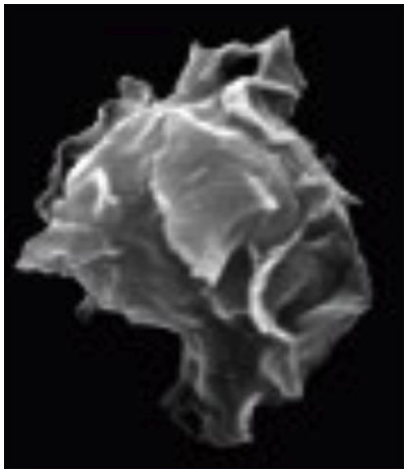
UPMC PARIS UNIVERSITAS

p2t physiologie physiopathologie thérapeutique ÉCOLE DOCTORALE

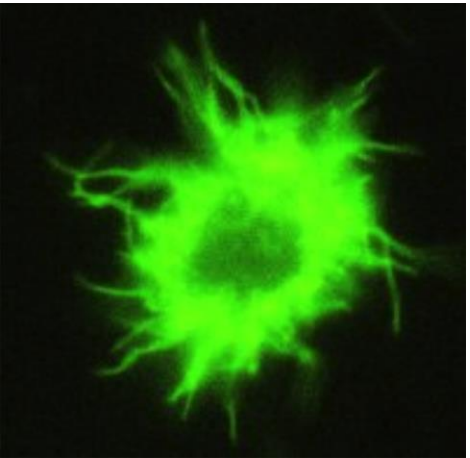
CRC CENTRE DE RECHERCHE DES CORDELIERS

Inserm Institut national de la santé et de la recherche médicale



Dendritic cells

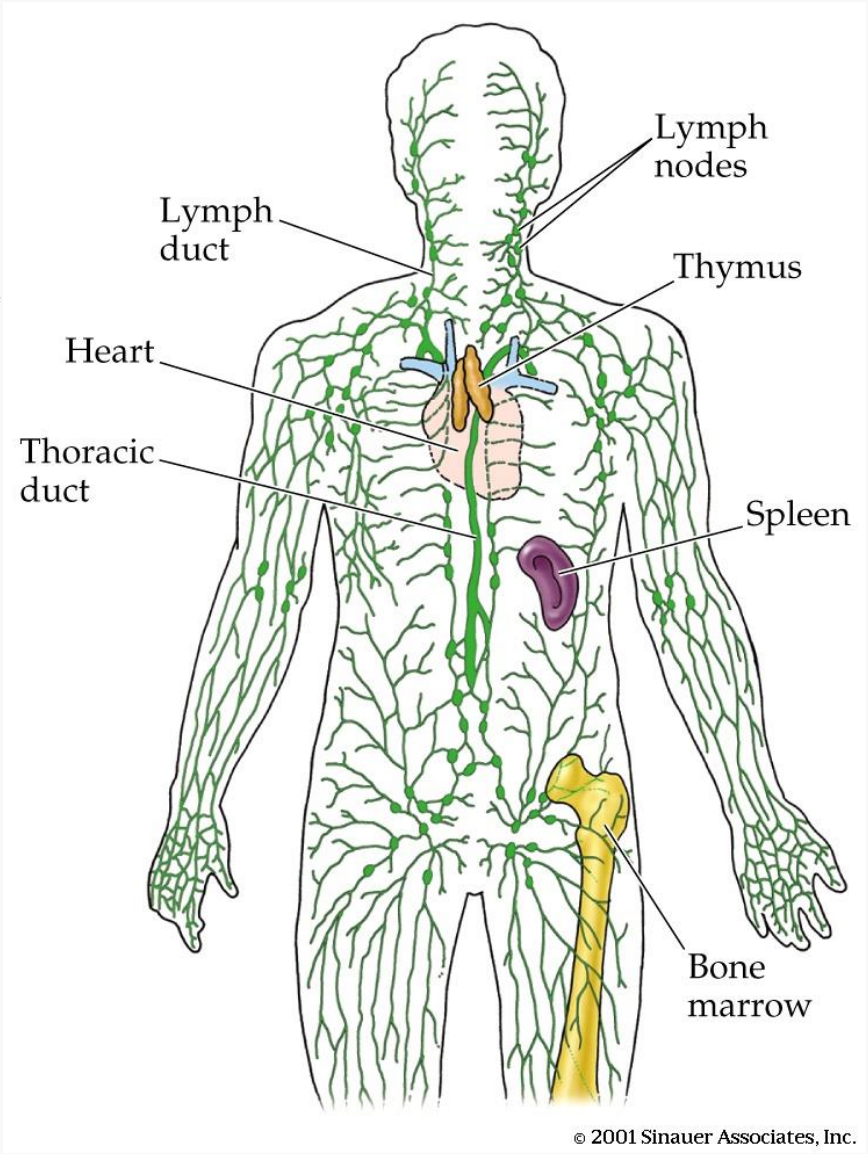
Emmanuel STEPHEN-VICTOR (PhD)



INSERM U 1138 - Centre de Recherche des Cordeliers, Paris

Pathogens

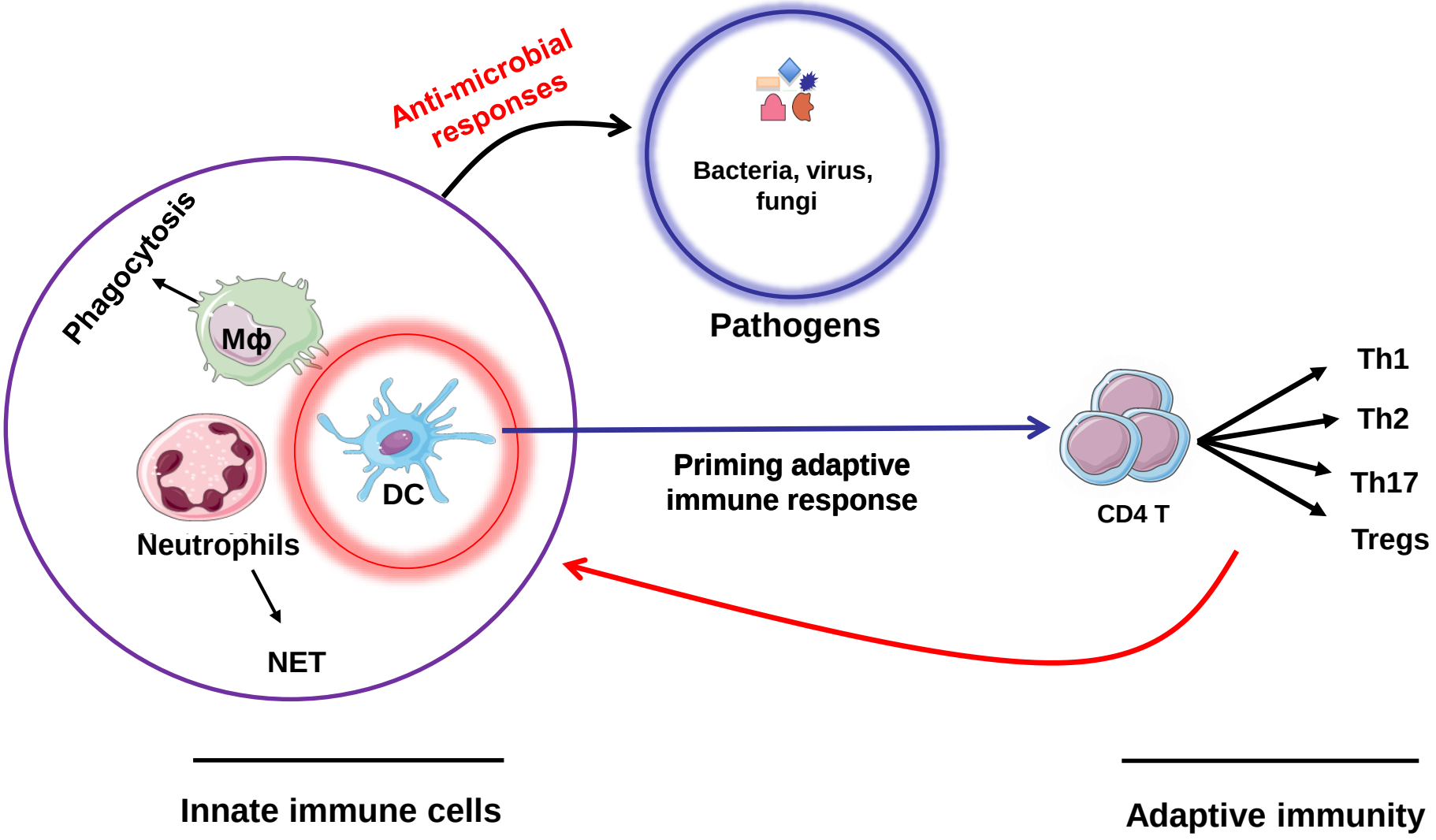
Bacteria, virus, fungi



⇒ **The pathogen-derived antigens need to be transported**

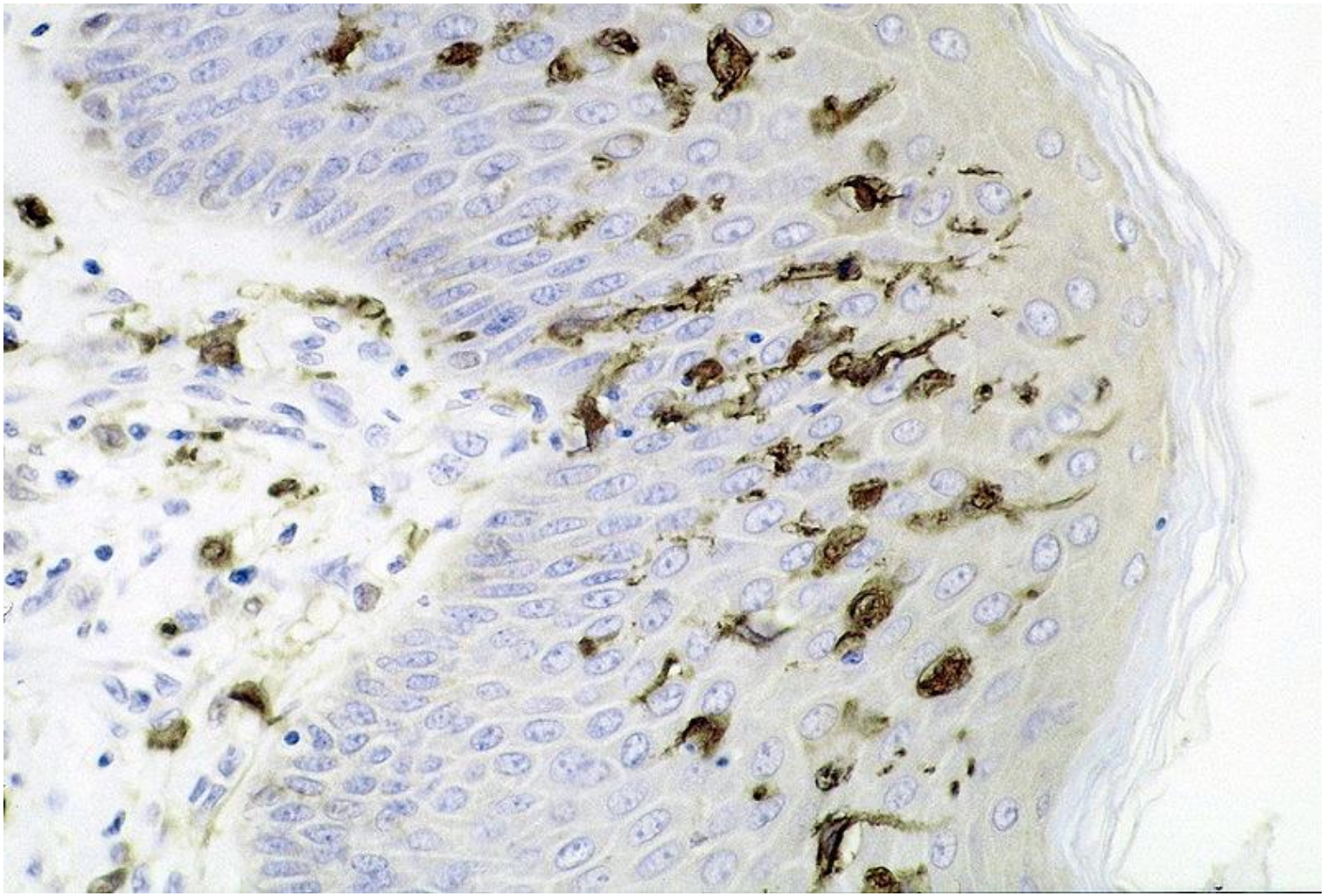
⇒ **adptaive immune response needs to be initiated**

Immunity and Anti-immunity – Host Vs Pathogen



Dendritic cells -Discovery

1868 Paul Langerhans => Branched skin cells at epidemis resembling neurons



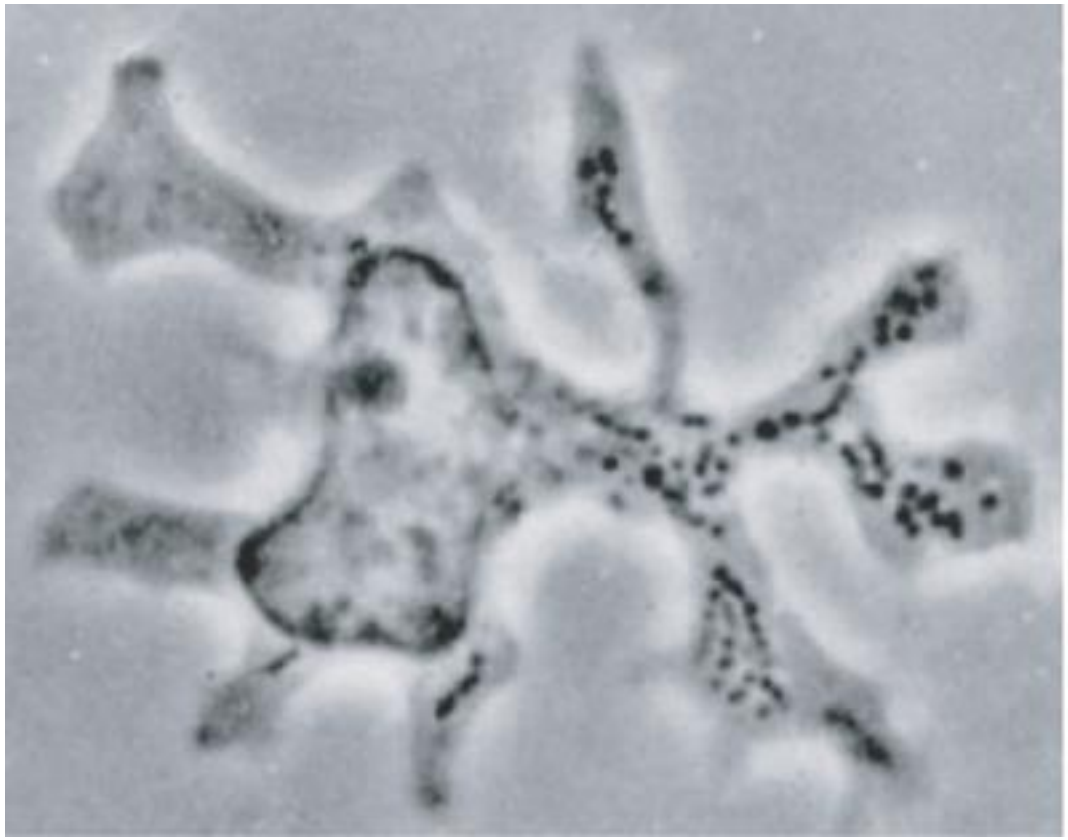
Dendritic cells -Discovery

1868 Paul Langerhans
=> Branched skin cells at epidemis resembling neurons

1973 Ralph Steinman

=> Dendritic cells from the spleen of mice

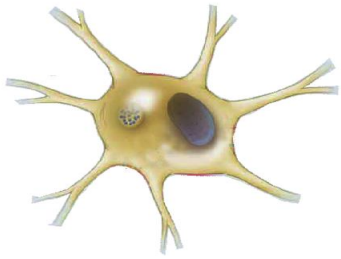
One of the first views of DCs in mouse spleen



- Infection
- Cancer
- Transplantation
- Autoimmunity and chronic inflammation
- Allergy
- Vaccines

Steinman RM et al. J Exp Med, 1973

Dendritic cells: Major professional antigen presenting cells



Critical for primary immune response

Large array of endocytic receptors **(not B cells)**

Large array of antigen presenting and co-stimulatory molecules **(not MØ)**

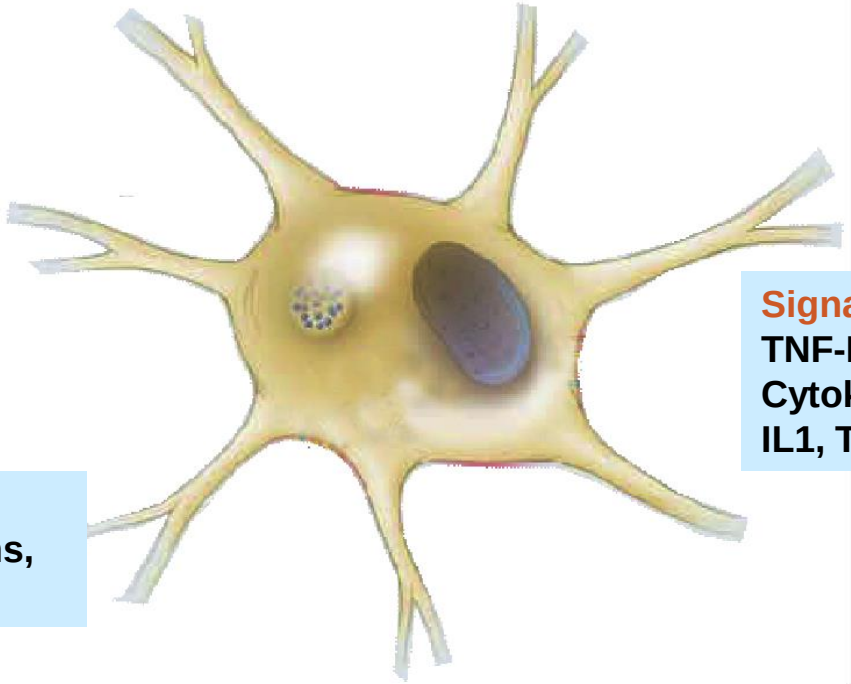
Ability to migrate to T cell areas for presentation **(not other APCs)**

