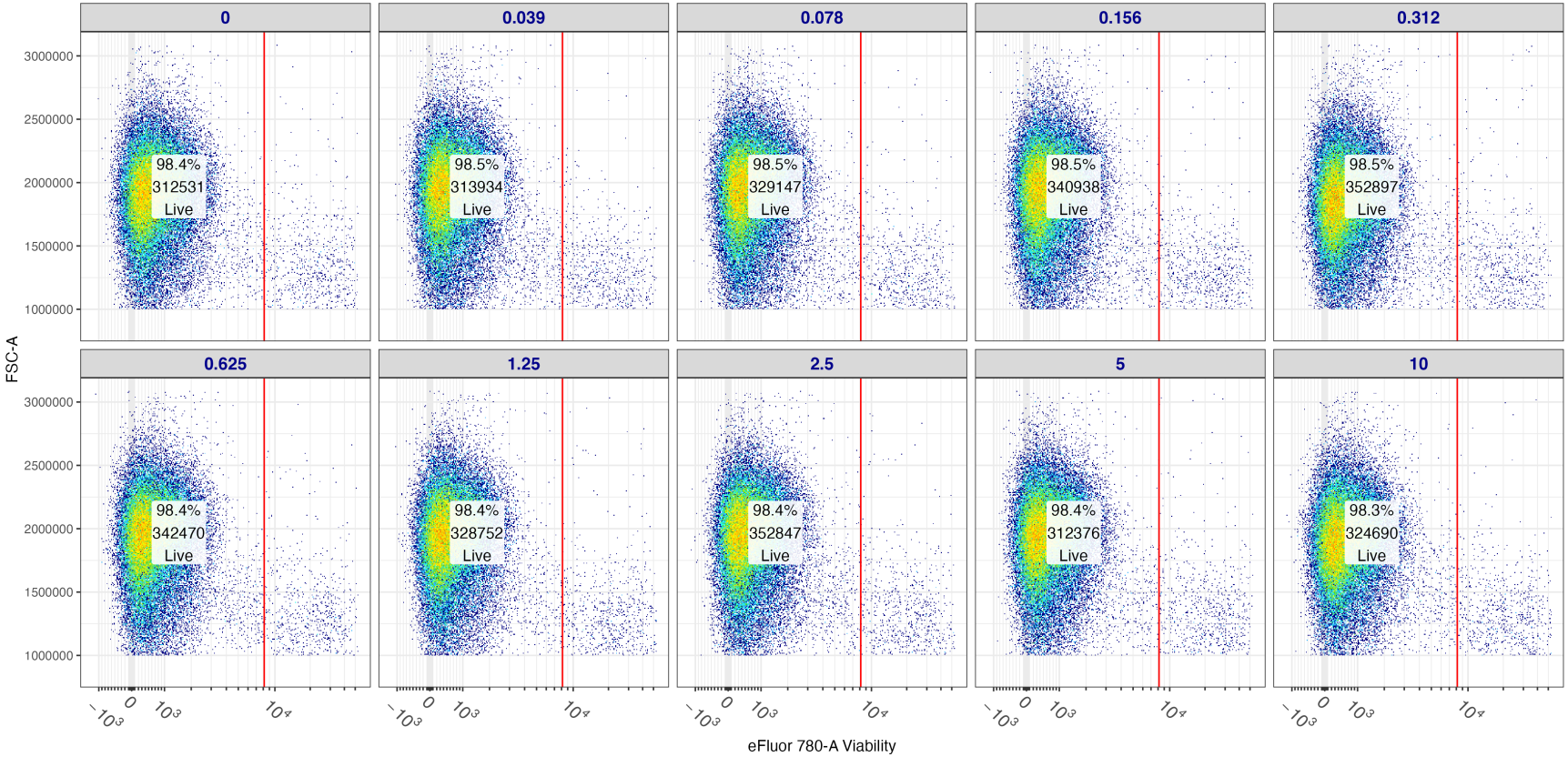
*Parent pop: Cells | CD27 BUV395-A || Cat: 563815 || BD Biosciences || Lot: 4330333 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: singletGate || args: prediction_level=0.99, wider_gate=FALSE || collapse: FALSE || groupBy: Dirname || pp: — || pp args: —*
*Compensation: —*

**CD27 BUV395-A : Live | EXP1460 | JD | Clone L128**

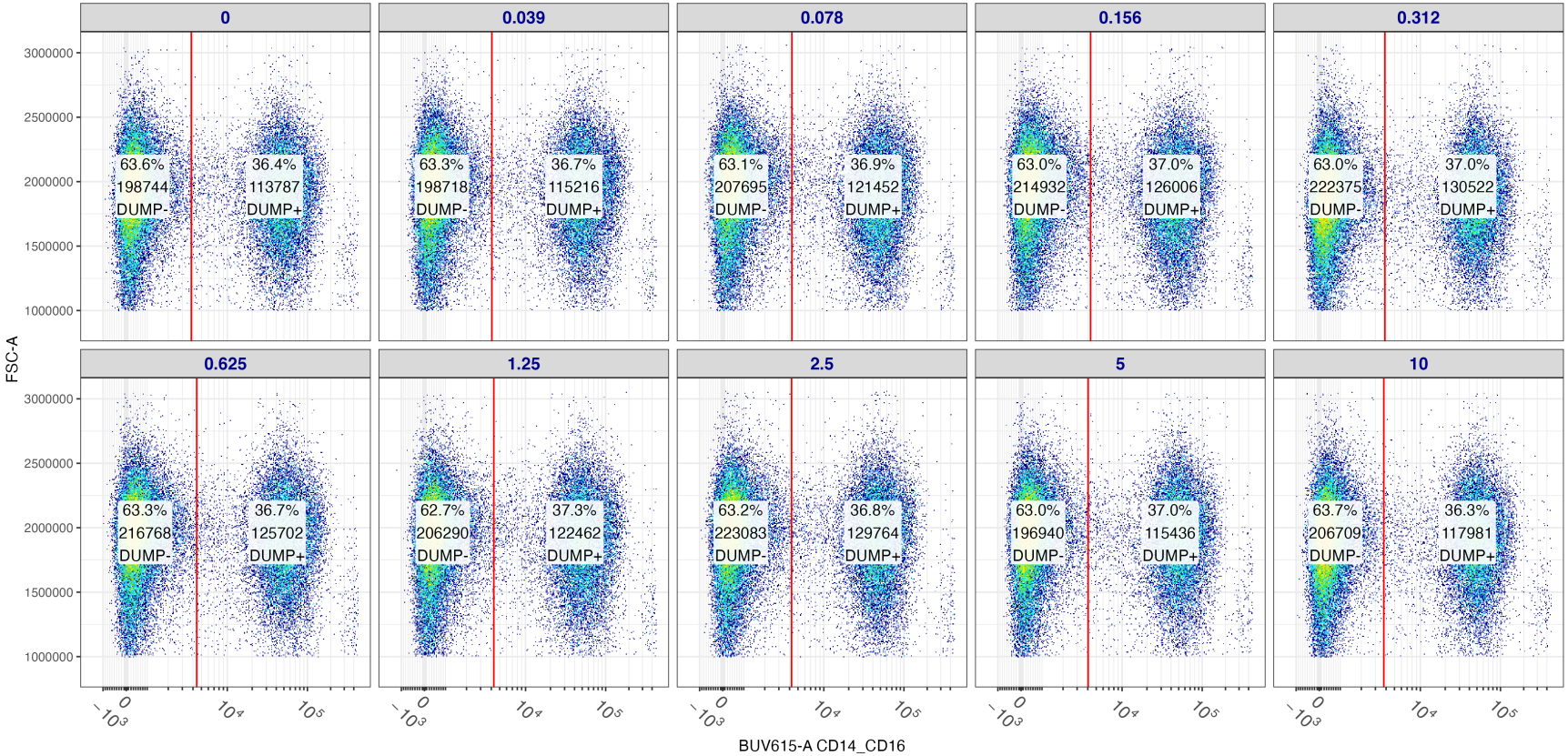
*Parent pop: Single_cells | CD27 BUV395-A || Cat: 563815 || BD Biosciences || Lot: 4330333 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: gate_mindensity || args: gate_range=NULL, adjust=1, min=0 || collapse: TRUE || groupBy: Dirname || pp: — || pp args: —*
*Compensation: —*

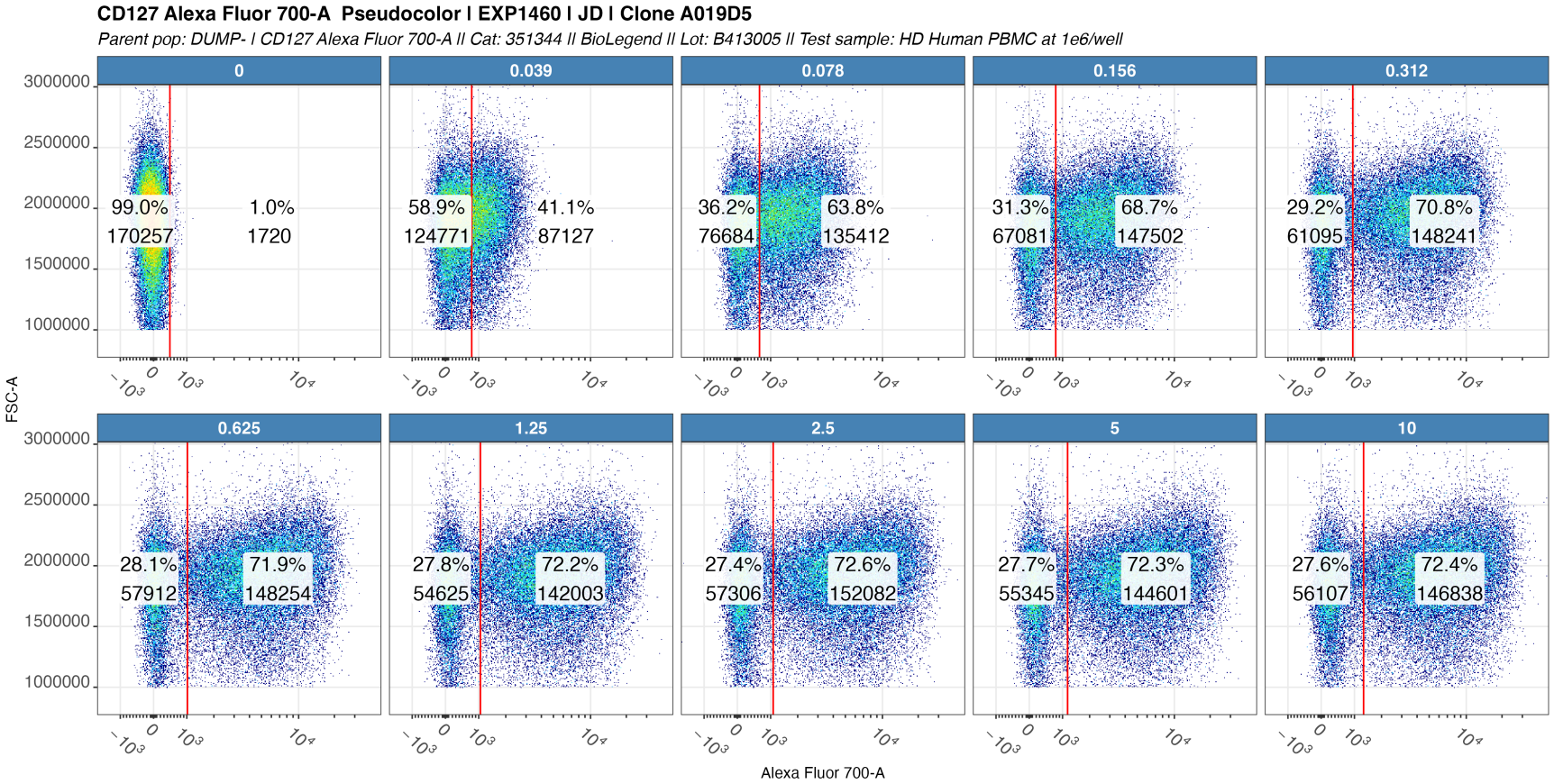
**CD27 BUV395-A : DUMP | EXP1460 | JD | Clone L128**

*Parent pop: Live | CD27 BUV395-A || Cat: 563815 || BD Biosciences || Lot: 4330333 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: gate_flowmeans || args: K=2, cluster='neg', quantile=0.99 || collapse: FALSE || groupBy: Dirname || pp: — || pp args: —*
*Compensation: —*

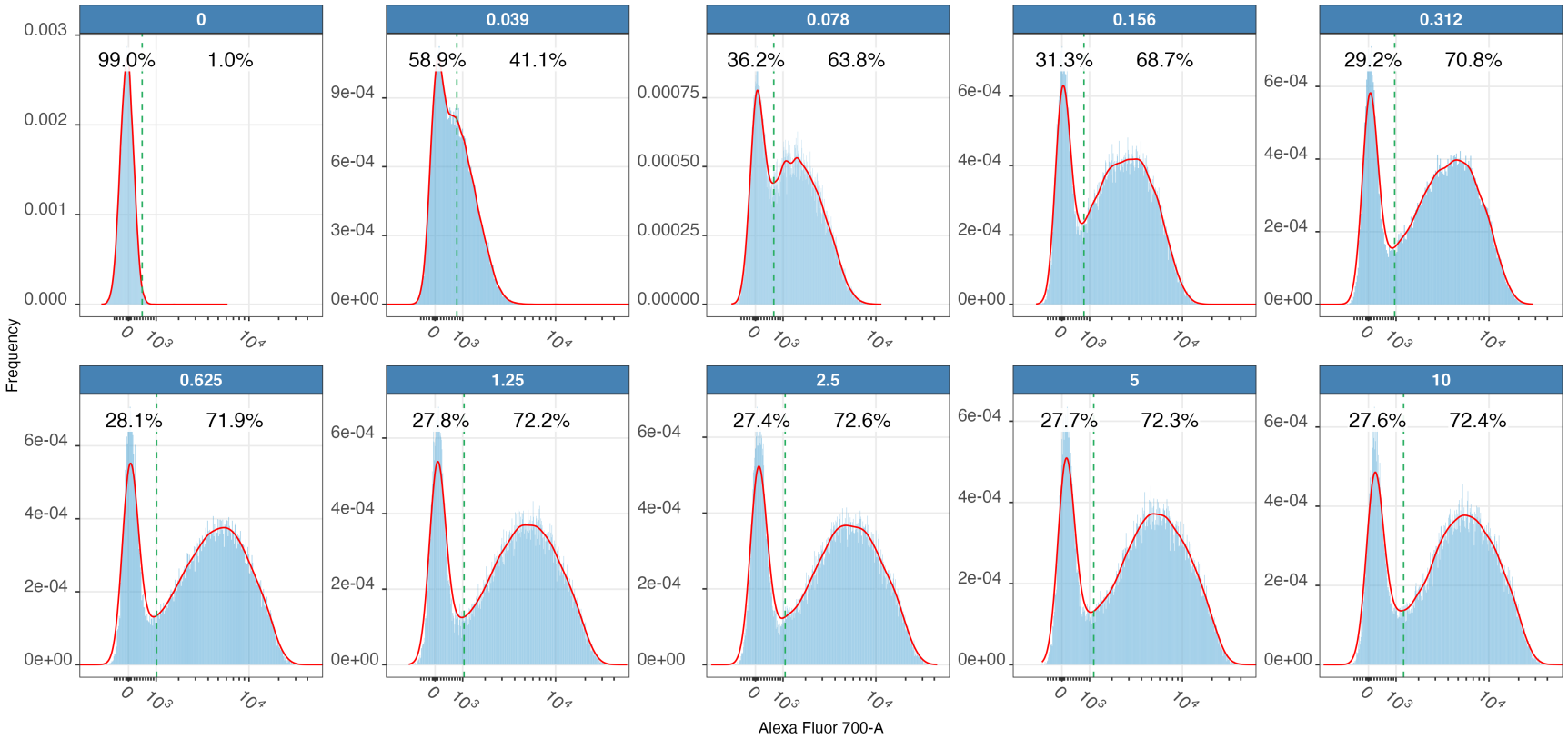
### 5.3 CD127




*Gating method: gate_mind_fmeans_costain || args: K=2, cluster='neg', quantile=0.99, gate_range=NULL, adjust=2 || collapse: FALSE || groupBy: Dirname || pp: pp_costain_floor || pp args: costain_quantile=0.99*
*Compensation: —*

