Centre d'Immunologie et des Maladies Infectieuses

Ontogeny, plasticity and physicochemical properties of human T cells

Martin LARSEN

www.Immulab.fr

INSERM U1135, CHU Pitié-Salpetrière, Paris, France

T-cell development in thymus

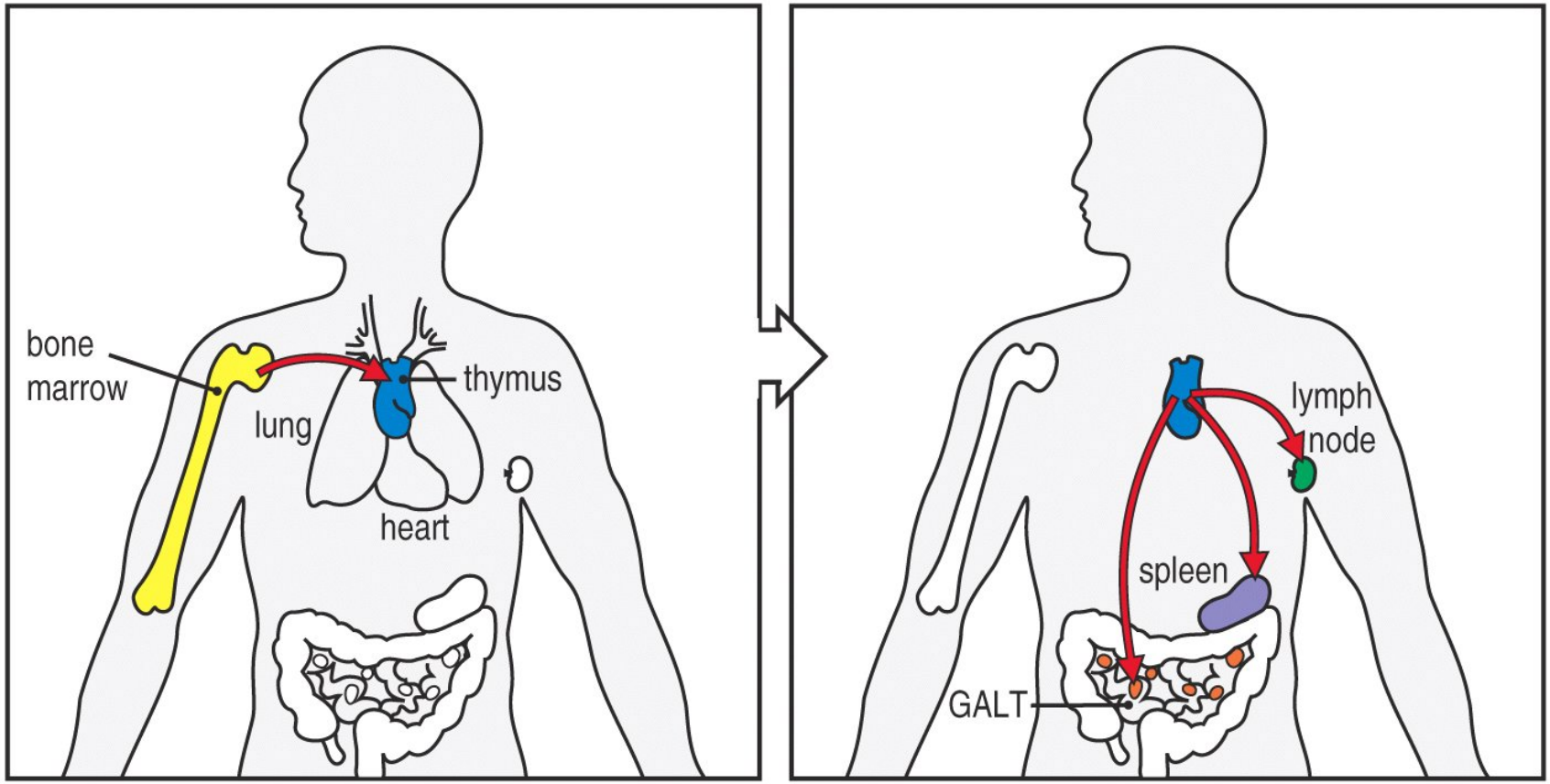


Figure 5-1 The Immune System, 2/e (© Garland Science 2005)

Outline of T-cell Development and Maturation

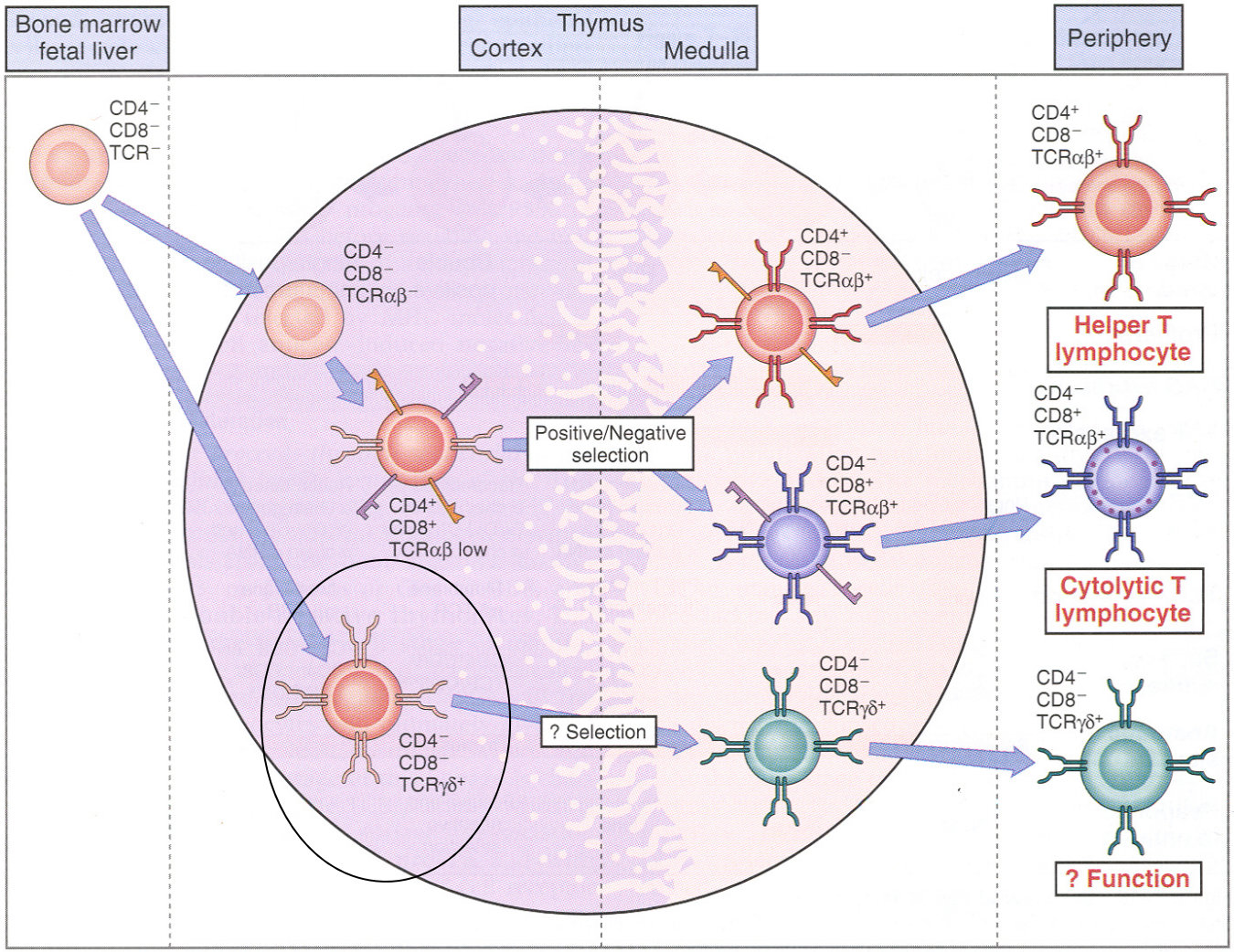
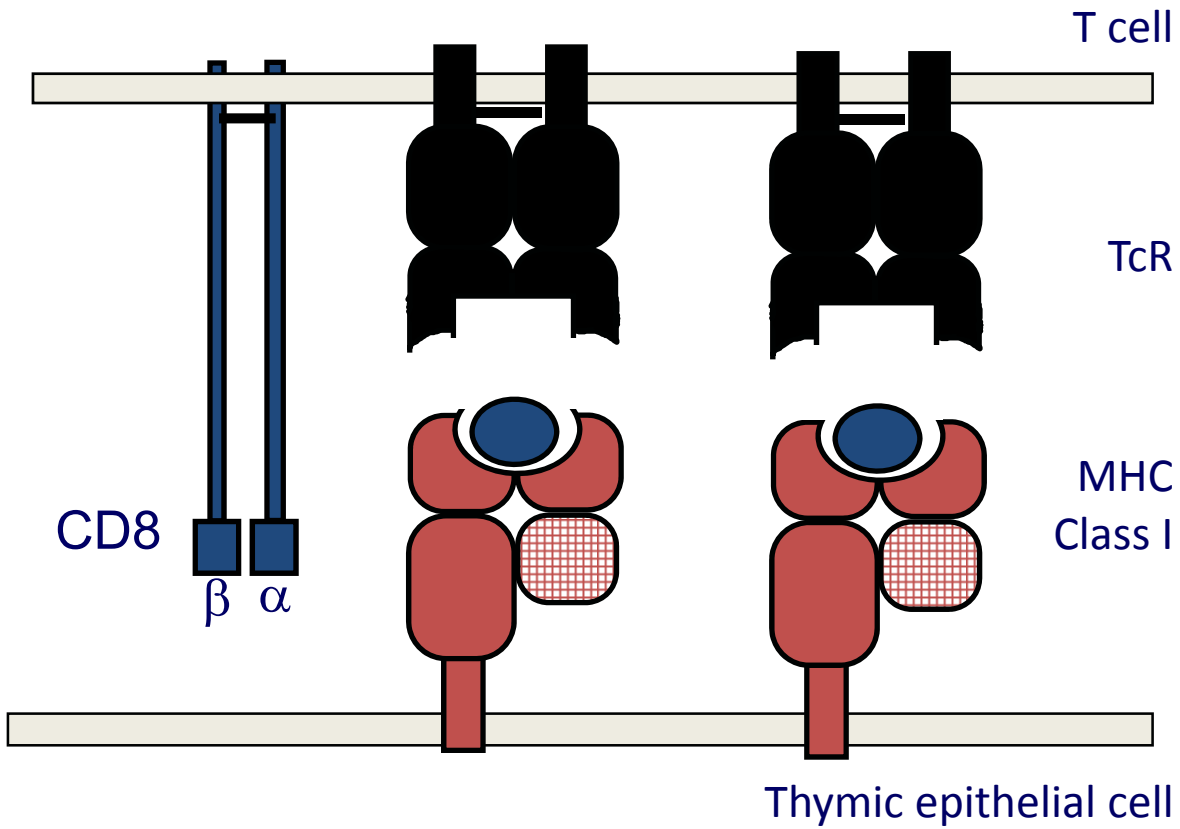


Figure 7–17 Stages of T cell maturation.
Events corresponding to each stage of T cell maturation from a bone marrow stem cell to a mature T lymphocyte are illustrated. Several surface markers in addition to those shown in the figure have been used to define distinct stages of T cell maturation. TCR, T cell receptor.

Removal of useless cells

Peptide is not recognised or irrelevant
Thymocyte receives no signal, fails to be positively selected and dies by apoptosis.