Some features of dendritic cells

Adhesion and co-stimulation
ICAM 1,3; B7-1, B7-2; CD58/LFA3

Secretion
IL-12, IL-10, TNF, chemokines

High levels of Ag presenting molecules
HLA I, II; CD1

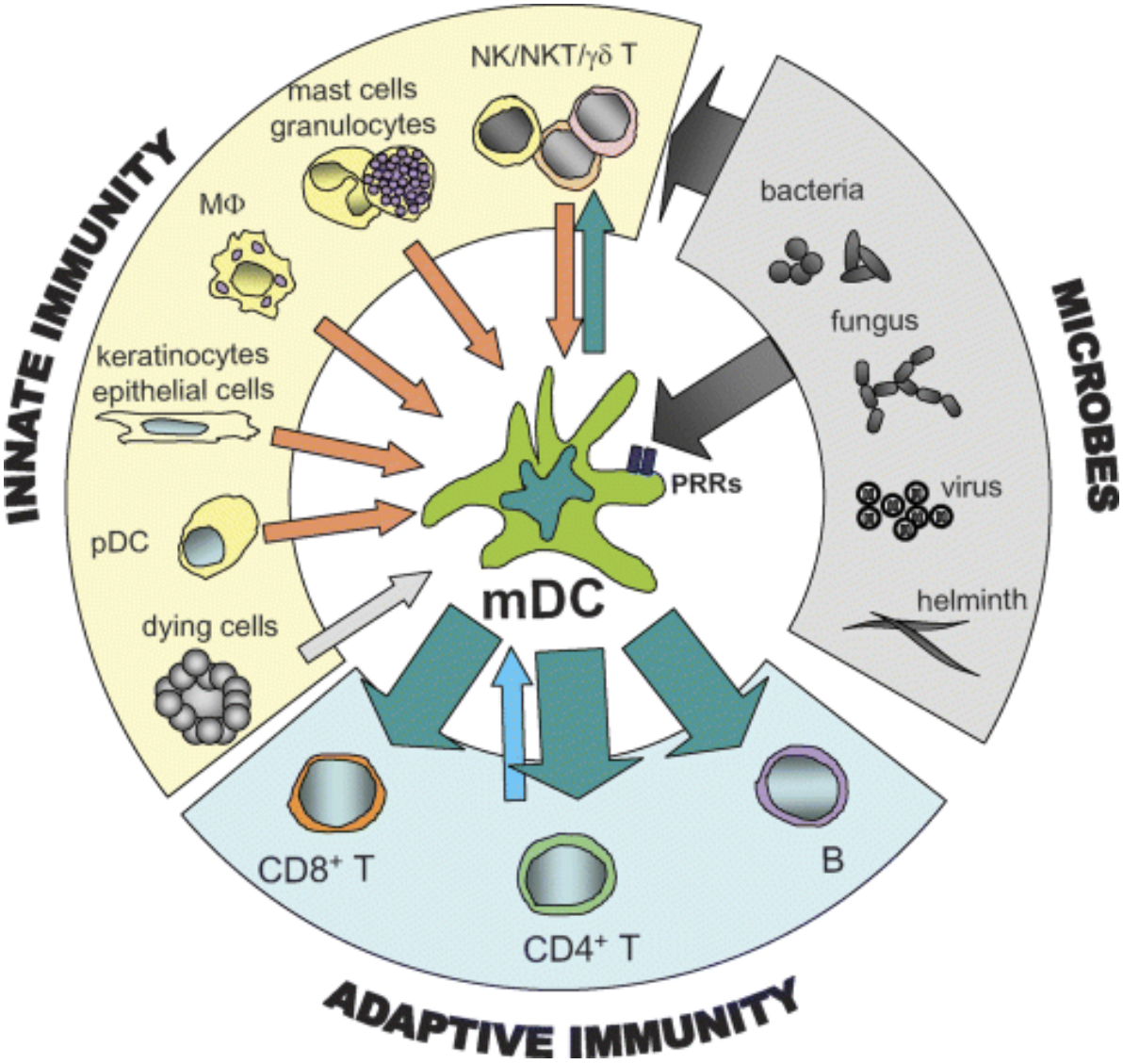


Signaling of DC
TNF-R, CD40, TLR
Cytokine R: GM-CSF, IL-4, IL1, TGF-b, IL-10

Receptors for Ag uptake
MMR, DC205, Langerin, integrins, FcγR TLR, CR

Receptors for pathogens
CD4, DC-SIGN, chemokine receptors

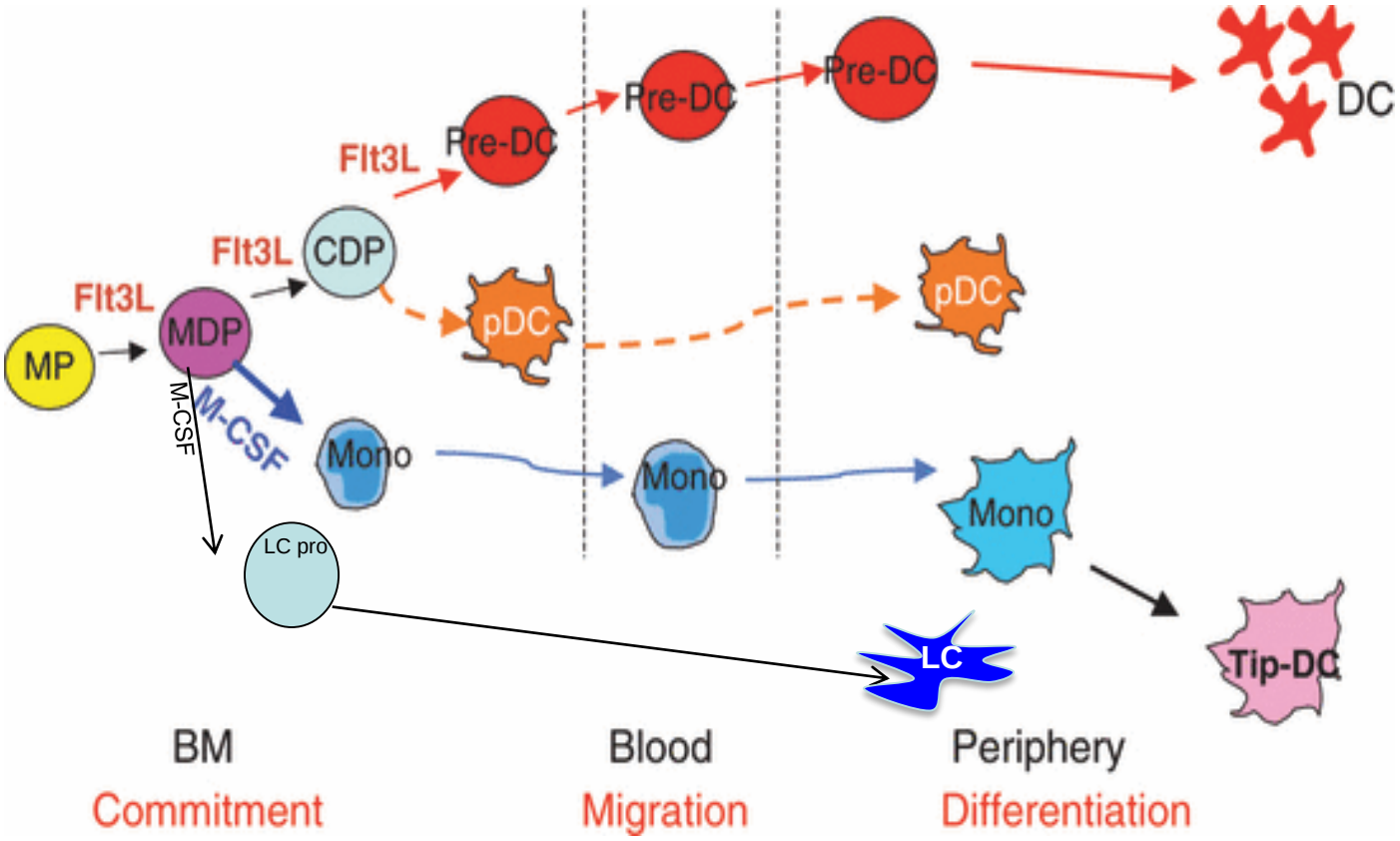
Dendritic cells in health and disease



Immunological Reviews
Volume 219, Issue 1, pages 118-142, 10 SEP 2007 DOI: 10.1111/j.1600-065X.2007.00551.x
http://onlinelibrary.wiley.com/doi/10.1111/j.1600-065X.2007.00551.x/full#f1

Ontogeny of dendritic cells

Origin and development of dendritic cells



MP: Myeloid progenitor, MDP: monocyte dendritic cell progenitor, CDP: committed DC progenitor

Flt3L: fms-like tyrosine kinase 3 ligand

Liu & Nussenzweig, Immunol Rev 2010

Subsets of dendritic cells

Dendritic cell subsets in health and disease

	Epidermal Langerhans cells	Dermal Interstitial DCs	Blood Myeloid DCs	Blood Plasmacytoid DCs
				
C-type lectin	Langerin	DC-SIGN Mannose Receptor	(DC-SIGN) (Mannose Receptor)	BDCA-2
Specific molecules	CD11c CD1a E-cadherin	CD11c CD1a/CD14 CD11b CD36 Factor XIIIa	CD11c	CD123 ILT7
TLRs	1, 2, 3, 6, (7), (10)	1, 2, 3, 4, 5, 6, 7, 8	1, 2, 3, 4, 5, 6, (7), 8, 10	1, 6, 7, 9, 10

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Volume 219, Issue 1, **pages 118-142, 10 SEP 2007 DOI: 10.1111/j.1600-065X.2007.00551.x**
http://onlinelibrary.wiley.com/doi/10.1111/j.1600-065X.2007.00551.x/full#f4

Human dendritic cell deficiency

Characteristic	DC deficiency syndrome		
	DCML deficiency	IRF8 mutation (K108E)	IRF8 mutation (T80A)
CD123⁺ plasmacytoid DCs	Population absent	Population absent	Normal
CD11c⁺ myeloid DCs	Population absent	Population absent	Normal
CD1c⁺ myeloid DCs	Population absent	Population absent	Reduced in number
CD141⁺ myeloid DCs	Population absent	Population absent	Normal
Monocytes	Severely reduced in number	Severely reduced in number	Normal
B cells	Reduced in number	Normal	Normal
Natural killer cells	Reduced in number	Normal	Normal
T cells	Normal	Normal	Normal
Defective cytokines	IL-12, IFNγ, IL-6, TNF	IL-12, IFNγ, IL-6, TNF	IL-12
Myeloproliferation	No	Yes	No
Numbers of CD34⁺ cells in the periphery	Increased	Increased	Not determined
Serum levels of FLT3L	50–100-fold elevation	2–3-fold elevation	Normal
Regulatory T cells	Reduced in number	Reduced in number	Not determined
GMPs and MLPs	GMPs reduced in number; MLPs absent	Populations expanded	Not determined
Pathogen susceptibility	*Mycobacterium* spp., fungi, viruses (HPV, influenza virus)	*Mycobacterium* spp., fungi, viruses	*Mycobacterium* spp.
Age of patient at diagnosis	7–60 years	<3 months	12–15 months
Inheritance pattern	Autosomal dominant or *de novo* mutation	Autosomal recessive	Autosomal dominant or *de novo* mutation

DC, dendritic cell; DCML, DC, monocyte, B and NK lymphoid; FLT3L, FMS-related tyrosine kinase 3 ligand; GMPs, granulocyte–macrophage progenitors; HPV, human papilloma virus; IFNγ, interferon-γ; IL, interleukin; IRF8, interferon regulatory factor 8; MLPs, multi-lymphoid progenitors; TNF, tumour necrosis factor.

Collin et al., Nature Rev Immunol 2011

Purification / preparation of dendritic cells

DCs – Low population

<1% Spleenocytes
<1% blood leukocytes
<1% tissues (skin, lungs)

Obtaining DC

1992 J Banchereau et C Caux (Lyon)

Bone marrow cells → DC human

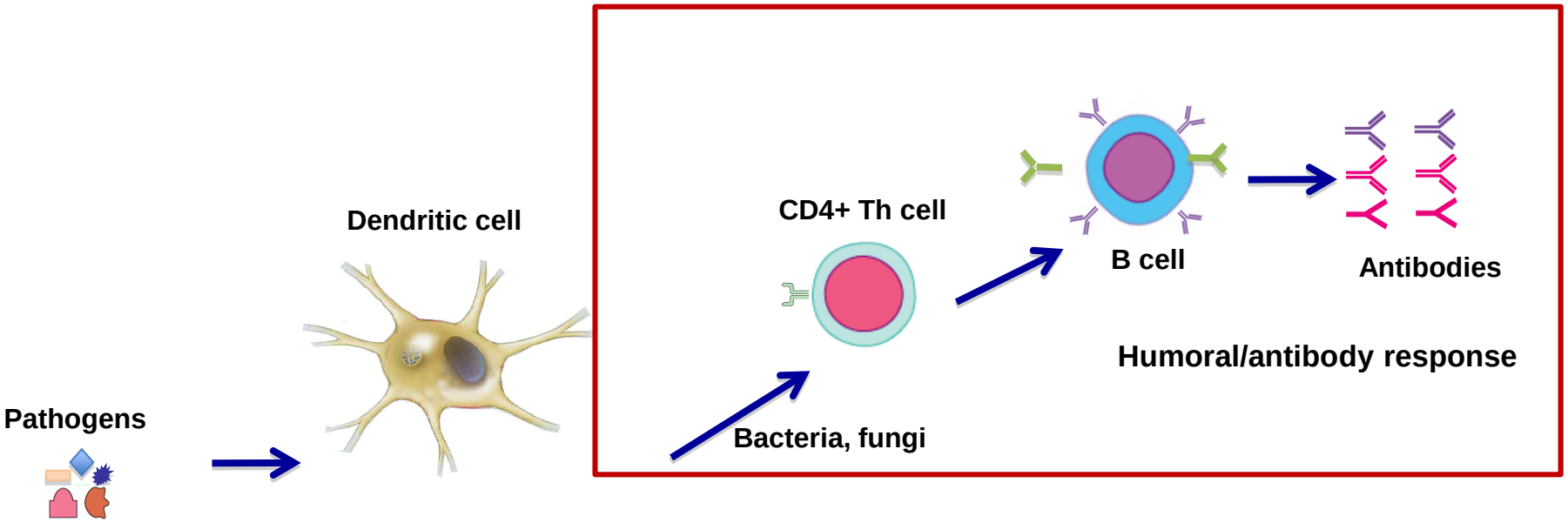
1992 R Steinman (NY)
K Inaba (Kyoto)

Splenic cells → DC murine

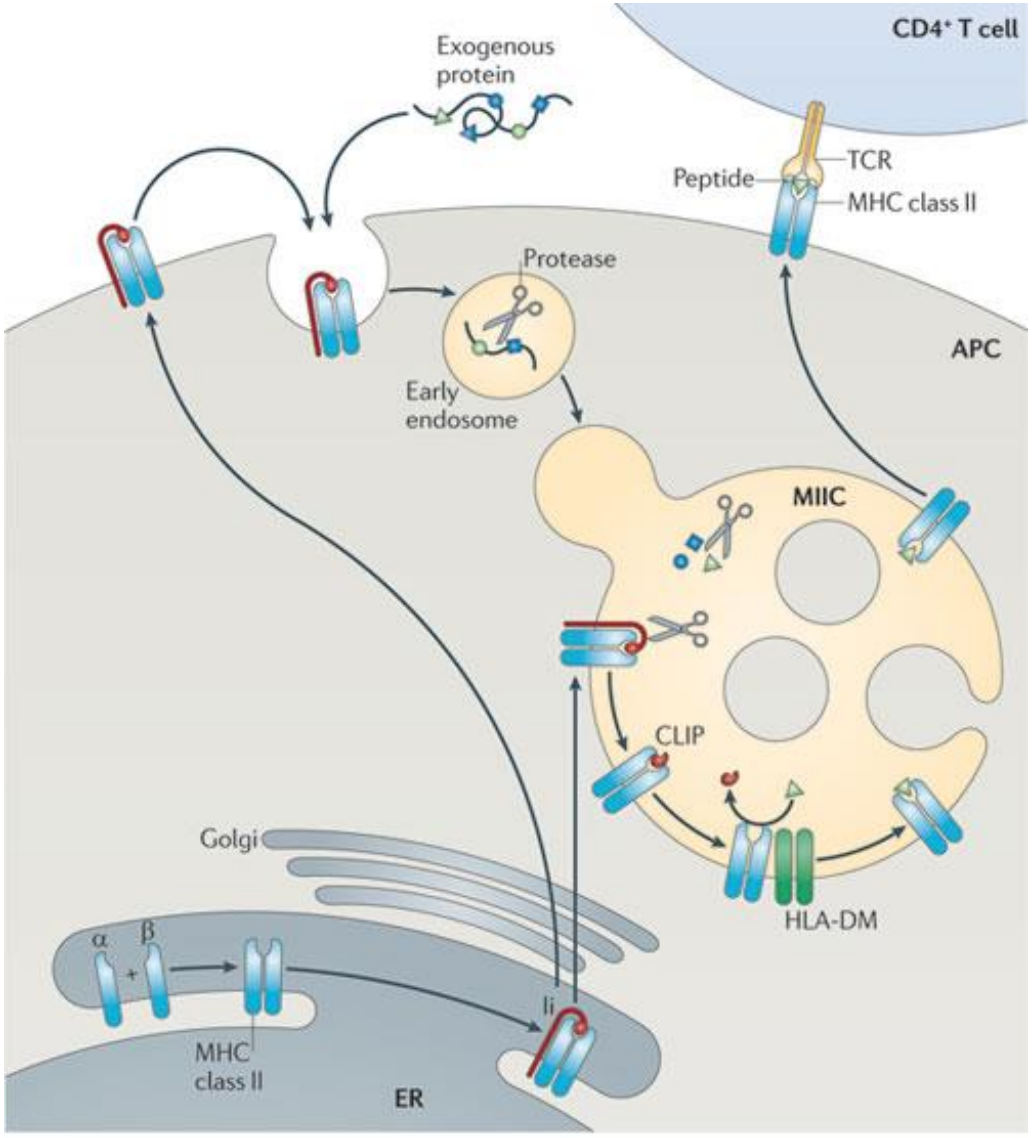
1994 A Lanzavecchia (Bellinzona, Suisse)

Blood monocytes → DC or macrophages

Host immune response to pathogens



MHC Class II presentation by Dendritic cells

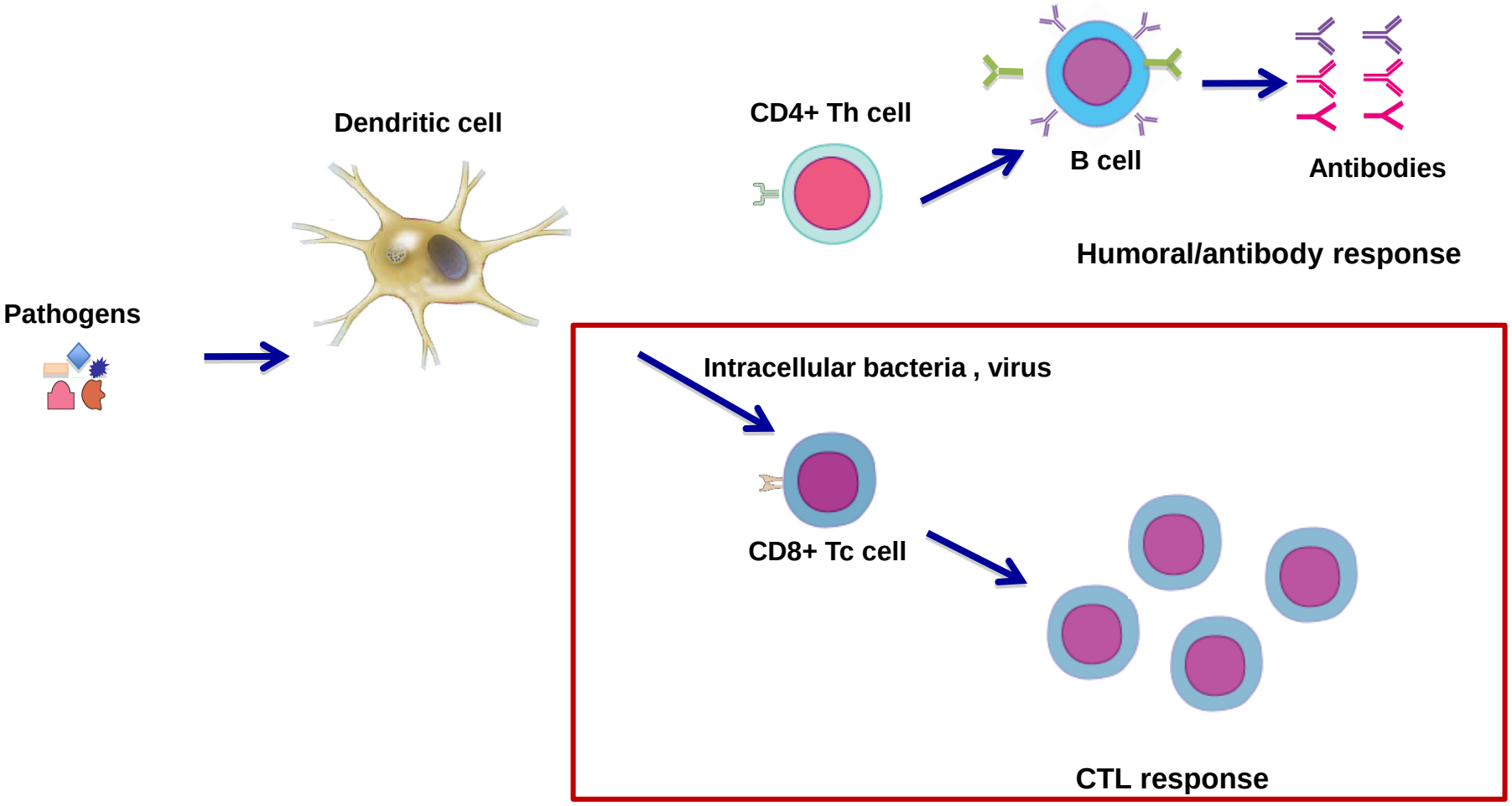


invariant chain (Ii)
MHC class II compartment (MIIC)
class II-associated Ii peptide (CLIP)