**CD127 Alexa Fluor 700-A Histogram | EXP1460 | JD | Clone A019D5**

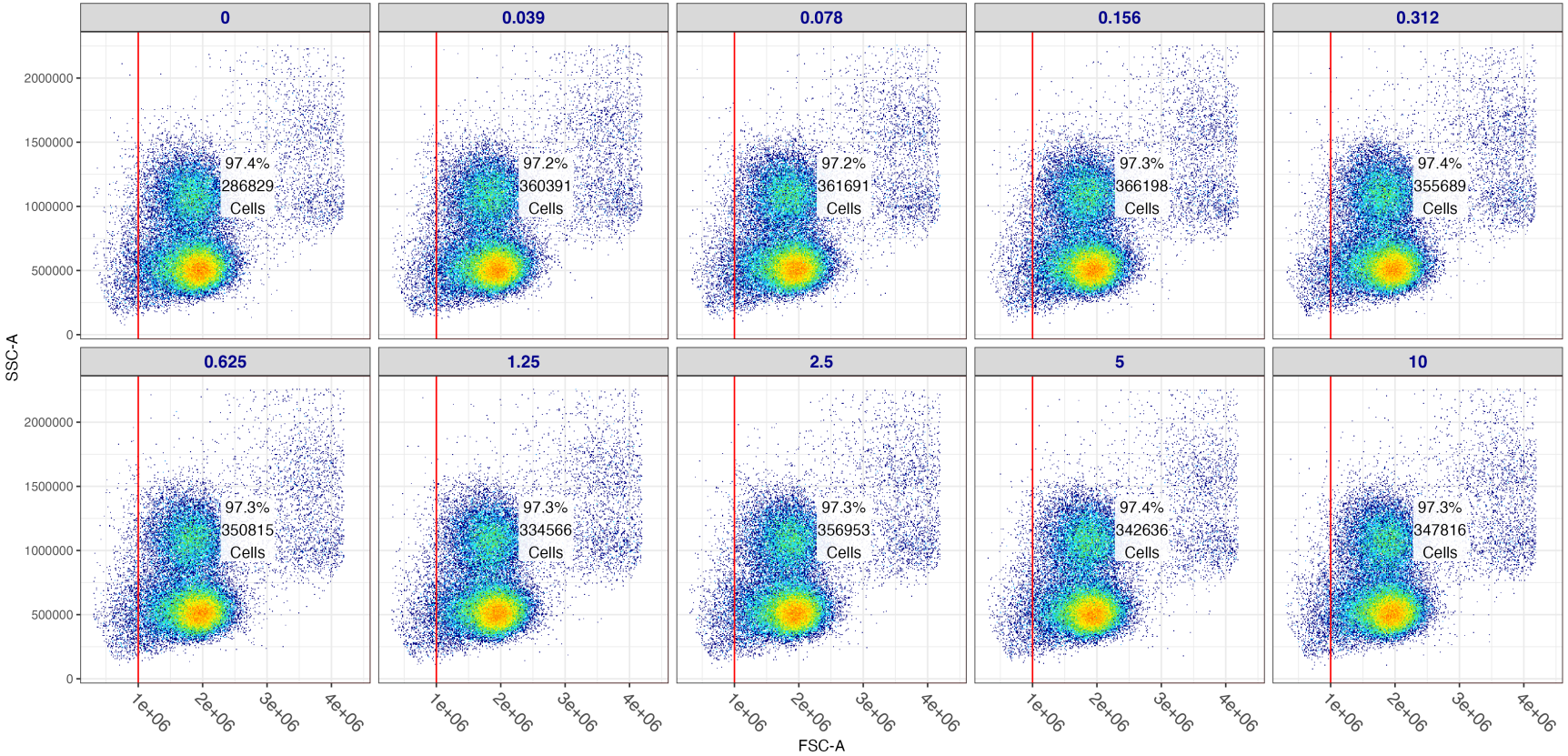
*Parent pop: DUMP- || CD127 Alexa Fluor 700-A || Cat: 351344 || BioLegend || Lot: B413005 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: gate_mind_fmeans_costain || args: K=2, cluster='neg', quantile=0.99, gate_range=NULL, adjust=2 || collapse: FALSE || groupBy: Dirname || pp: pp_costain_floor || pp args: costain_quantile=0.99*
*Compensation: —*

**CD127 Alexa Fluor 700-A : Cells | EXP1460 | JD | Clone A019D5**

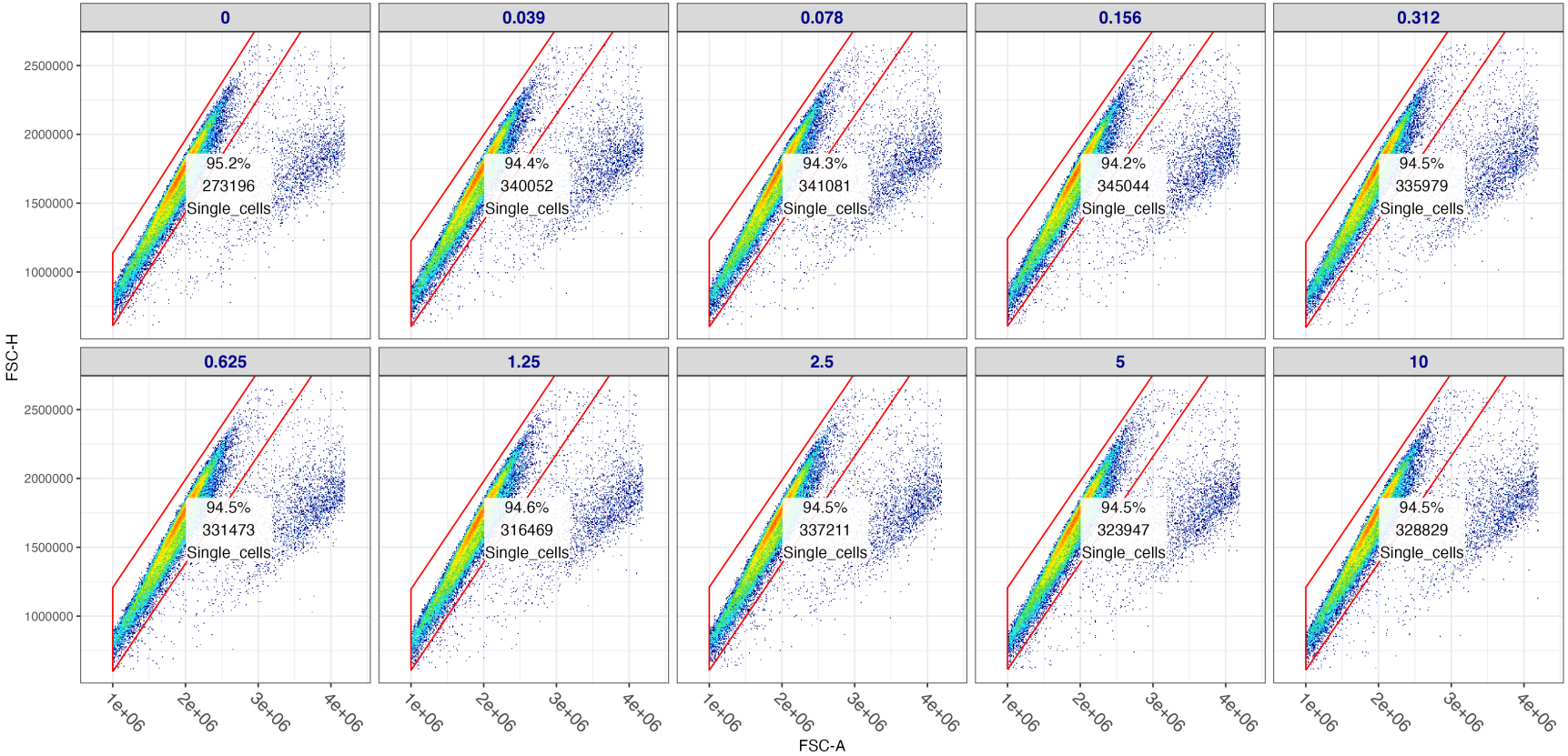
*Parent pop: root | CD127 Alexa Fluor 700-A || Cat: 351344 || BioLegend || Lot: B413005 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: boundary || args: min=c(1e6,-Inf), max=c(Inf,Inf) || collapse: TRUE || groupBy: Dirname || pp: — || pp args: —*
*Compensation: —*

**CD127 Alexa Fluor 700-A : Single_cells | EXP1460 | JD | Clone A019D5**

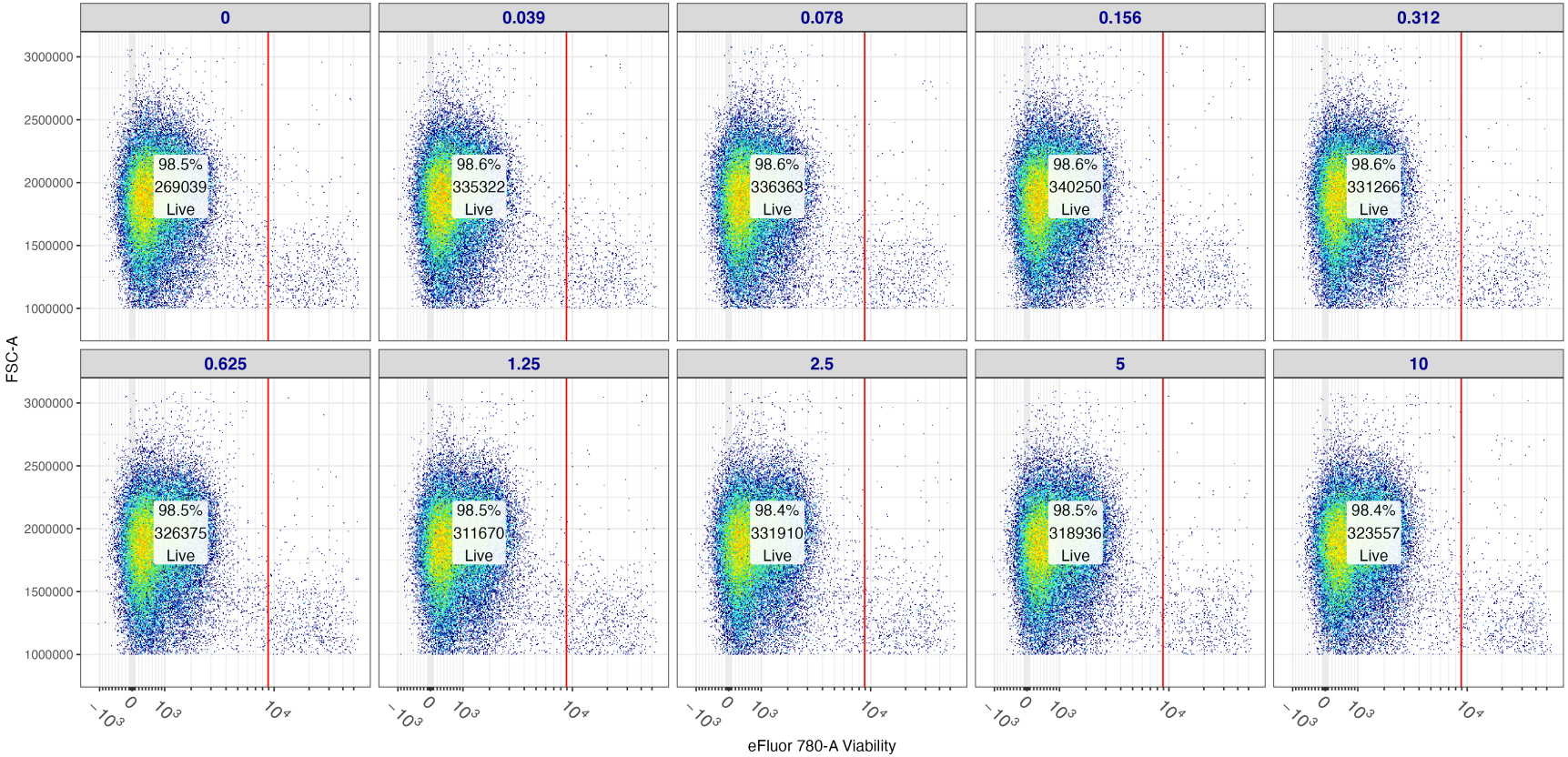
*Parent pop: Cells | CD127 Alexa Fluor 700-A || Cat: 351344 || BioLegend || Lot: B413005 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: singletGate || args: prediction_level=0.99, wider_gate=FALSE || collapse: FALSE || groupBy: Dirname || pp: — || pp args: —*
*Compensation: —*

