SUPPLEMENTARY FIGURE LEGENDS

Figure E1: Abundance of $CD4^+GATA3^+$ Th2 cells in IgG4-RD patients

(A) Representative plot for GATA-3 expression in $CD4^+CD45RA^-$ gated cells from patients with IgG4-RD compared with a healthy control.

(B) The frequency of $CD4^+GATA\text{-}3^+$ TH2 cells in atopic and non-atopic subsets of IgG4-RD subjects compared to healthy controls (p-values for unpaired t-tests are shown; error bars represent the standard deviation).

Figure E2. Expanded $CD4^+CD27^-CD62L^-$ cells are $CD45RO^+$

CD45RO levels on $CD4^+CD27^-CD62L^-$ from 4 index patients P2, P12, P19 and P25

Fig E3: Abundance of regulatory T cells in peripheral blood of IgG4-RD patients

The numbers of $CD4^+CD45RO^+CD39^+CD25^+Foxp3^+$ Treg cells in peripheral blood of IgG4-RD subjects. P values are based on the Mann-Whitney test.

Figure E4. In vitro culture of $CD4^+$ CTLs from PBMCs of an IgG4-RD subject

Flow-sorted $CD4^+SLAMF7^+$ CTLs from an IgG4-RD patient were stimulated with anti-human CD3 and anti-human CD28 beads in presence of recombinant human IL-2 (20 ng/mL) and their phenotype was checked after 2 weeks of culture.

Figure E5: $CD4^+$ CTLs show surface expression of CD8α

Gating strategy to depict the CD8α expression on $CD4^+T\text{-}bet^+$ CTLs from a representative patient (P25).

Figure E6. CD4+ CTLs from IgG4-RD patients are functional killer cells

The cytotoxicity of in vitro expanded $CD4^+$ CTLs derived from two subjects (P11 and P31) and $CD4^+CD45RO^+$ cells derived from a healthy control against an allogeneic EBV-transformed B cell target cell line was measured after 12 hours of co-culture with or

without anti-CD3 (10 μ g/mL) using Annexin V staining at varying $CD4^+$ CTL: target ratios.

Figure E7. The TCR Vβ repertoire of expanded T_{EM} cells in IgG4-RD

(A) Cumulative distribution of clone frequencies in $CD4^+$ T_{EM} cells from the 4 IgG4-RD subjects and 4 healthy controls in (A**)**. The minimum number of clones accounting for 10% (D10) and 50% (D50) of the clonal diversity are shown in the table below.

(B) Flow cytometry gating scheme used to validate the dominant expanded T_{EM} clones identified by next-generation sequencing (subject P25 shown).

(C) Vβ-specific antibodies were used to compare the frequencies of the dominant T_{EM} clones identified in subjects P25, P28 and P31, marked with a red boundary in (A), with the background frequencies found in controls and non-T_{EM} cells in the respective patients.

Figure E8. Increased frequency of $CD4^+SLAMF7^+$ CTLs in peripheral blood of IgG4-RD patients

Frequency of total $CD4^+SLAMF7^+$ CTLs in IgG4-RD subjects (n = 101) compared to healthy controls (n = 35). **Mann-Whitney test, $p < 0.05$.

Figure E9: Abundance of $CD4^+SLAMF7^+$ CTLs in atopic vs non-atopic IgG4-RD patients

The frequency of $CD4^+SLAMF7^+$ CTLs in atopic and non-atopic subsets of IgG4-RD subjects compared to healthy controls (p-values for unpaired t-tests are shown; error bars represent the standard deviation).

Figure E10. Expansion of $CD4^+SLAMF7^+$ CTLs in disease lesions of IgG4-RD subjects.

(A) Immunofluorescence staining of $CD4^+SLAMF7^+$ CTLs in the affected tissues of IgG4-RD subjects (lacrimal gland biopsy from P3, lymph node biopsy from P11, submandibular salivary gland P25, laryngeal biopsy from P27, nasal palate biopsy from P31, retroperitoneal biopsy from P40 and nasal septum biopsy from P43). CD4 (red), DAPI (blue) and SLAMF7 (green) staining are shown.
(B) Immunofluorescence staining of an affected submandibular salivary gland from an IgG4RD subject (P25) for CD4, Vβ17 (red) and SLAMF7 (green) shown both individually and as an overlay.

Figure E11. $CD4^+SLAMF7^+$ CTLs express granzyme A in disease lesions of IgG4-RD subjects.

(A) Immunofluorescence staining for granzyme A on $CD4^+SLAMF7^+$ CTLs in the affected tissues of IgG4-RD subjects (lymph node biopsy from P11, nasal palate biopsy from P31, retroperitoneal biopsy from P32). CD4 (red), DAPI (blue), SLAMF7 (green) and Granzyme A (magenta) stainings are shown.

Figure E12. Expanded $CD4^+$ CTLs clones from IgG4-RD patients secrete IFN-γ

Intracellular staining for IFN-γ and IL-4 in expanded clones of $CD4^+$ CTLs identified using Vβ and T-bet staining from two IgG4-RD patients after restimulation with PMA (100 ng/mL) and ionomycin (100 ng/mL).

Figure E13. Expanded $CD4^+$ CTLs from IgG4-RD patients secrete IL-1β.

IL-1β-producing $CD4^+$ cells in the tissues of IgG4-RD subjects and controls. CD4 (red), DAPI (blue) and IL-1β (green) staining are shown.

Figure E14. Expanded $CD4^+$ CTLs from IgG4-RD patients secrete TGF-s a

TGF- β 1-producing $CD4^+GzmA^+$ cells in the salivary gland tissues of 5 IgG4-DS subjects and 5 controls (SS patients). CD4 (red), TGF- β 1(white), DAPI (blue) and Granzyme A (green) staining are shown.

Figure E15. The rituximab target CD20 is not expressed on $CD4^+$ $SLAMF7^+$ CTLs

Flow cytometry revealed no detectable CD20 on CD4+CTLs. Three subjects (P25, P27 and P49 are shown).

Figure E16. Expansion of $CD4^+SLAMF7^+$ CTLs in subjects with systemic sclerosis.

Expansion of $CD4^+SLAMF7^+$ CTLs in patients with systemic sclerosis (n = 17). Patients with diffuse scleroderma (n = 8) and limited scleroderma (n = 9) are also shown separately. Boxplots display the 25^{th} to 75^{th} percentiles. P values are based on the Mann-Whitney test.

SUPPLEMENTARY TABLE E1:
Clinical features of scleroderma patients studied.

Type	Time since diagnosis	Organ involvement	Treatment	Serologies
Diffuse	40 months	Raynaud's, interstitial lung disease, skin	Mycophenolate mofetil	ANA (pattern unknown), RNP, Scl70
Diffuse	12 months	Skin, esophageal dysfunction, GERD, sclerodactyly	Hydroxychloroquine	ANA (speckled), Ro, La
Limited	120 months	Raynaud's, cutaneous & lingual telangiectasias, digital ulcers, bowel hypomotility	Calcium channel blockers, sildenafil	ANA (centromere)
Diffuse	108 months	Erosive synovitis (RA), sclerodactyly, telangiectasias, myopericarditis, restrictive lung disease	Methotrexate	ANA (nuclear), Ro, RF, CCP
Limited	126 months	Raynaud's, gastric antral vascular ectasia, facial telangiectasias, microscopic colitis	None	ANA (centromere)
Limited	84 months	Raynaud's, esophageal dysfunction, GERD	None	ANA (centromere), dsDNA (low titer)
Diffuse	36 months	Raynaud's, sclerodactyly, interstitial lung disease, inflammatory myopathy	Glucocorticoids, azathioprine	ANA (nucleolar)
Diffuse	60 months	Interstitial lung disease, mild pulmonary hypertension, skin thickening, sclerodactyly, capillary loop dropout	None	ANA (speckled)
Limited	180 months	Raynaud's, facial telangiectasias	Hydroxychloroquine	ANA (centromere)
Limited	36 months	Interstitial lung disease, GERD		ANA (centromere)
Limited	132 months	morphea, secondary Sjogren's	None	ANA (centromere)
Limited	24 months	Raynaud's, cutaneous telangiectasias	None	ANA (centromere)
Limited	Unknown	Raynaud's, narrowing of oral aperture, sclerodactyly	None	ANA (speckled)
Diffuse	4 months	Skin thickening	Methotrexate	ANA (homogeneous)
Limited	84 months	Raynaud's, calcinosis cutis, telangiectasias, esophageal dysmotility	None	ANA (centromere)