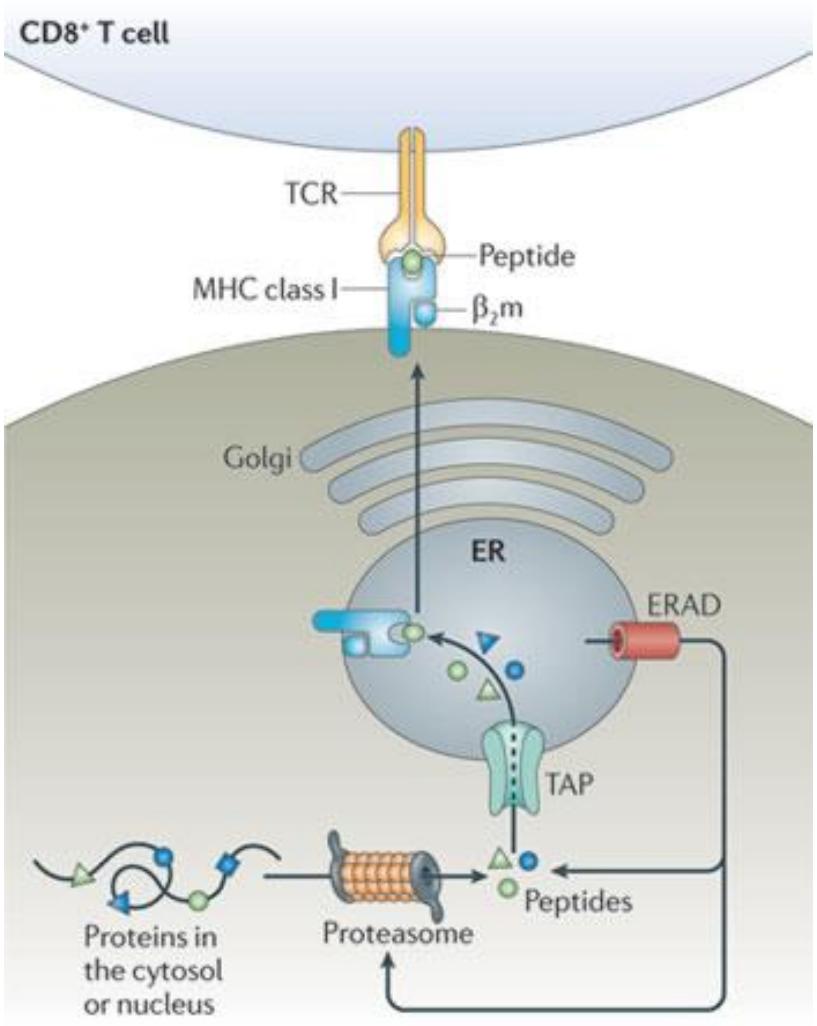
Nature Reviews | Immunology

Neefjes et al 2011

Host immune response to pathogens



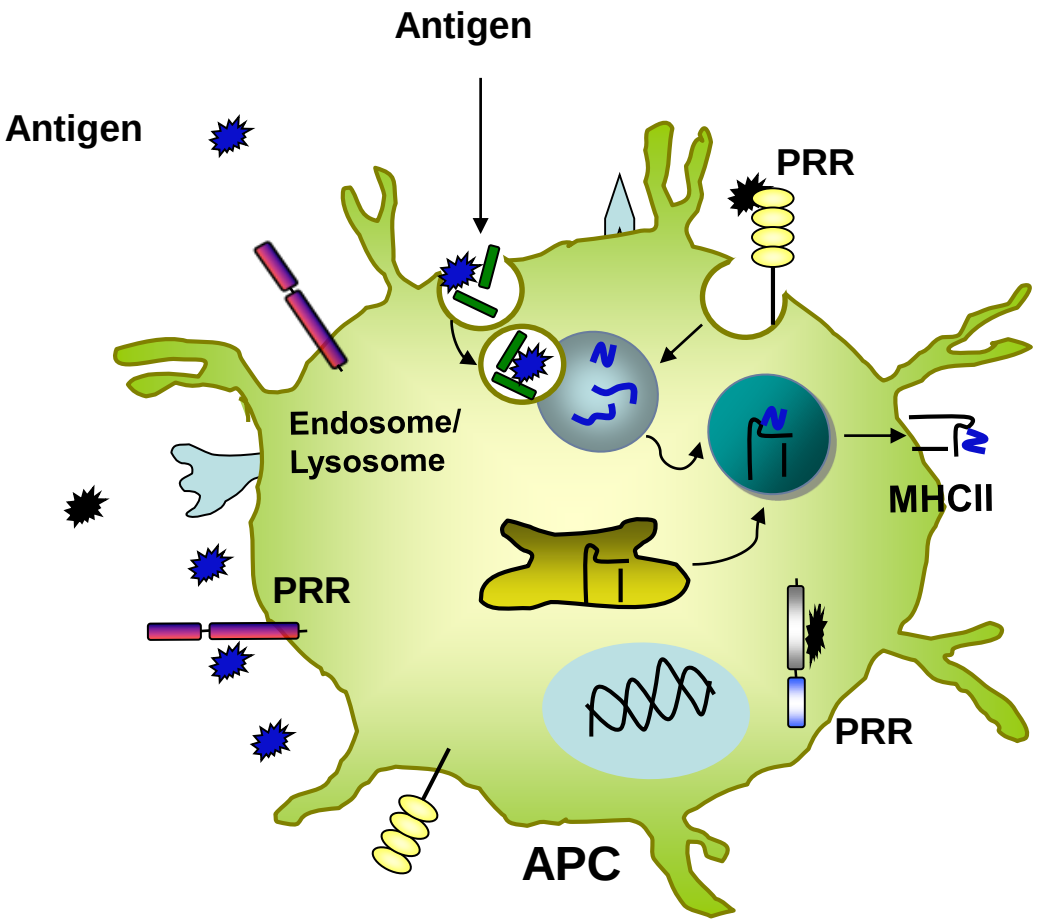
MHC Class I presentation by Dendritic cells



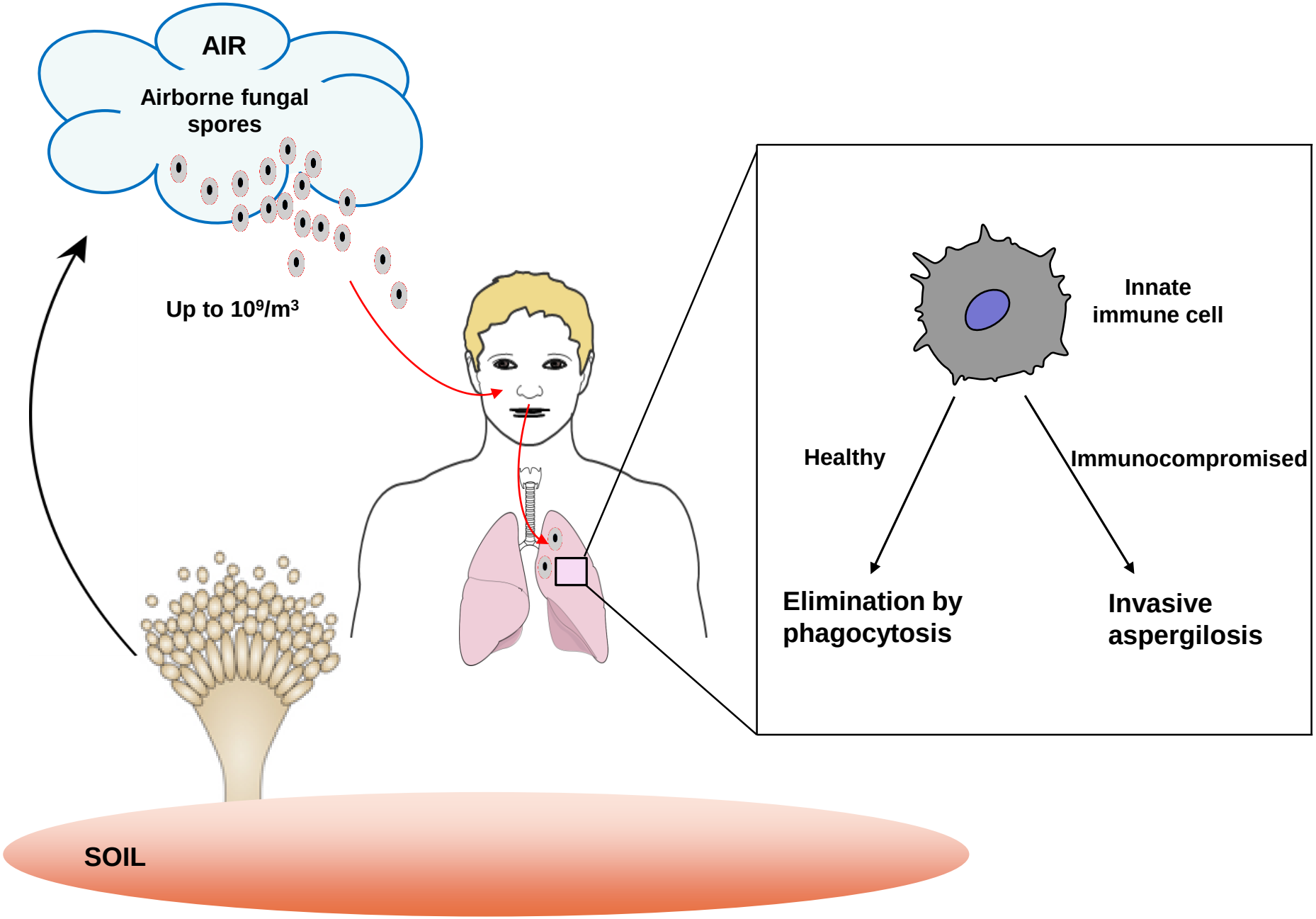
TAP:transporter associated with antigen presentation,
ER: endoplasmic reticulum

Neefjes et al 2011

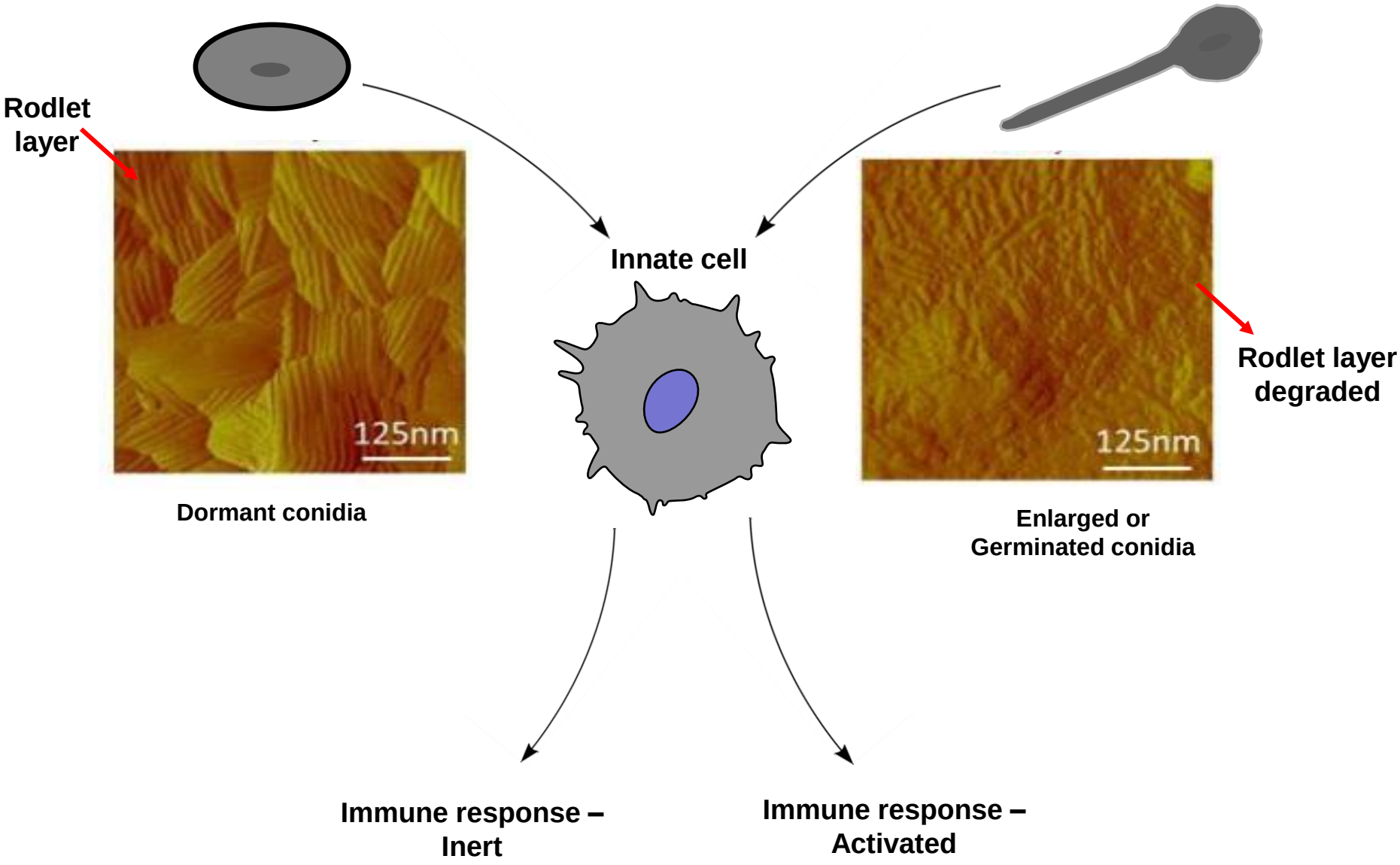
CD4+T cell immune response to pathogens



A. fumigatus


AIR
Airborne fungal spores
Up to $10^9/m^3$
Innate immune cell
Healthy
Immunocompromised
Elimination by phagocytosis
Invasive aspergilosis
SOIL

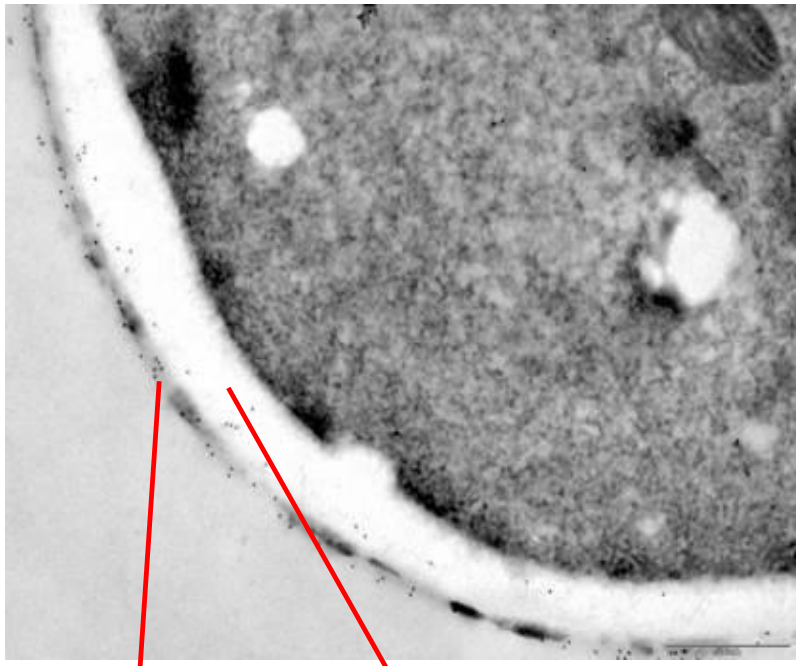
A. fumigatus sensing by innate cells – Role of Rodlet



Aimanianda, Bayry* et al Nature. 2009*

A. fumigatus – structural organization of the conidia

Electron micrograph of fixed conidia



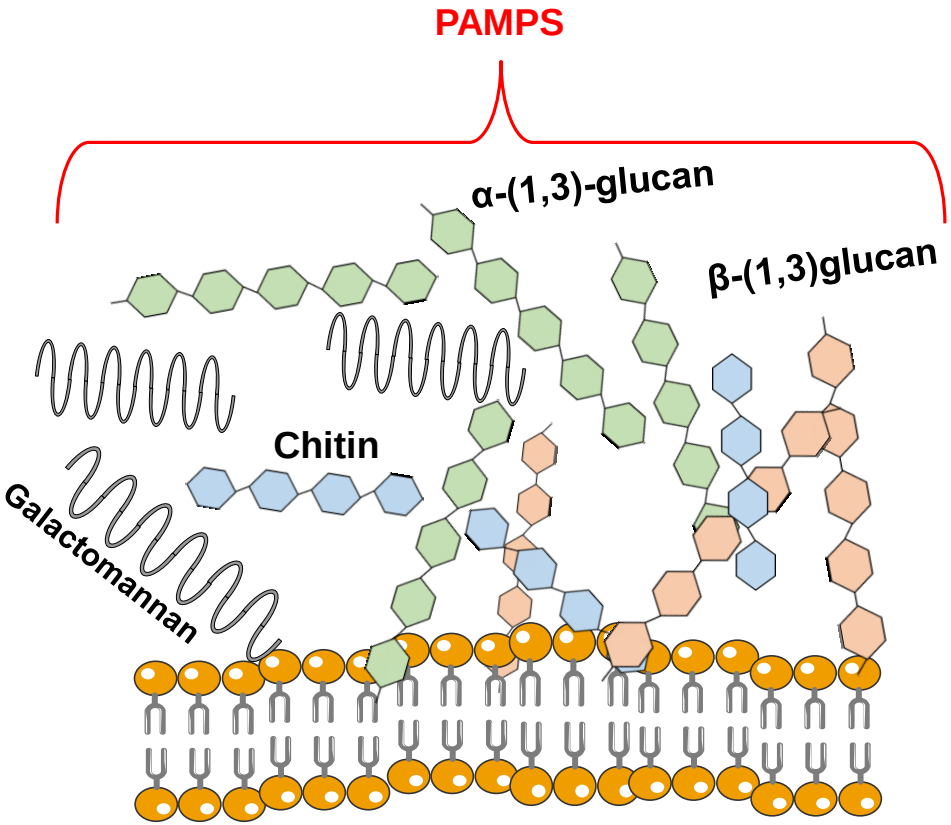
Courtesy: Beauvais A

Rodlet layer

Polysaccharides

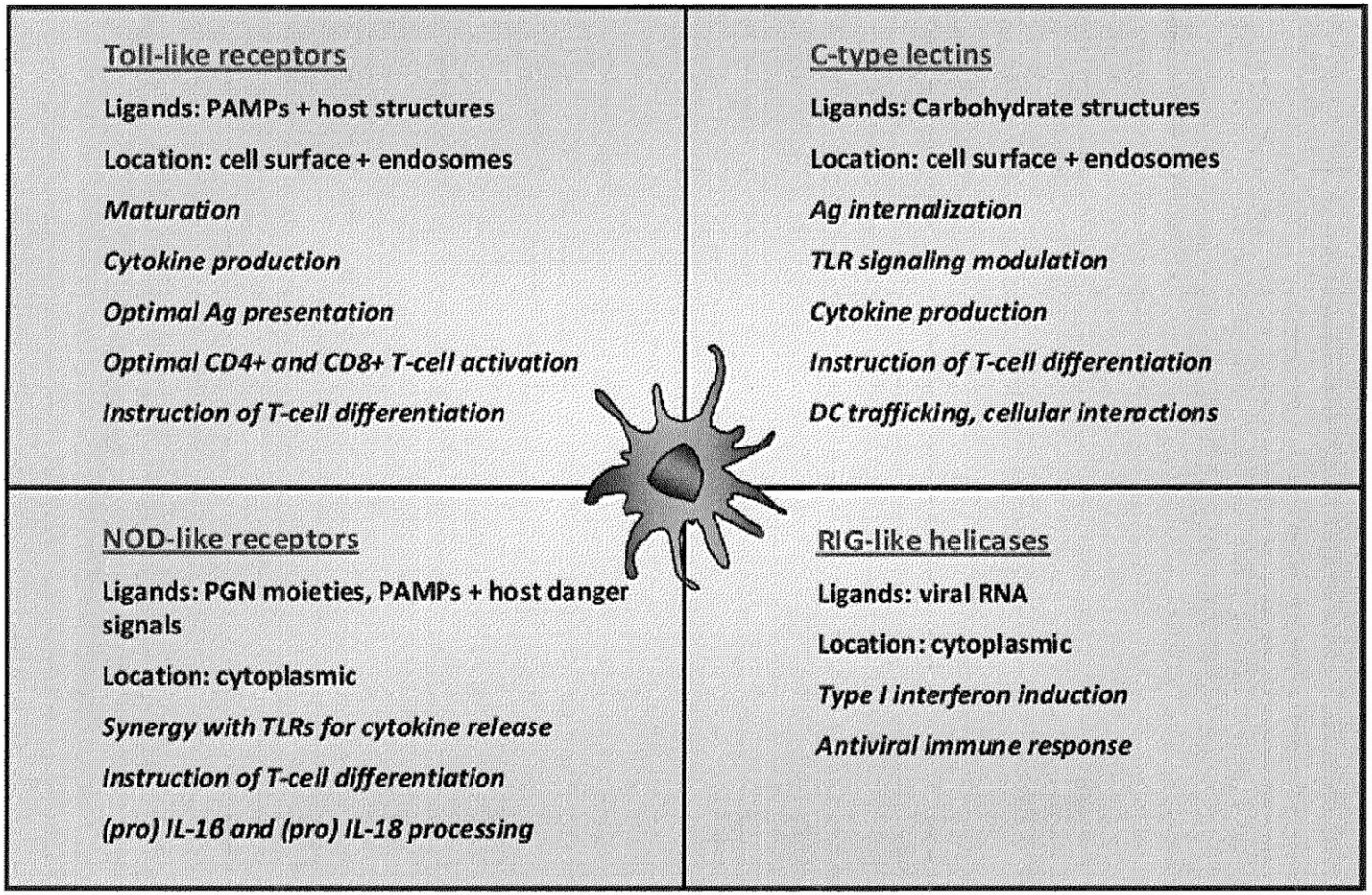
Proteins

Lipids



Exposure of polysaccharides during germination

PRRs on dendritic cells

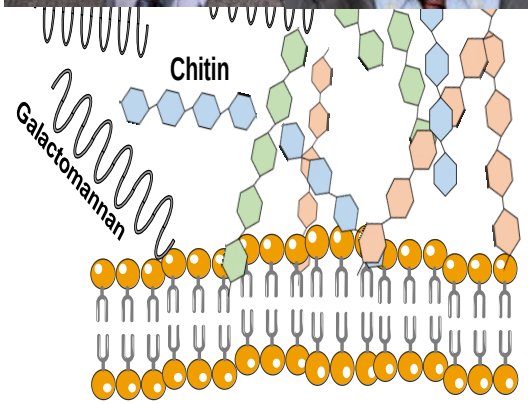


General characteristics of the four main pattern recognition receptor families expressed by DCs. The most important effects of PRR triggering are given in italics. Abbreviations: *PAMP*, pathogen-associated molecular pattern; *Ag*, antigen; *PGN*, peptidoglycan