**CD127 Alexa Fluor 700-A : Live | EXP1460 | JD | Clone A019D5**

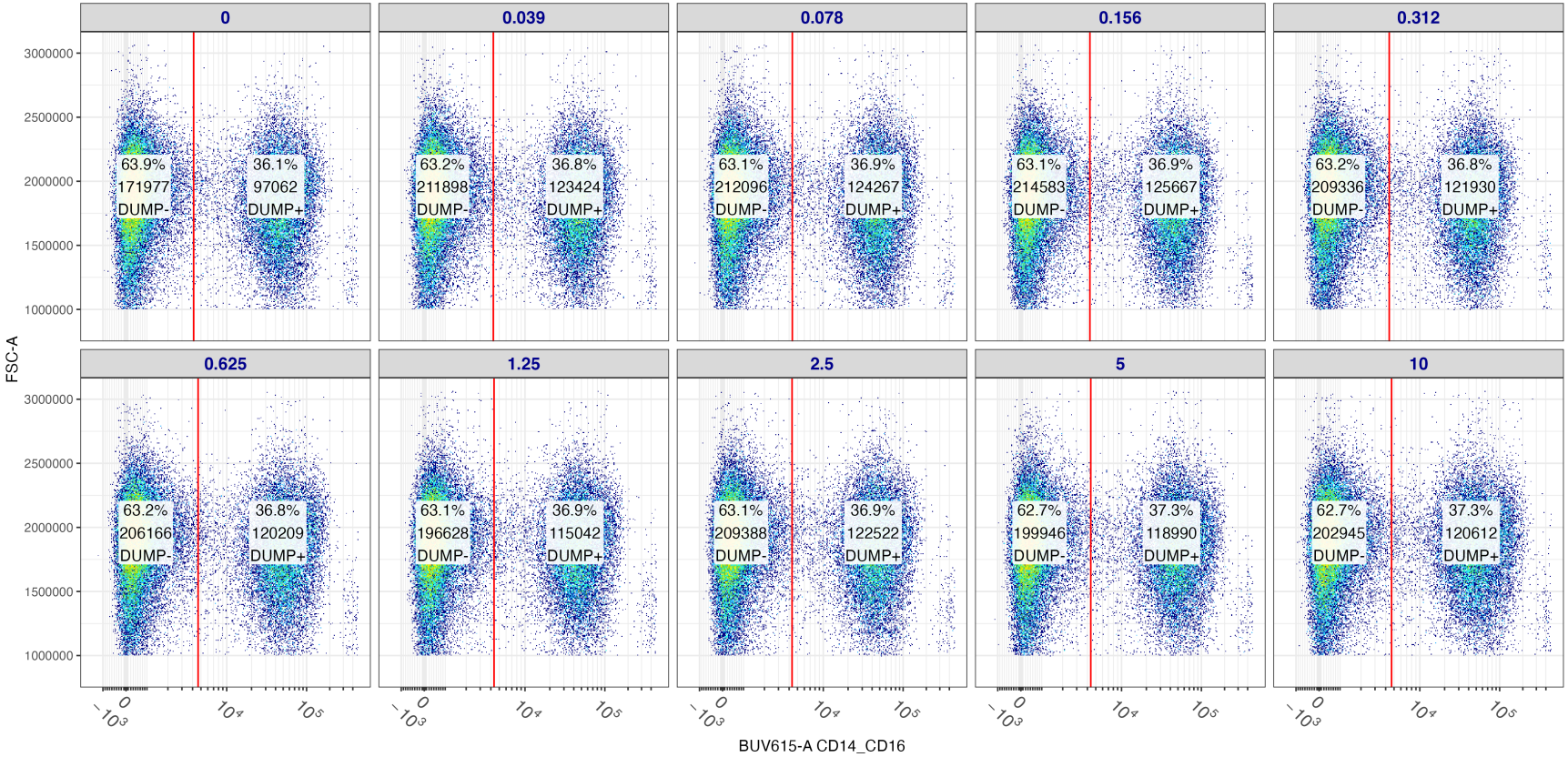
*Parent pop: Single_cells | CD127 Alexa Fluor 700-A || Cat: 351344 || BioLegend || Lot: B413005 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: gate_mindensity || args: gate_range=NULL, adjust=1, min=0 || collapse: TRUE || groupBy: Dirname || pp: — || pp args: — Compensation: —*

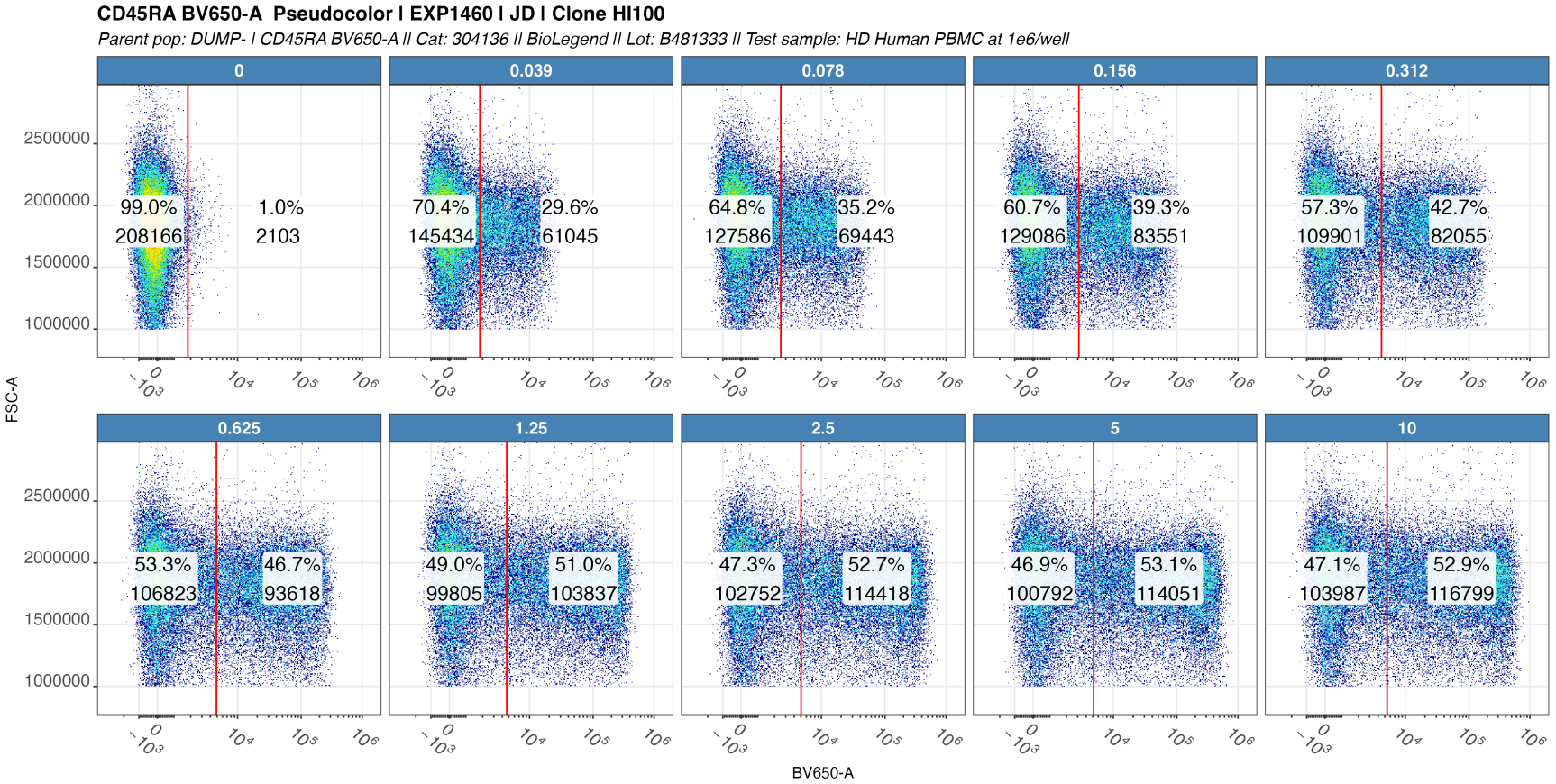
**CD127 Alexa Fluor 700-A : DUMP I EXP1460 I JD I Clone A019D5**

*Parent pop: Live I CD127 Alexa Fluor 700-A II Cat: 351344 II BioLegend II Lot: B413005 II Test sample: HD Human PBMC at 1e6/well*




*Gating method: gate_flowmeans II args: K=2, cluster='neg', quantile=0.99 II collapse: FALSE II groupBy: Dirname II pp: — II pp args: —*
*Compensation: —*

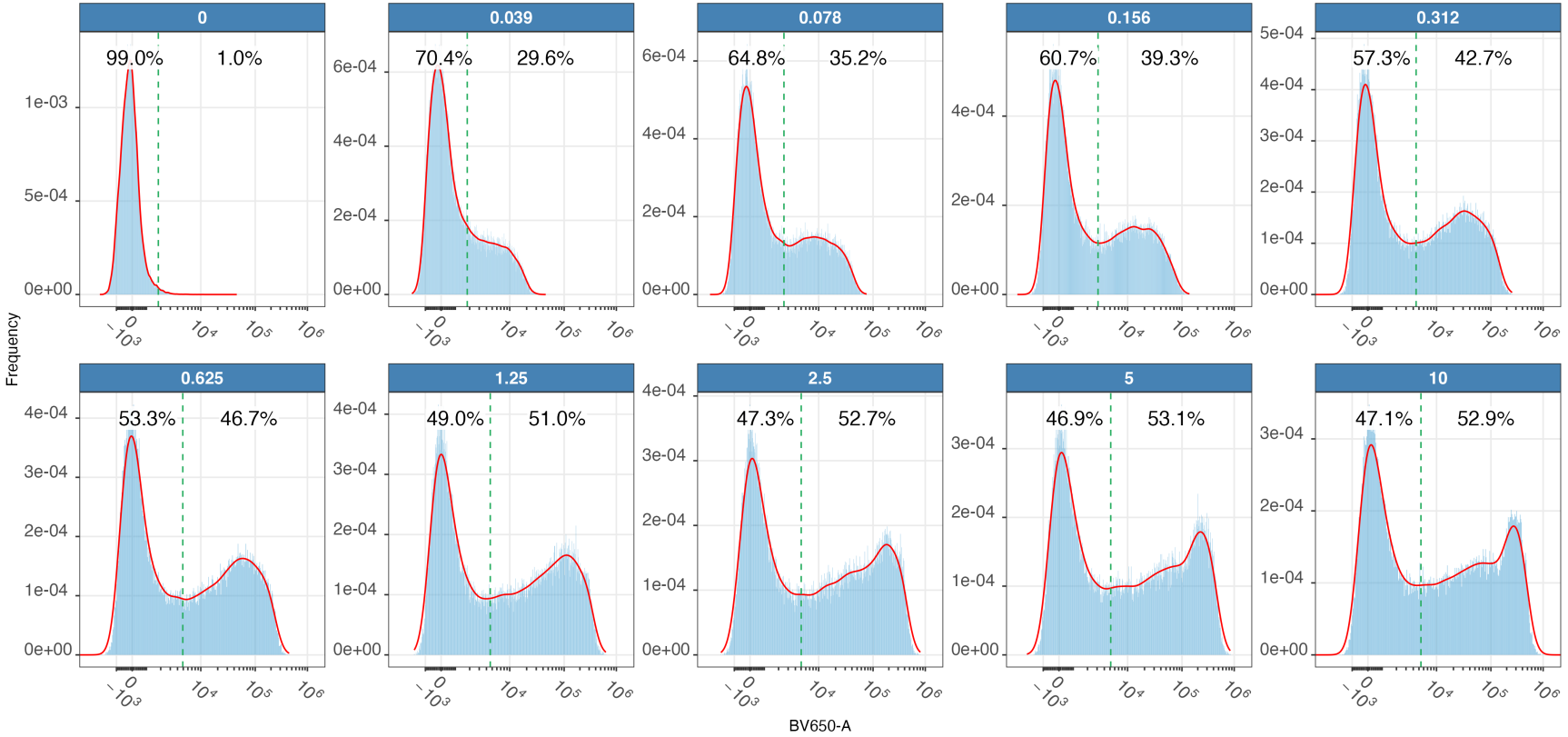
### 5.4 CD45RA




*Gating method: gate_mind_fmeans_costain || args: K=2, cluster='neg', quantile=0.99, gate_range=NULL, adjust=2 || collapse: FALSE || groupBy: Dirname || pp: pp_costain_floor || pp args: costain_quantile=0.99*
*Compensation: —*

**CD45RA BV650-A Histogram | EXP1460 | JD | Clone HI100**

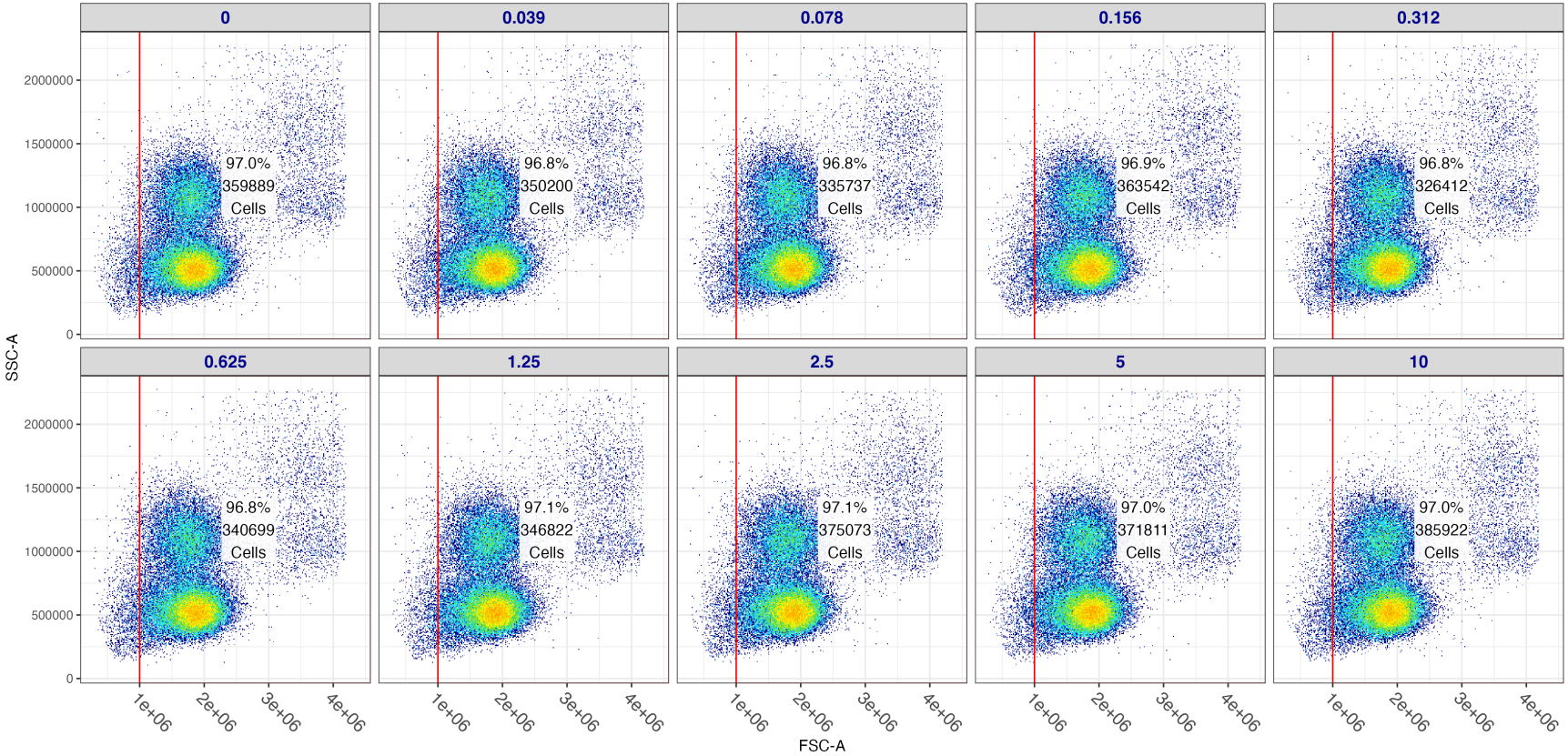
*Parent pop: DUMP- || CD45RA BV650-A || Cat: 304136 || BioLegend || Lot: B481333 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: gate_mind_fmeans_costain || args: K=2, cluster='neg', quantile=0.99, gate_range=NULL, adjust=2 || collapse: FALSE || groupBy: Dirname || pp: pp_costain_floor || pp args: costain_quantile=0.99*
*Compensation: —*