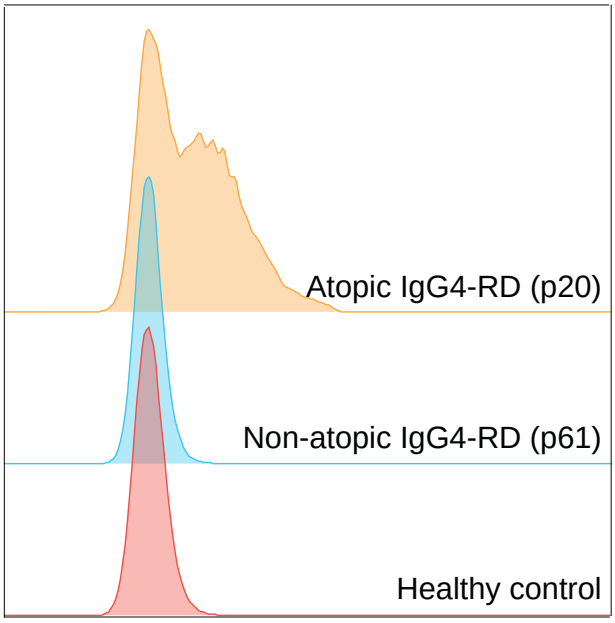
Diffuse	24 months	Diffuse skin thickening, sclerodactyly, Raynaud's, hypopigmentation, gastrointestinal dysmotility, diffuse rash, photosensitivity, alopecia,	Hydroxychloroquine, prednisone, azathioprine	ANA (pattern unknown)

286
287

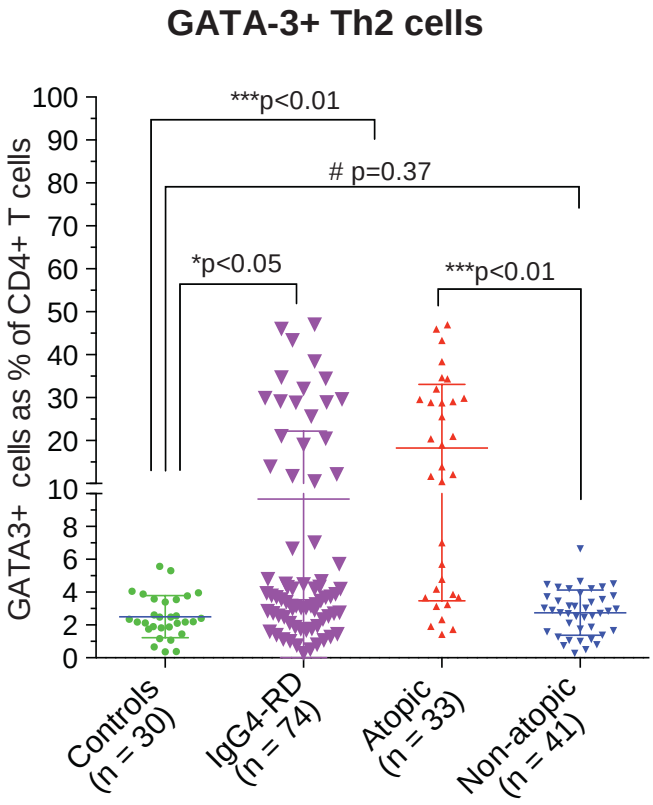
Supplementary table E2. Categorization of IgG4-RD patients into Atopic vs Non-atopic.

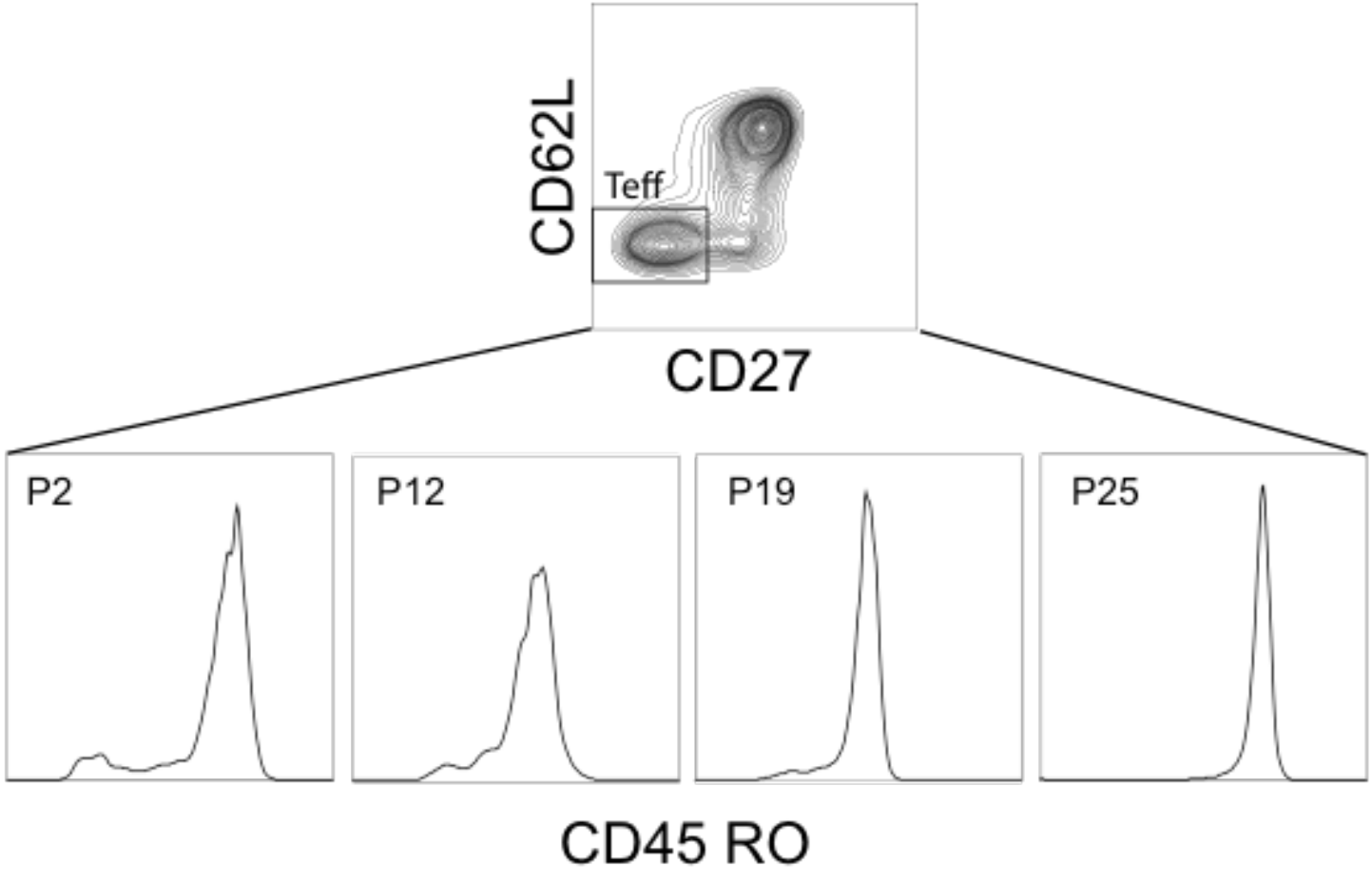
	Atopic patients (n=33)	Non-atopic patients (n=41)	All patients (n=74)
Mean age in years (range)	57.9 (30-84)	62.75 (33-88)	60.6 (30-88)
Male / Female ratio	2.3	1.732	2.1
Atopic symptoms, n (%)			
Rhinitis	18 (55%)		
Conjunctivitis	5 (7%)		
Asthma	9 (27%)		
Hives	3 (12%)		
Oral allergic syndrome	1 (3%)		
Eczema	2 (6%)		
Hay fever	2 (6%)		
Anaphylaxis	0		
Laboratory analysis, mean (range)			
Eosinophils (cells/mL) (< 500 cells/μL)	615 (40-1850)	316 (10-2000)	445 (10-1850)
Serum IgE (IU/mL) (< 100 IU/mL)	446 (5-1860)	236 (5-4570)	333 (5-1860)
Serum IgG4 (mg/dL) (< 121 mg/dL)	716 (11-4780)	273 (3-2200)	471 (3-4780)
Eosinophilia	(15/33)(45%)	(7/41)(17%)	(22/74)(17%)
Elevated IgE	(20/33)(61%)	(6/41)(15%)	(26/74)(15%)
Elevated IgG4	(26/33)(79%)	(12/41)(29%)	(38/74)(29%)