PRRS and DCs – 2011 Noble prize

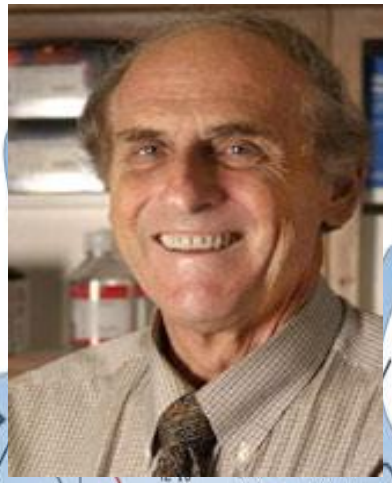
Bruce Beutler

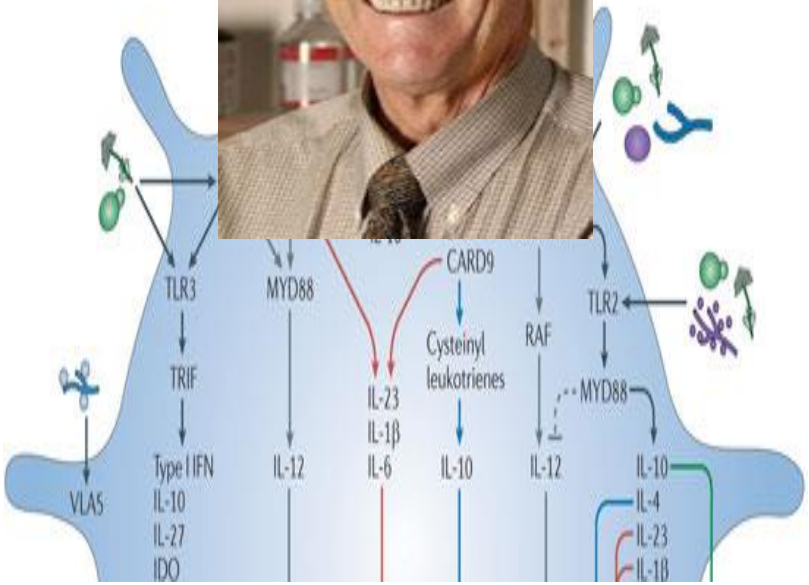
Jules Hoffmann



Toll receptors

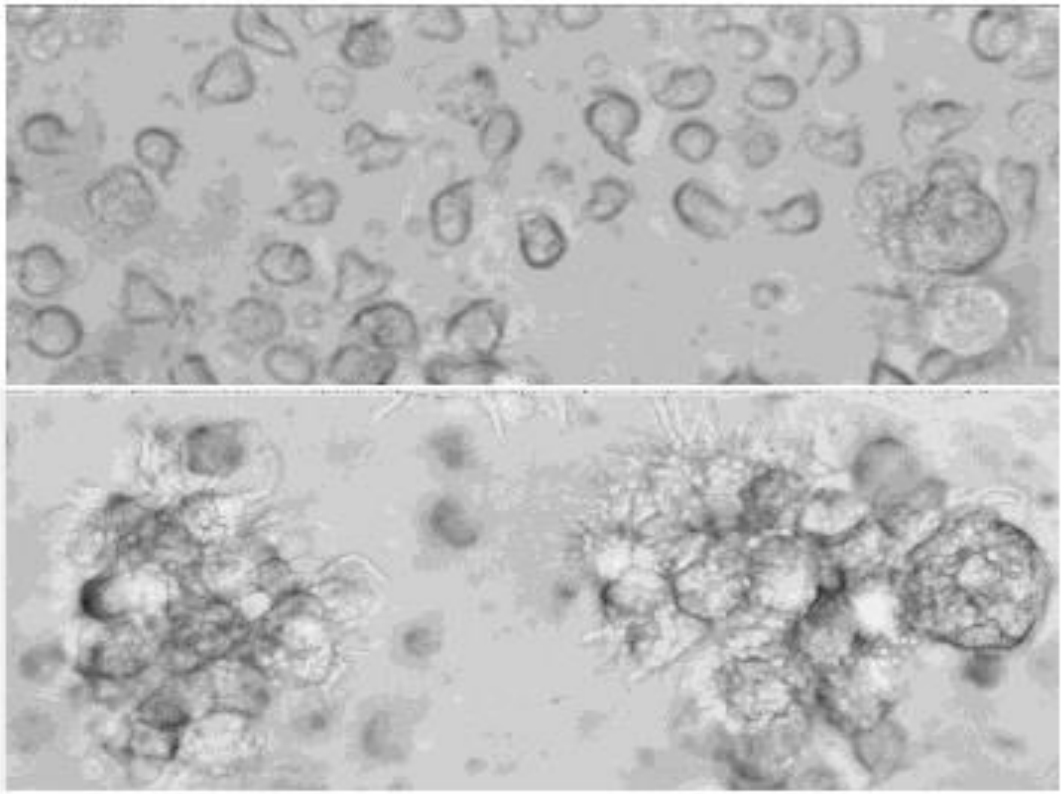
Ralph Seinman





Dendritic cells

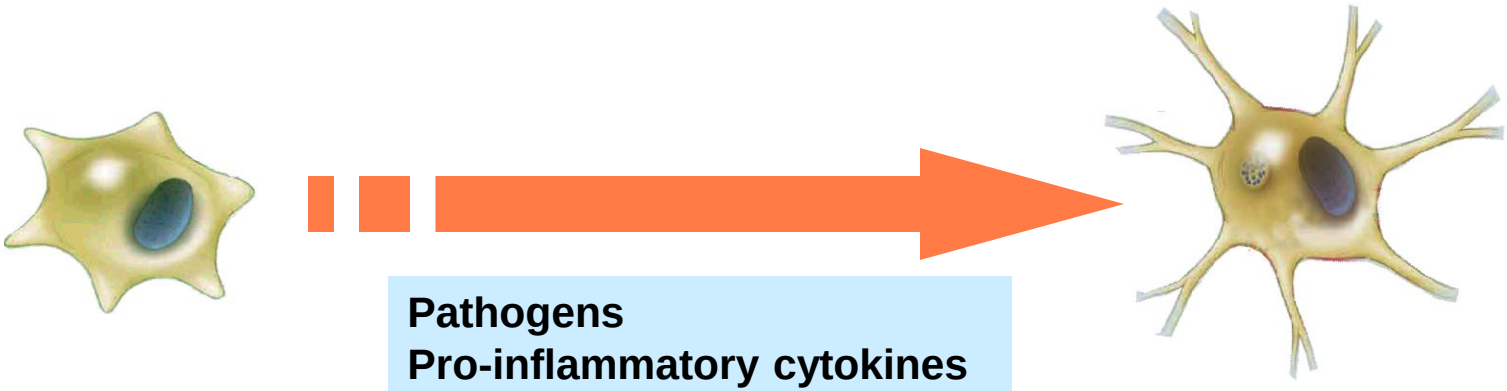
Changes during dendritic cell maturation



Immature DC

Mature DC

Changes during dendritic cell maturation



Pathogens
Pro-inflammatory cytokines
T cells

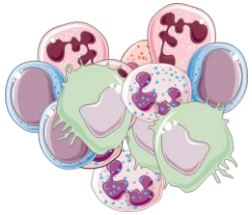
Immature DC

Mature DC

High intracellular HLA II
High endocytosis,
Low CD54, 58, 80, 86
Low CD40,
Low CD83
Low granule antigens

High surface HLA II
Low endocytosis
High CD54, 58, 80, 86
High CD40,
High CD83
Production of inflammatory cytokines

Methodology



PBMC

CD14
Monocytes

GM-CSF & IL-4
5 days

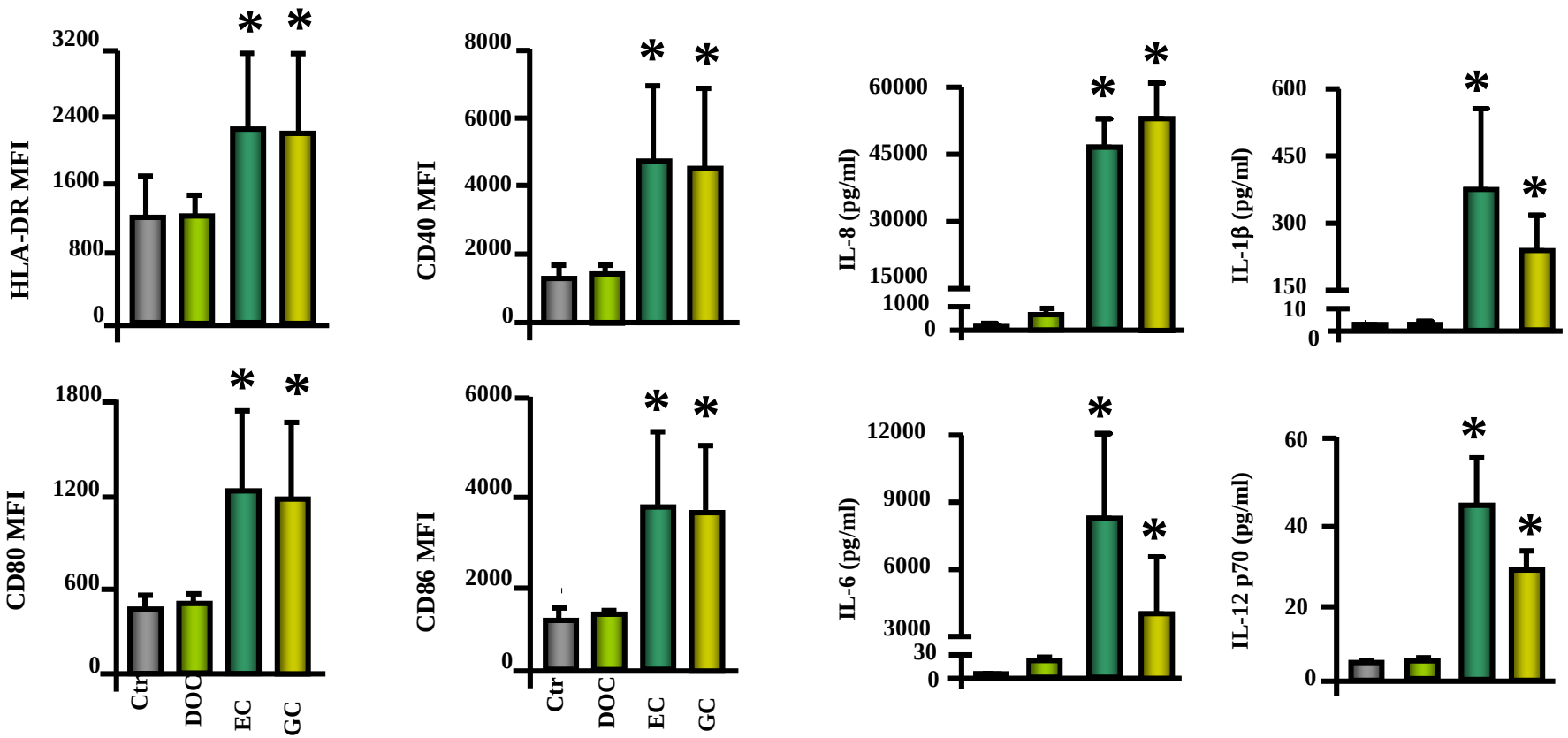
MoDC

A. fumigatus
(morphotypes)
Dormant, Enlarged,
Germinated conidia

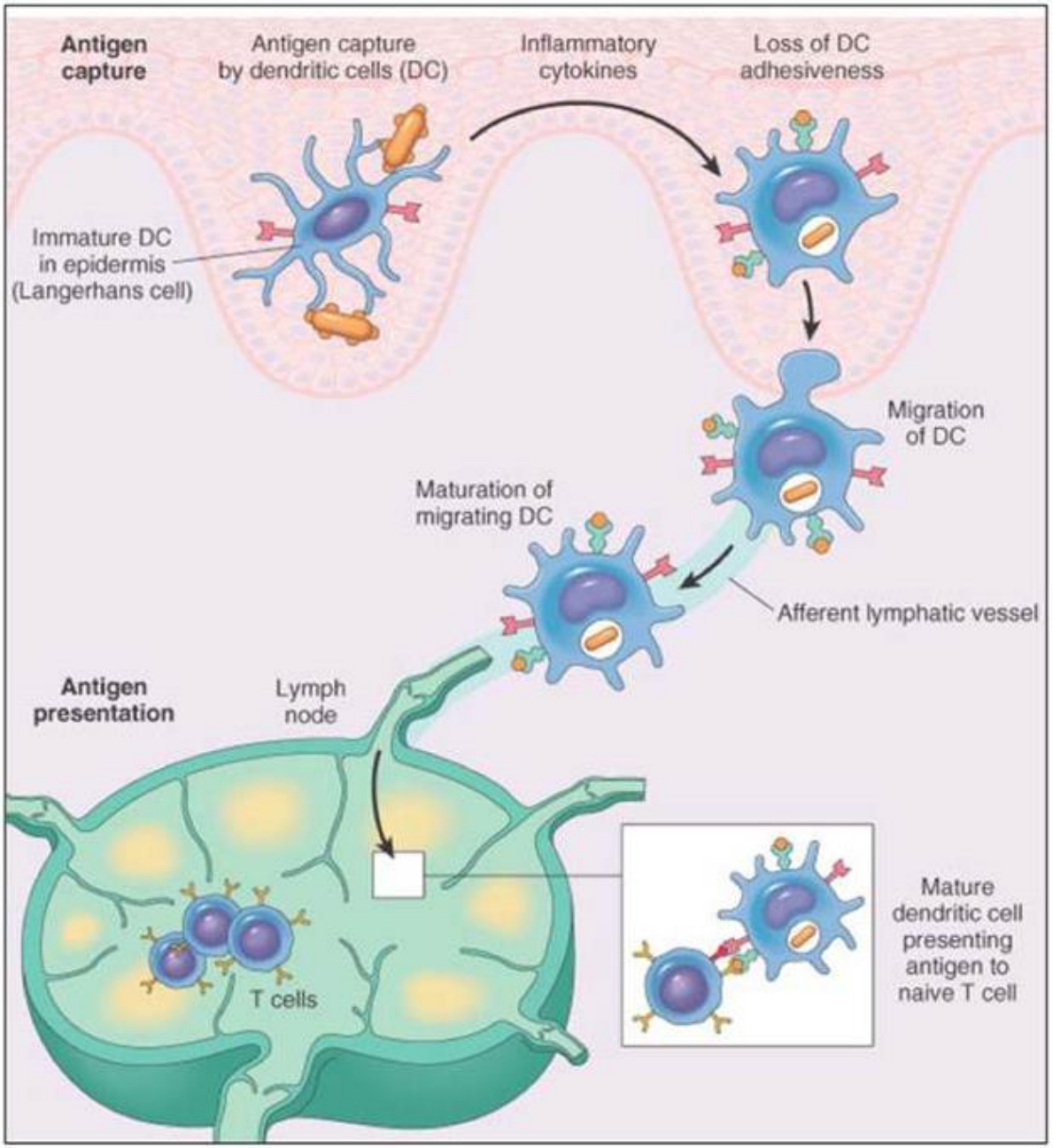
48 hrs

DC responses

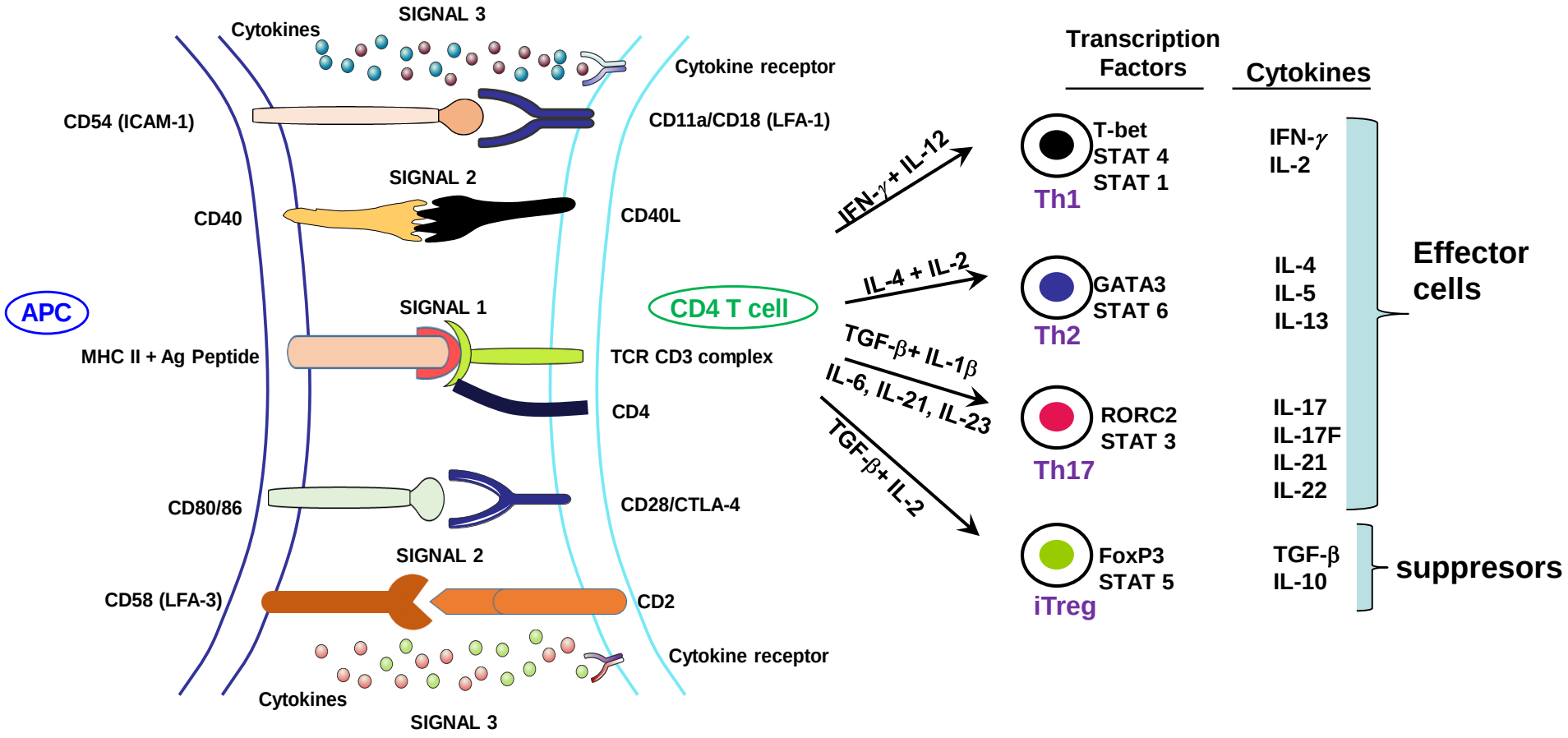
Enlarged and germinated conidia but not dormant conidia induce maturation and activation of human dendritic cells



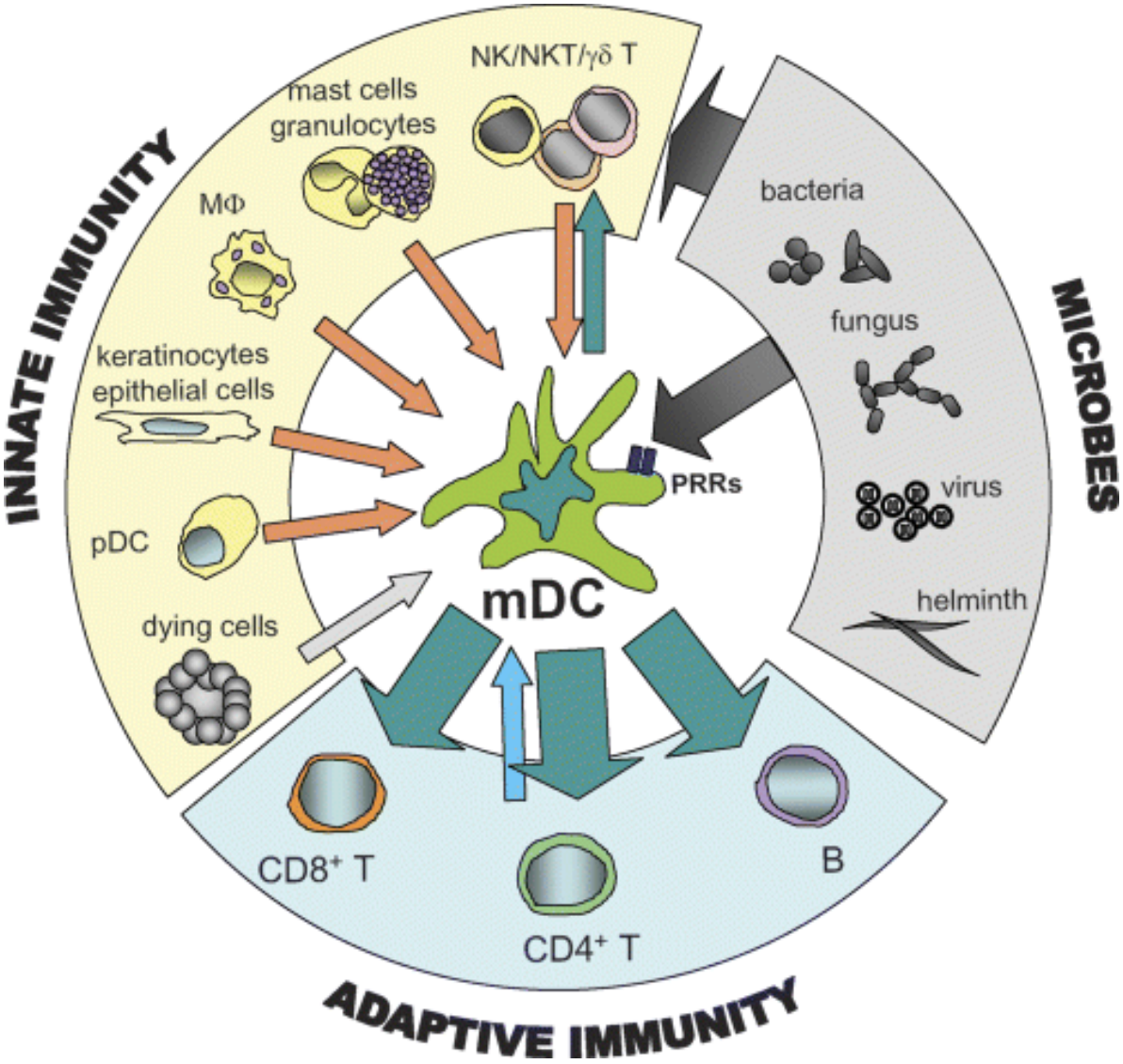
Migration of dendritic cells to draining lymph nodes