**CD45RA BV650-A : Cells | EXP1460 | JD | Clone HI100**

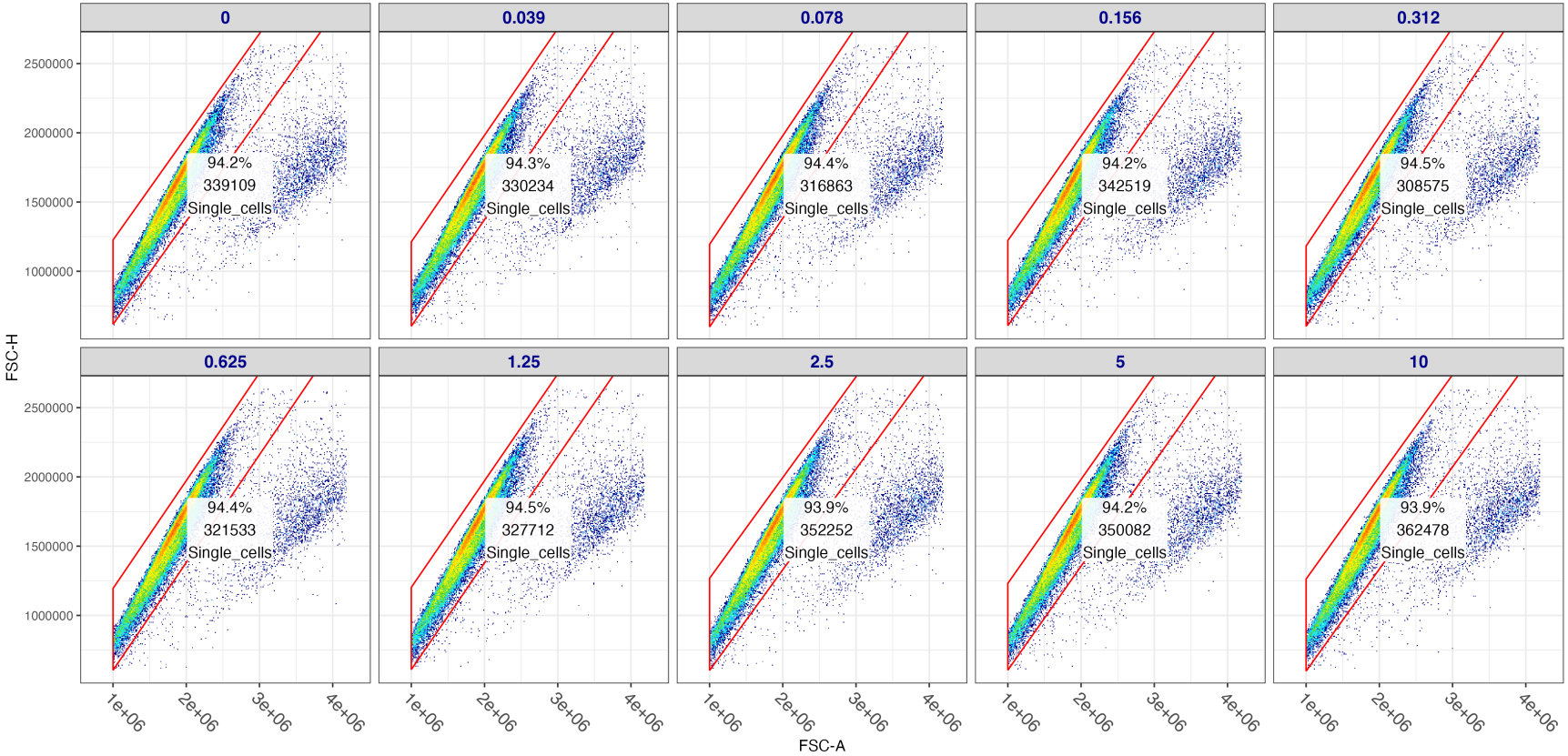
*Parent pop: root | CD45RA BV650-A || Cat: 304136 || BioLegend || Lot: B481333 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: boundary || args: min=c(1e6,-Inf), max=c(Inf,Inf) || collapse: TRUE || groupBy: Dirname || pp: — || pp args: —*
*Compensation: —*

**CD45RA BV650-A : Single_cells | EXP1460 | JD | Clone HI100**

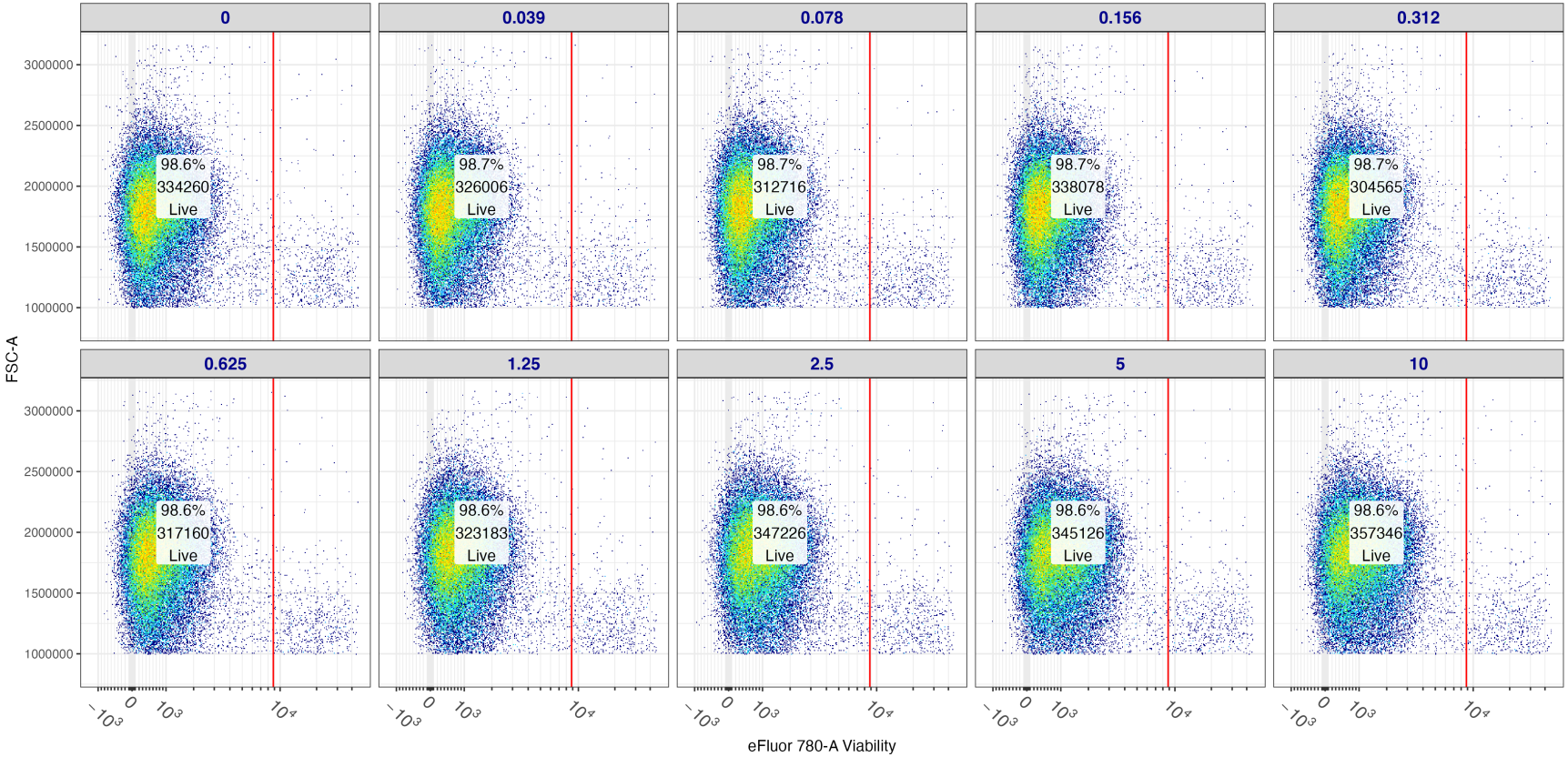
*Parent pop: Cells | CD45RA BV650-A || Cat: 304136 || BioLegend || Lot: B481333 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: singletGate || args: prediction_level=0.99, wider_gate=FALSE || collapse: FALSE || groupBy: Dirname || pp: — || pp args: —*
*Compensation: —*

**CD45RA BV650-A : Live | EXP1460 | JD | Clone HI100**

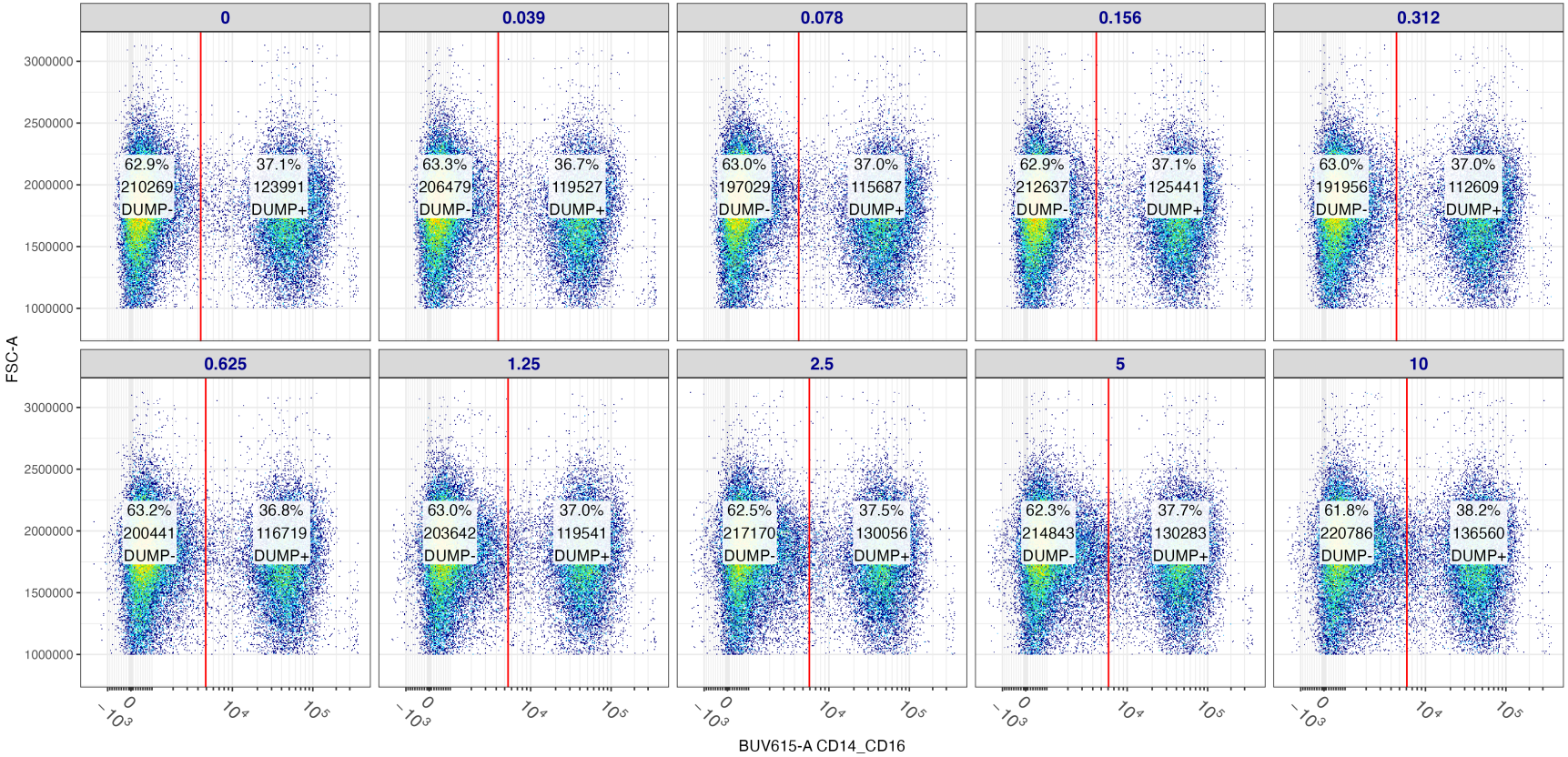
*Parent pop: Single_cells | CD45RA BV650-A || Cat: 304136 || BioLegend || Lot: B481333 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: gate_mindensity || args: gate_range=NULL, adjust=1, min=0 || collapse: TRUE || groupBy: Dirname || pp: — || pp args: — Compensation: —*