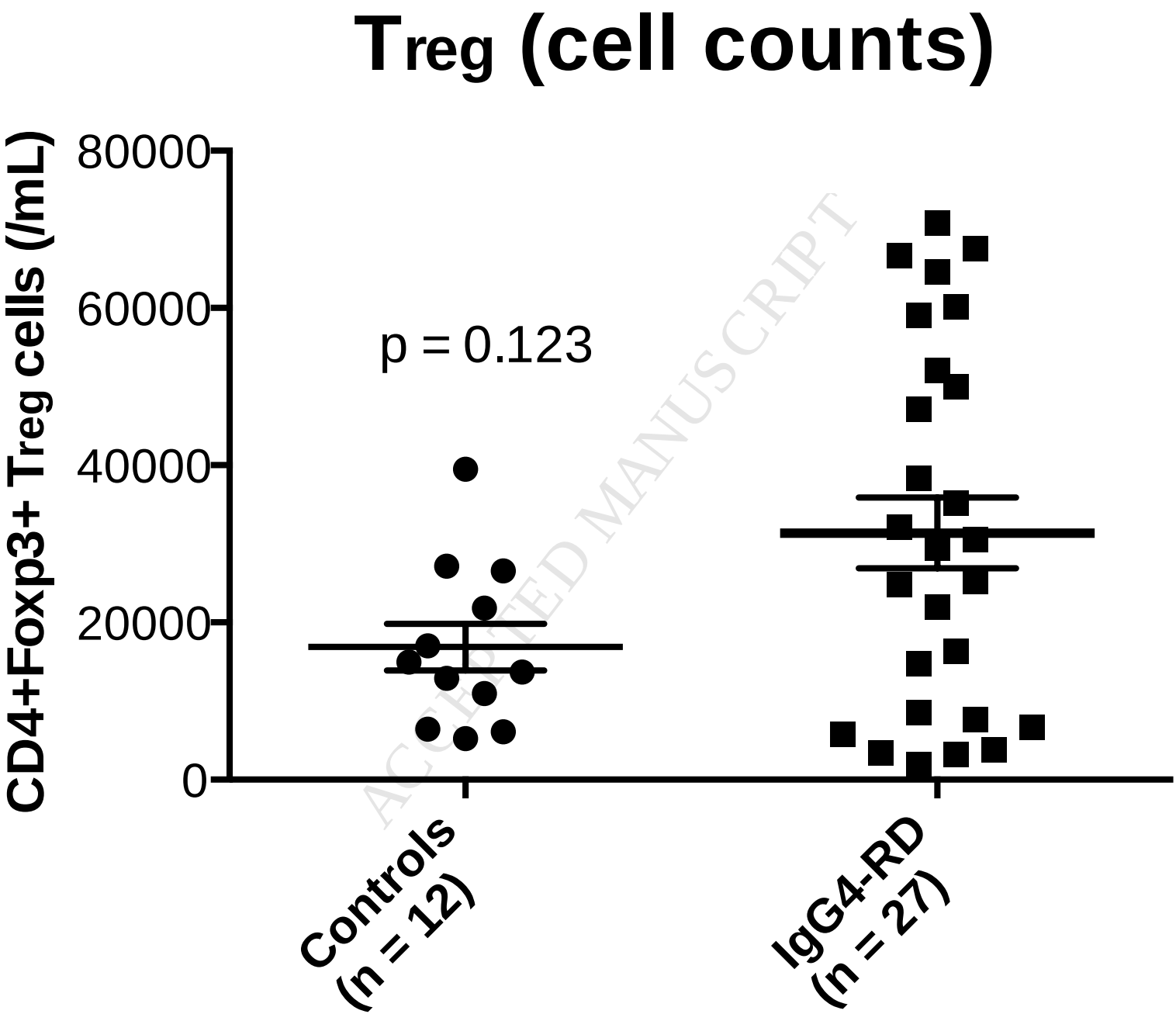
A

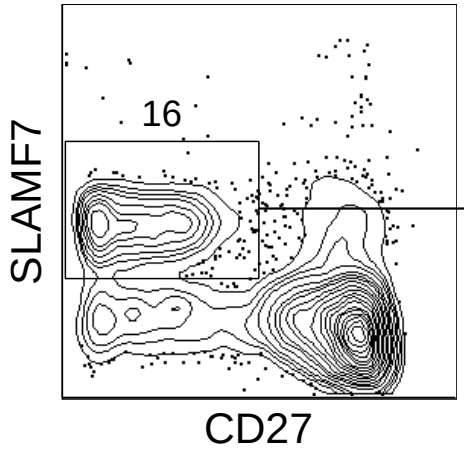

CD4+CD45RA- gated
Atopic IgG4-RD (p20)
Non-atopic IgG4-RD (p61)
Healthy control

B


GATA-3+ Th2 cells
GATA3+ cells as % of CD4+ T cells
100
90
80
70
60
50
40
30
20
10
8
6
4
2
0
***p<0.01
p=0.37
*p<0.05
***p<0.01
Controls (n = 30)
IgG4-RD (n = 74)
Atopic (n = 33)
Non-atopic (n = 41)