CD4+T cell immune response to pathogens



Dendritic cells in health and disease



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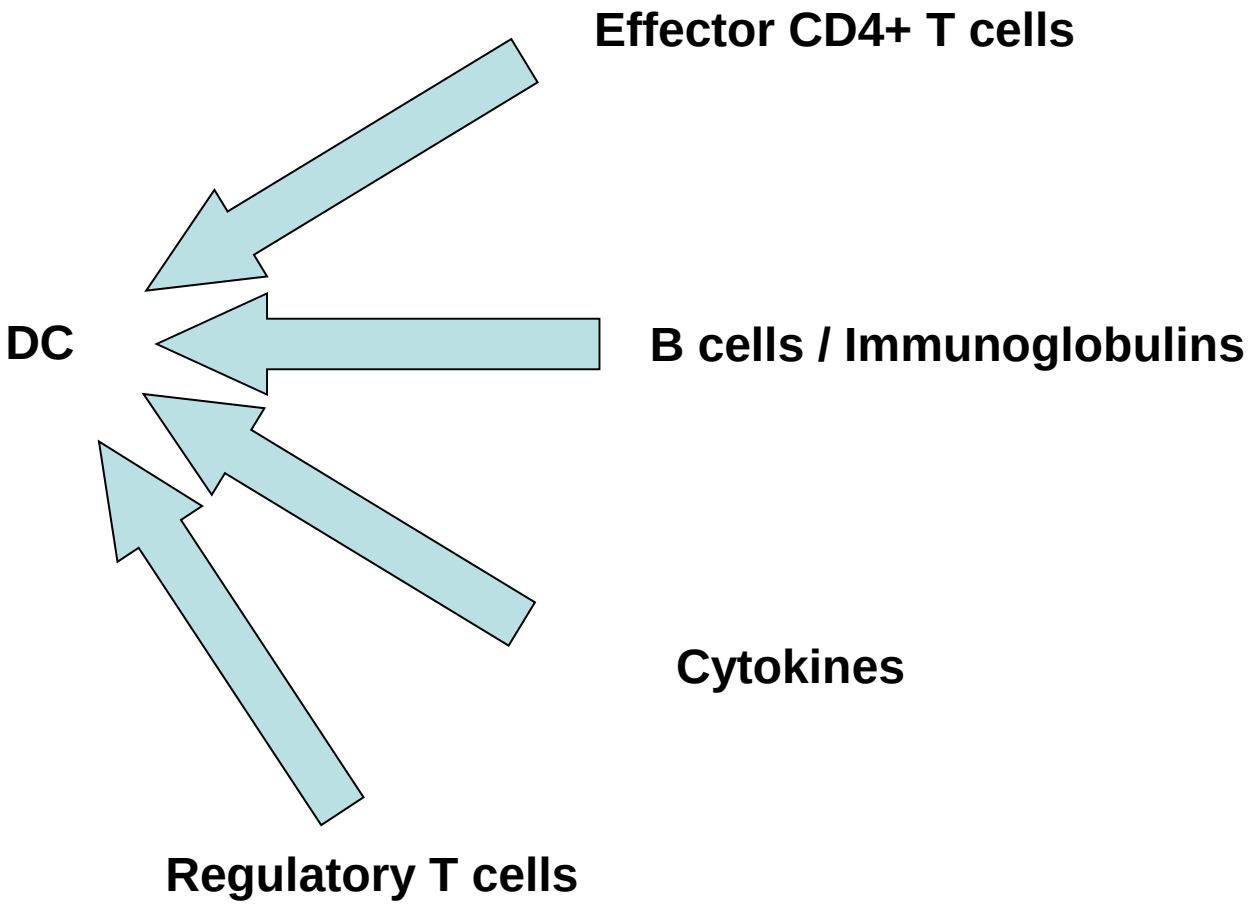
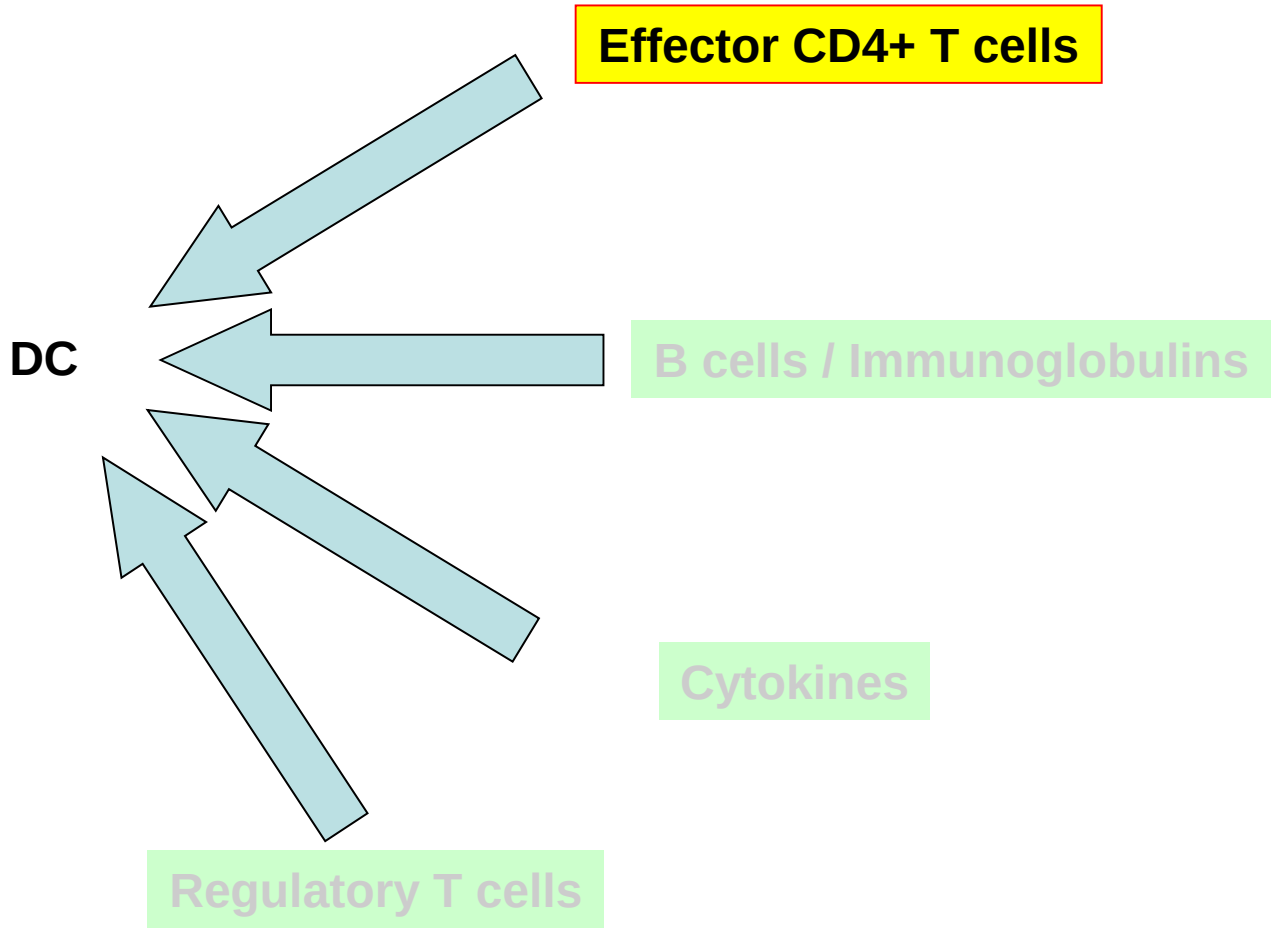
http://onlinelibrary.wiley.com/doi/10.1111/j.1600-065X.2007.00551.x/full#f1

Regulation of Dendritic cell functions by immune compartment

Regulation of Dendritic cell functions by immune compartment

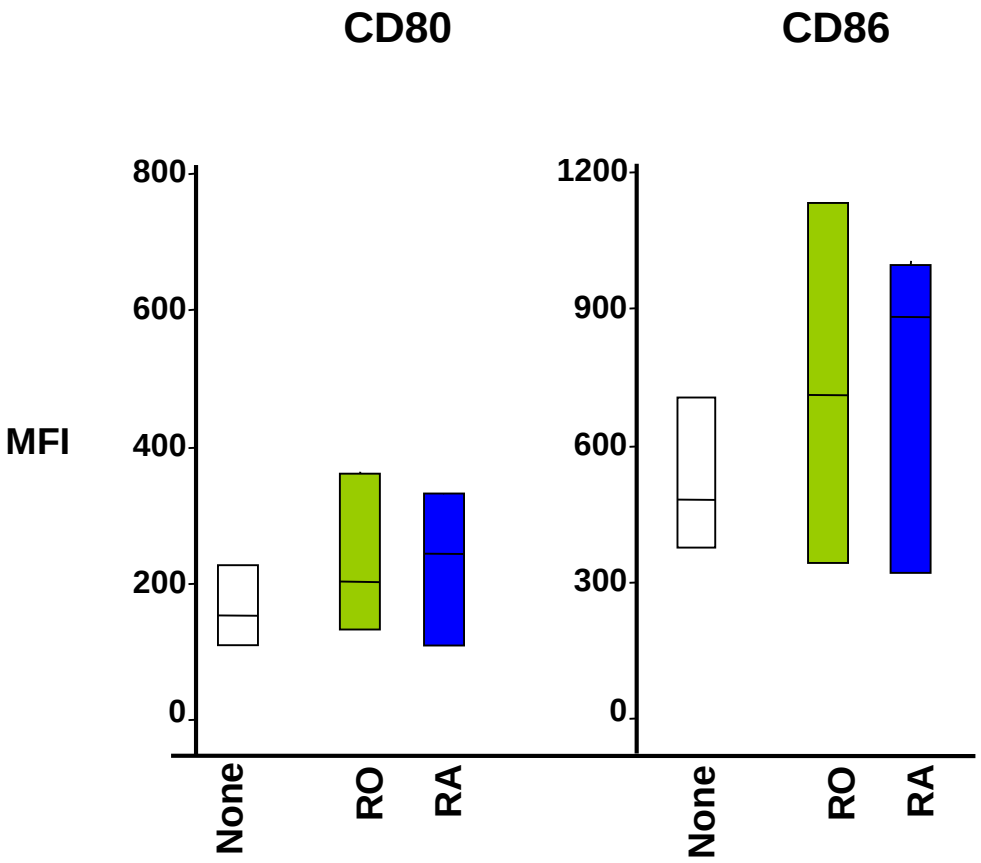


Regulation of Dendritic cell functions by immune compartment

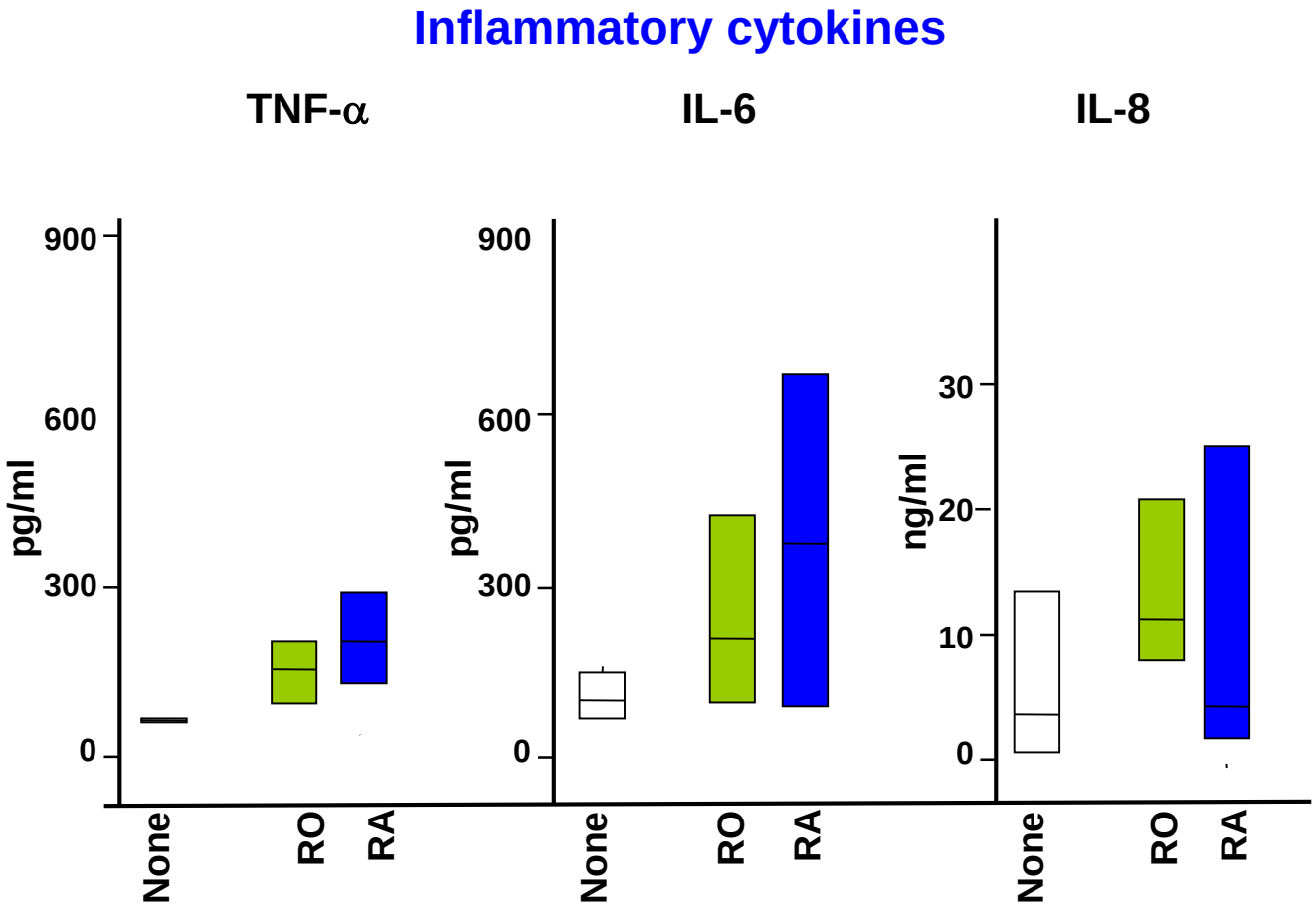


T cells induce activation of dendritic cells

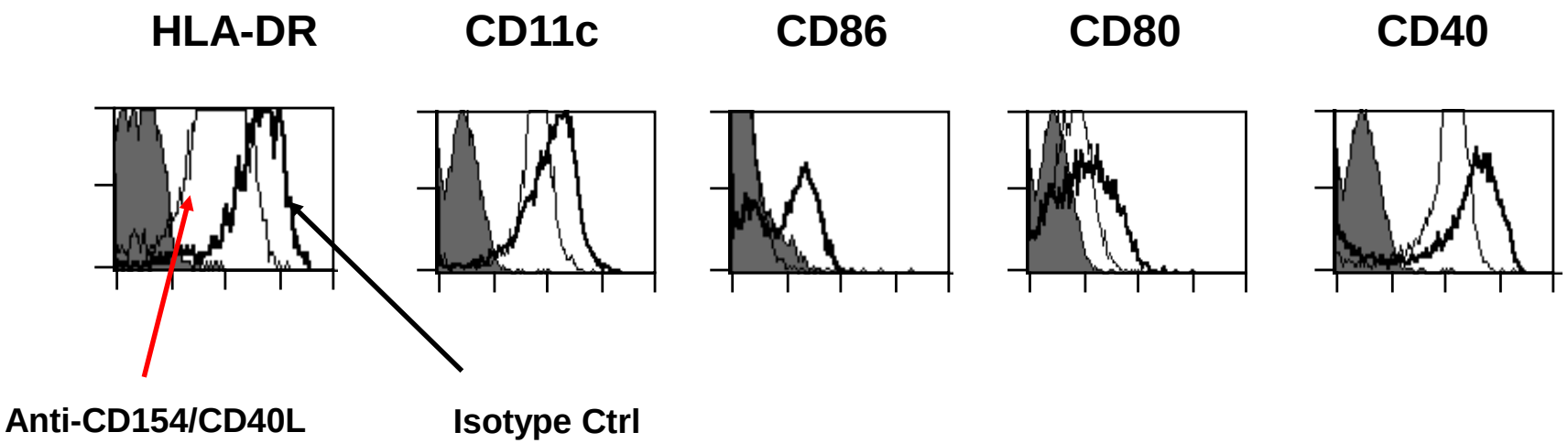
Co-stimulatory molecules



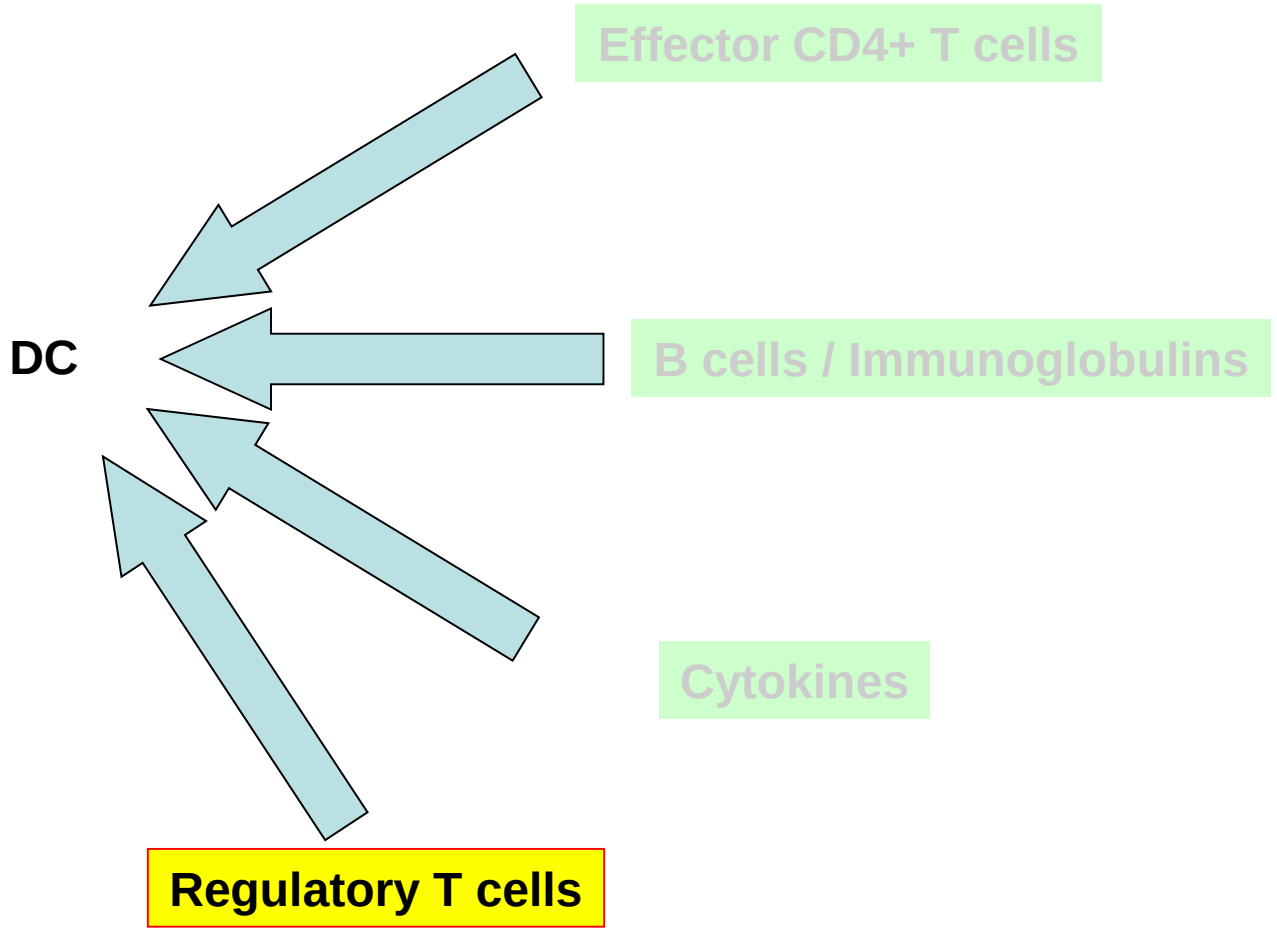
T cells induce activation of dendritic cells