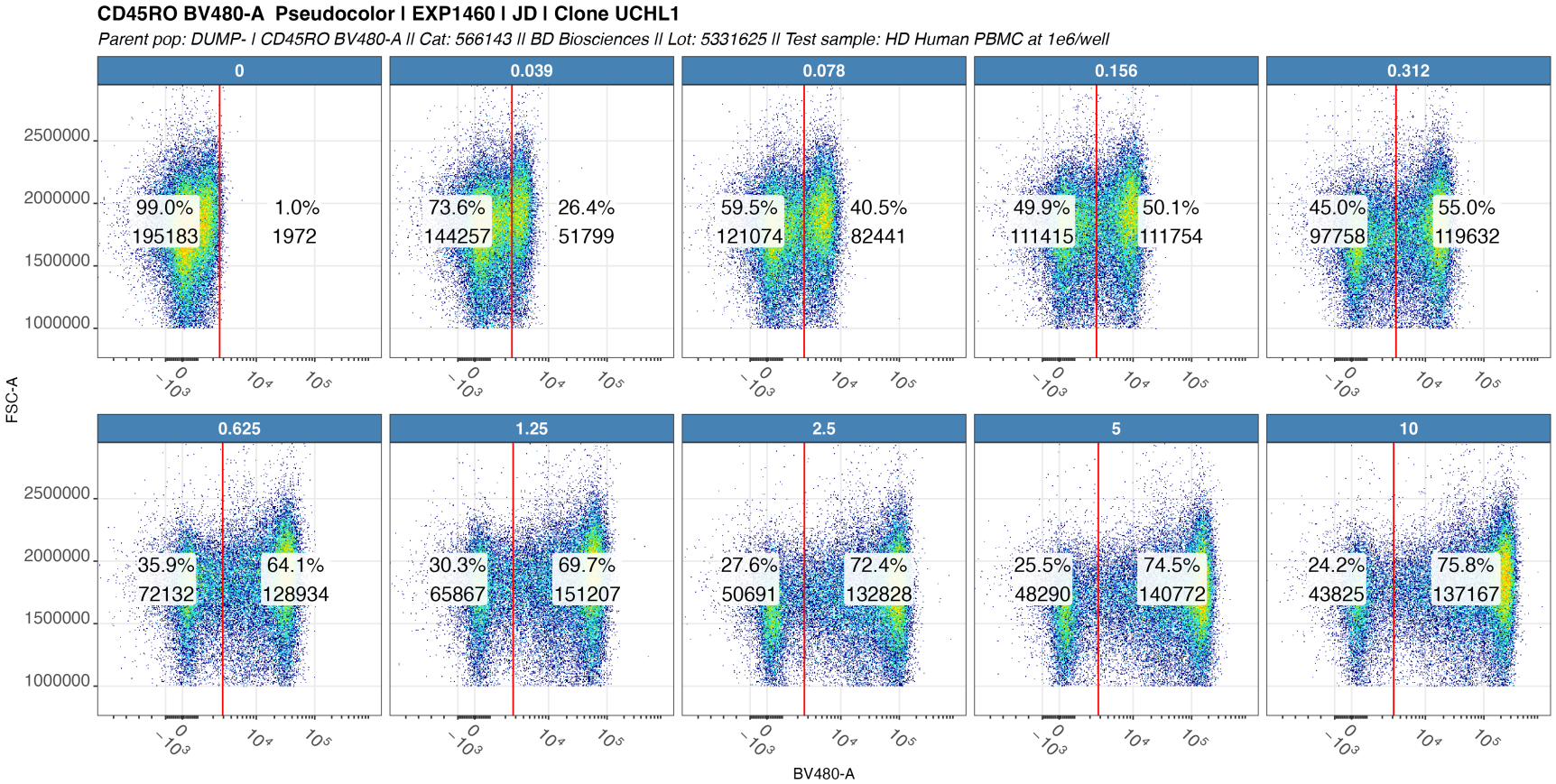
**CD45RA BV650-A : DUMP | EXP1460 | JD | Clone HI100**

*Parent pop: Live | CD45RA BV650-A || Cat: 304136 || BioLegend || Lot: B481333 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: gate_flowmeans || args: K=2, cluster='neg', quantile=0.99 || collapse: FALSE || groupBy: Dirname || pp: — || pp args: —*
*Compensation: —*

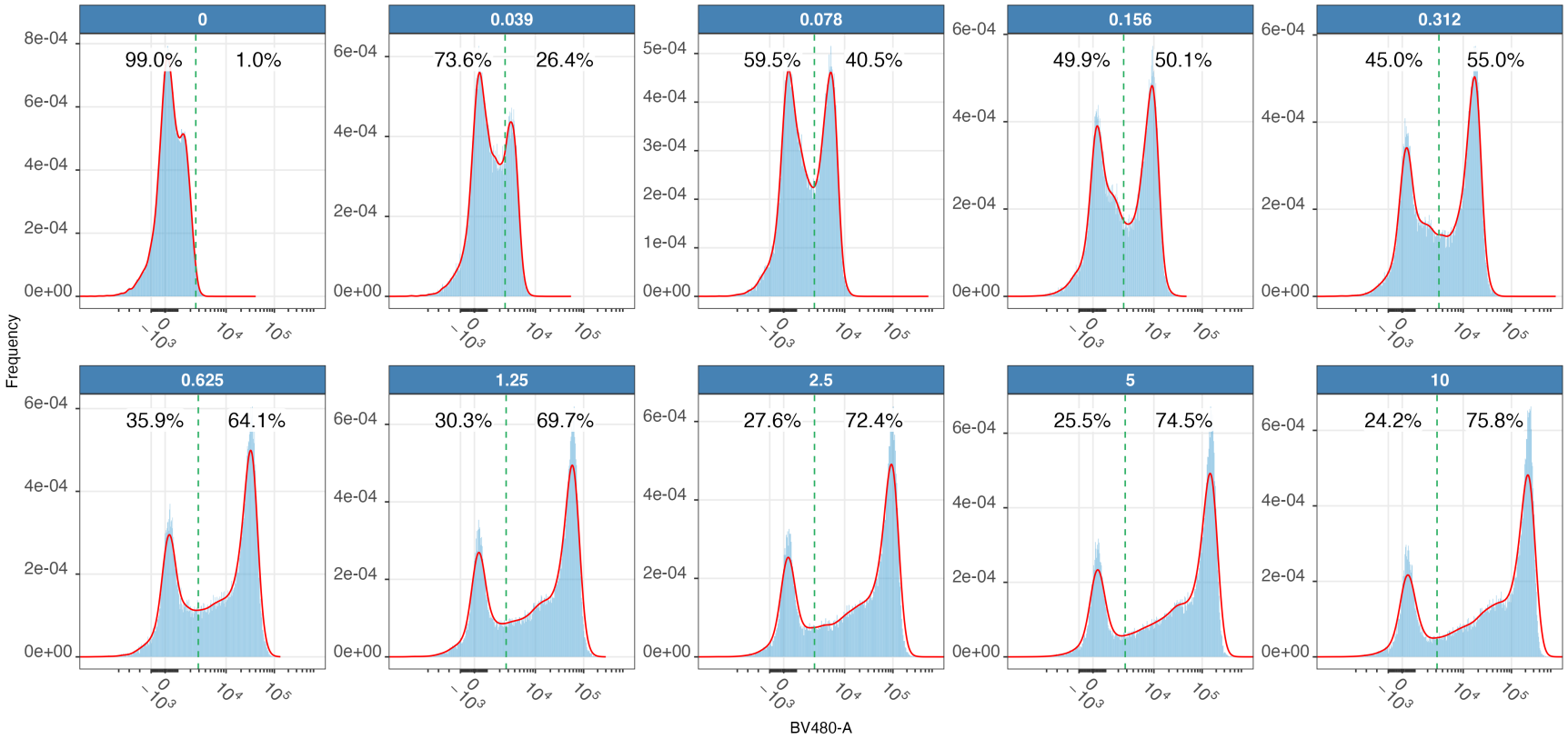
### 5.5 CD45RO




*Gating method: gate_mind_fmeans_costain || args: K=2, cluster='neg', quantile=0.99, gate_range=NULL, adjust=2 || collapse: FALSE || groupBy: Dirname || pp: pp_costain_floor || pp args: costain_quantile=0.99*
*Compensation: —*

**CD45RO BV480-A Histogram | EXP1460 | JD | Clone UCHL1**

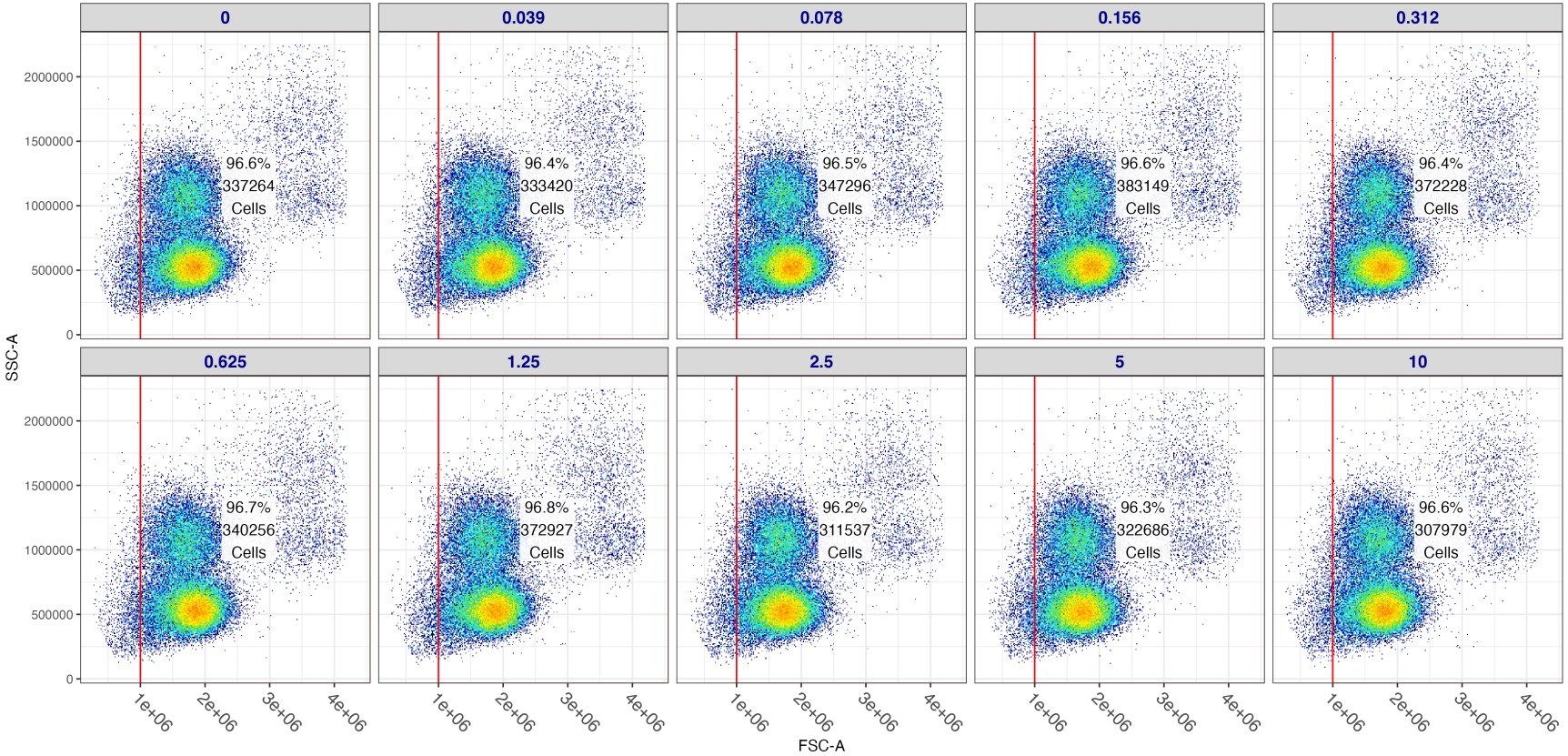
*Parent pop: DUMP- || CD45RO BV480-A || Cat: 566143 || BD Biosciences || Lot: 5331625 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: gate_mind_fmeans_costain || args: K=2, cluster='neg', quantile=0.99, gate_range=NULL, adjust=2 || collapse: FALSE || groupBy: Dirname || pp: pp_costain_floor || pp args: costain_quantile=0.99*
*Compensation: —*

**CD45RO BV480-A : Cells | EXP1460 | JD | Clone UCHL1**

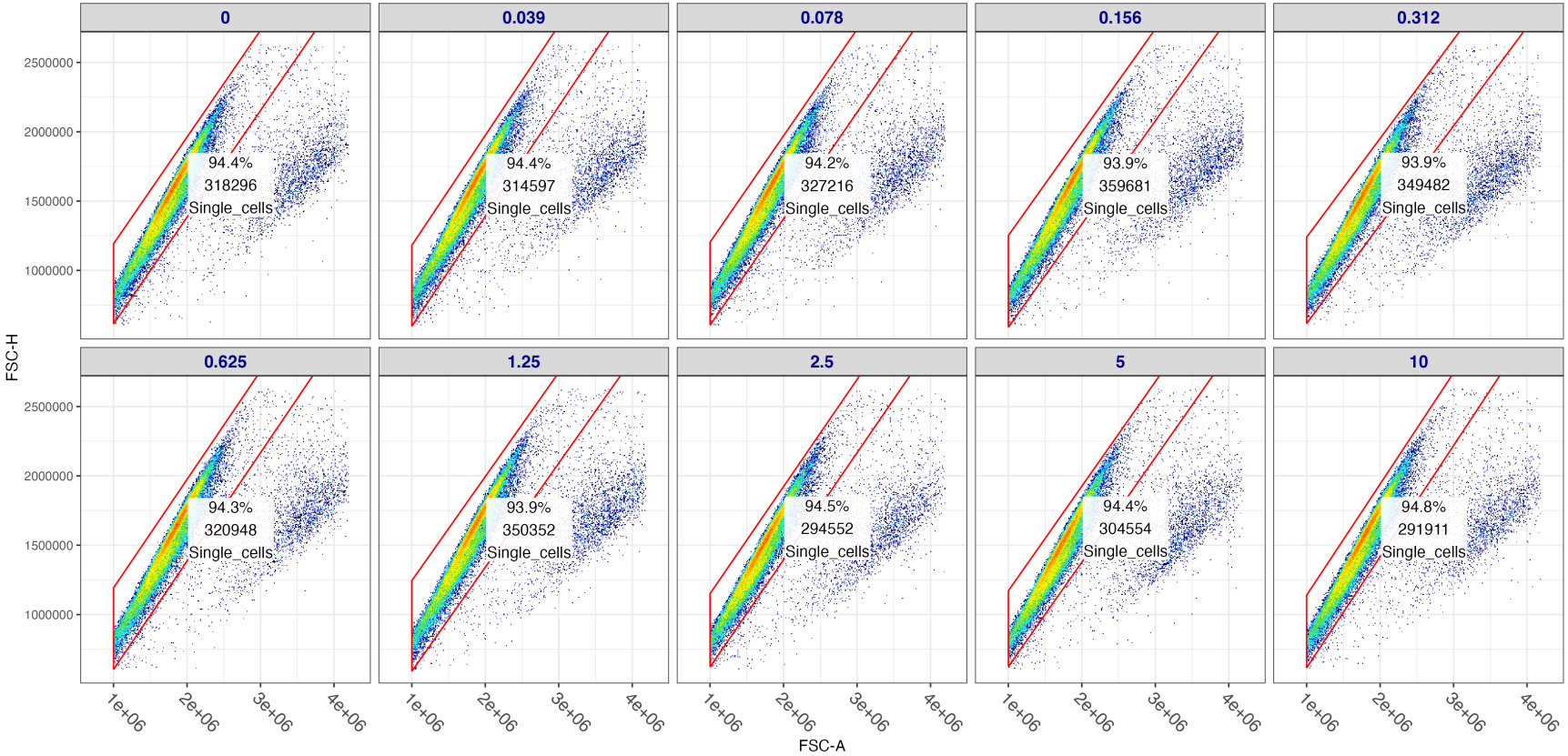
*Parent pop: root | CD45RO BV480-A || Cat: 566143 || BD Biosciences || Lot: 5331625 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: boundary || args: min=c(1e6,-Inf), max=c(Inf,Inf) || collapse: TRUE || groupBy: Dirname || pp: — || pp args: —*
*Compensation: —*