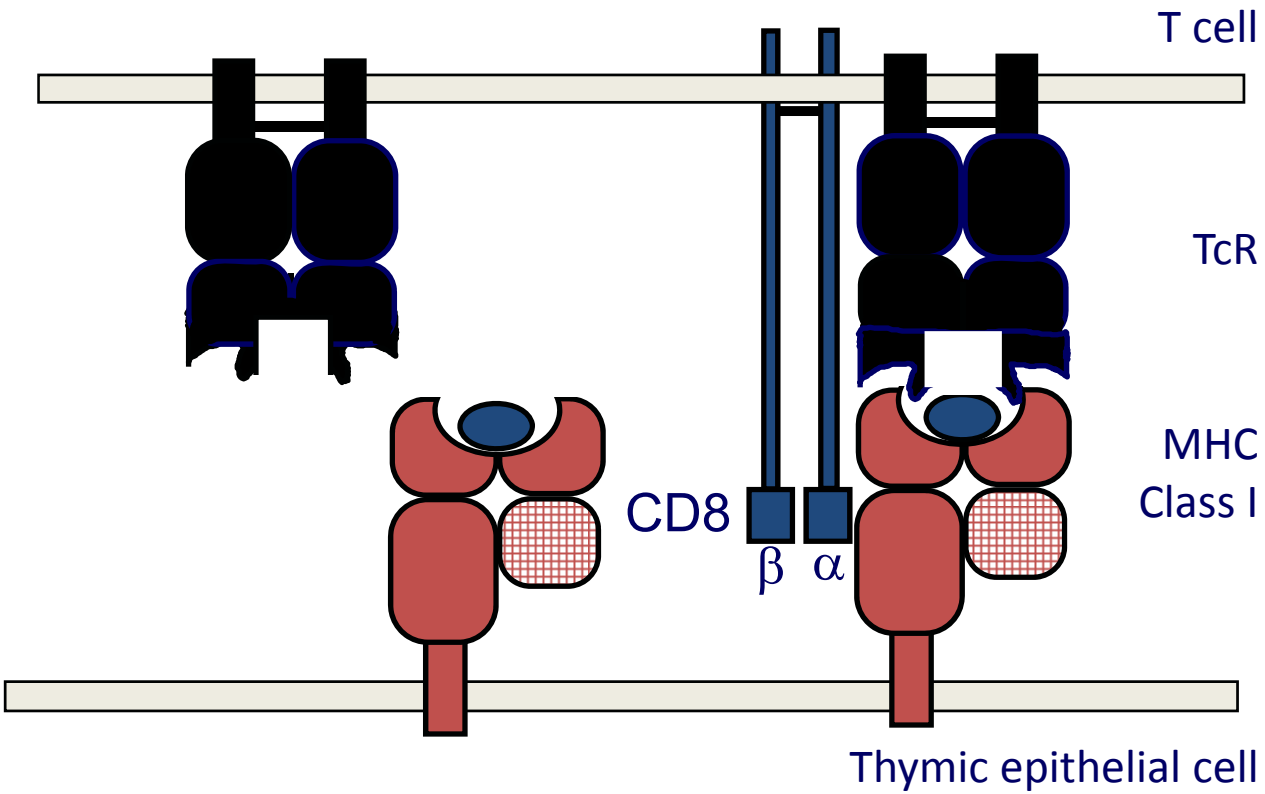
WEAK OR NO SIGNAL



Positive selection

Peptide is a partial agonist
Thymocyte receives a partial signal and is rescued from apoptosis
i.e. the cell is positively selected to survive and mature.

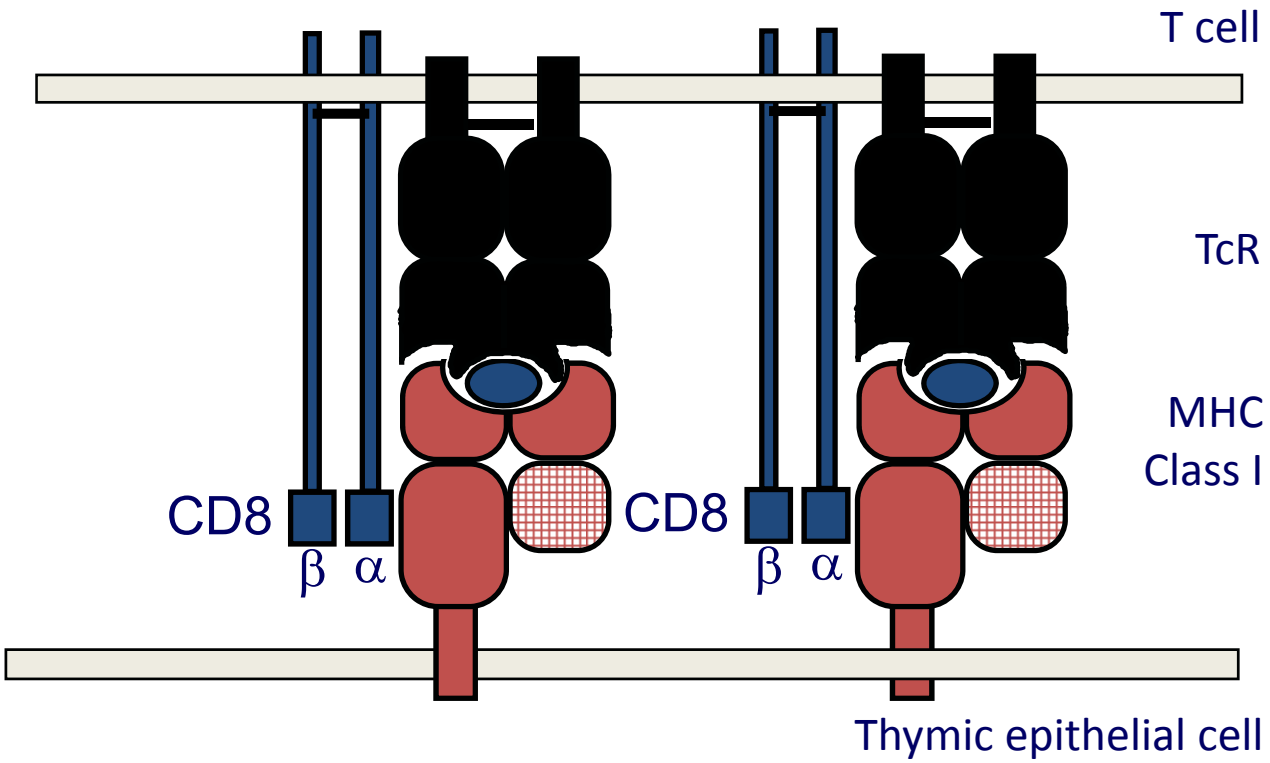
PARTIAL SIGNAL



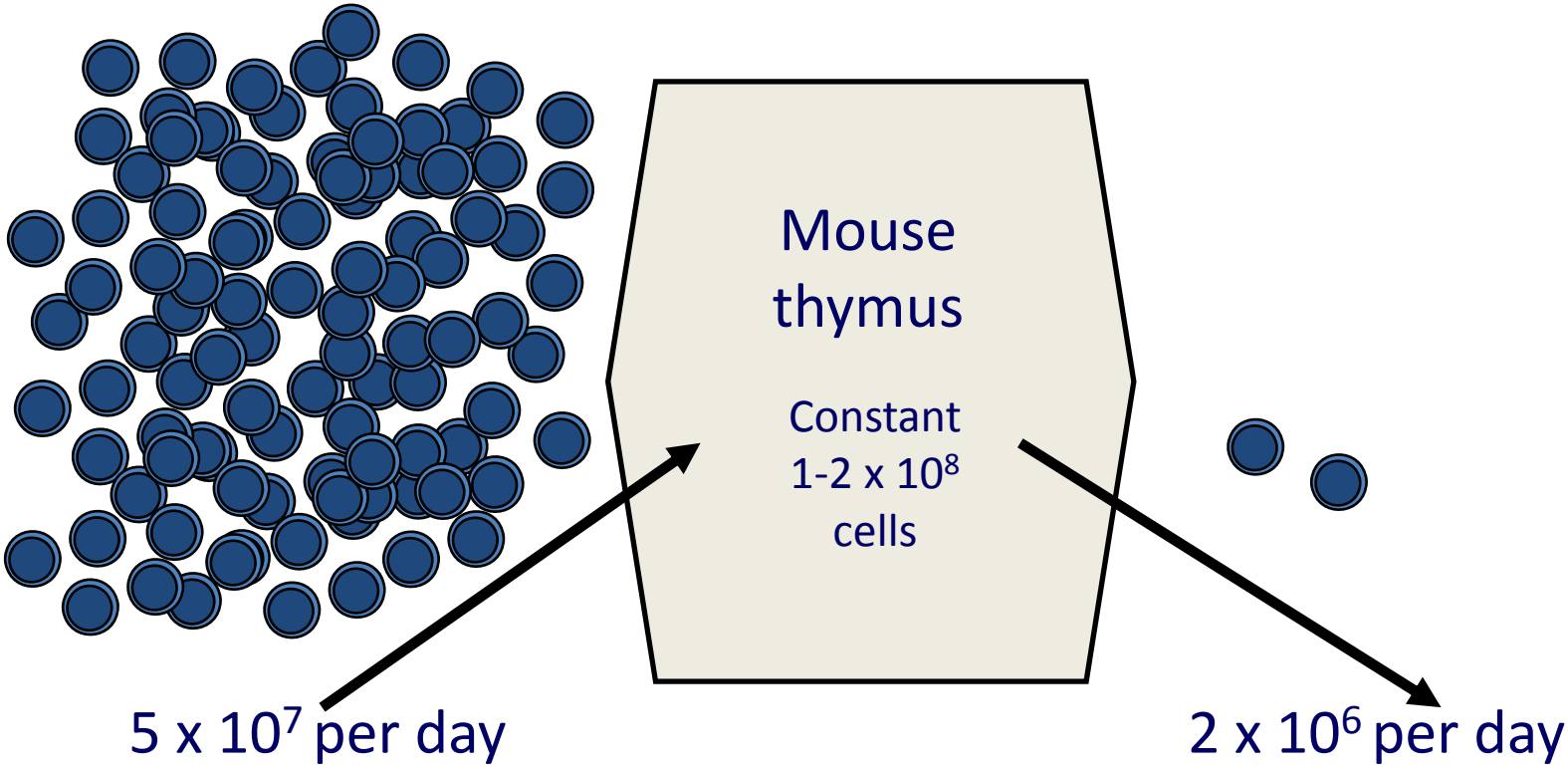
Negative selection

Peptide is an agonist
Thymocyte receives a powerful signal and undergoes apoptosis
i.e. the cell is negatively selected and dies.