Stimulation of dendritic cells by CD40L (CD154) on T cells leads to maturation



Regulation of Dendritic cell functions by immune compartment

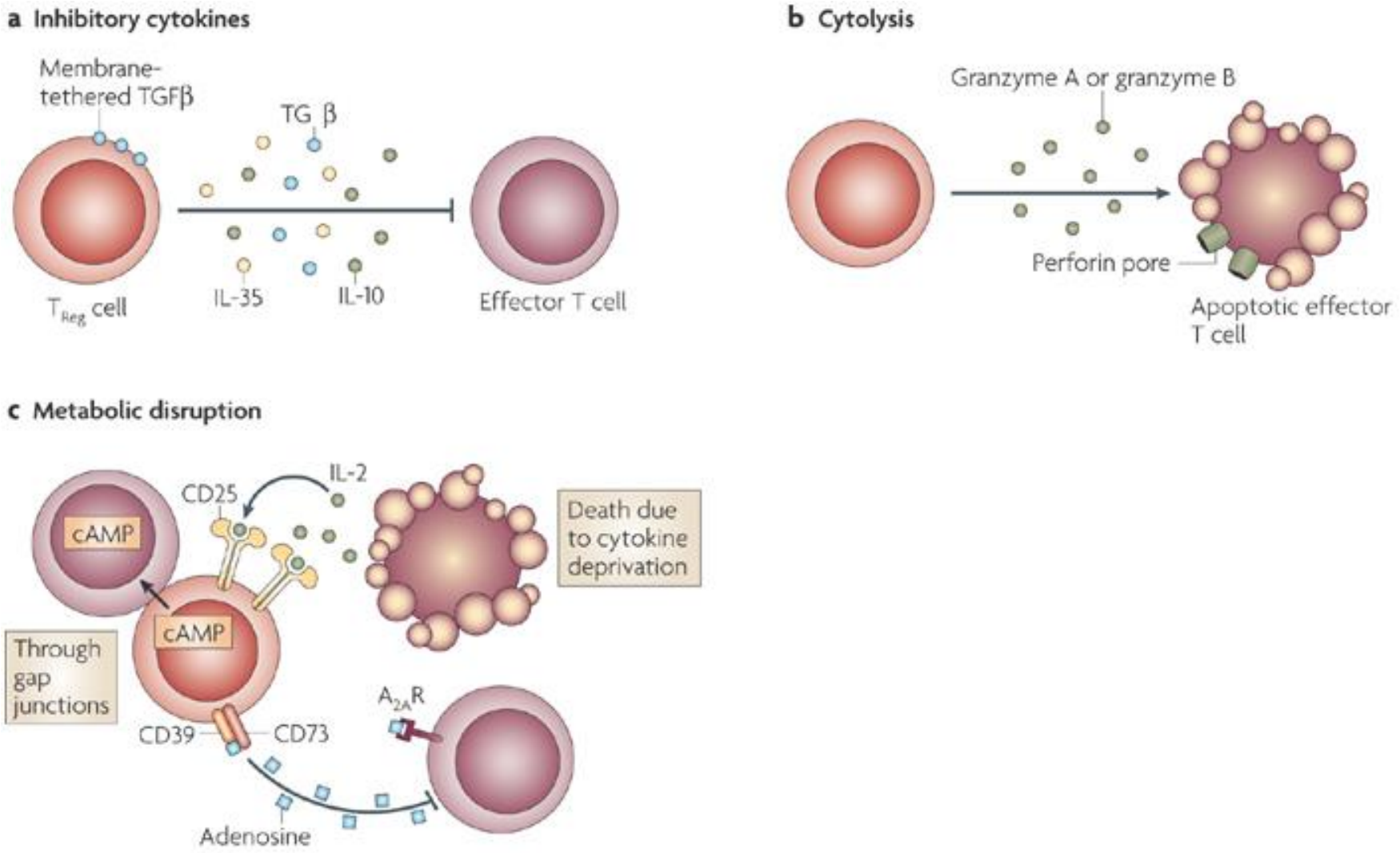


Dendritic cells and regulatory T cells

CD4+CD25^{+} regulatory T cells in human circulation

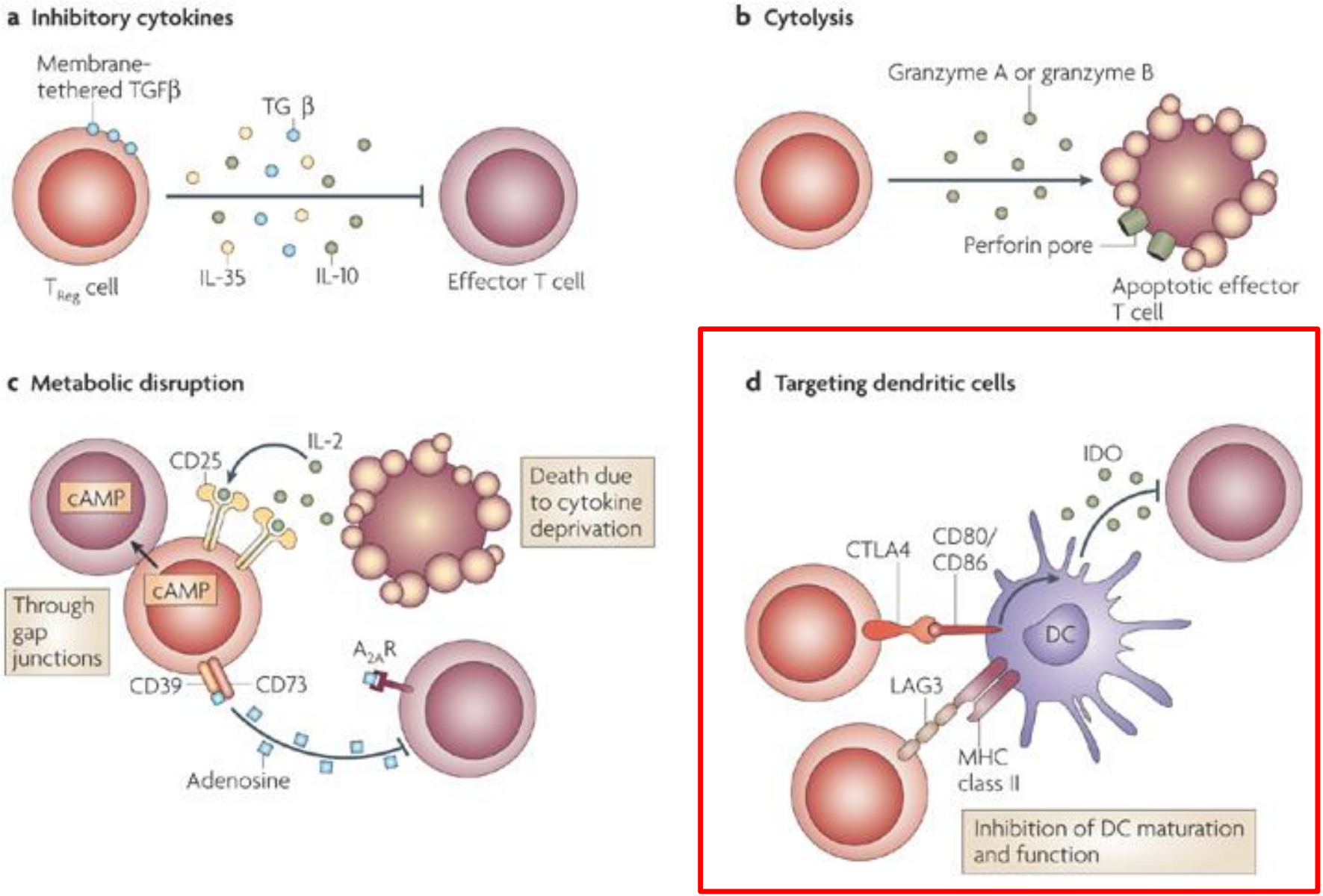
- **Represent ~2-4% of total PBMCs**
- **Express CD25**
- ***FoxP3* (transcription factor forkhead)**
- **Intracellular CTLA-4 Expression**
- **Immunosuppressors**

CD4+CD25+ regulatory T cells: Mechanisms of suppression



Vignali et al., 2008; Nat Rev Immunol

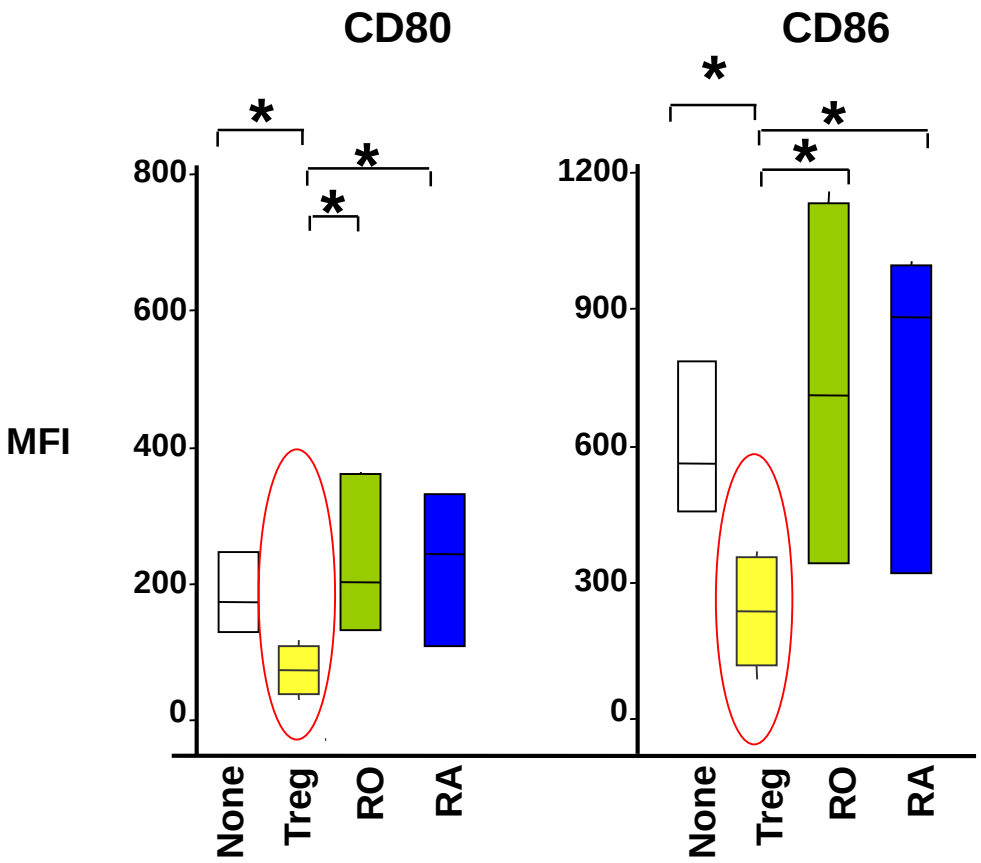
CD4+CD25+ regulatory T cells: Mechanisms of suppression



Vignali et al., 2008; Nat Rev Immunol

CD4+CD25+ regulatory T cells inhibit TLR-mediated activation of human dendritic cells

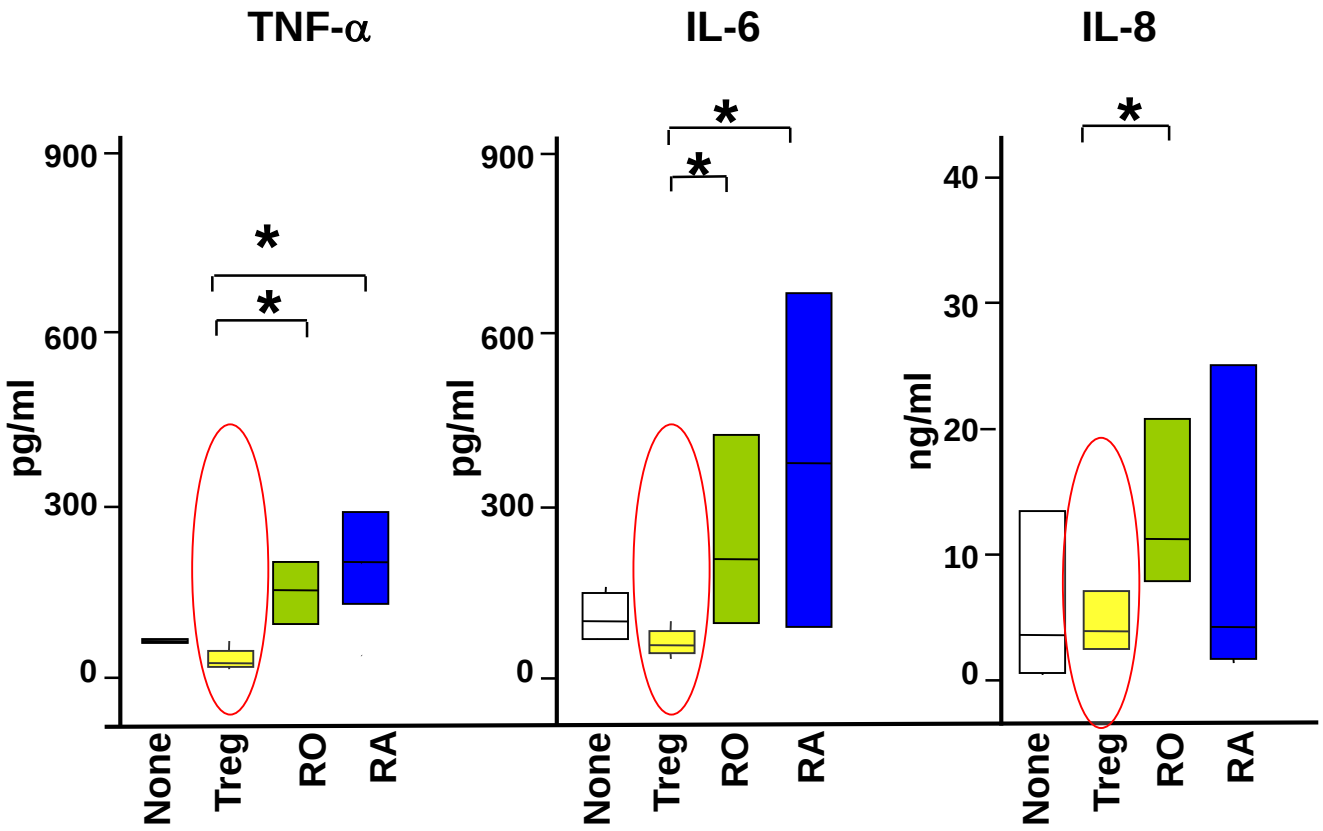
Co-stimulatory molecules



J Immunol 2004
J Immunol 2007
Arthritis Rheum 2007
Nature Rev Immunol 2008
Nature 2008
Arthritis Rheum 2009
Nature Rev Rheumatol 2009
Am J Pathol 2009
Vaccine 2011

CD4+CD25+ regulatory T cells inhibit TLR-mediated activation of human dendritic cells

Inflammatory cytokines



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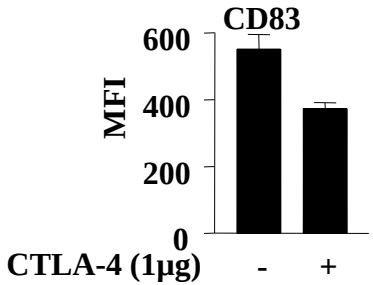
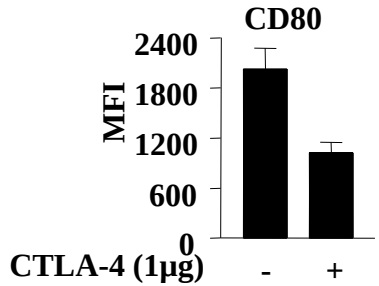
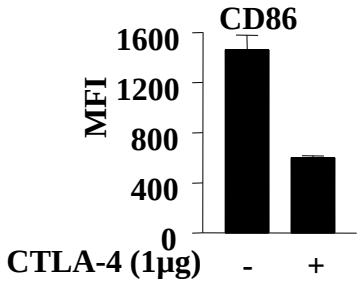
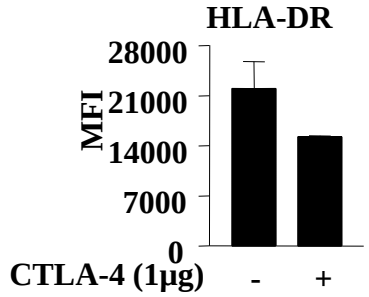
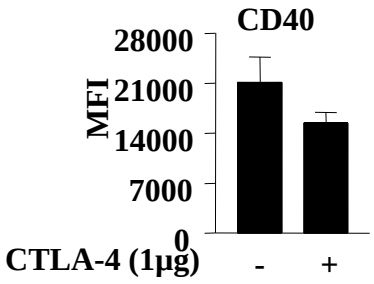
CTLA-4 is the major molecule implicated in the CD4+CD25+ regulatory T cell-mediated inhibition of dendritic cells

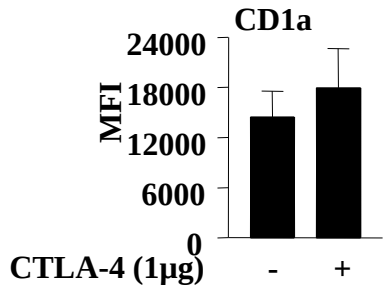




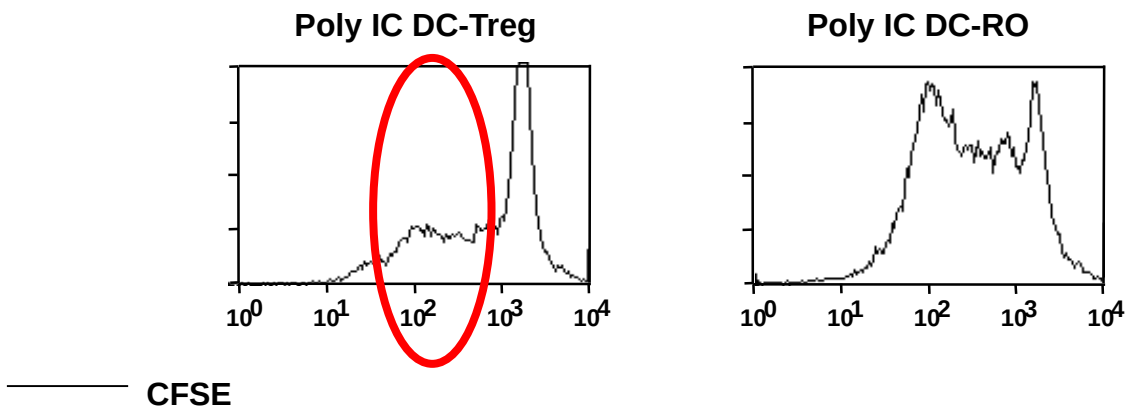






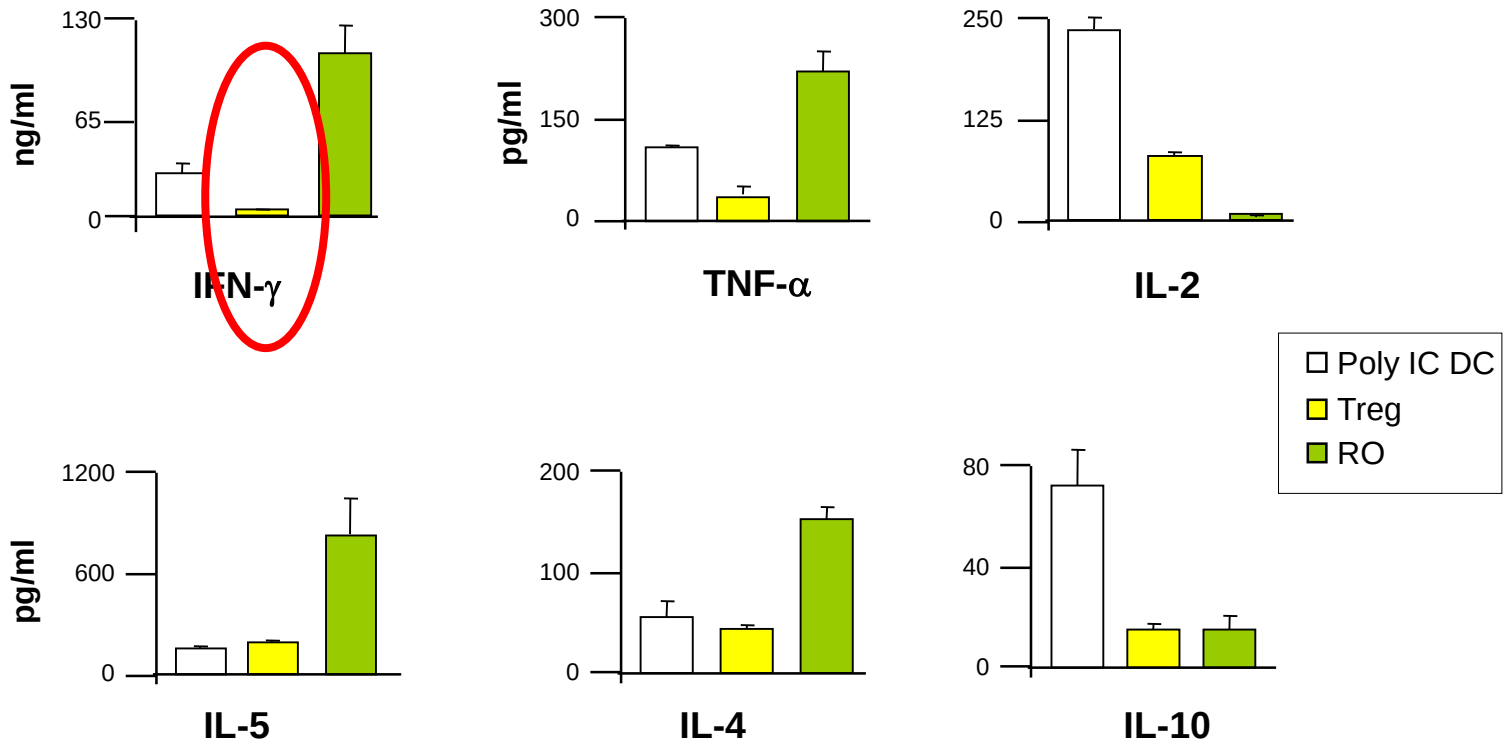


CD4+CD25+ regulatory T cells inhibit T cell proliferation mediated by TLR-activated human dendritic cells