CD62L
Teff
CD27
P2
P12
P19
P25
CD45 RO





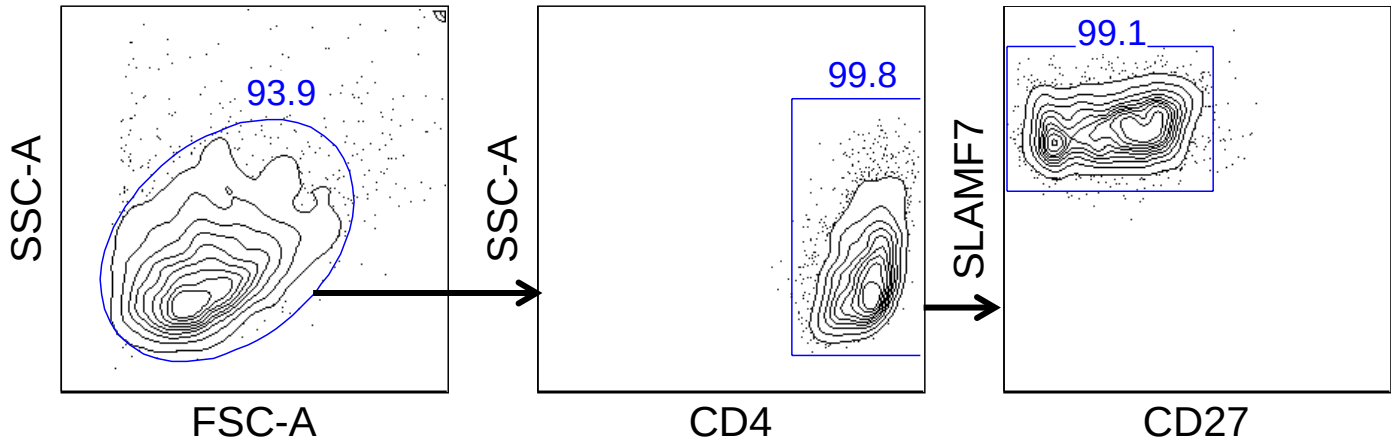
FACS SORT

↓

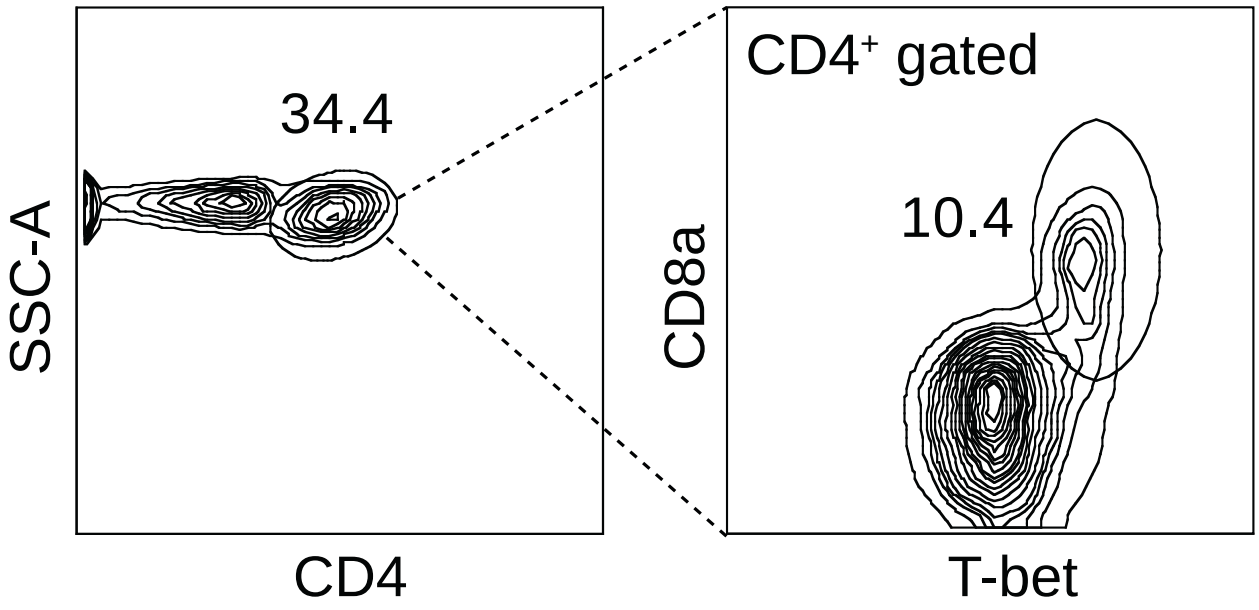
αCD3 + αCD28

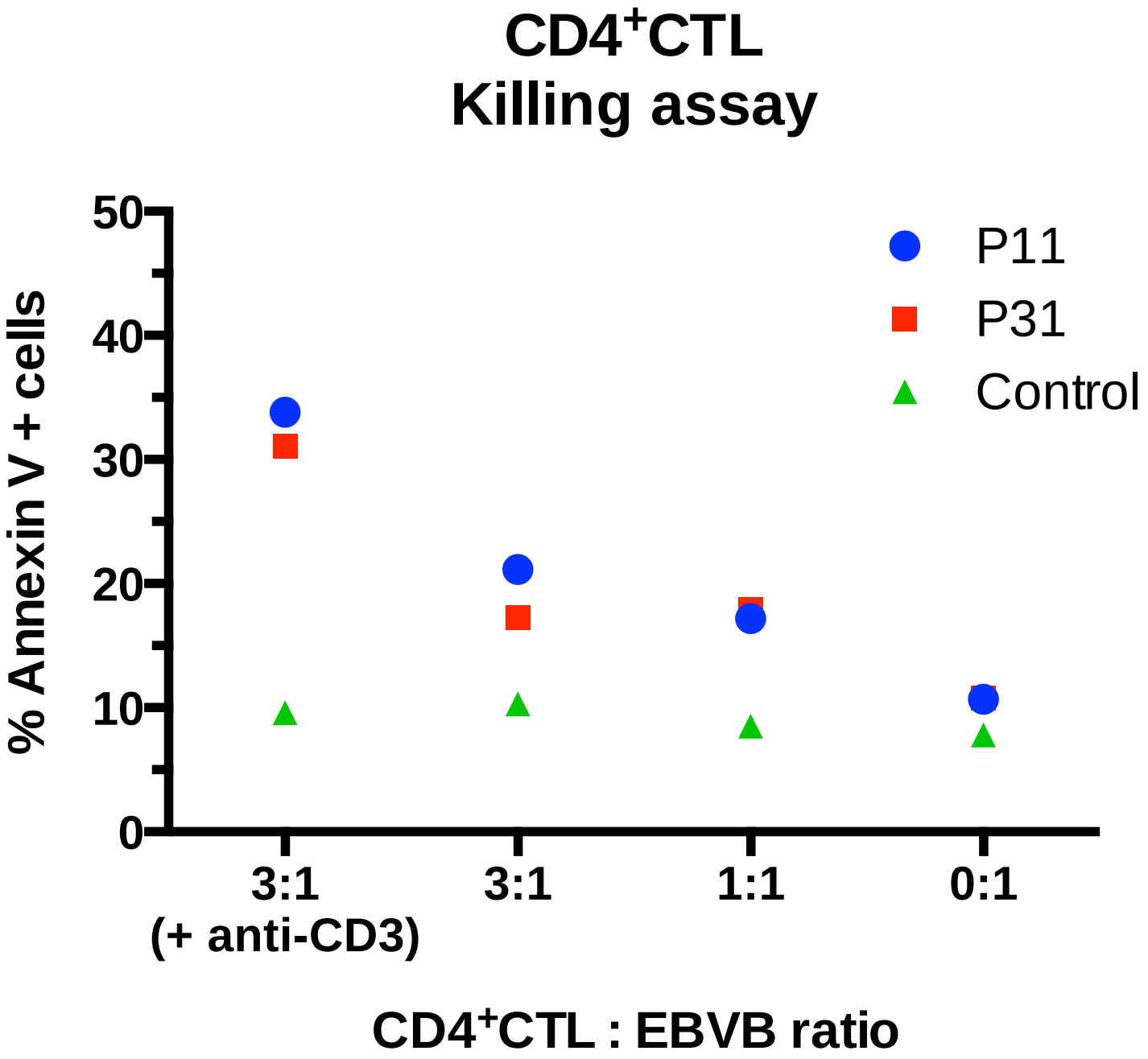
↓

2 WEEK CULTURE IN IL-2

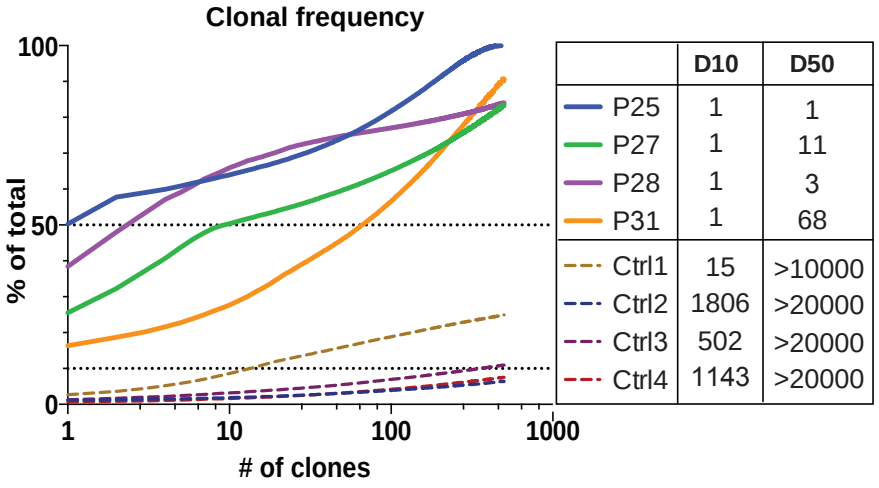


~ 50 FOLD INCREASE IN CELL NUMBER


34.4
SSC-A
CD4
CD4+ gated
10.4
CD8a
T-bet

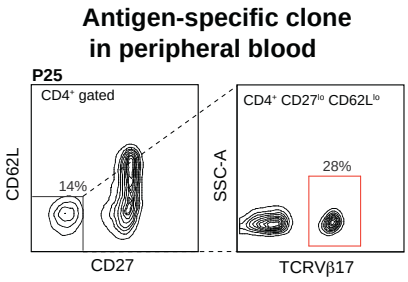

CD4+CTL
Killing assay
% Annexin V + cells
50
40
30
20
10
0
P11
P31
Control
3:1
(+ anti-CD3)
3:1
1:1
0:1
CD4+CTL : EBVB ratio

A

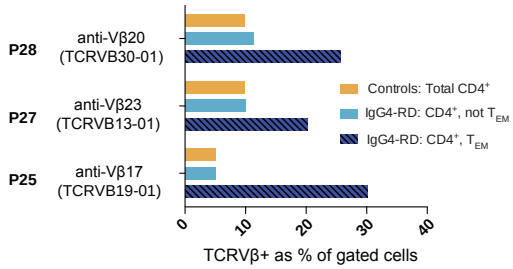


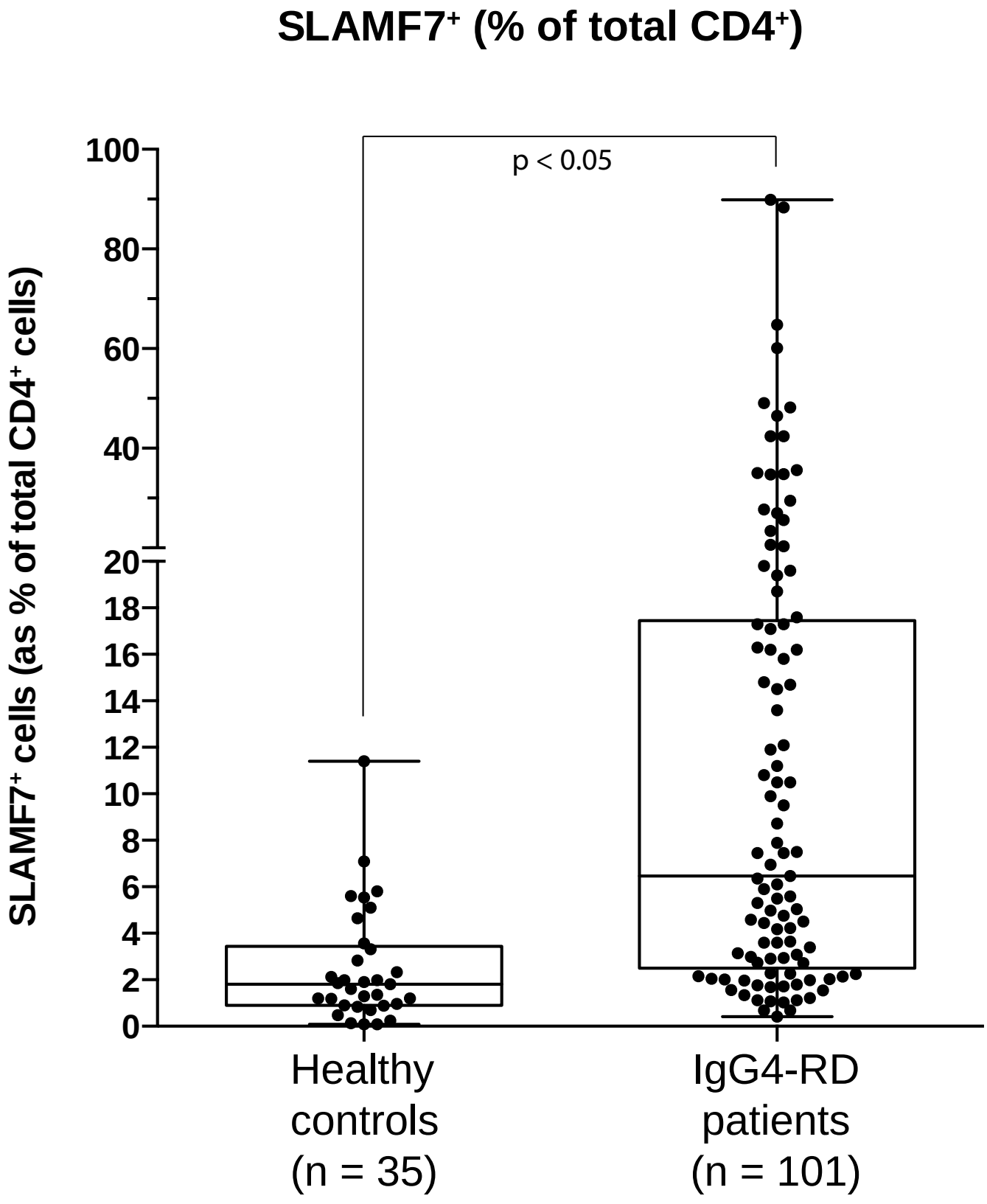
	D10	D50
P25	1	1
P27	1	11
P28	1	3
P31	1	68
Ctrl1	15	>10000
Ctrl2	1806	>20000
Ctrl3	502	>20000
Ctrl4	1143	>20000