FULL SIGNAL



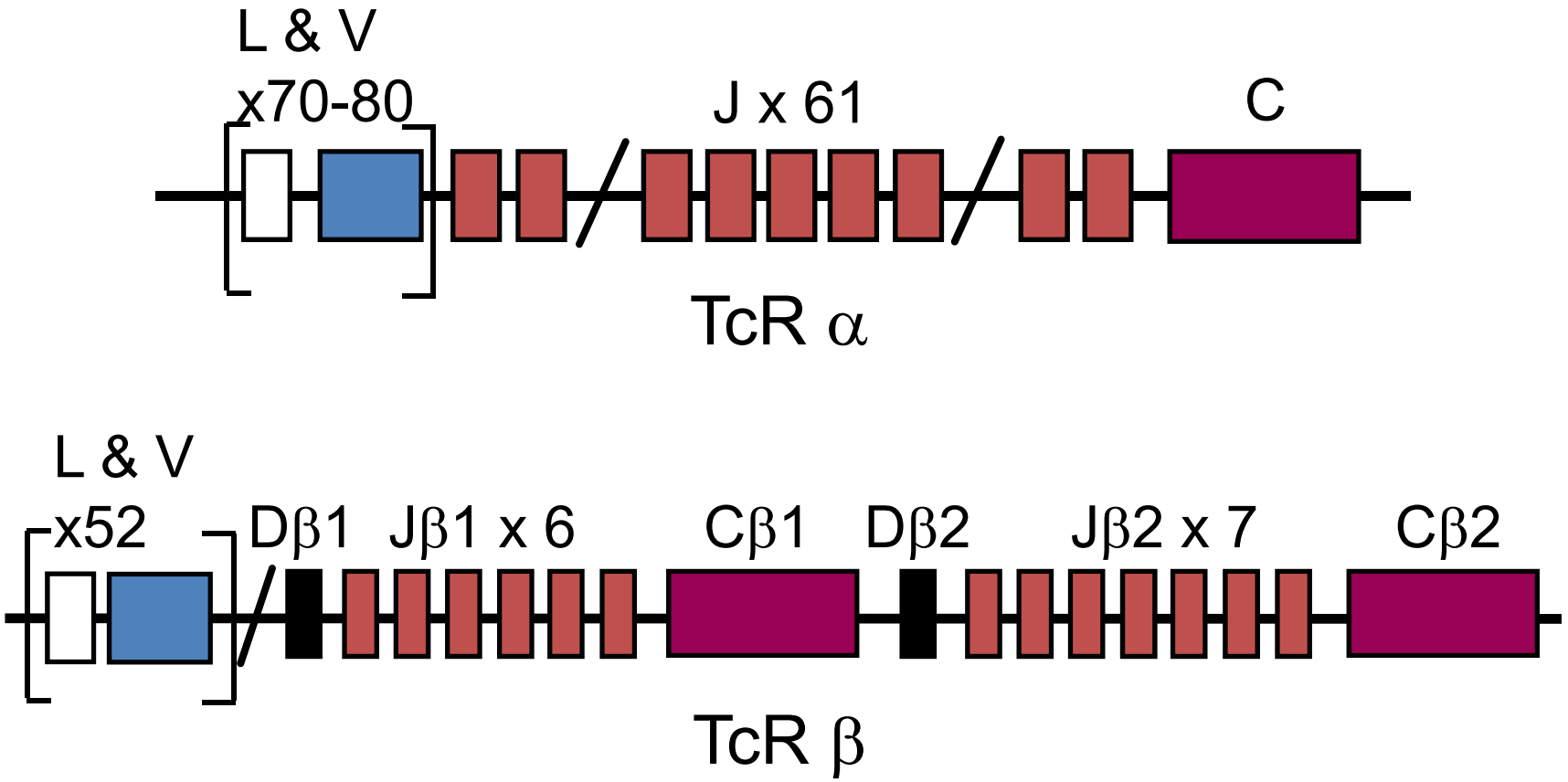
T cells mature in the thymus but most die there.



98% of cells die in the thymus without inducing any inflammation or any change in the size of the thymus.

Thymic macrophages phagocytose apoptotic thymocytes.

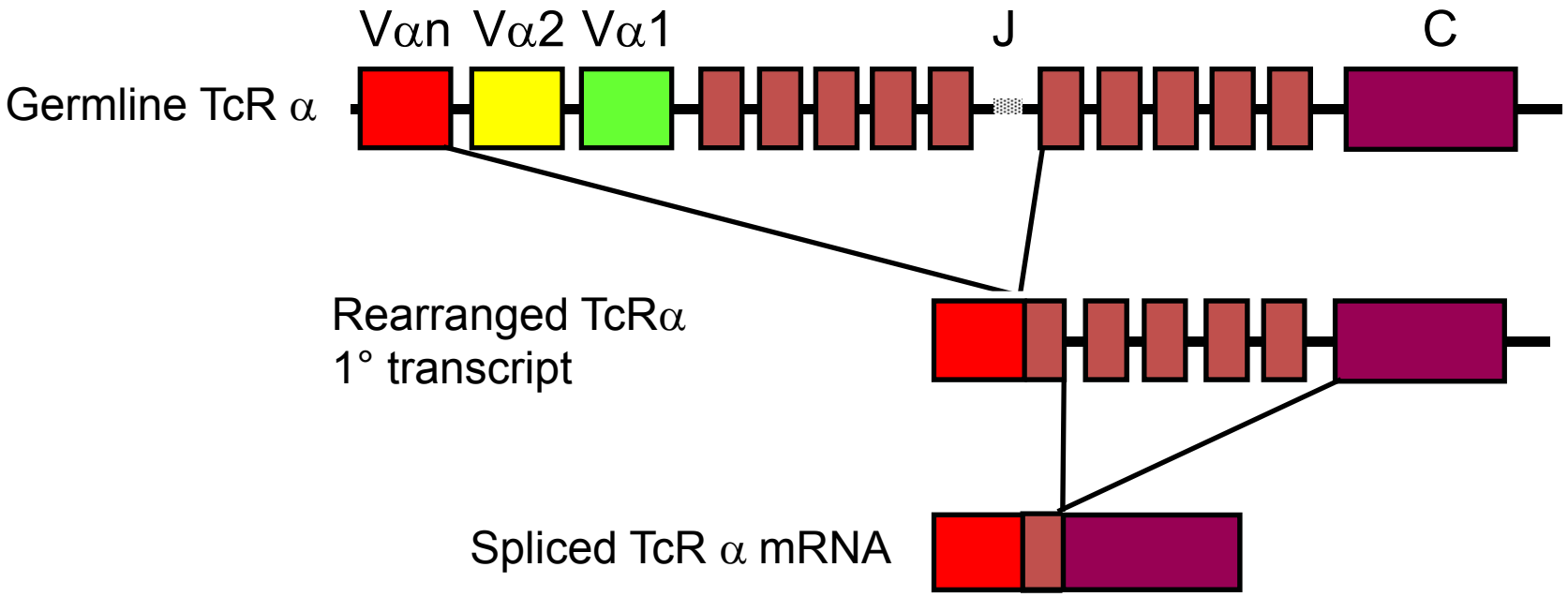
Organisation of TcR genes



TcR genes segmented into V, (D), J & C elements
(VARIABLE, DIVERSITY, JOINING & CONSTANT)
Closely resemble Ig genes (α~IgL and β~IgH)