**CD45RO BV480-A : Single_cells | EXP1460 | JD | Clone UCHL1**

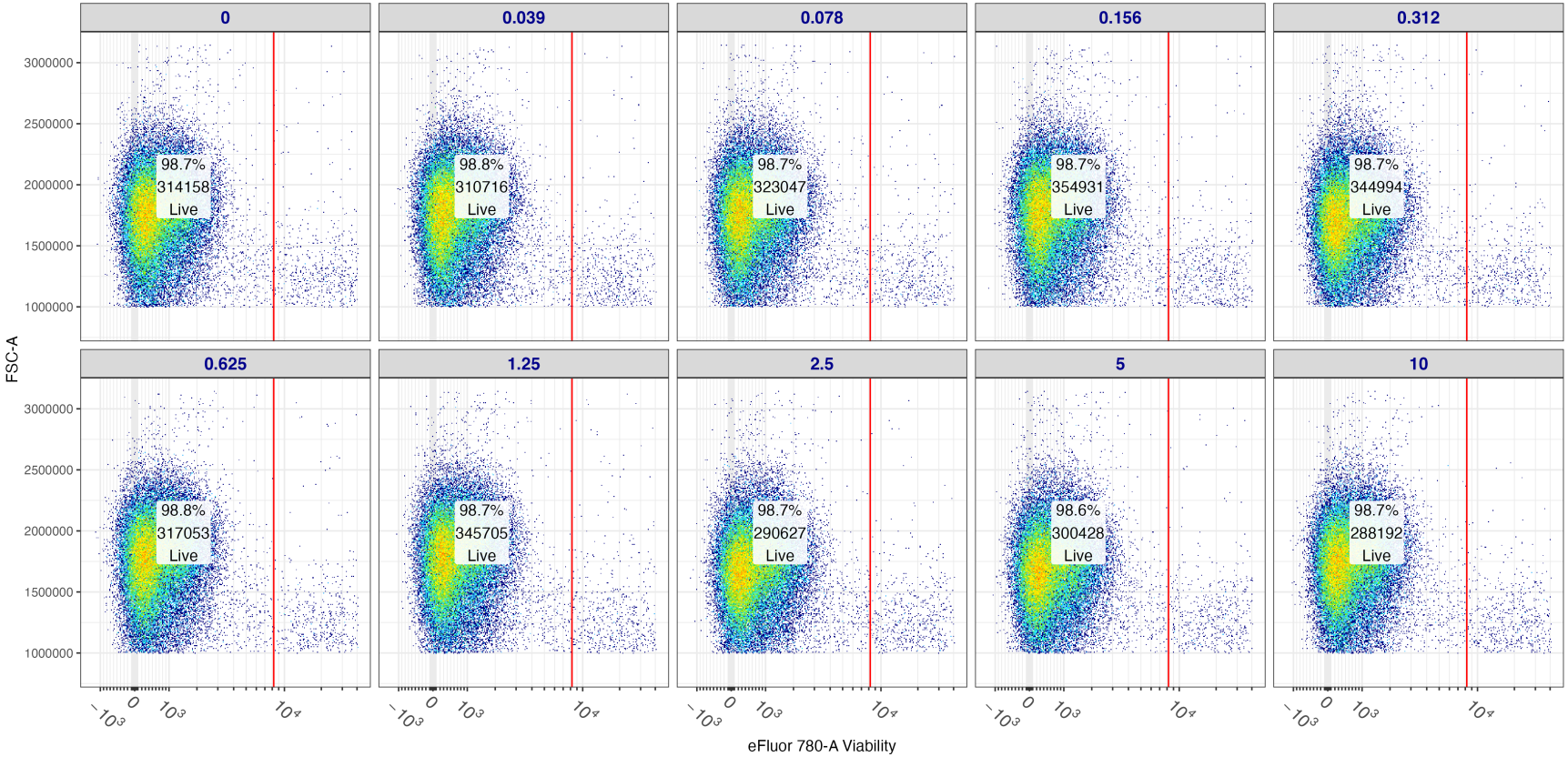
*Parent pop: Cells | CD45RO BV480-A || Cat: 566143 || BD Biosciences || Lot: 5331625 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: singletGate || args: prediction_level=0.99, wider_gate=FALSE || collapse: FALSE || groupBy: Dirname || pp: — || pp args: —*
*Compensation: —*

**CD45RO BV480-A : Live | EXP1460 | JD | Clone UCHL1**

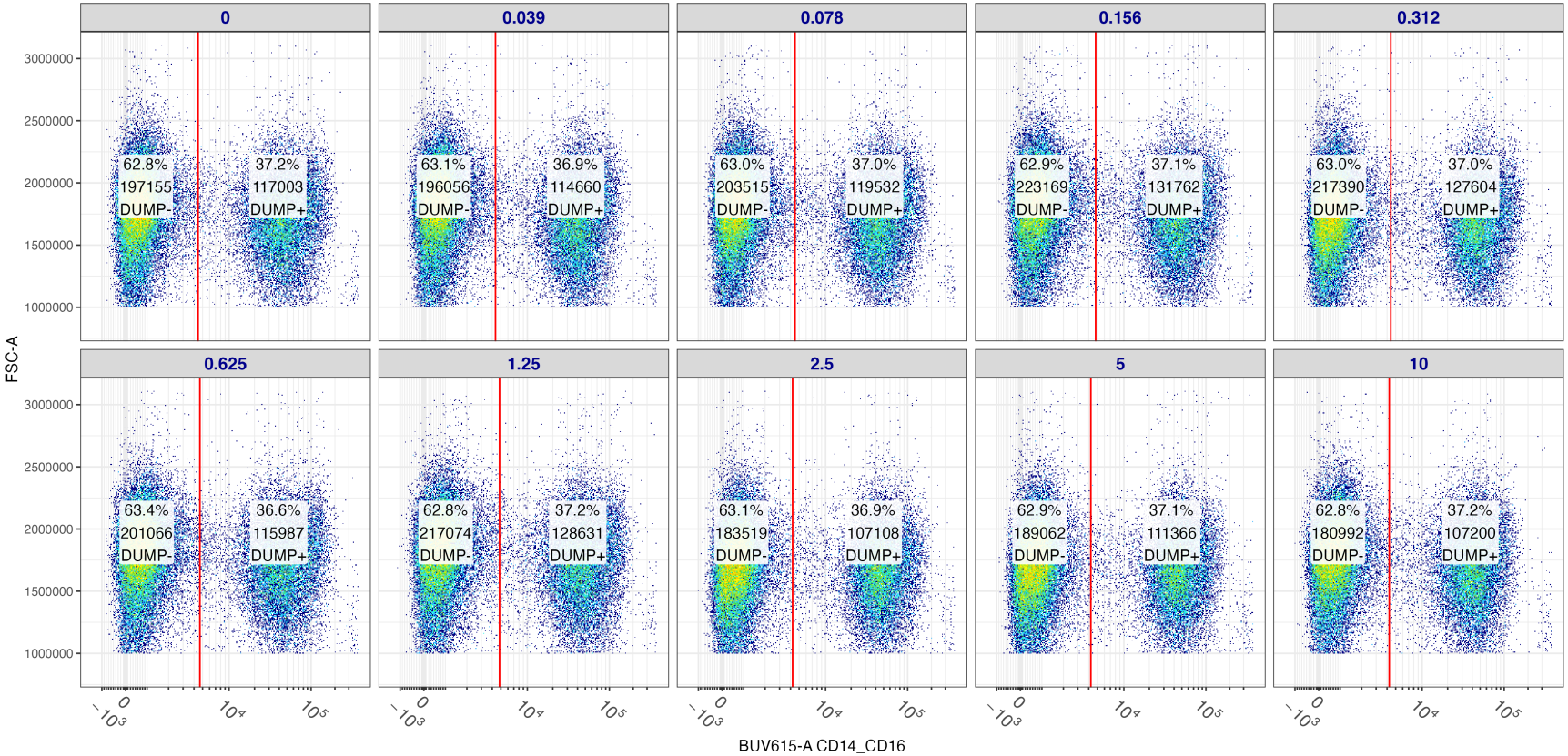
*Parent pop: Single_cells | CD45RO BV480-A || Cat: 566143 || BD Biosciences || Lot: 5331625 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: gate_mindensity || args: gate_range=NULL, adjust=1, min=0 || collapse: TRUE || groupBy: Dirname || pp: — || pp args: — Compensation: —*

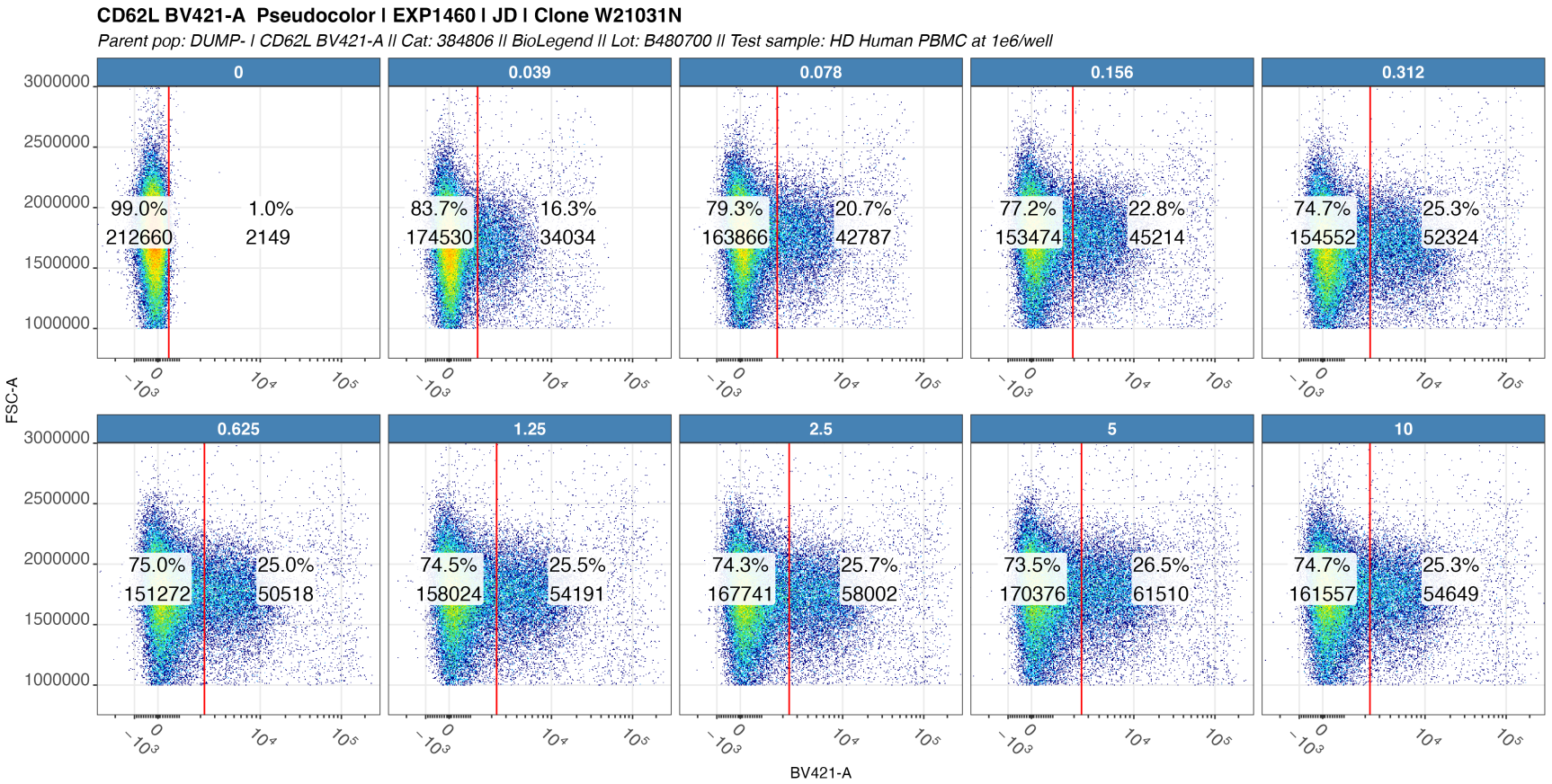
**CD45RO BV480-A : DUMP | EXP1460 | JD | Clone UCHL1**

*Parent pop: Live | CD45RO BV480-A || Cat: 566143 || BD Biosciences || Lot: 5331625 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: gate_flowmeans || args: K=2, cluster='neg', quantile=0.99 || collapse: FALSE || groupBy: Dirname || pp: — || pp args: —*
*Compensation: —*

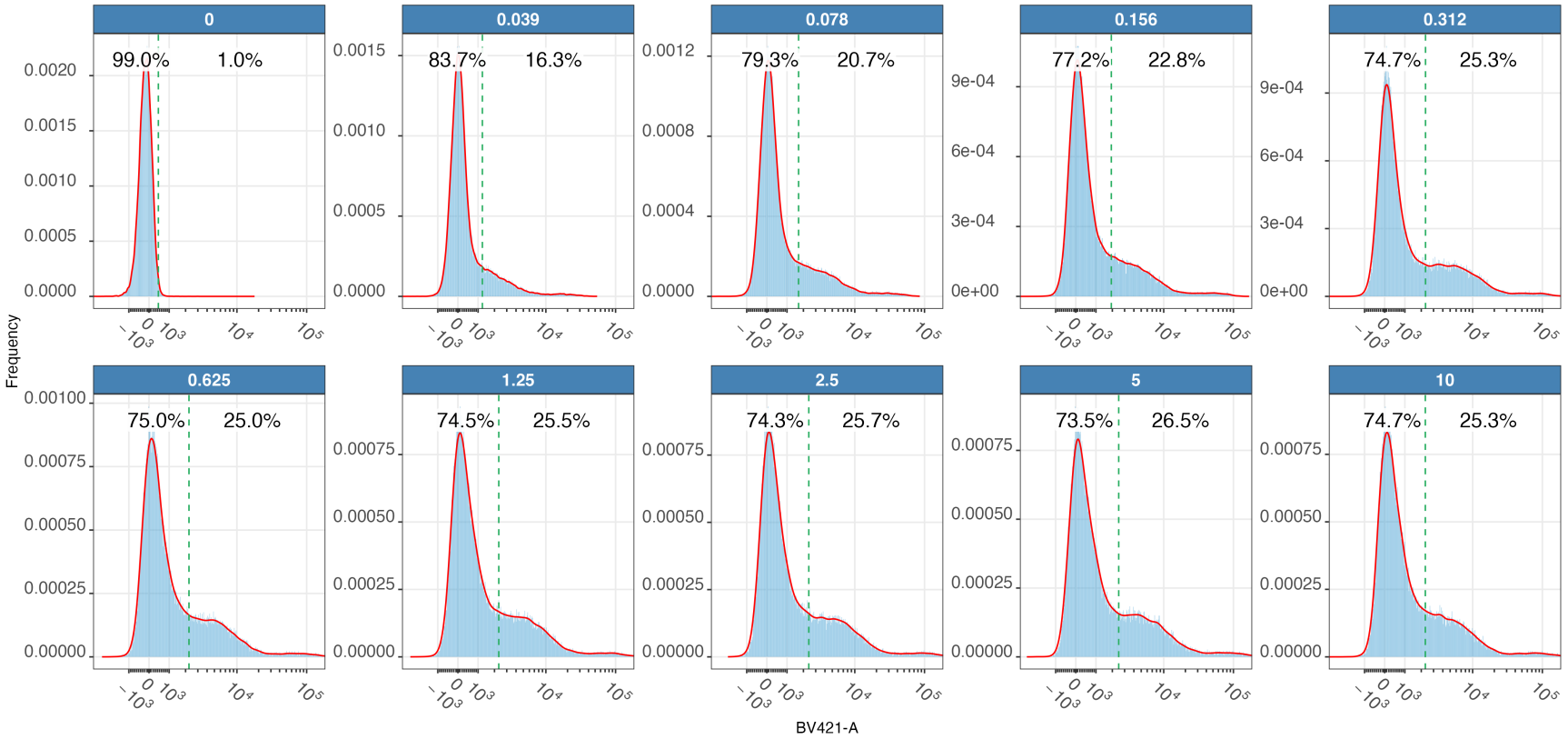
### 5.6 CD62L




*Gating method: gate_mind_fmeans_costain || args: K=2, cluster='neg', quantile=0.99, gate_range=NULL, adjust=2 || collapse: FALSE || groupBy: Dirname || pp: pp_costain_floor || pp args: costain_quantile=0.99*
*Compensation: —*