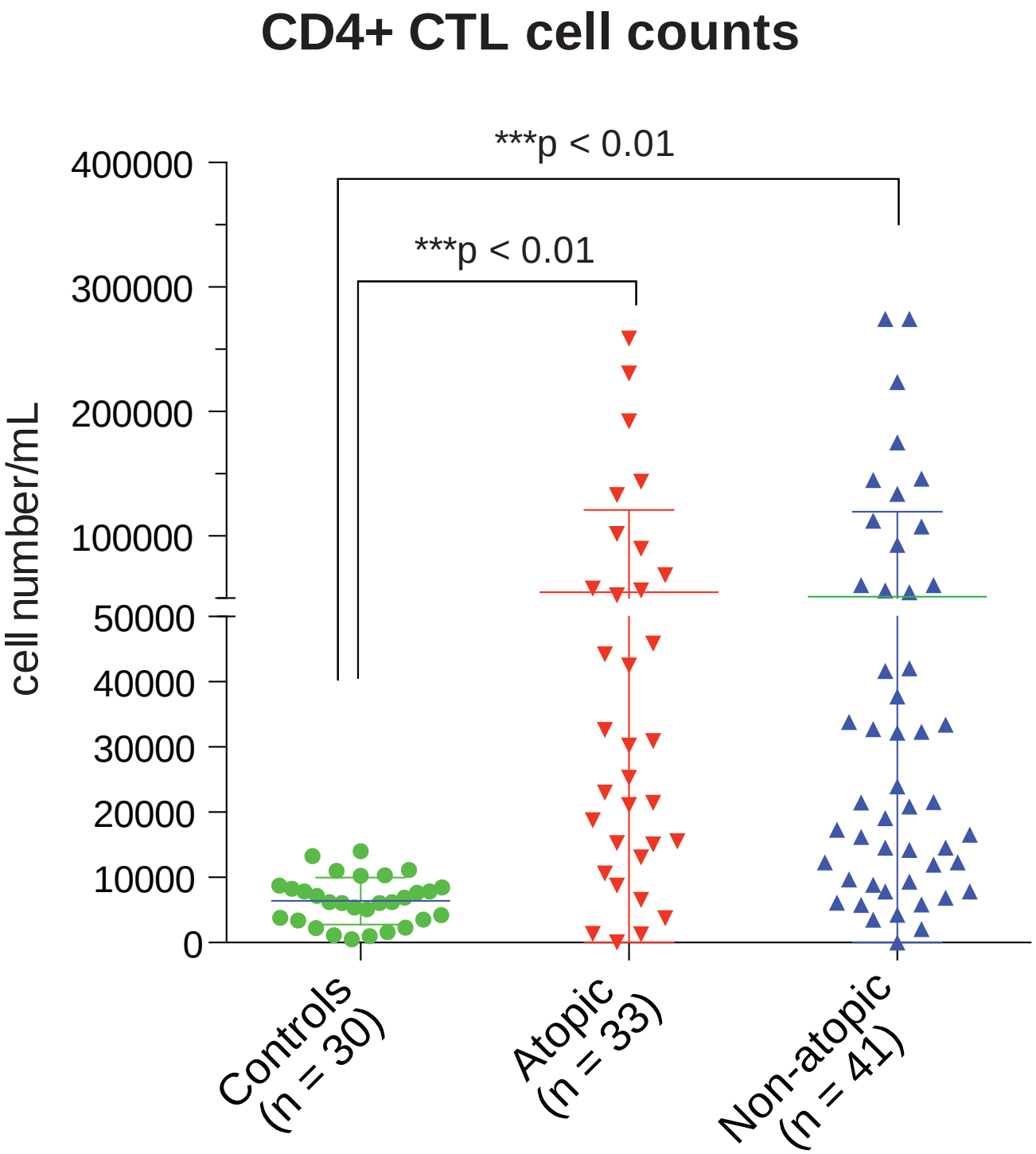
B

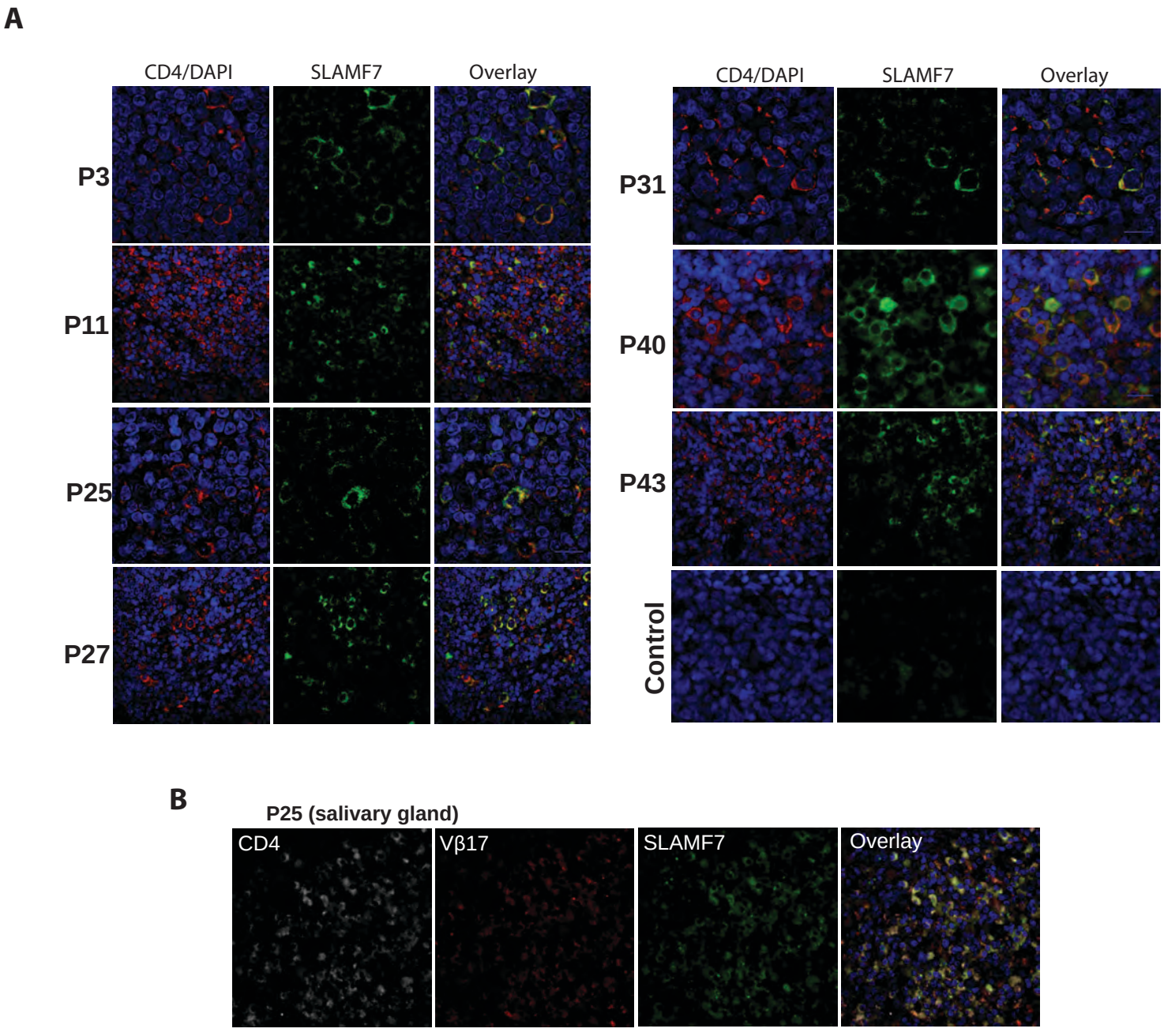


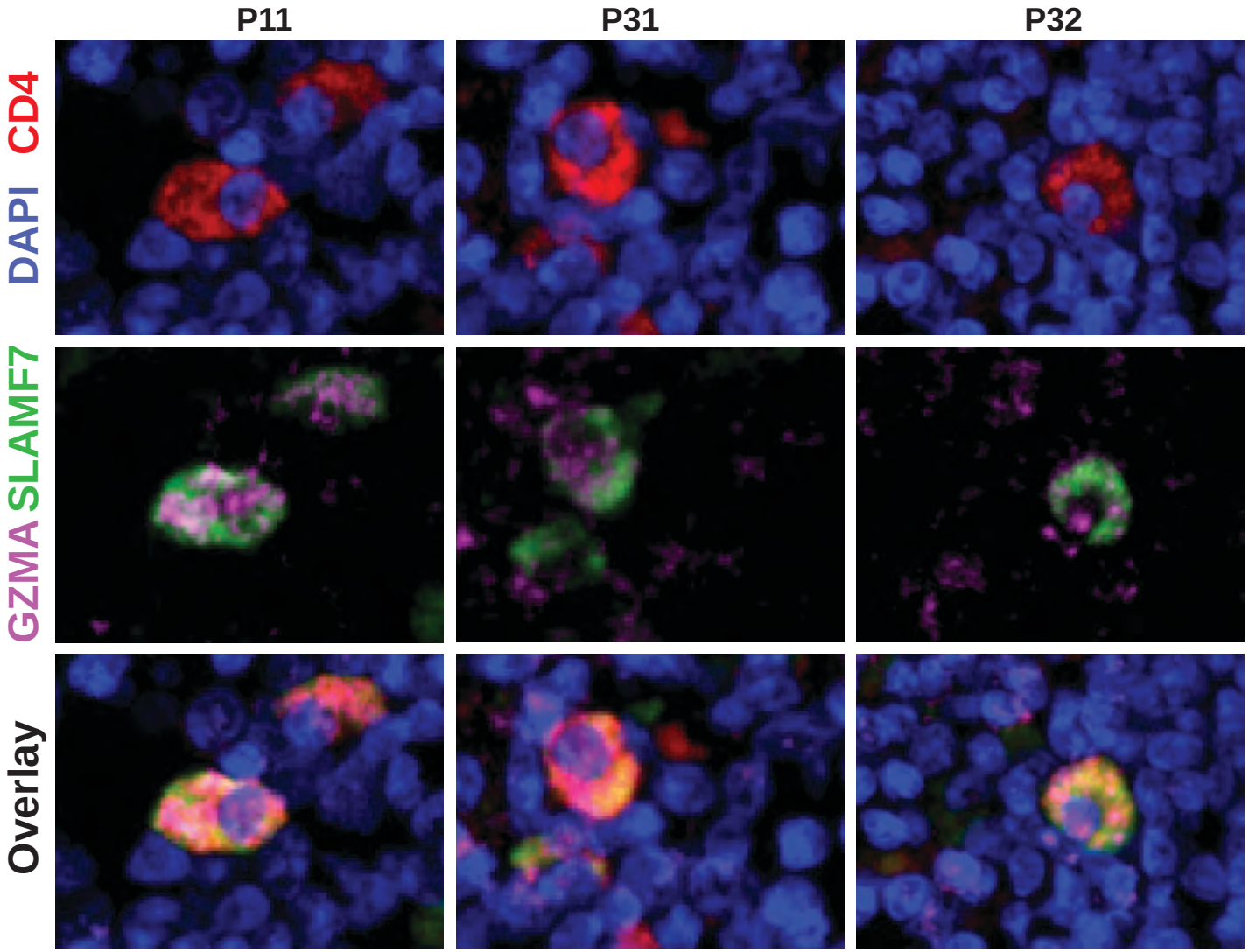
C

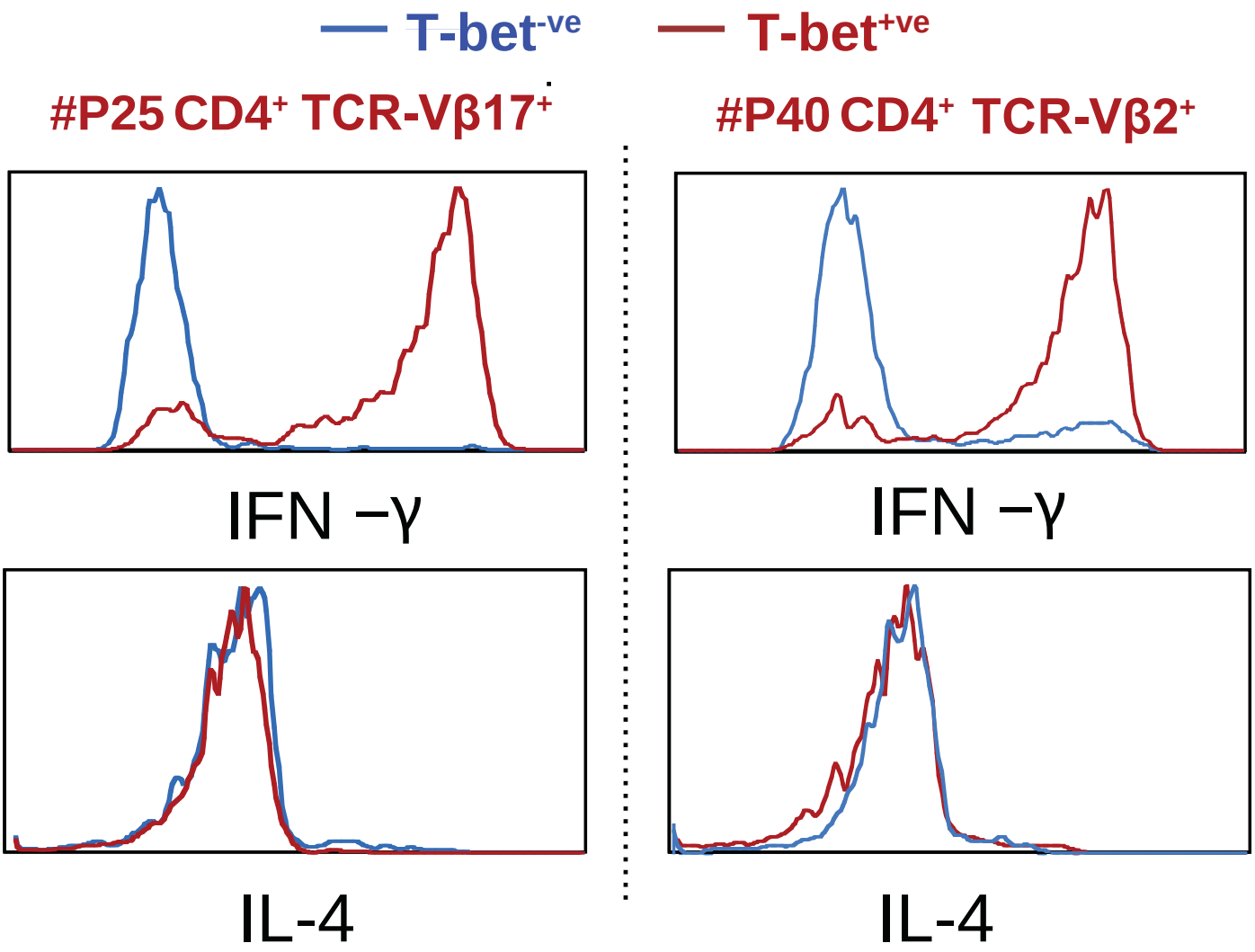


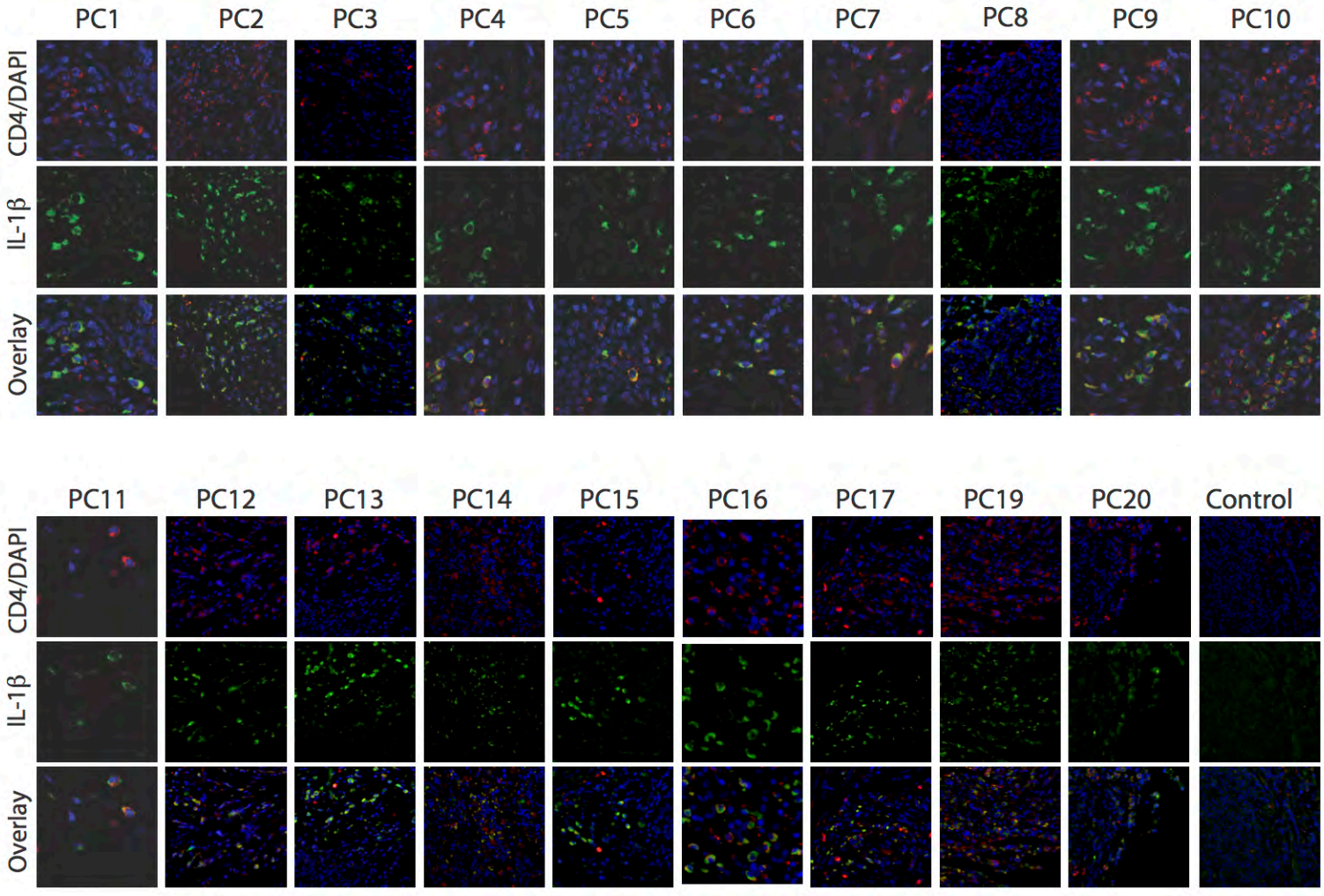

SLAMF7+ (% of total CD4+)
SLAMF7+ cells (as % of total CD4+ cells)
p < 0.05
100
80
60
40
20
18
16
14
12
10
8
6
4
2
0
Healthy controls (n = 35)