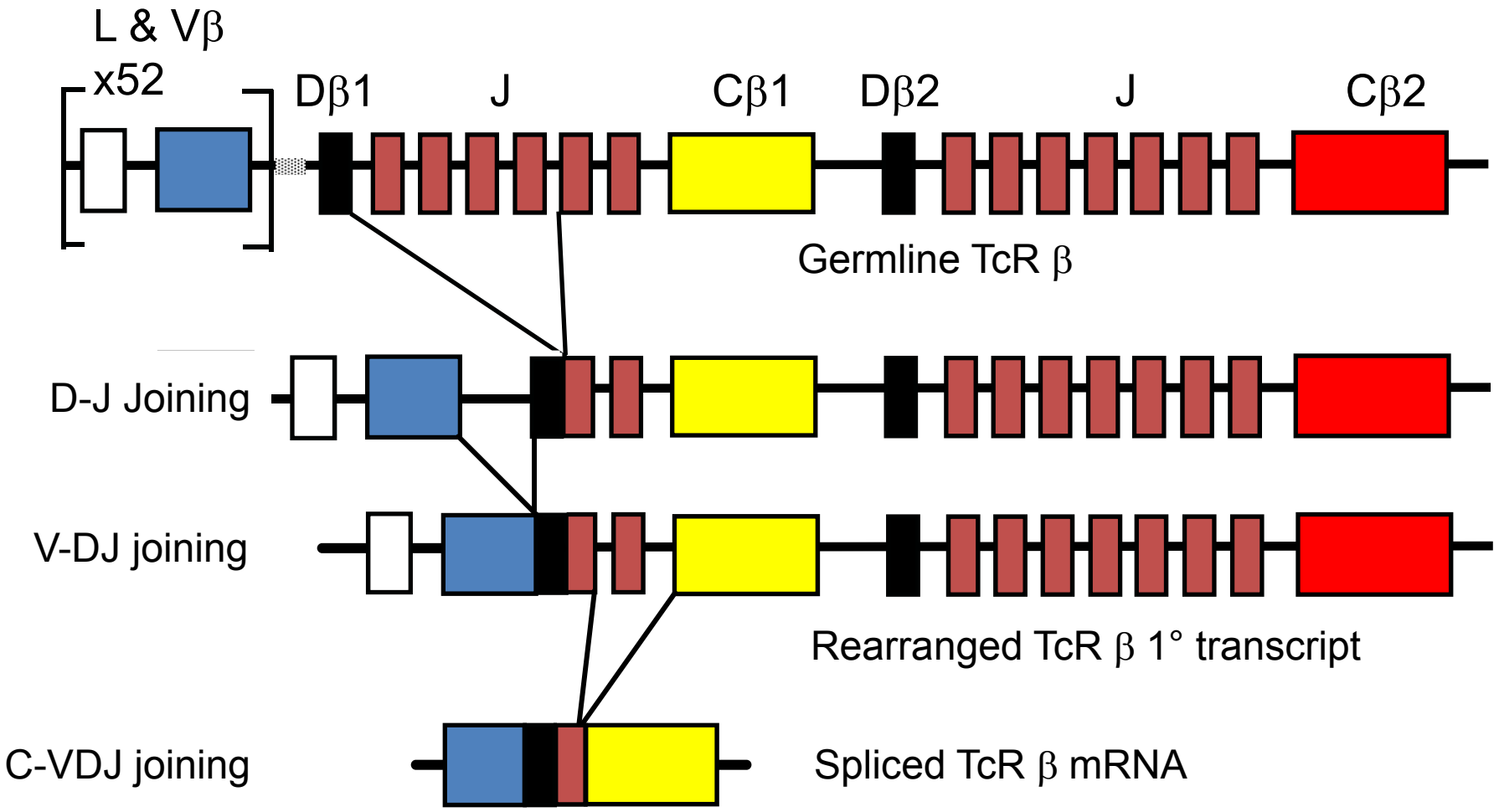
This example shows the mouse TcR locus

TcR α gene rearrangement by SOMATIC RECOMBINATION



Rearrangement very similar to the IgL chains

TcR β gene rearrangement SOMATIC RECOMBINATION



Estimate of the number of human TcR and Ig

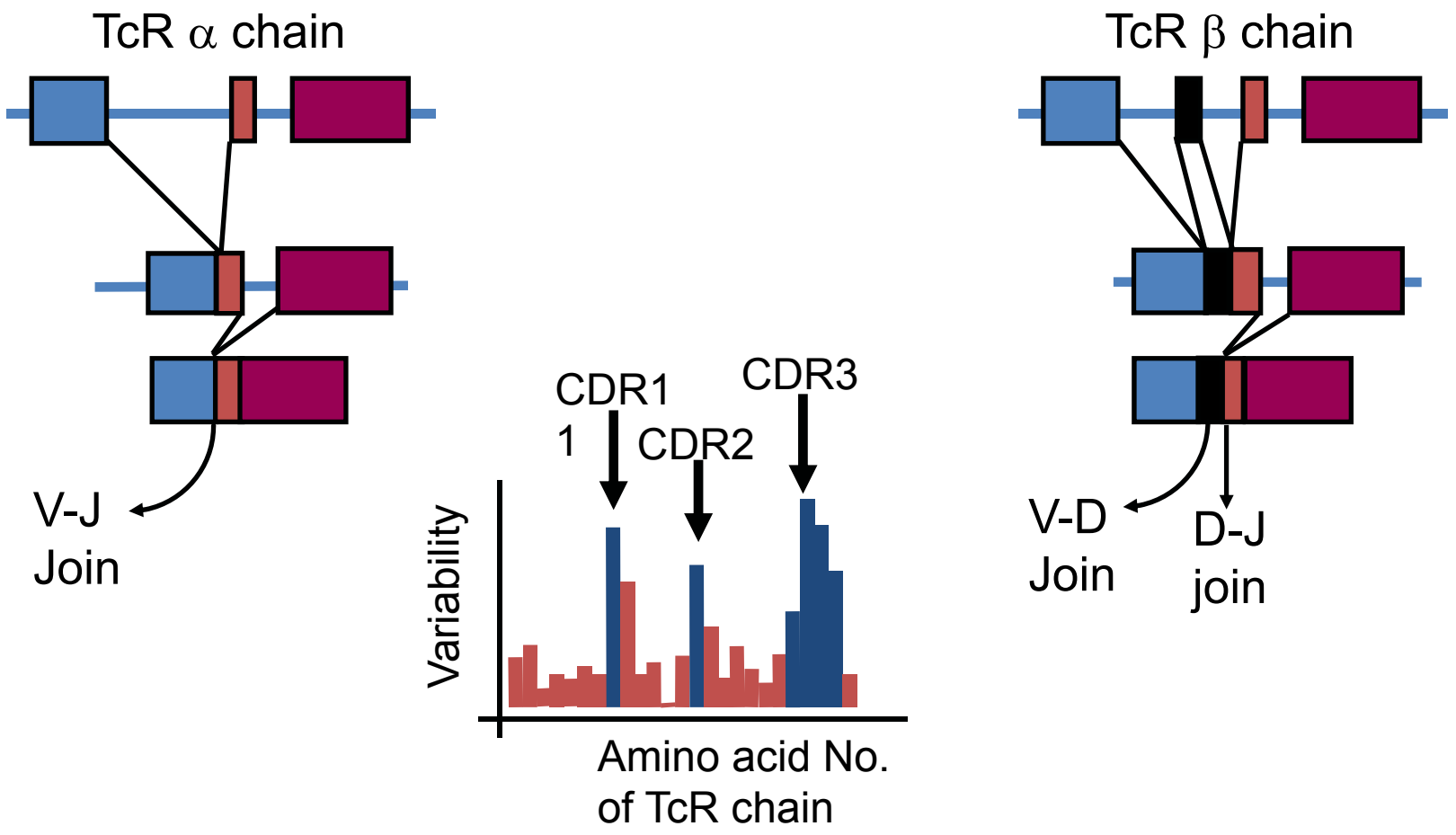
Excluding somatic hypermutation

Element	Immunoglobulin		αβ TcR	
	H	κ & λ	β	α
Variable segments	40	59	52	~70
Diversity segments	27	0	2	0
D segments in all 3 frames	Yes	-	Yes	-
Joining segments	6	9	13	61
Joints with N & P nucleotides	2	(1)*	2	1
No. of V gene pairs	2360		3640	
Junctional diversity	$\sim 10^{13}$		$\sim 10^{13}$	
Total diversity	$\sim 10^{16}$**		$\sim 10^{16}$	

* Only half of human κ chains have N & P regions

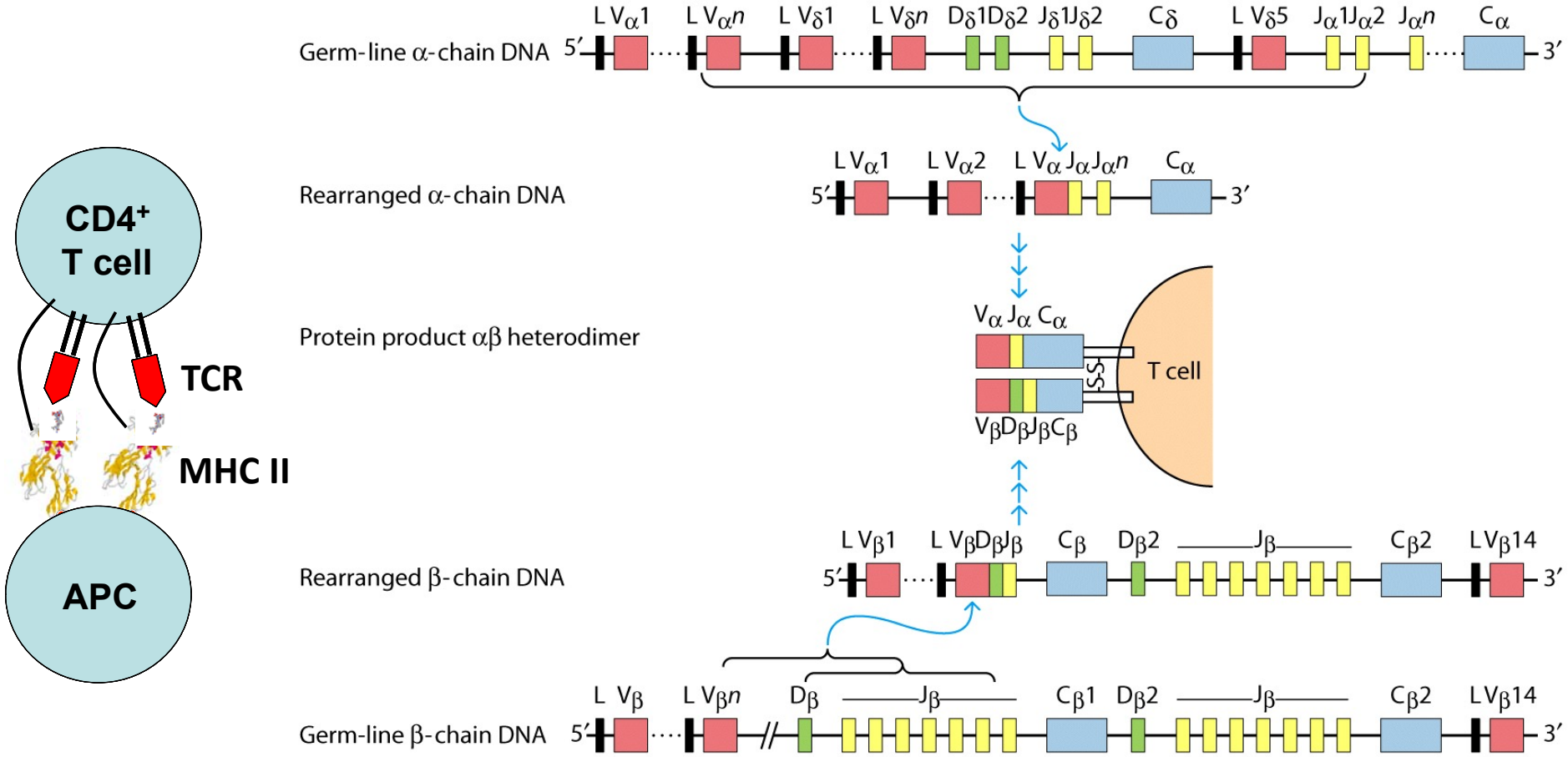
**No of distinct receptors increased further by somatic hypermutation