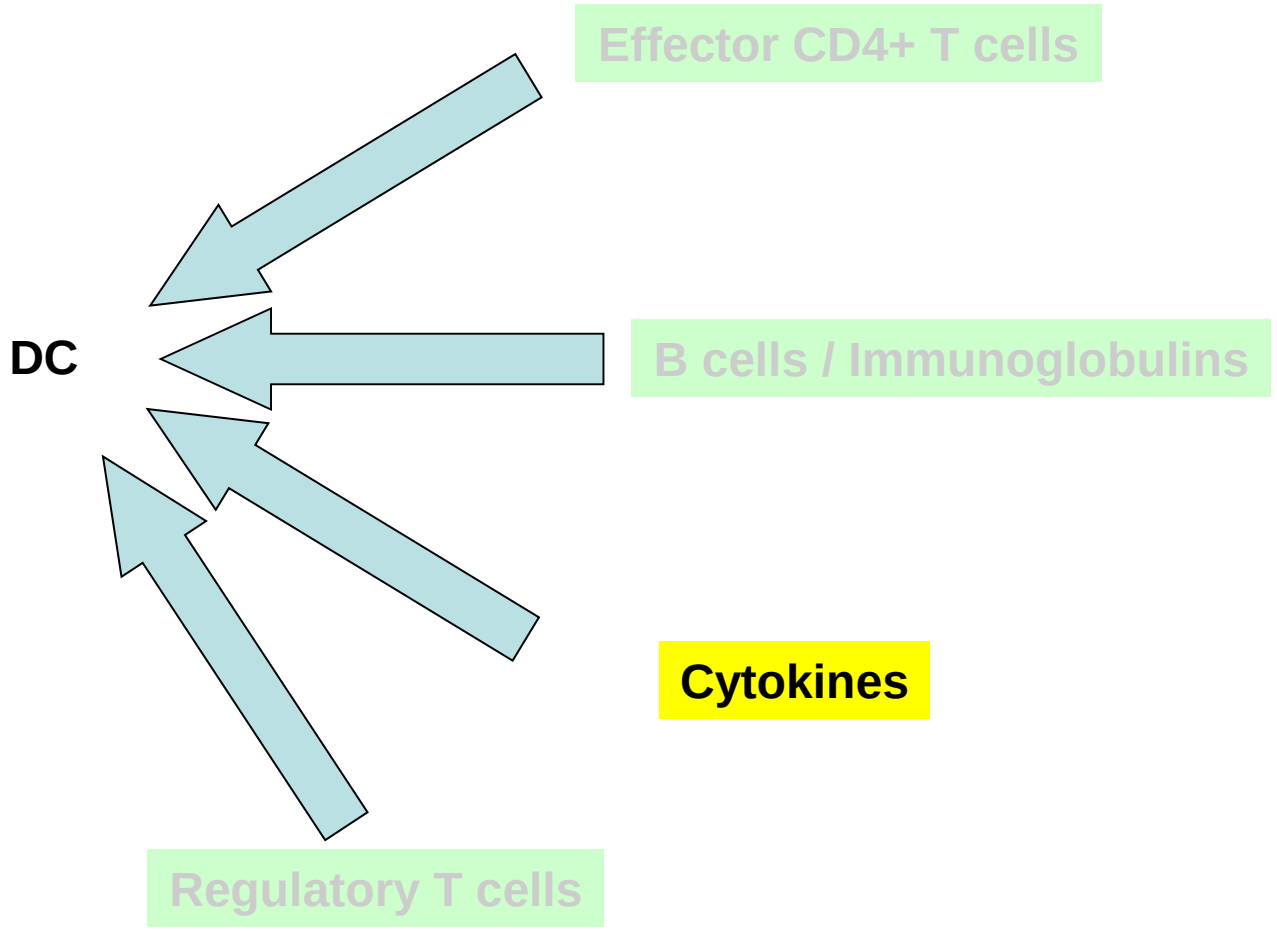
Bayry et al., J Immunol. 2007

CD4+CD25+ regulatory T cells inhibit DC-mediated IFN-responses

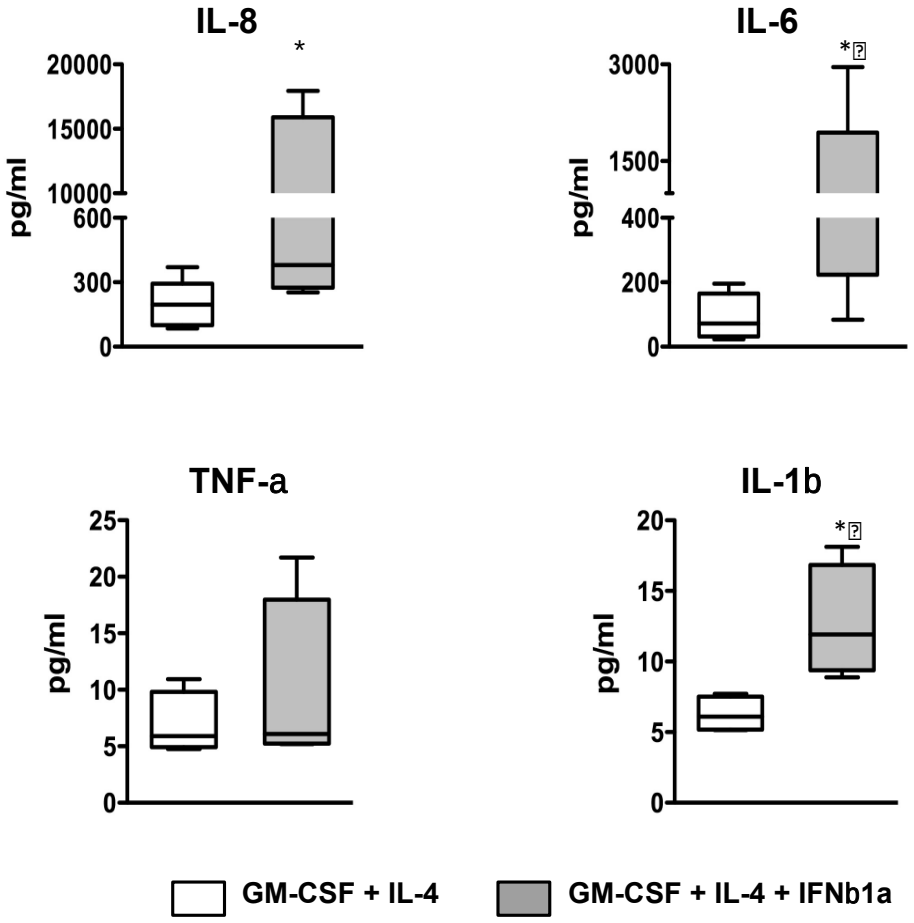


Bayry et al., J Immunol. 2007

Regulation of Dendritic cell functions by immune compartment



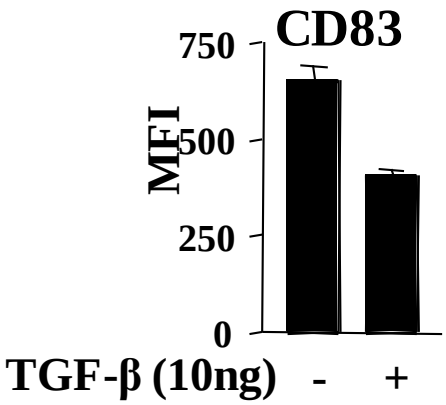
Regulation of Dendritic cell functions by immune compartment

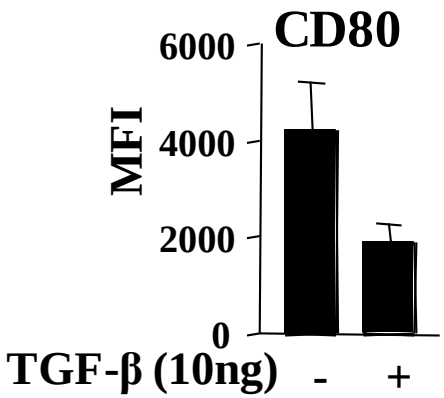


Type I interferon:
Promotion of activation of DC

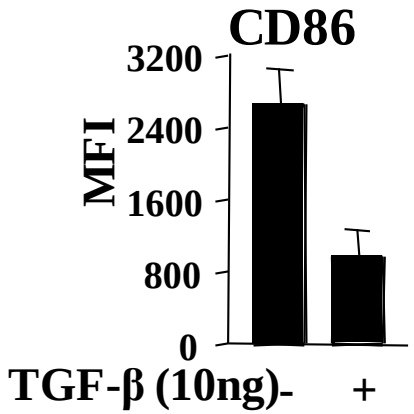
Maddur et al., Arthrits Rheum 2010

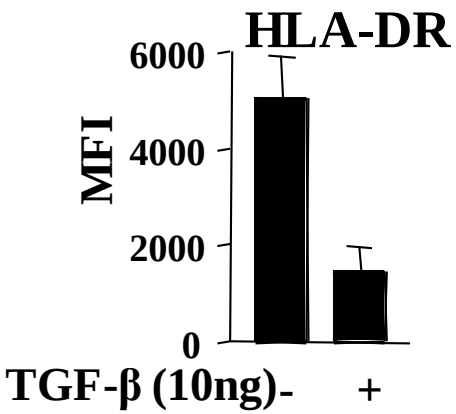
Regulation of Dendritic cell functions by immune compartment



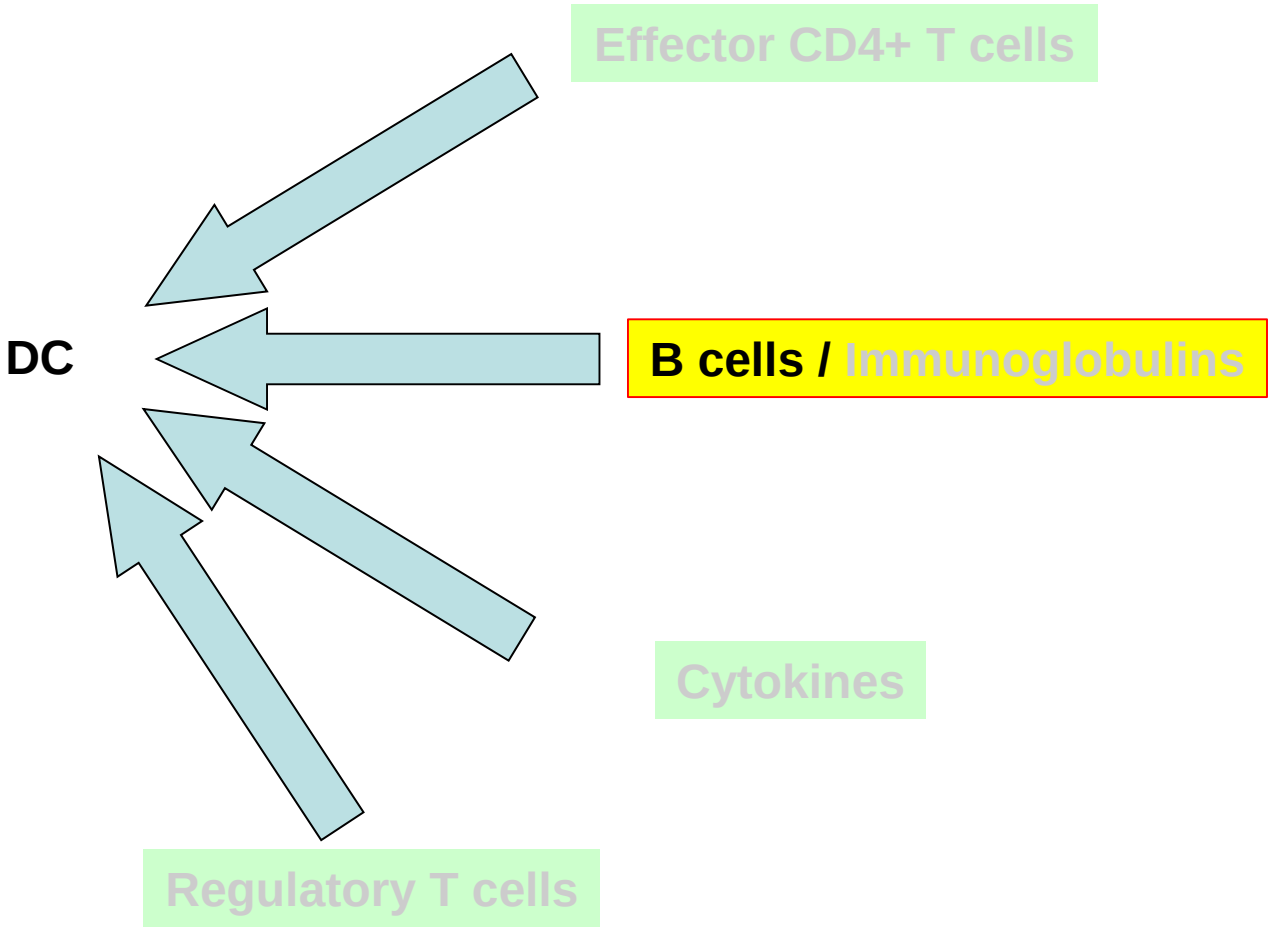


TGF-β: inhibit the DC maturation

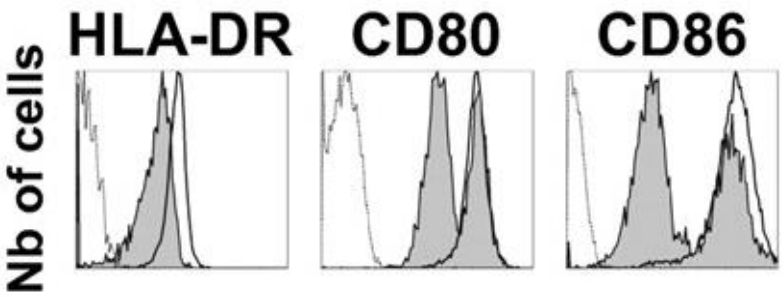




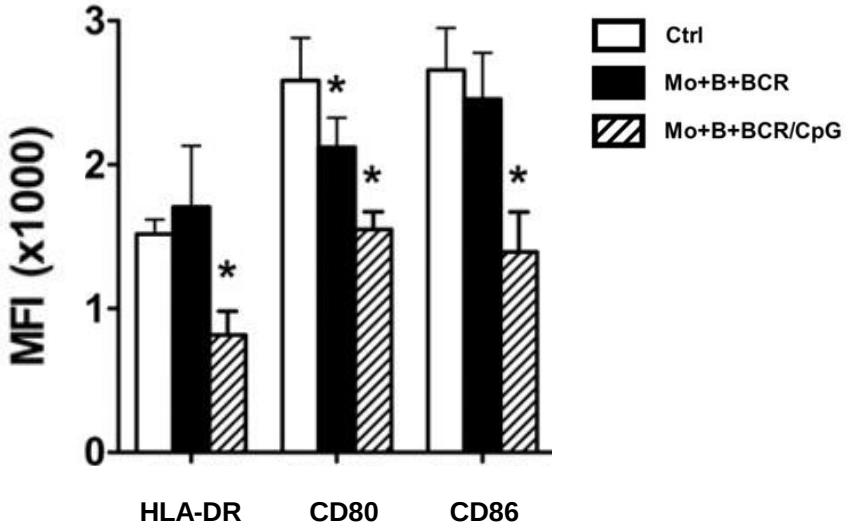
Regulation of Dendritic cell functions by immune compartment



Regulatory B cells slow down the complete maturation of iDCs into mDCs.