**CD62L BV421-A Histogram | EXP1460 | JD | Clone W21031N**

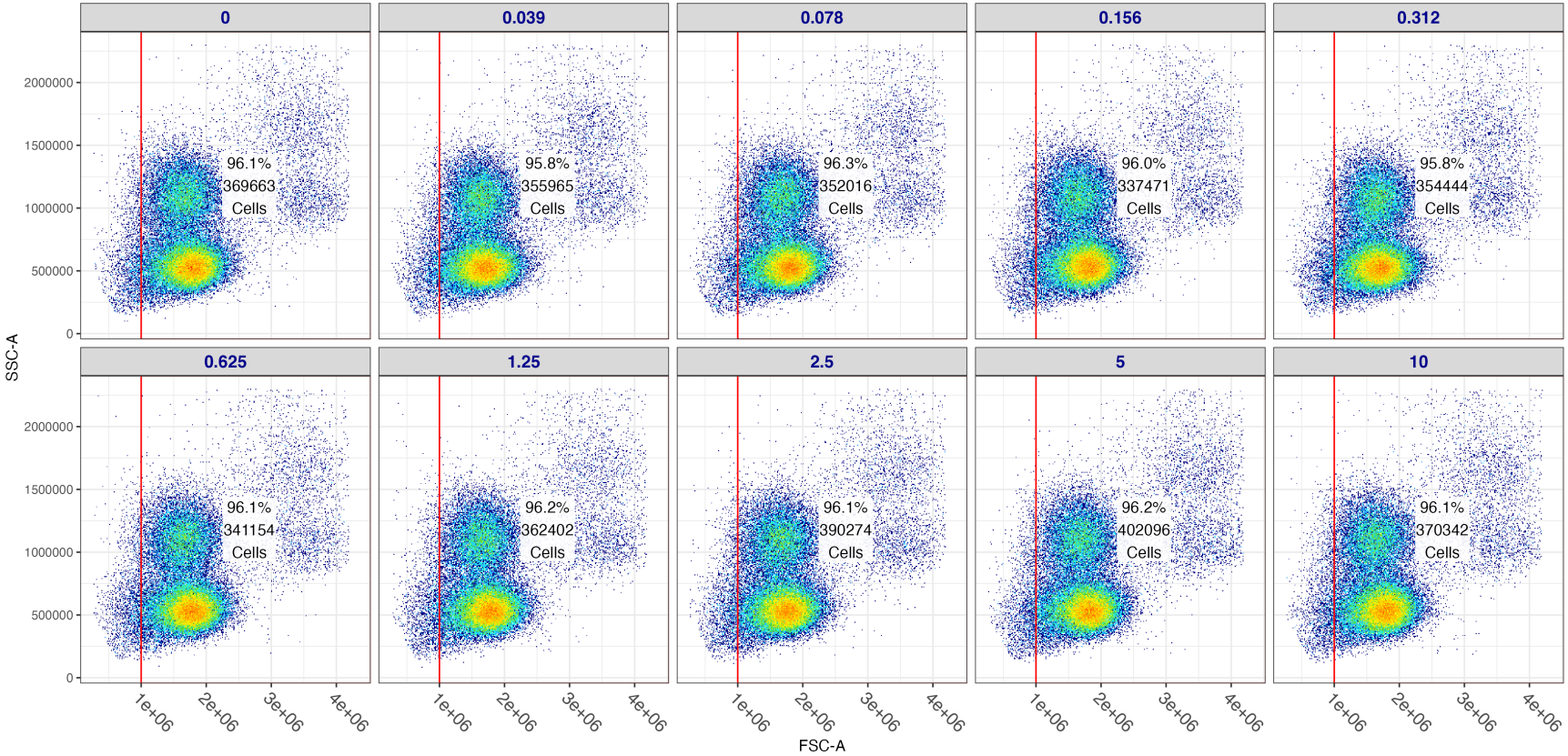
*Parent pop: DUMP- || CD62L BV421-A || Cat: 384806 || BioLegend || Lot: B480700 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: gate_mind_fmeans_costain || args: K=2, cluster='neg', quantile=0.99, gate_range=NULL, adjust=2 || collapse: FALSE || groupBy: Dirname || pp: pp_costain_floor || pp args: costain_quantile=0.99*
*Compensation: —*

**CD62L BV421-A : Cells | EXP1460 | JD | Clone W21031N**

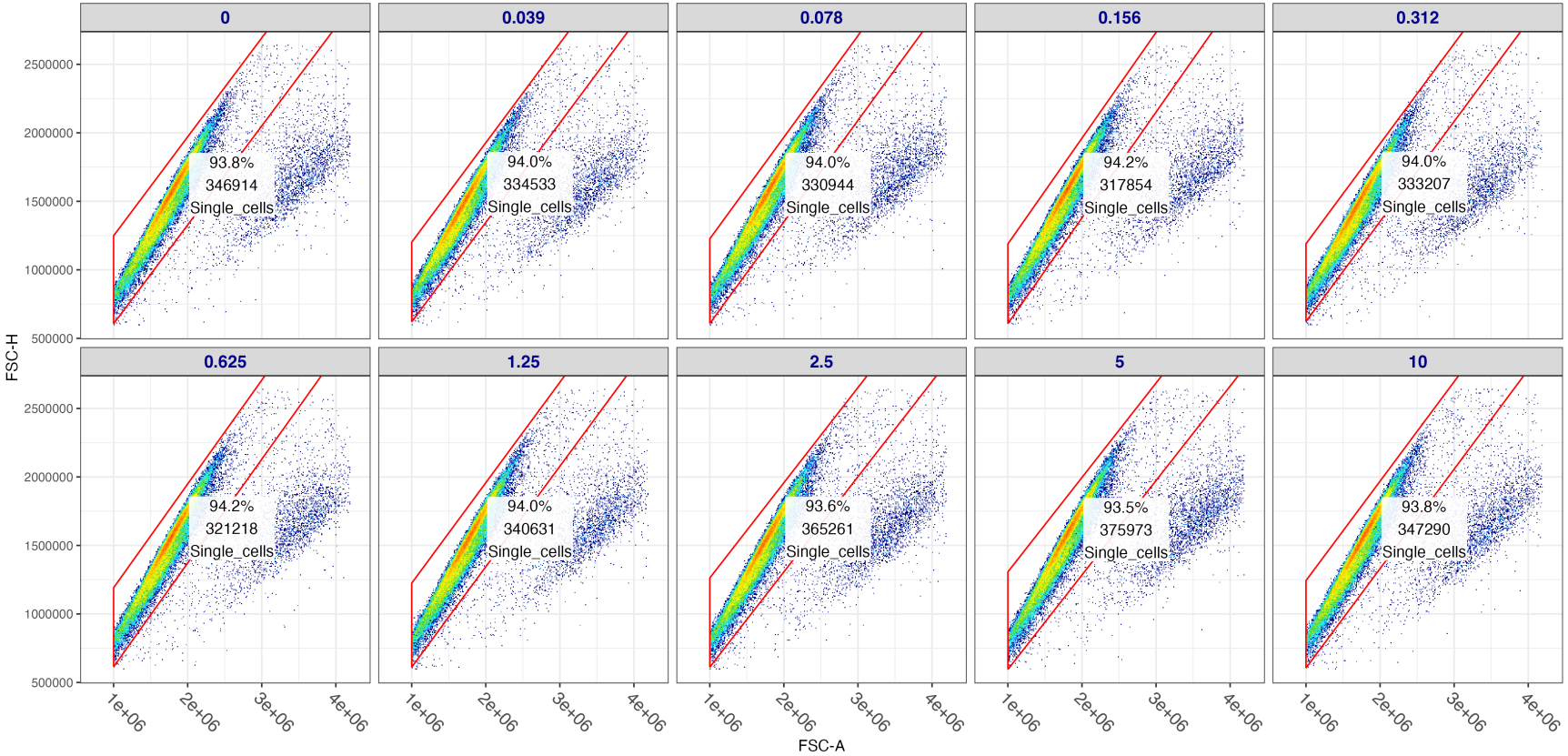
*Parent pop: root | CD62L BV421-A || Cat: 384806 || BioLegend || Lot: B480700 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: boundary || args: min=c(1e6,-Inf), max=c(Inf,Inf) || collapse: TRUE || groupBy: Dirname || pp: — || pp args: —*
*Compensation: —*

**CD62L BV421-A : Single_cells | EXP1460 | JD | Clone W21031N**

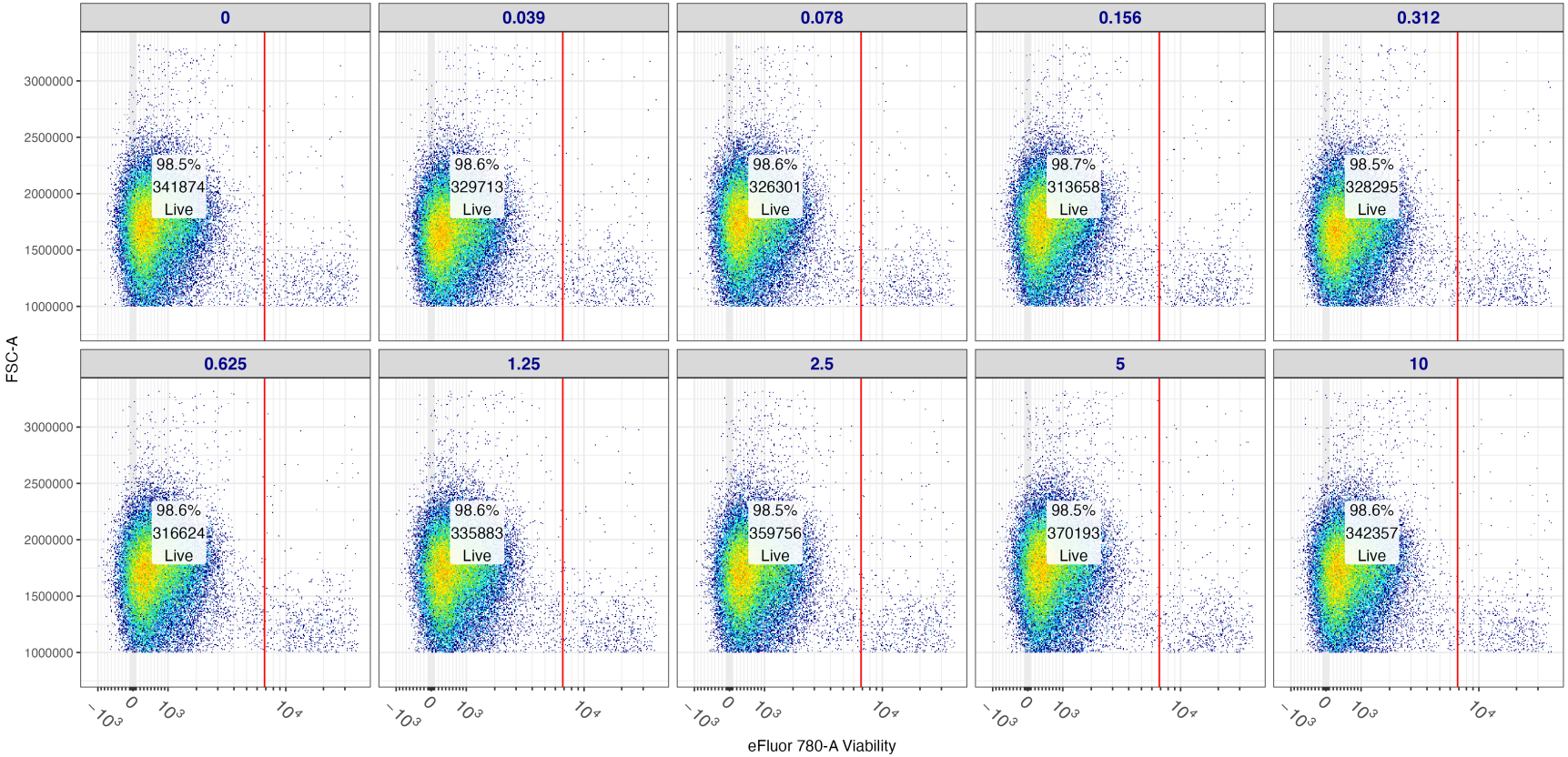
*Parent pop: Cells | CD62L BV421-A || Cat: 384806 || BioLegend || Lot: B480700 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: singletGate || args: prediction_level=0.99, wider_gate=FALSE || collapse: FALSE || groupBy: Dirname || pp: — || pp args: —*
*Compensation: —*