Location of junctional diversity



CDR = Complemantarity determining region

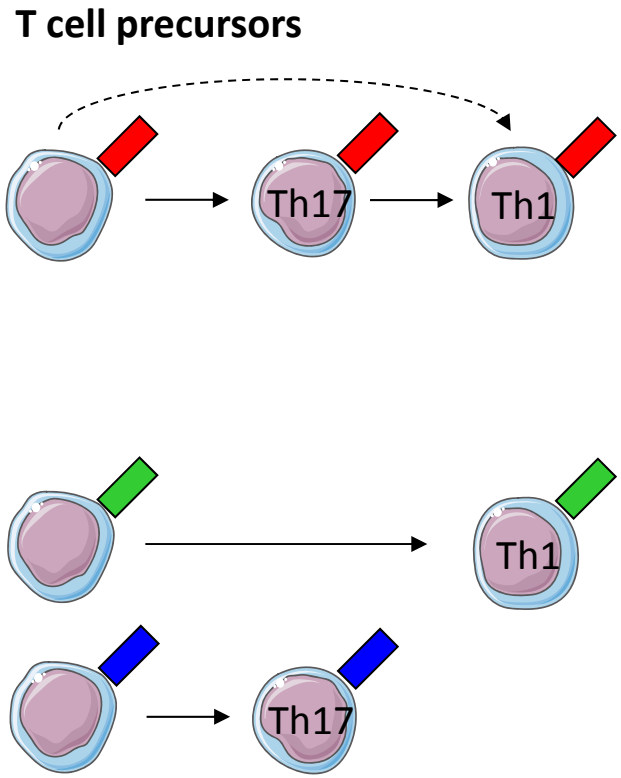
T cell receptor specificity



Larsen *et al* EJI 2011

Plasticity of human CD4$^+$ T cells

TCR finger printing



Plasticity

Overlapping TCR repertoires between Th subsets

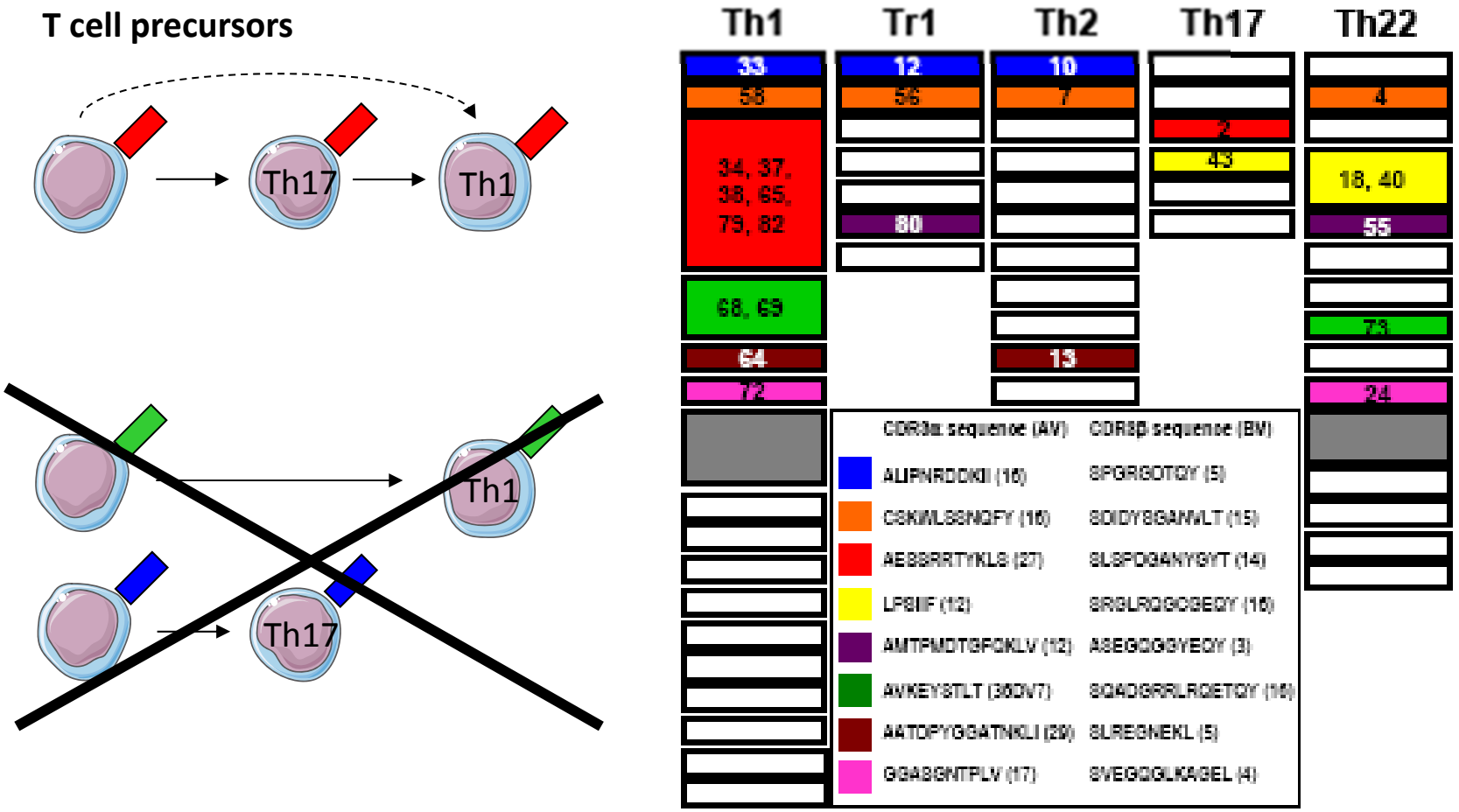
No Plasticity

Distinct TCR repertoires between Th subsets

Larsen *et al* EJI 2011

Plasticity of human CD4+ T cells

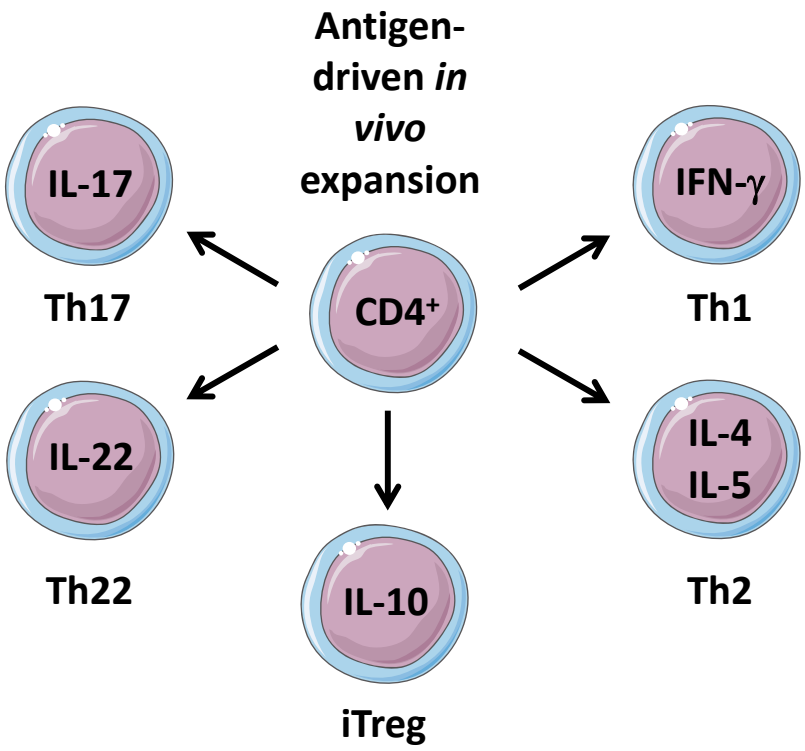
TCR finger printing



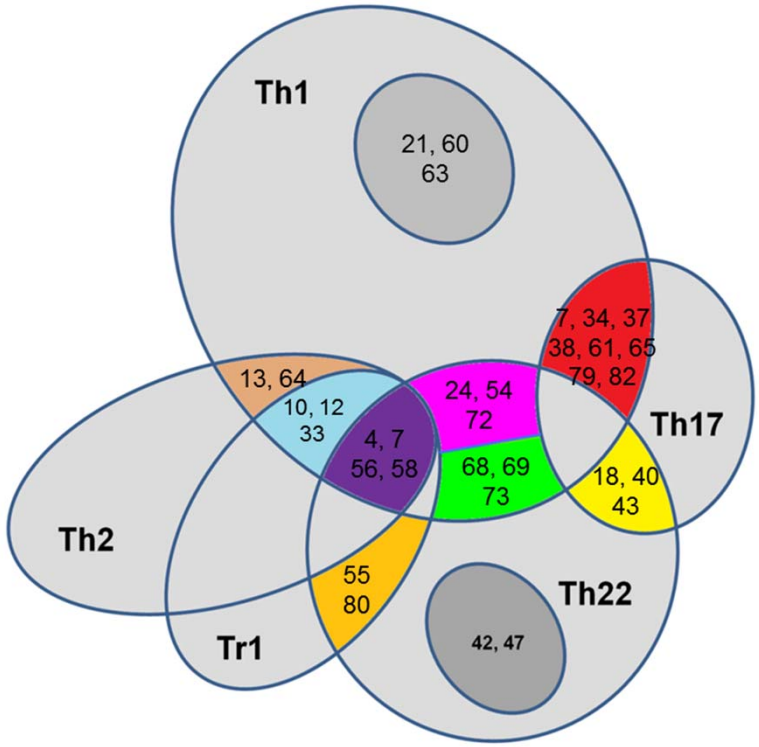
Larsen *et al* EJI 2011

Plasticity of human CD4+ T cells

A



B

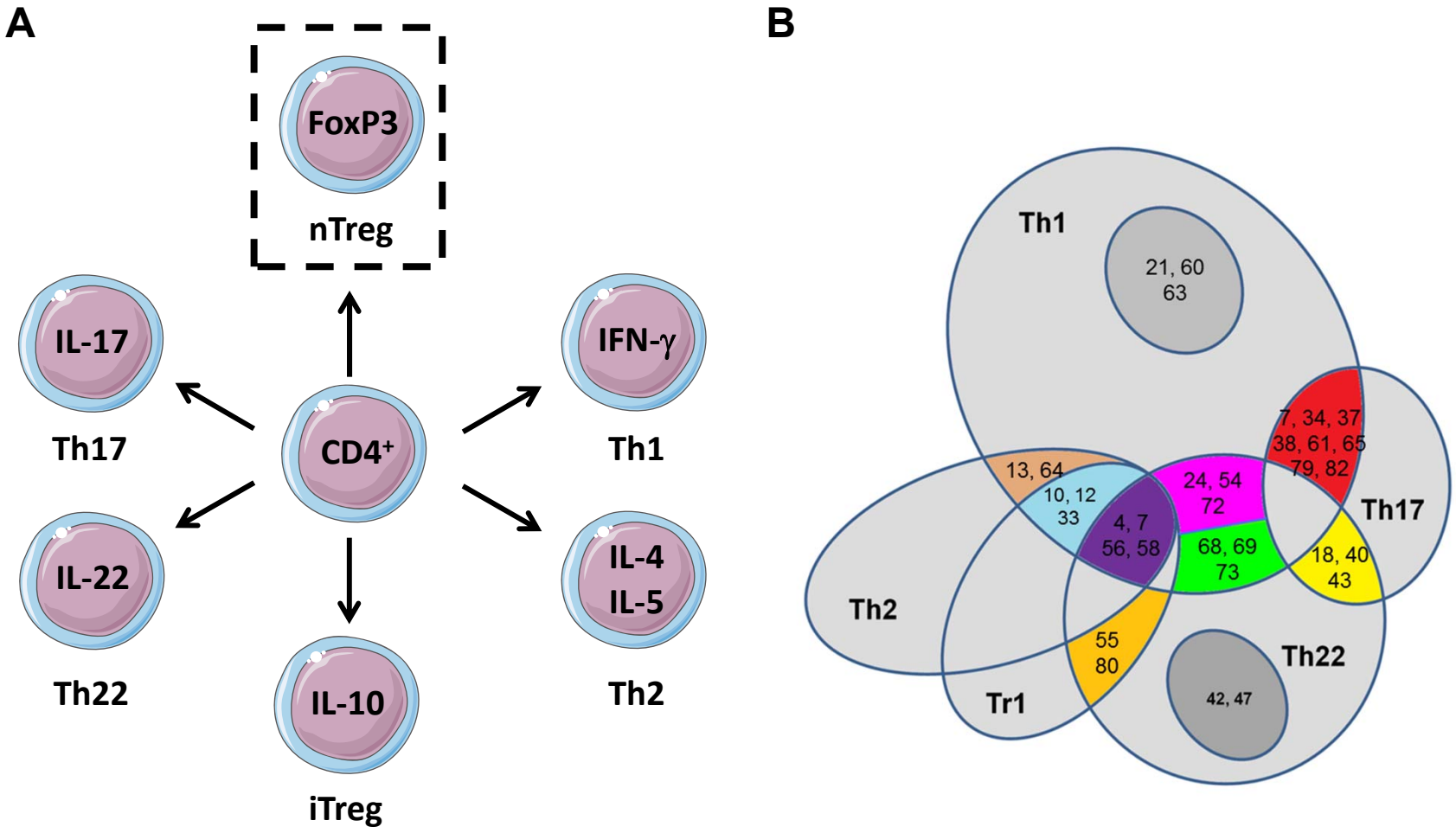


Conclusion

Peripheral Naive T cell precursors are able to adopt all Th profiles irrespective of antigen specificity

Larsen *et al* EJI 2011

Plasticity of human CD4+ T cells



To be elucidated….

Overlap between conventional T cells and thymus-derived regulatory T cells (nTregs)?

Larsen *et al* EJI 2011

Thymic selection of conventional T cells versus Tregs

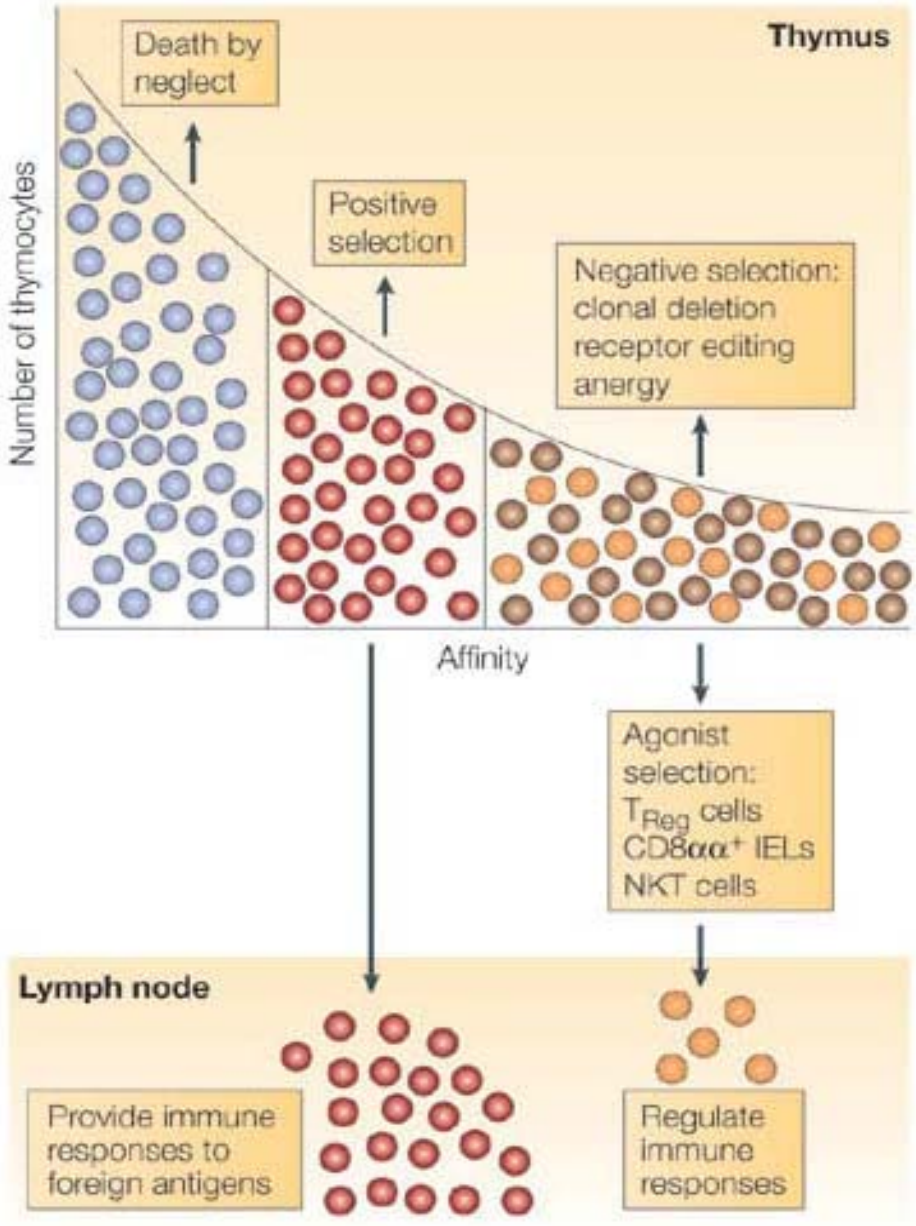
Naturally occuring Tregs are derived from thymocytes with “high” affinity to self that escape elimination by negative selection through TGF-β signalling..

Jordan et al Nat Immunol 2001

Ooyang et al Immunity 2010

However, some studies indicate that Tregs can develop from T cells lacking a TCR. Thus suggesting that other factors than affinity plays a role.