IgG4-RD patients (n = 101)

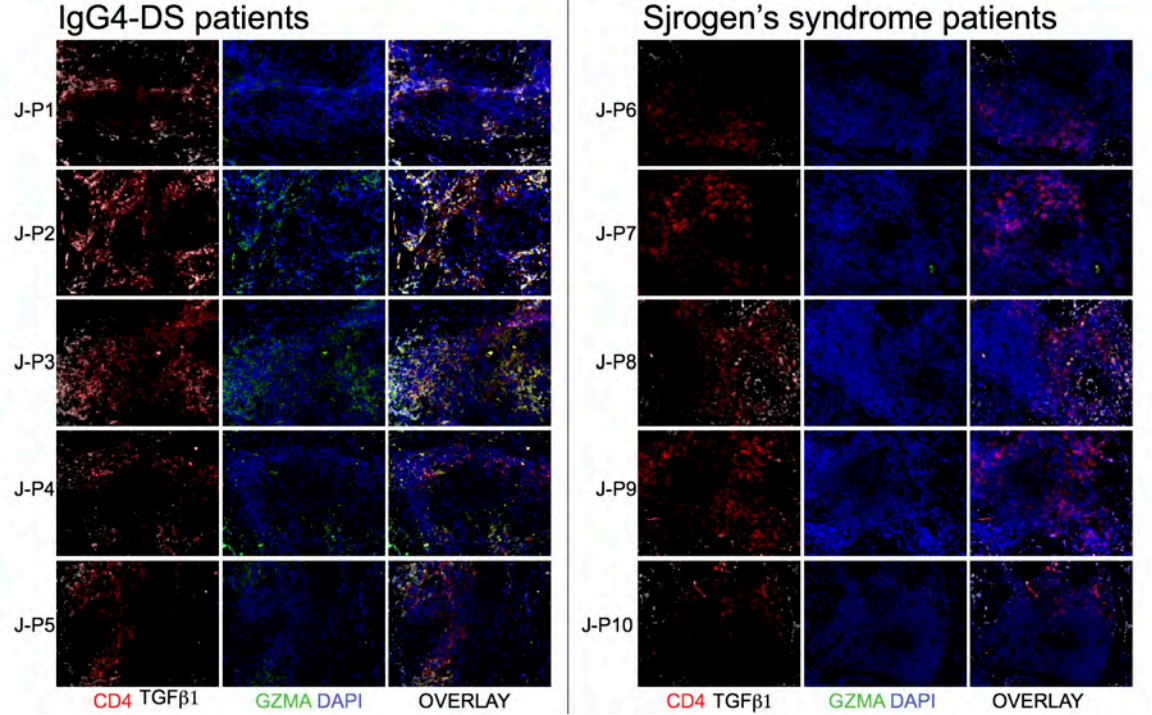

CD4+ CTL cell counts
***p < 0.01
***p < 0.01
cell number/mL
400000
300000
200000
100000
50000
40000
30000
20000
10000
0
Controls (n = 30)
Atopic (n = 33)
Non-atopic (n = 41)

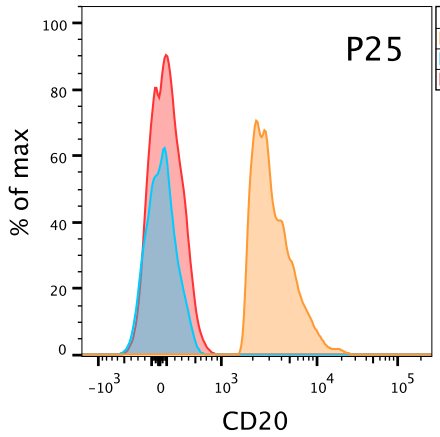

A
CD4/DAPI
SLAMF7
Overlay
P3
P11
P25
P27
P31
P40
P43
Control
B