**CD62L BV421-A : Live | EXP1460 | JD | Clone W21031N**

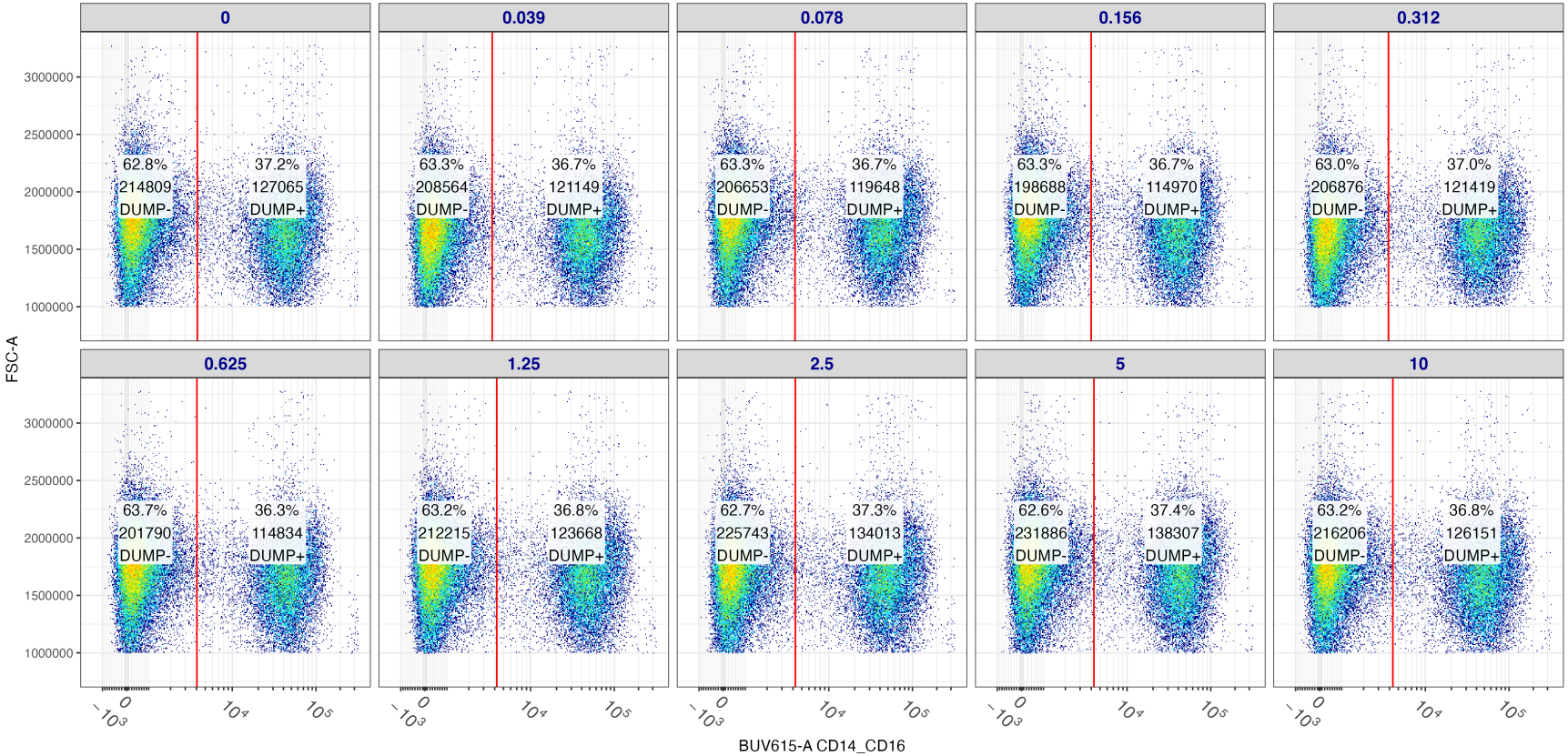
*Parent pop: Single_cells | CD62L BV421-A || Cat: 384806 || BioLegend || Lot: B480700 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: gate_mindensity || args: gate_range=NULL, adjust=1, min=0 || collapse: TRUE || groupBy: Dirname || pp: — || pp args: — Compensation: —*

**CD62L BV421-A : DUMP | EXP1460 | JD | Clone W21031N**

*Parent pop: Live | CD62L BV421-A || Cat: 384806 || BioLegend || Lot: B480700 || Test sample: HD Human PBMC at 1e6/well*




*Gating method: gate_flowmeans || args: K=2, cluster='neg', quantile=0.99 || collapse: FALSE || groupBy: Dirname || pp: — || pp args: —*
*Compensation: —*

# 6 Info

Table 3: Info sheet: script-level settings of this run

| Variable | Value | Source |
|---|---|---|
| **General** | | |
| Template_Version | 3.6.8 | Info sheet |
| working_directory | (not set) | not set |
| compensation | (not set) | not set |
| miflowcyt_export (L) | FALSE | Info sheet |
| dashboard_dir | (not set) | not set |
| titration_export_dir | (not set) | not set |
| dashboard_tabbed_by | (not set) | not set |
| MM_R2_threshold | 0.9 | Info sheet |
| Downsample_to (N) | (not set) | not set |
| flowjo_transformation_arg | channelRange=9000, pos=4.5, neg=0, widthBasis=-10 | Info sheet |
| keywords | (not set) | not set |
| **TiterPubs** | | |
| titerpubs_export | TRUE | Info sheet |
| scrub_confidential | TRUE | Info sheet |
| Organization/Affiliation | (not set) | not set |
| Submitter_Name | (not set) | not set |
| Submitter_ORCID | (not set) | not set |
| Submitter_Email | <redacted-email> | Info sheet |
| Quality_Control_Measures | (not set) | not set |
| Collection_Software | (not set) | not set |
| Target_Species | Human | Info sheet |
| Cell_Type_Fallback | (not set) | not set |
| Sample_Details | (not set) | not set |
| Tissue_Fallback | (not set) | not set |
| References | https://doi.org/10.5281/zenodo.24181307 | Info sheet |
| **Protocol** | | |
| Incubation_Time | 30 | Info sheet |
| Incubation_Temperature_C | 4 | Info sheet |
| **Equipment** | | |
| Equipment_Make | (not set) | not set |
| Equipment_Model | (not set) | not set |
| **Provenance** | | |
| master_panel_inventory_path | Master_Panel_Inventory.xlsx | run folder |
| **Protocol** | | |
| Preparation_Protocol | (not set) | not set |
| Protocol_reference | https://www.biolegend.com/en-us/protocols/intracellular-flow-cytometry-staining-protocol | Info sheet |

Table 3: Info sheet: script-level settings of this run *(continued)*

| Variable | Value | Source |
|---|---|---|
| **TiterPubs** | | |
| scrub_eln_placeholder | EXP1460 | Info sheet |

# 7 Session Information

**Script version:** ACTA v3.6.11

**Render timestamp:** 2026-11-12 20:33:24 PST

**Operator:** JD

**Layout workbook:** 1460_Ab_Titration_Instructions.xlsx

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- Session info -------------------------------------------------------------
 setting  value
 version  R version 4.5.1 (2025-06-13)
 os       macOS Tahoe 26.7.2
 system   aarch64, darwin20
 ui       X11
 language (EN)
 collate  C.UTF-8
 ctype    C.UTF-8
 tz       America/Los_Angeles
 date     2026-11-12
 pandoc   3.9.0.2 @ /opt/homebrew/bin/ (via rmarkdown)
 quarto   NA

- Packages -----------------------------------------------------------------
 package       * version   date (UTC) lib source
 ACTA          * 3.6.11    2026-11-13 [1] Bioconductor
 askpass         1.2.1     2024-10-04 [2] CRAN (R 4.5.0)
 BH            * 1.90.0-1  2025-12-14 [2] CRAN (R 4.5.2)
 Biobase         2.70.0    2025-10-29 [2] Bioconductor 3.22 (R 4.5.1)
 BiocGenerics    0.56.0    2025-10-29 [2] Bioconductor 3.22 (R 4.5.1)
 BiocManager   * 1.30.27   2025-11-14 [2] CRAN (R 4.5.2)
 bslib           0.11.0    2026-05-16 [2] CRAN (R 4.5.2)
 cachem          1.1.0     2024-05-16 [2] CRAN (R 4.5.0)
 cellranger      1.1.0     2016-07-27 [2] CRAN (R 4.5.0)
 cli             3.6.6     2026-04-09 [2] CRAN (R 4.5.2)
 cytolib         2.22.0    2025-10-29 [2] Bioconductor 3.22 (R 4.5.1)
 data.table      1.18.4    2026-05-06 [2] CRAN (R 4.5.2)
 DEoptimR        1.2-0     2026-06-07 [2] CRAN (R 4.5.2)
 digest          0.6.39    2025-11-19 [2] CRAN (R 4.5.2)
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diptest       * 0.77-2    2025-08-20 [2] CRAN (R 4.5.0)
dplyr         * 1.2.1     2026-04-03 [2] CRAN (R 4.5.2)
evaluate        1.0.5     2025-08-27 [2] CRAN (R 4.5.0)
farver          2.1.2     2024-05-13 [2] CRAN (R 4.5.0)
fastmap         1.2.0     2024-05-15 [2] CRAN (R 4.5.0)
feature         1.2.16    2026-05-11 [2] CRAN (R 4.5.2)
flowClust       3.48.0    2025-10-29 [2] Bioconductor 3.22 (R 4.5.1)
flowCore      * 2.22.1    2026-01-01 [2] https://bioc-release.r-universe.dev (R 4.5.2)
flowMeans     * 1.70.0    2025-10-29 [2] Bioconductor 3.22 (R 4.5.1)
flowWorkspace * 4.22.1    2026-01-01 [2] https://bioc-release.r-universe.dev (R 4.5.2)
generics        0.1.4     2025-05-09 [2] CRAN (R 4.5.0)
ggcyto        * 1.39.1    2026-02-25 [2] Github (RGLab/ggcyto@e5ed286)
ggplate       * 0.4.0     2026-09-01 [2] CRAN (R 4.5.2)
ggplot2       * 4.0.3     2026-04-22 [2] CRAN (R 4.5.2)
glue            1.8.1     2026-04-17 [2] CRAN (R 4.5.2)
graph           1.88.1    2025-12-08 [2] Bioconductor 3.22 (R 4.5.2)
gridExtra       2.3       2017-09-09 [2] CRAN (R 4.5.0)
gtable          0.3.6     2024-10-25 [2] CRAN (R 4.5.0)
hexbin          1.28.5    2024-11-13 [2] CRAN (R 4.5.0)
htmltools       0.5.9     2025-12-04 [2] CRAN (R 4.5.2)
jquerylib       0.1.4     2021-04-26 [2] CRAN (R 4.5.0)
jsonlite        2.0.0     2025-03-27 [2] CRAN (R 4.5.0)
kableExtra    * 1.4.0     2024-01-24 [2] CRAN (R 4.5.0)
KernSmooth      2.23-26   2025-01-01 [2] CRAN (R 4.5.1)
knitr         * 1.51      2025-12-20 [2] CRAN (R 4.5.2)
ks              1.15.2    2026-05-09 [2] CRAN (R 4.5.2)
labeling        0.4.3     2023-08-29 [2] CRAN (R 4.5.0)
lattice         0.22-9    2026-02-09 [2] CRAN (R 4.5.2)
lifecycle       1.0.5     2026-01-08 [2] CRAN (R 4.5.2)
lubridate     * 1.9.5     2026-02-04 [2] CRAN (R 4.5.2)
magrittr        2.0.5     2026-04-04 [2] CRAN (R 4.5.2)
Matrix          1.7-5     2026-03-21 [2] CRAN (R 4.5.2)
matrixStats     1.5.0     2025-01-07 [2] CRAN (R 4.5.0)
mclust        * 6.1.2     2025-10-31 [2] CRAN (R 4.5.0)
mvtnorm         1.4-1     2026-06-06 [2] CRAN (R 4.5.2)
ncdfFlow      * 2.56.0    2025-10-29 [2] Bioconductor 3.22 (R 4.5.1)
openCyto      * 2.22.0    2025-10-29 [2] Bioconductor 3.22 (R 4.5.1)
openxlsx      * 4.2.8.1   2025-10-31 [2] CRAN (R 4.5.0)
otel            0.2.0     2025-08-29 [2] CRAN (R 4.5.0)
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pcaPP           2.0-5     2024-08-19 [2] CRAN (R 4.5.0)
pdftools        3.9.0     2026-05-14 [2] CRAN (R 4.5.2)
pillar          1.11.1    2025-09-17 [2] CRAN (R 4.5.0)
pkgconfig       2.0.3     2019-09-22 [2] CRAN (R 4.5.0)
plyr            1.8.9     2023-10-02 [2] CRAN (R 4.5.0)
pracma          2.4.6     2025-10-22 [2] CRAN (R 4.5.0)
purrr           1.2.2     2026-04-10 [2] CRAN (R 4.5.2)
qpdf            1.4.1     2025-07-02 [2] CRAN (R 4.5.0)
R6              2.6.1     2025-02-15 [2] CRAN (R 4.5.0)
ragg          * 1.5.2     2026-03-23 [2] CRAN (R 4.5.2)
RBGL            1.86.0    2025-10-29 [2] Bioconductor 3.22 (R 4.5.1)
RColorBrewer    1.1-3     2022-04-03 [2] CRAN (R 4.5.0)
Rcpp            1.1.1-1.1 2026-04-24 [2] CRAN (R 4.5.2)
readxl        * 1.5.0     2026-05-16 [2] CRAN (R 4.5.2)
Rgraphviz       2.54.0    2025-10-29 [2] Bioconductor 3.22 (R 4.5.1)
rlang           1.2.0     2026-04-06 [2] CRAN (R 4.5.2)
rmarkdown       2.31      2026-03-26 [2] CRAN (R 4.5.2)
robustbase      0.99-7    2026-02-05 [2] CRAN (R 4.5.2)
RProtoBufLib    2.22.0    2025-10-29 [2] Bioconductor 3.22 (R 4.5.1)
rrcov           1.7-7     2025-04-21 [2] CRAN (R 4.5.0)
rstudioapi    * 0.19.0    2026-06-11 [2] CRAN (R 4.5.2)
S4Vectors       0.48.1    2026-04-04 [2] https://bioc-release.r-universe.dev (R 4.5.3)
S7              0.2.2     2026-04-22 [2] CRAN (R 4.5.2)
sass            0.4.10    2025-04-11 [2] CRAN (R 4.5.0)
scales          1.4.0     2025-04-24 [2] CRAN (R 4.5.0)
sessioninfo   * 1.2.4     2026-06-04 [2] CRAN (R 4.5.2)
stringi         1.8.7     2025-03-27 [2] CRAN (R 4.5.0)
stringr       * 1.6.0     2025-11-04 [2] CRAN (R 4.5.0)
svglite         2.2.2     2025-10-21 [2] CRAN (R 4.5.0)
systemfonts     1.3.2     2026-03-05 [2] CRAN (R 4.5.2)
textshaping     1.0.5     2026-03-06 [2] CRAN (R 4.5.2)
this.path     * 2.8.0     2026-04-10 [2] CRAN (R 4.5.2)
tibble          3.3.1     2026-01-11 [2] CRAN (R 4.5.2)
tidyr         * 1.3.2     2025-12-19 [2] CRAN (R 4.5.2)
tidyselect      1.2.1     2024-03-11 [2] CRAN (R 4.5.0)
timechange      0.4.0     2026-01-29 [2] CRAN (R 4.5.2)
vctrs           0.7.3     2026-04-11 [2] CRAN (R 4.5.2)
viridisLite     0.4.3     2026-02-04 [2] CRAN (R 4.5.2)
withr           3.0.2     2024-10-28 [2] CRAN (R 4.5.0)
```

```
 xfun            0.58      2026-06-01 [2] CRAN (R 4.5.2)
 XML             3.99-0.23 2026-03-20 [2] CRAN (R 4.5.2)
 xml2            1.5.2     2026-01-17 [2] CRAN (R 4.5.2)
 yaml            2.3.12    2025-12-10 [2] CRAN (R 4.5.2)
 zip             3.0.0     2026-06-10 [2] CRAN (R 4.5.2)

 [1] ~/ACTA_release/lib3611
 [2] /Library/Frameworks/R.framework/Versions/4.5-arm64/Resources/library
 * -- Packages attached to the search path.

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```