Tuovinen et al JI 2008



Physicochemical differences between Tconv and Treg TCRs

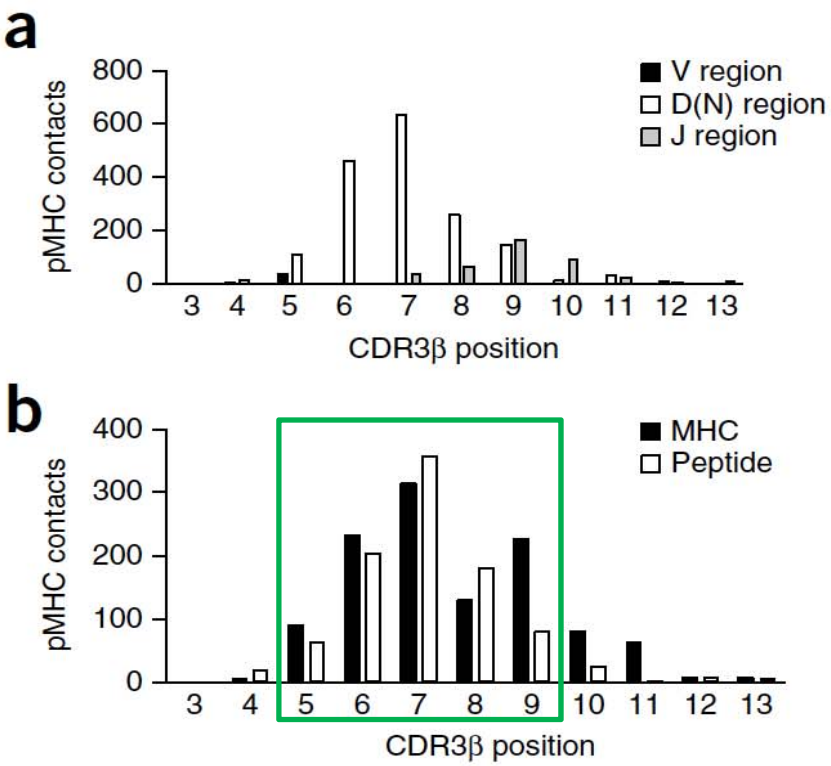
nature immunology

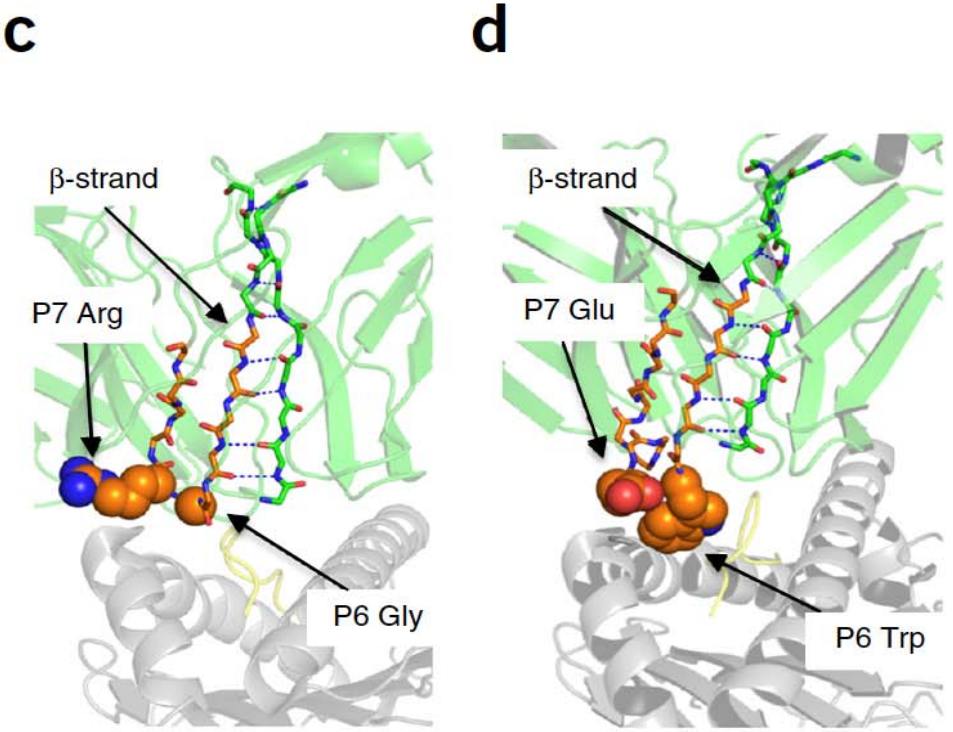
Hydrophobic CDR3 residues promote the development of self-reactive T cells

Brian D Stadinski[1], Karthik Shekhar[2], Iria Gómez-Touriño[3], Jonathan Jung[1], Katsuhiro Sasaki[1], Andrew K Sewell[4], Mark Peakman[3], Arup K Chakraborty[5–10] & Eric S Huseby[1]

Studies of individual T cell antigen receptors (TCRs) have shed some light on structural features that underlie self-reactivity. However, the general rules that can be used to predict whether TCRs are self-reactive have not been fully elucidated. Here we found that the interfacial hydrophobicity of amino acids at positions 6 and 7 of the complementarity-determining region CDR3β robustly promoted the development of self-reactive TCRs. This property was found irrespective of the member of the β-chain variable region (V_β) family present in the TCR or the length of the CDR3β. An index based on these findings distinguished $V_\beta 2^+$, $V_\beta 6^+$ and $V_\beta 8.2^+$ regulatory T cells from conventional T cells and also distinguished $CD4^+$ T cells selected by the major histocompatibility complex (MHC) class II molecule I-A^{g7} (associated with the development of type 1 diabetes in NOD mice) from those selected by a non–autoimmunity-promoting MHC class II molecule I-A^b. Our results provide a means for distinguishing normal T cell repertoires versus autoimmunity-prone T cell repertoires.

Physicochemical differences between Tconv and Treg TCRs



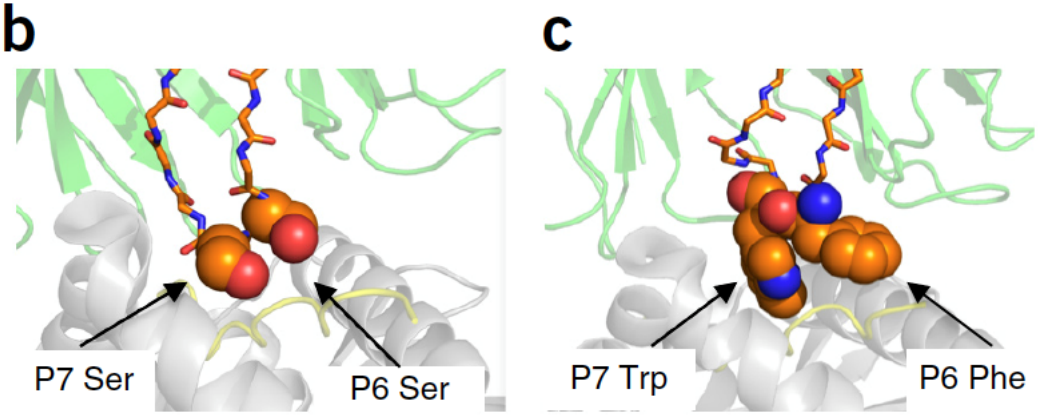


a

Two self-reactive TCRβs:
Specificity: HLA I-E (MHC class II)

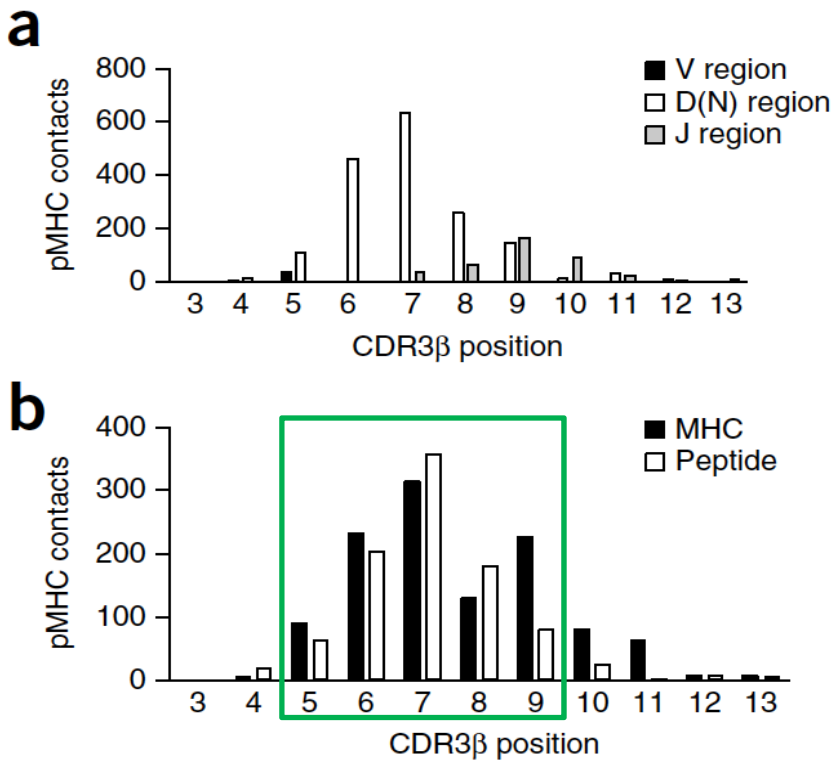
	CDR3β position												
	1	2	3	4	5	6	7	8	9	10	11	12	13
YAe62β	C	A	S	G	D	**F**	**W**	G	D	T	L	Y	F
B3K506β	C	A	S	I	D	**S**	**S**	G	N	T	L	Y	F

YAe62b: high affinity (eliminated by neg selection)
B3K506b: low affinity (differentiate to naïve cells)