P25 (salivary gland)
CD4
Vβ17
SLAMF7
Overlay


P11
P31
P32
DAPI CD4
GZMA SLAMF7
Overlay

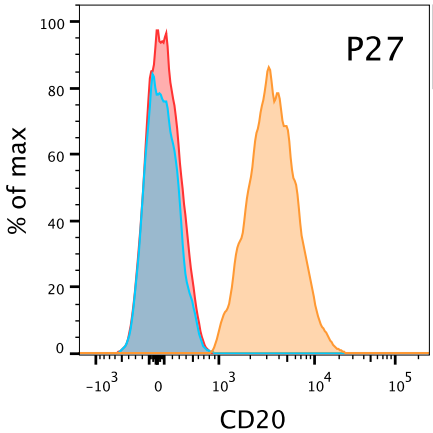

T-bet-ve
T-bet+ve
#P25 CD4+ TCR-Vβ17+
#P40 CD4+ TCR-Vβ2+
IFN −γ
IFN −γ
IL-4
IL-4


PC1
PC2
PC3
PC4
PC5
PC6
PC7
PC8
PC9
PC10
CD4/DAPI
IL-1β
Overlay
PC11
PC12
PC13
PC14
PC15
PC16
PC17
PC19
PC20
Control
CD4/DAPI
IL-1β
Overlay