Stadinski et al. Nat Immunol 2016

Hydrophobicity differences between Tconv and Treg TCRs



a

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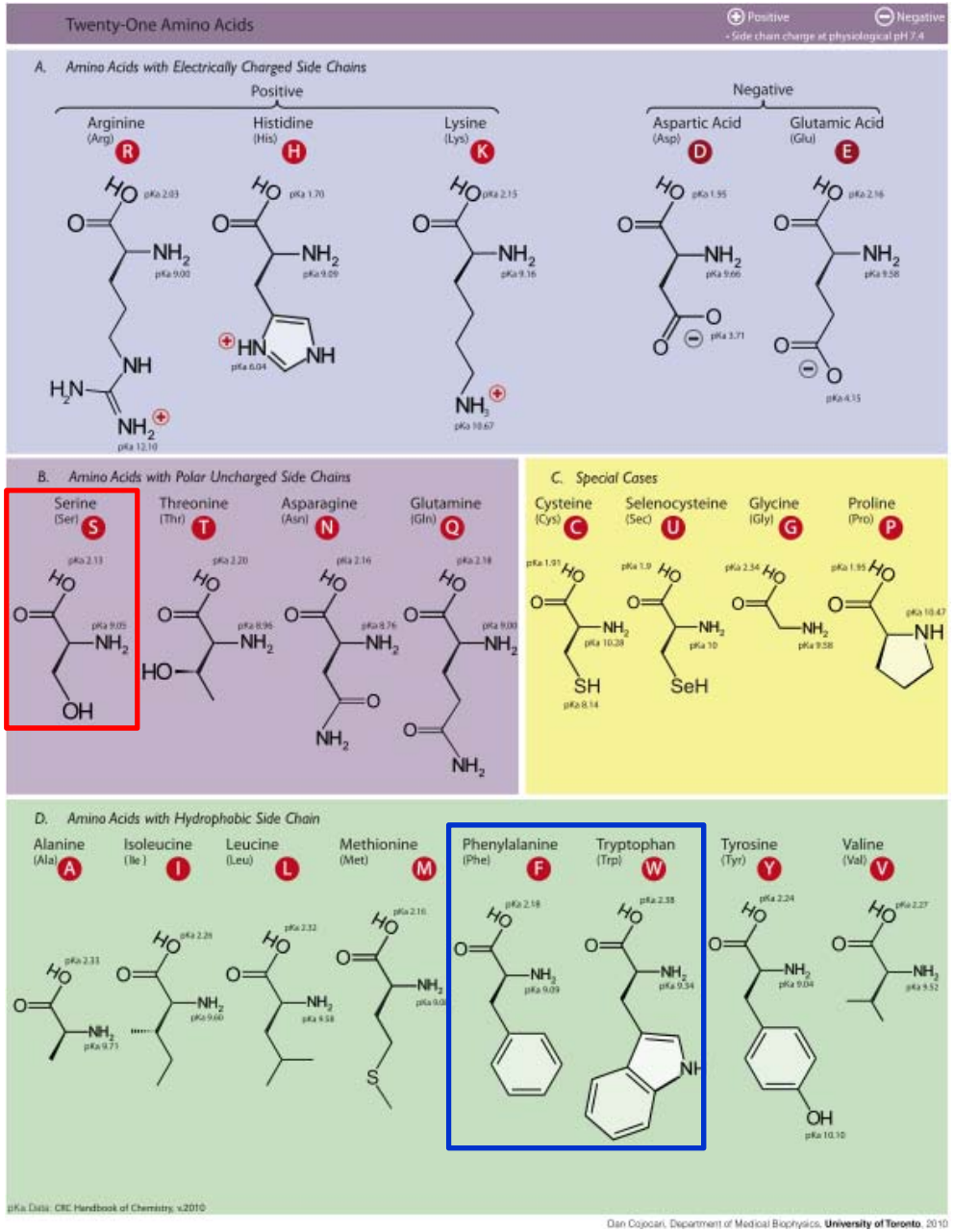
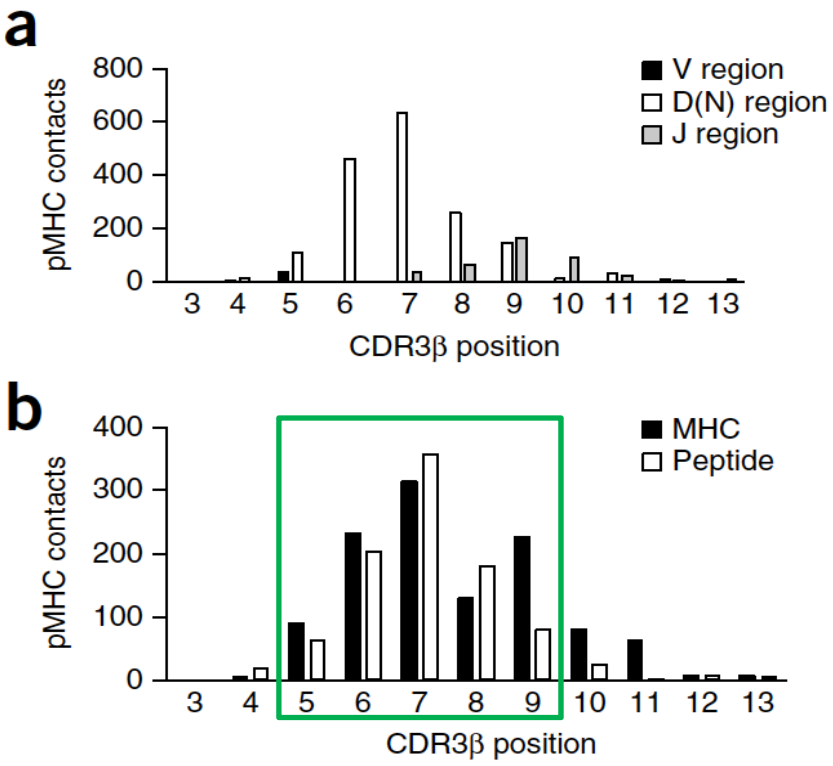
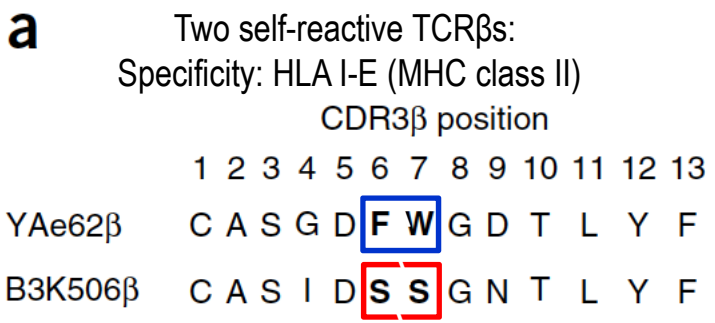


Stadinski et al. Nat Immunol 2016

Physicochemical differences between Tconv and Treg TCRs





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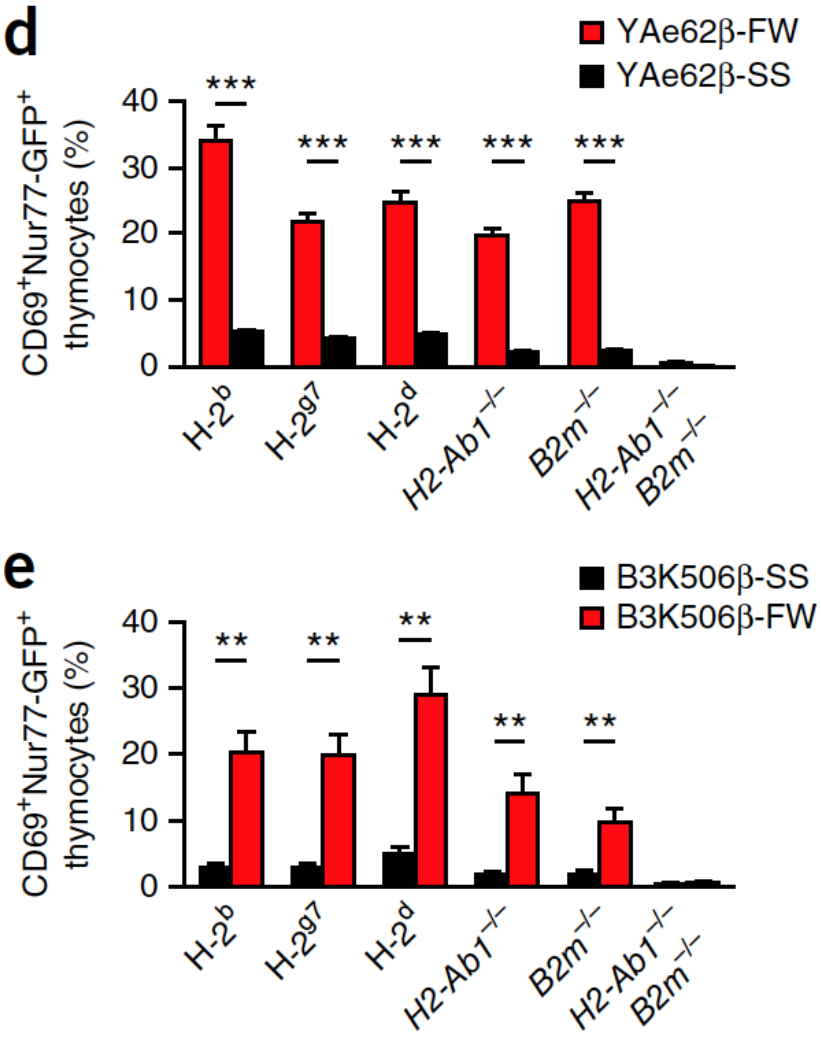
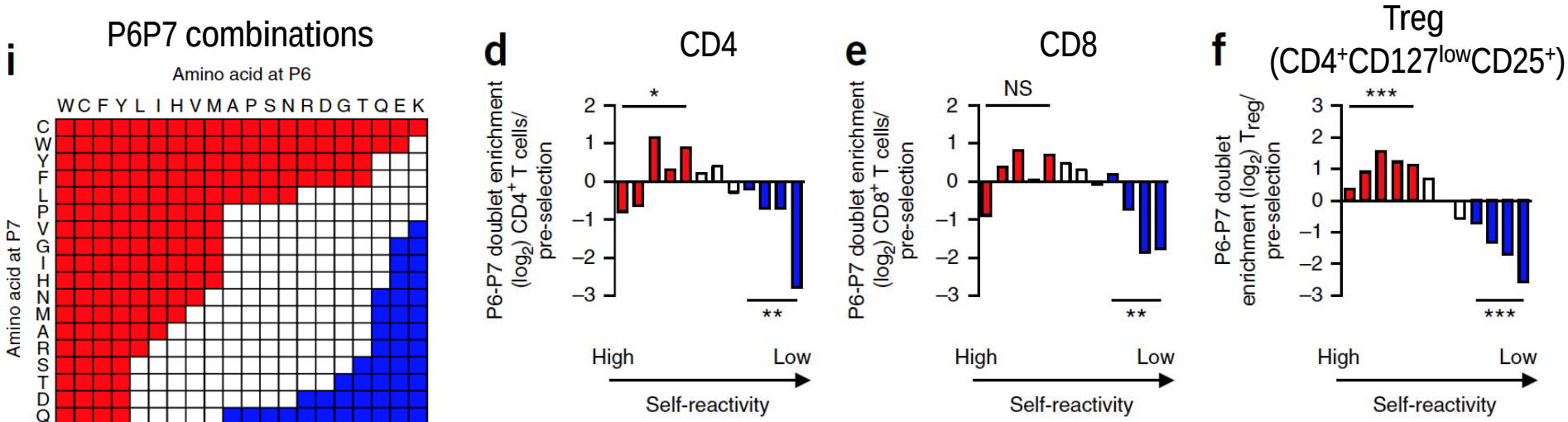


➢ Swapping P6P7 leads to inversion of self-reactivity

Stadinski et al. Nat Immunol 2016

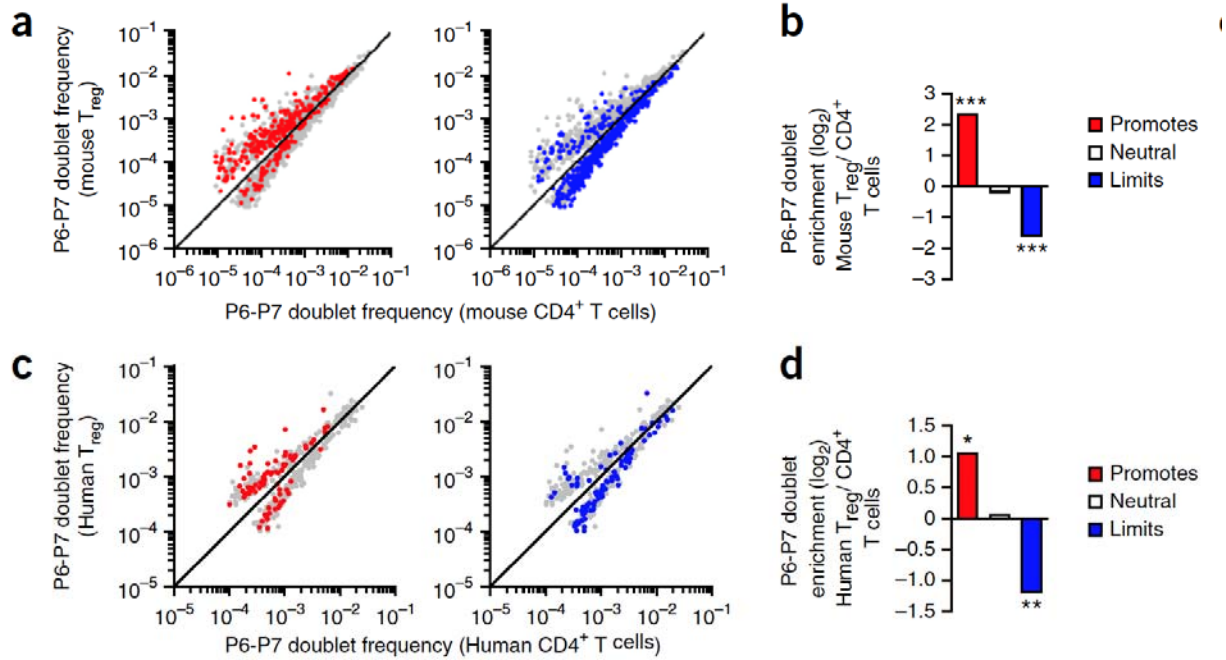
Physicochemical differences between Tconv and Treg TCRs



- Tregs select positively for promotors of self-reactivity (**red**)

Mouse

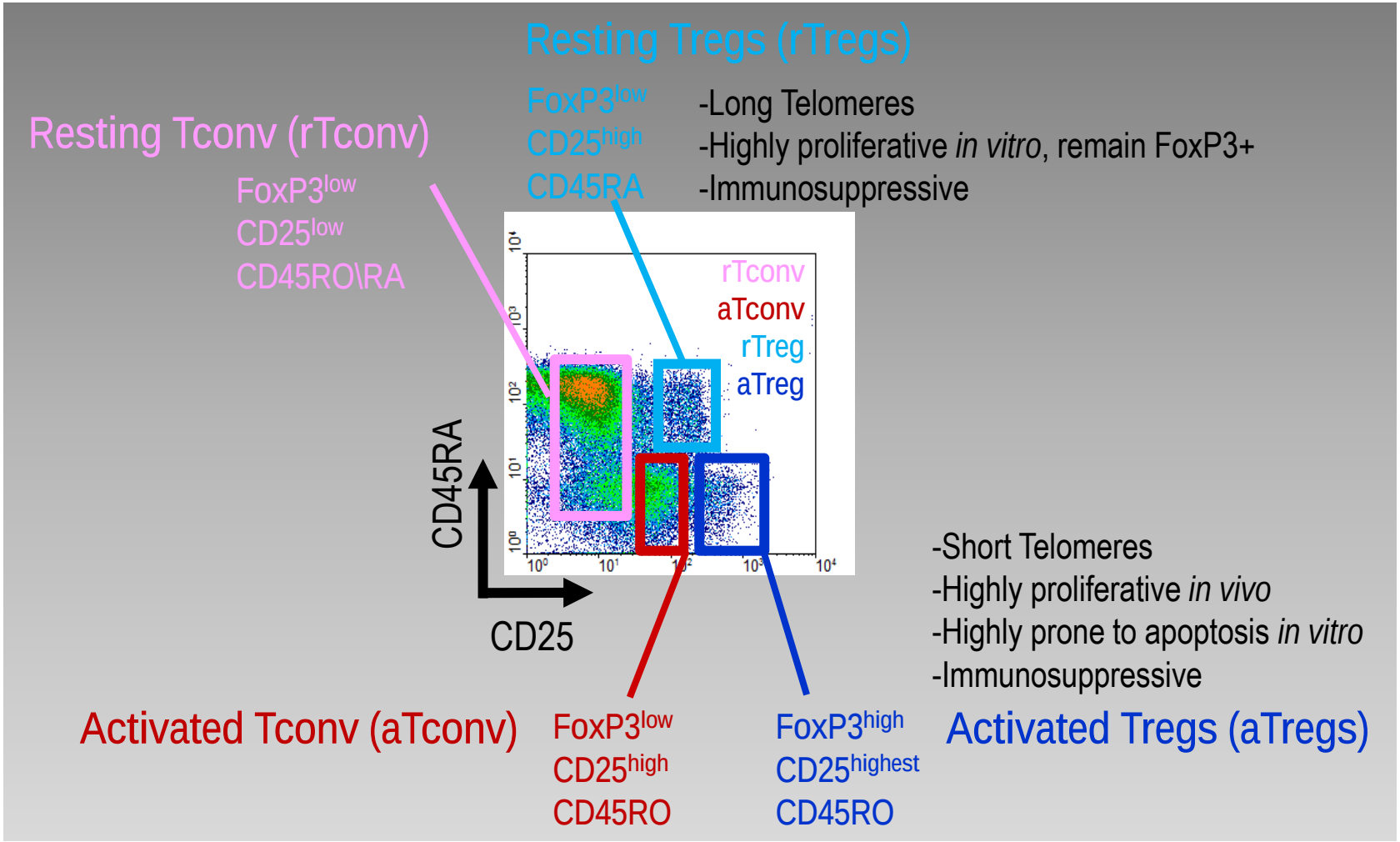
Human



- Mouse and less so human Tregs favor self-reactive promotor P6P7

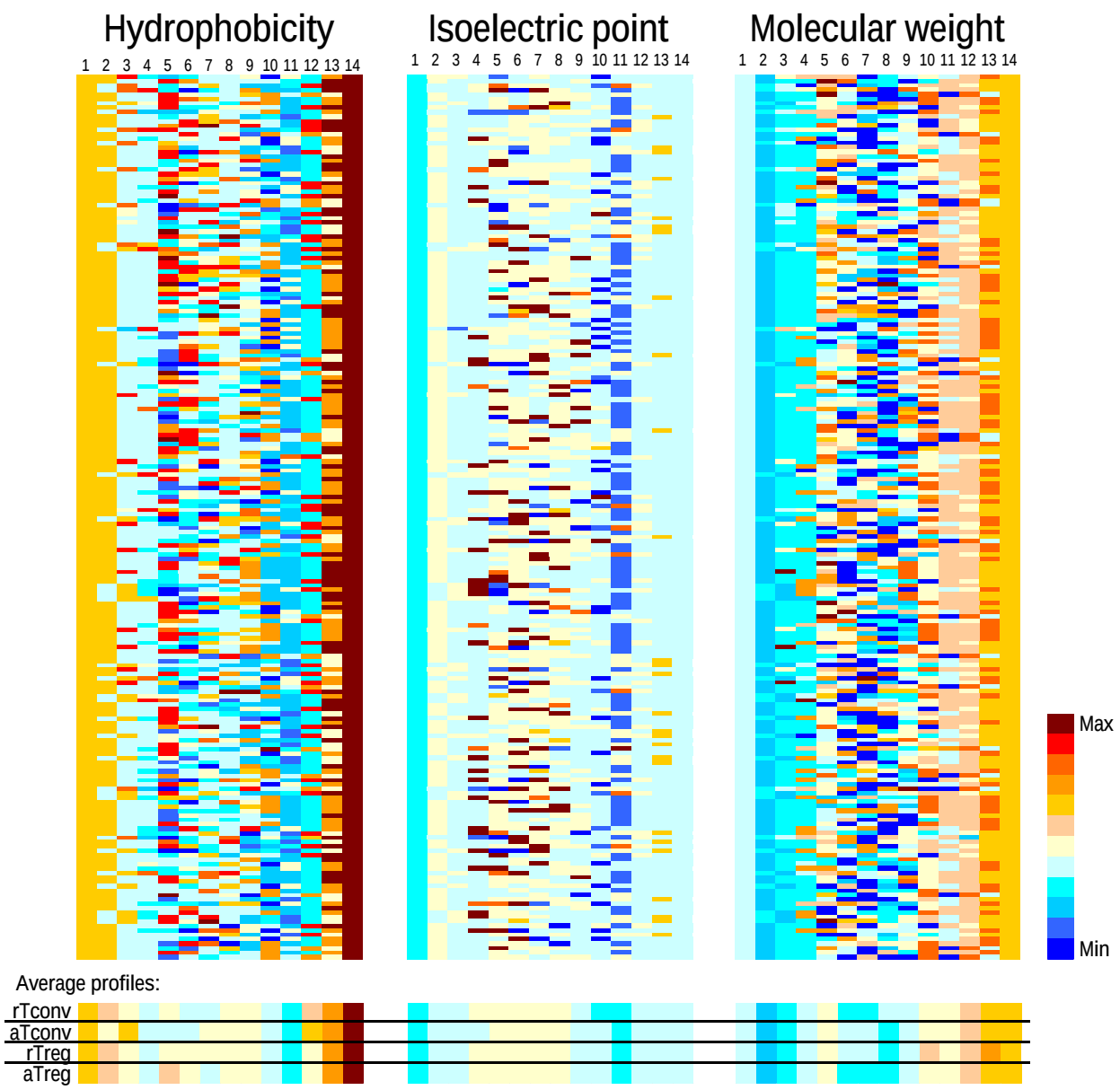
Stadinski et al. Nat Immunol 2016

Sorting of conventional and regulatory CD4+ T cells



Miyara *et al.* JEM 2006; Miyara *et al.* Immunity 2009

Physicochemical differences between Tconv and Treg TCRs



14mer analysis

Funky Cells

www.FunkyCells.com

Funky Cells Tool Box

- Subset differences are primarily associated with hydrophobicity

NTW

Conclusions & Perspectives

- Conventional CD4+ T cells display functional plasticity at the clonal level.

- It is still not determined if thymus-derived natural Tregs and Tconv are clonally related.

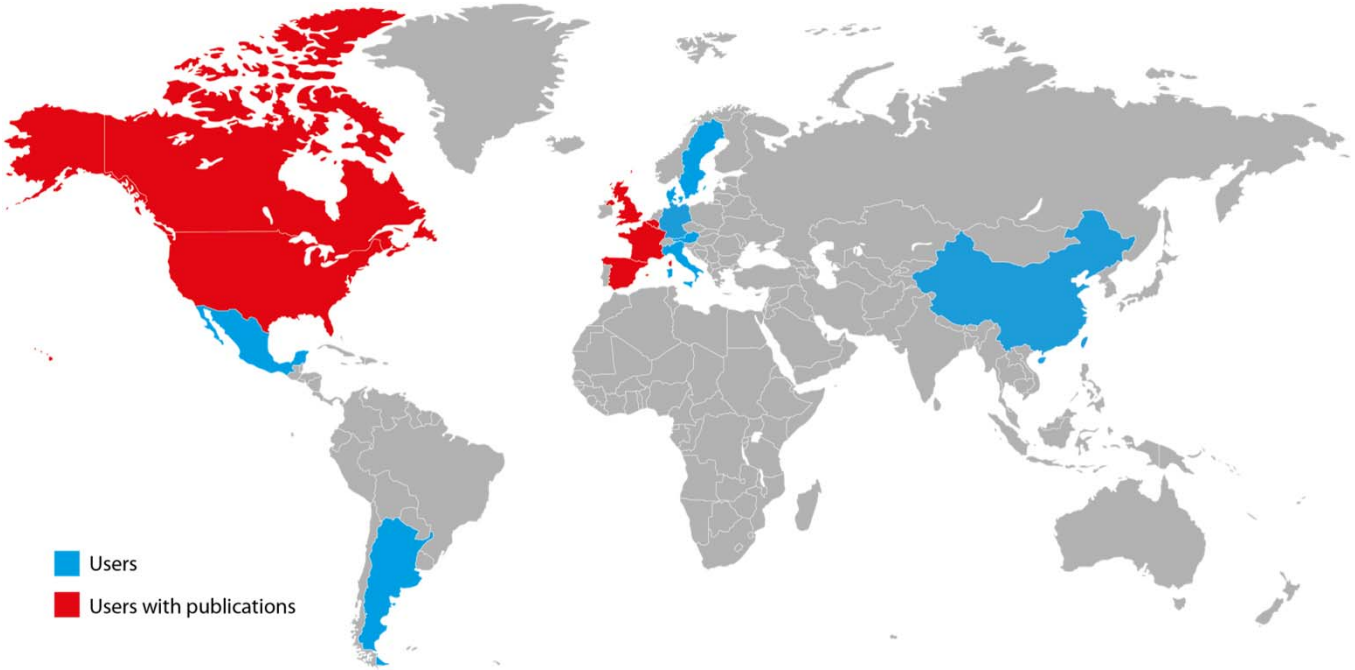
- Tconv and Treg subsets differ in physicochemical properties primarily through alterations in hydrophobicity.
  - Hydrophobicity: Tregs > Tconv

- Our data do not confirm that P6 and P7 are particularly hydrophobic in Tregs. Indeed P5 and less so P6 seems to be more general sites of alteration.

- The result is confirmed in three individuals. Of note, the predictive signal is low and cannot be used at the clonal level (only population level).

- A larger number of individuals should be analyzed to identify inter-individual variation (e.g. plot the correlation coefficient of each variable with PLS component 1 in a scatter plot to identify cross-individual robust predictive variables .

Funky Cells Tool Box software



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