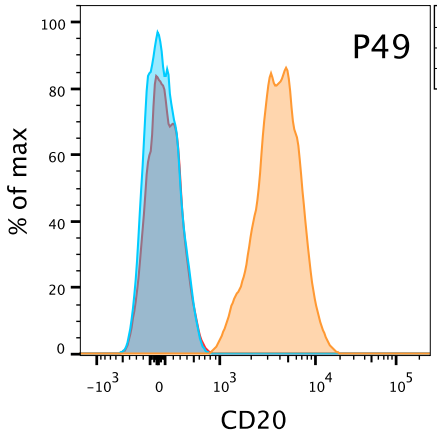

IgG4-DS patients
Sjrogen's syndrome patients
J-P1
J-P2
J-P3
J-P4
J-P5
J-P6
J-P7
J-P8
J-P9
J-P10
CD4 TGFβ1
GZMA DAPI
OVERLAY
CD4 TGFβ1
GZMA DAPI
OVERLAY



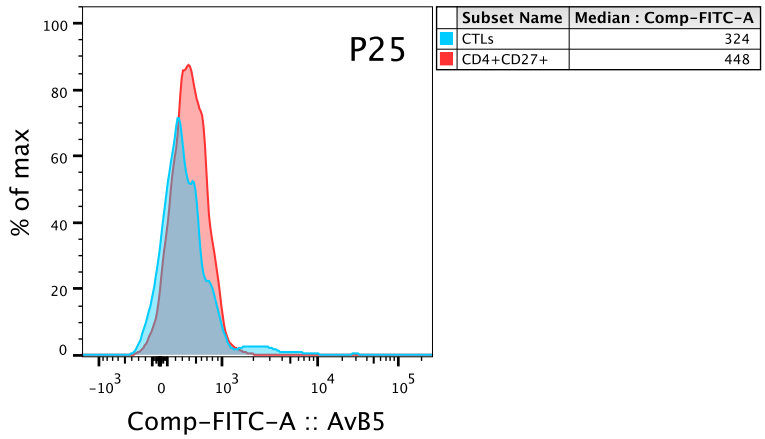
	Subset Name	Median : Comp-FITC-A
	B cells	1664
	CTLs	324
	CD4+CD27+	448



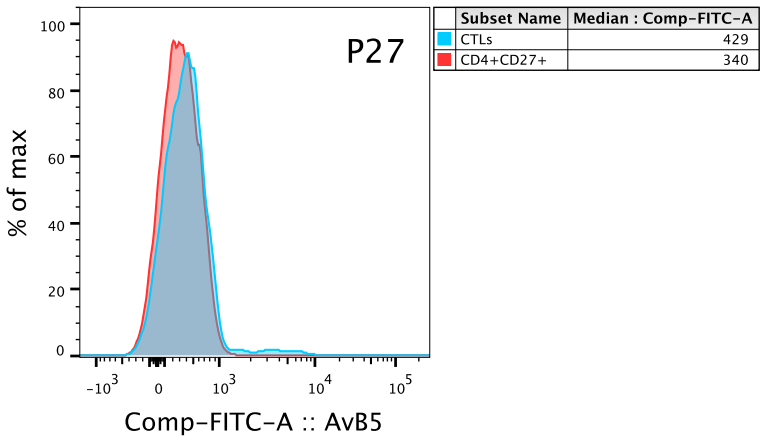
	Subset Name	Median : Comp-FITC-A
	B cells	1168
	CTLs	429
	CD4+CD27+	340



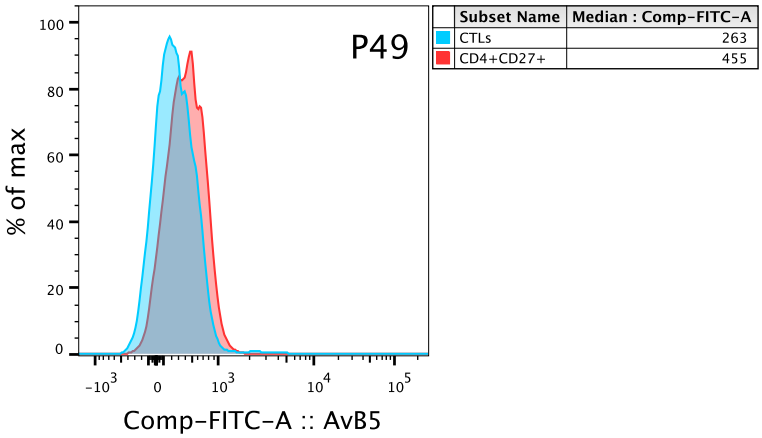
	Subset Name	Median : Comp-FITC-A
	B cells	1112
	CTLs	263
	CD4+CD27+	455



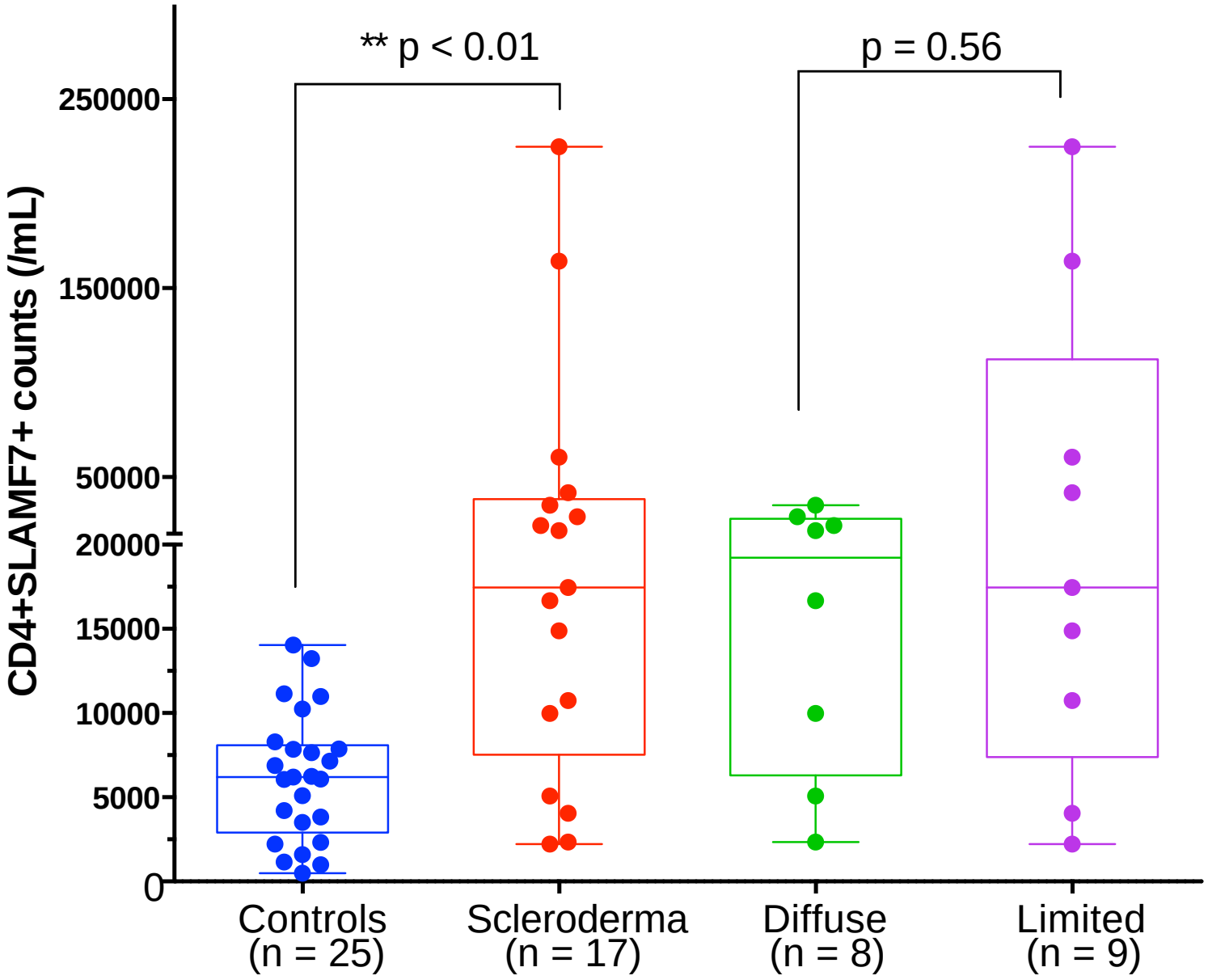
Subset Name	Median : Comp-FITC-A
CTLs	324
CD4+CD27+	448



Subset Name	Median : Comp-FITC-A
CTLs	429
CD4+CD27+	340



Subset Name	Median : Comp-FITC-A
CTLs	263
CD4+CD27+	455


** p < 0.01
p = 0.56
CD4+SLAMF7+ counts (/mL)
250000
150000
50000
20000
15000
10000
5000
0
Controls
(n = 25)
Scleroderma
(n = 17)
Diffuse
(n = 8)
Limited
(n = 9)