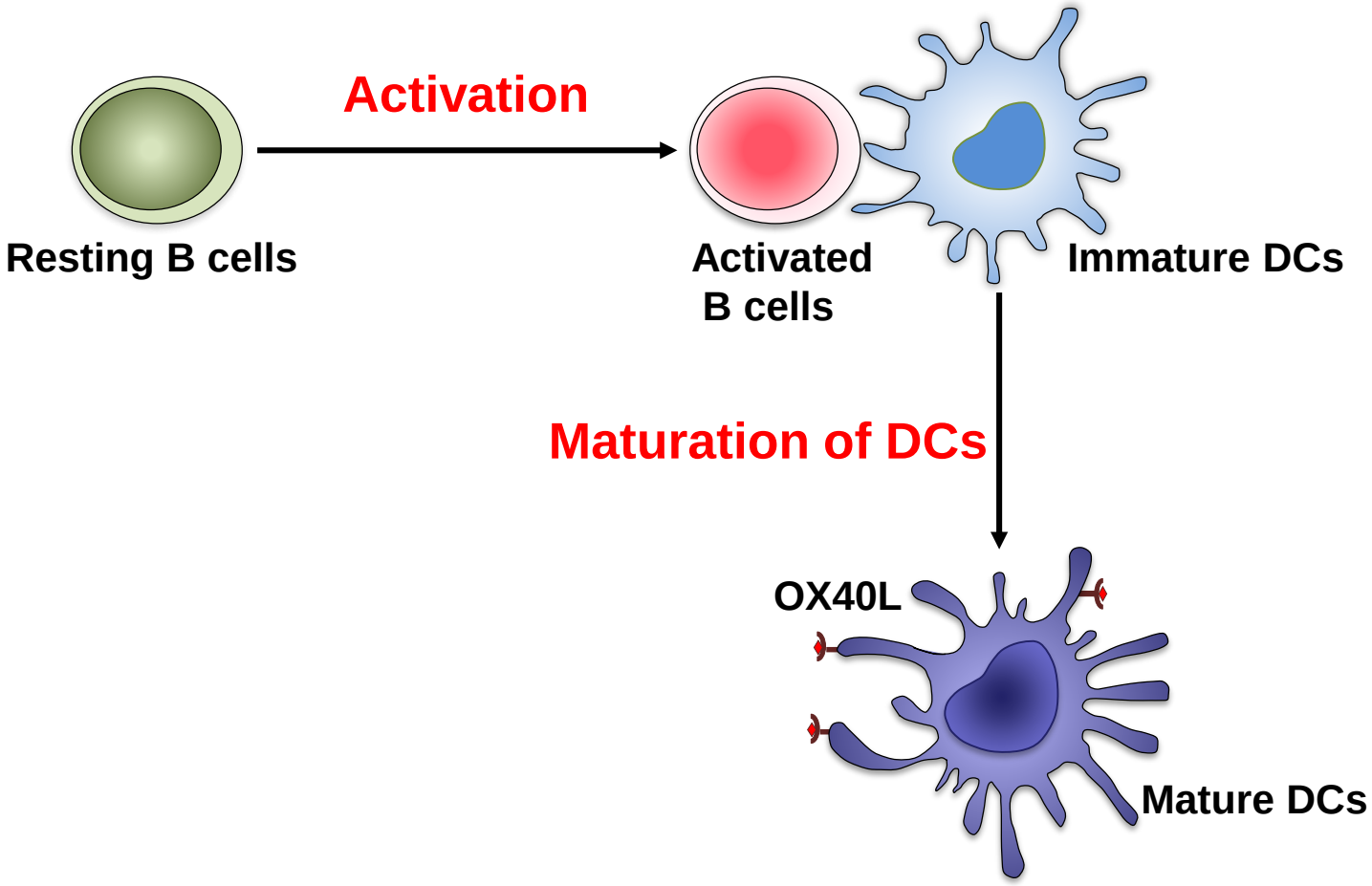


B cell stimulation:
CD40L+CpG-ODN 2006



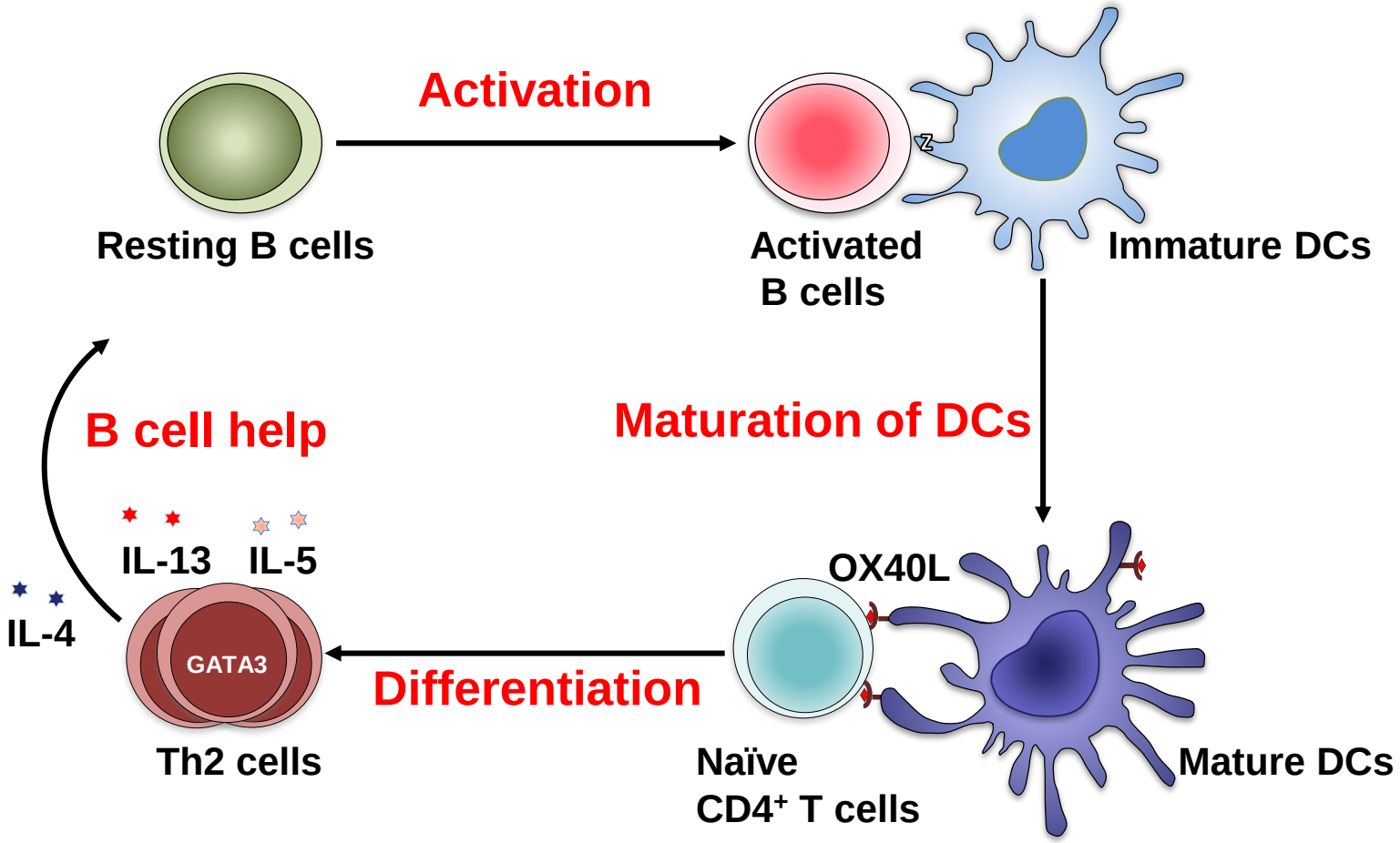
Morva A et al. Blood 2012;119:106-114
Maddur M S et al. Blood 2012;119:3863-3864

Activated B cells induce dendritic cell maturation and induce Th2 polarization by inducing OX-40 ligand



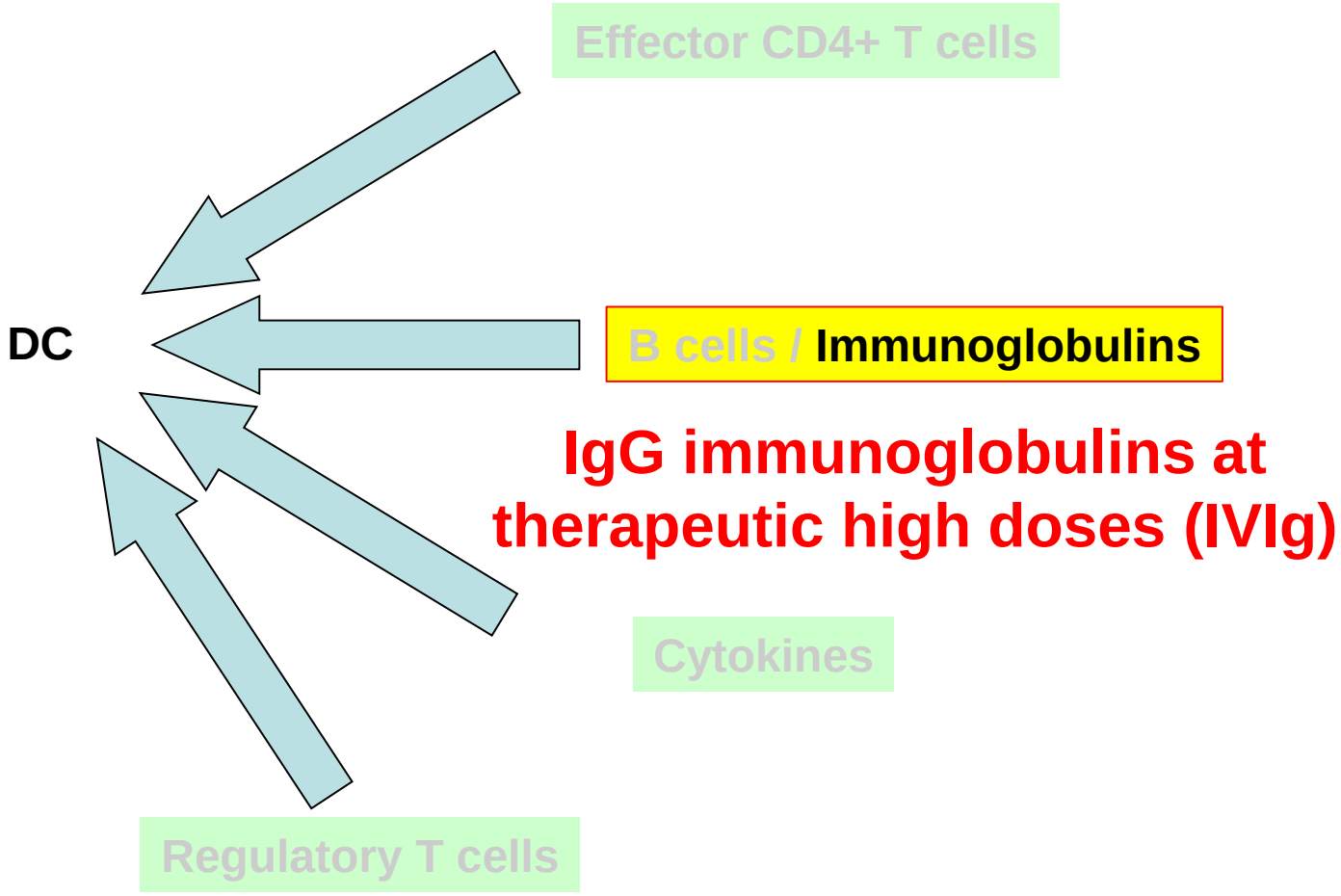
Maddur et al., Nature Commun 2014

Activated B cells induce dendritic cell maturation and induce Th2 polarization by inducing OX-40 ligand



Maddur et al., Nature Commun 2014

Regulation of Dendritic cell functions by immune compartment



Regulation of dendritic cells by circulating antibodies: Intravenous Immunoglobulin (IVIg)

Intravenous Immunoglobulin (IVIg)

- **Therapeutic preparation of pooled normal polyspecific human IgG obtained from large numbers of healthy donors**

Contains

Antibodies to non-self (foreign antigens)

Antibodies to self antigens

Antibodies to antibodies (anti-ids)

Intravenous Immunoglobulin (IVIg)

Polyclonal IgG – plasma pool – thousands of healthy donors

Primary Immunodeficiencies

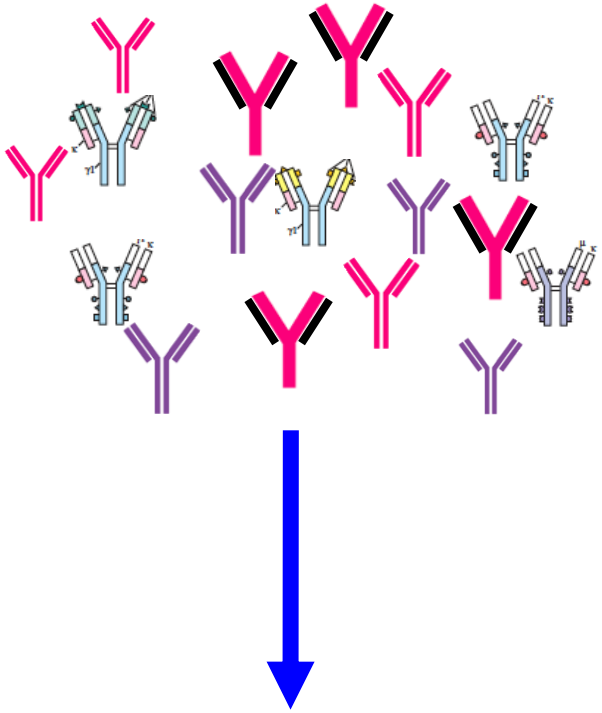
CVID,

XLA

Secondary Immunodeficiencies

BM transplantation,

Multiple myeloma



Autoimmune and inflammatory diseases

Immune thrombocytopenic purpura (ITP)

Guillain-Barré syndrome (GBS)

Kawasaki disease (KD)

CIDP

Dermatomyositis (DM)

Polymyositis (PM)

Psoriasis

Systemic lupus erythematosus (SLE)

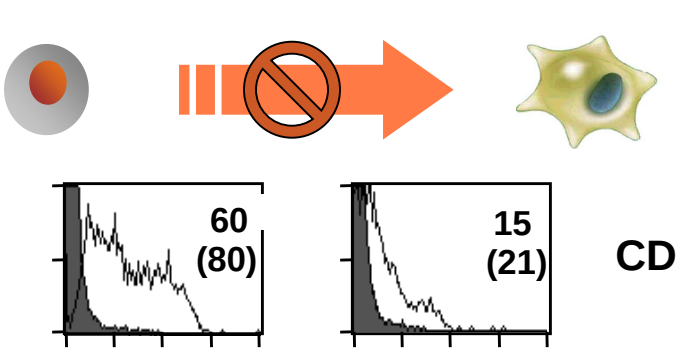
ANCA-associated vasculiits

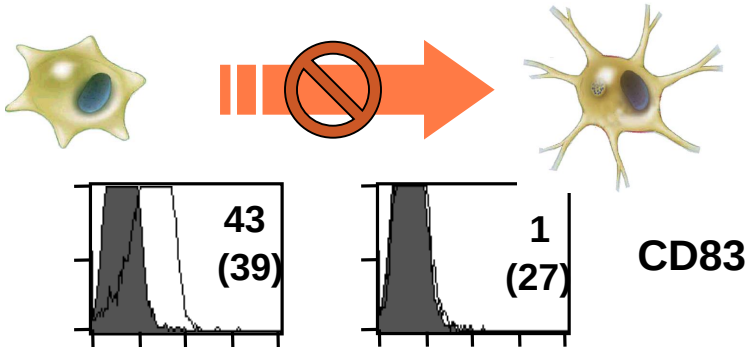
Myasthenia gravis (MG)

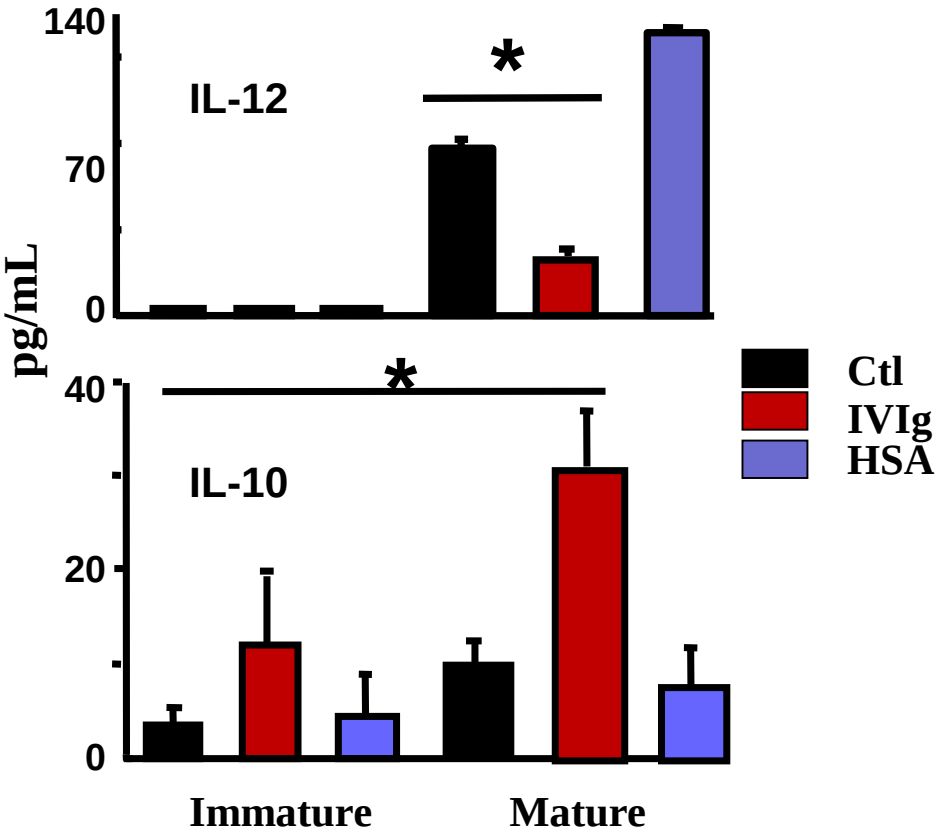
Transplant rejection

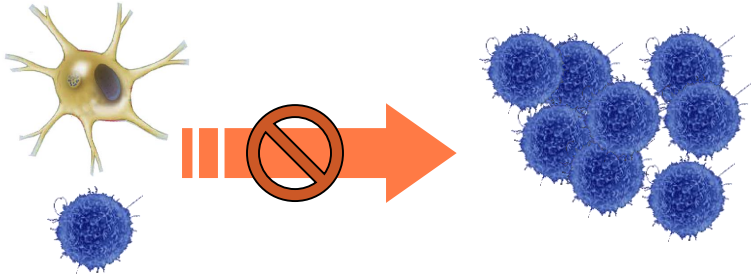
Wide - divergent therapeutic applications

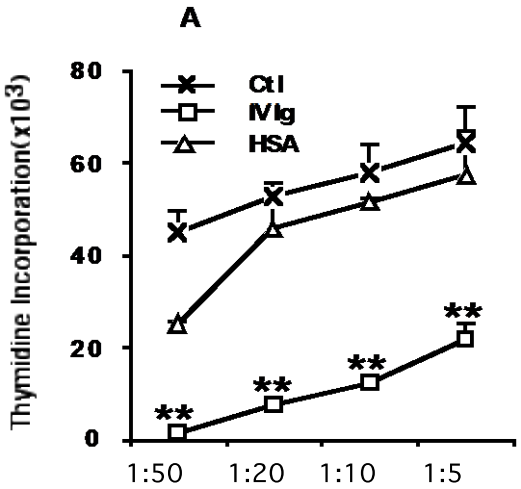
Regulation of maturation and function of dendritic cells by IVIg at therapeutic concentration





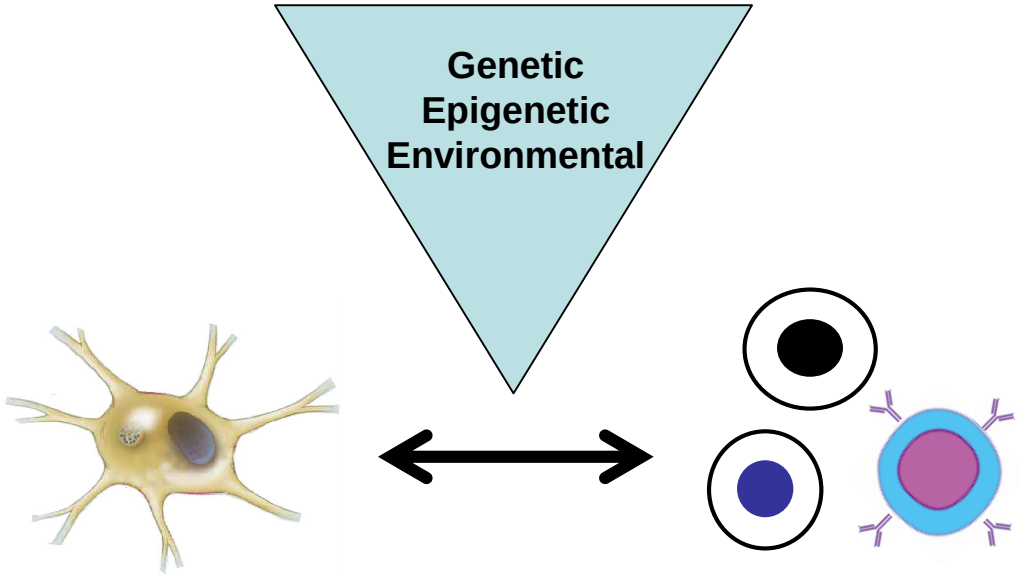






Blood 2003, Arthritis Rheum 2003, PNAS 2009

Contribution of DC to autoimmunity

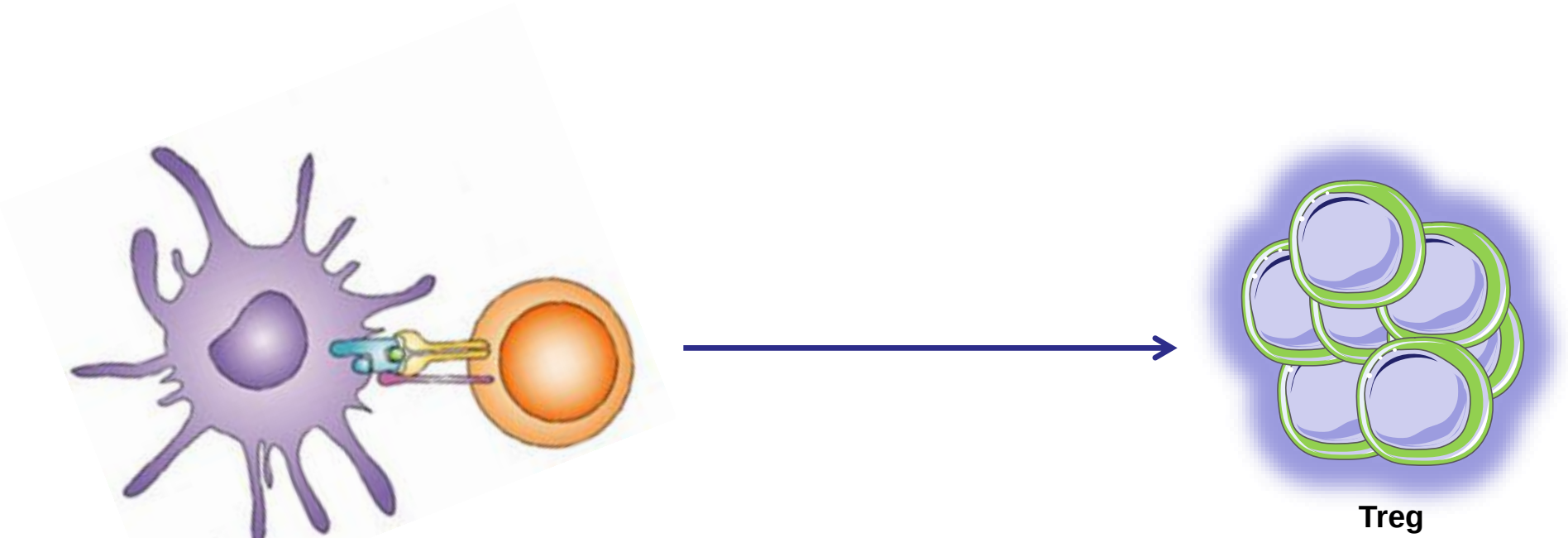


Cytokines/chemokines
Ag presentation
Co-stimulatory molecules
Migratory properties

Effector /pathogenic T cells
Treg number and function

Dendritic cells prevent autoimmunity by expansion of regulatory T cells

Regulatory T cell expansion by dendritic cells



Treg

Transforming growth factor-β (TGF-β)

Cyclooxygenase-2 (COX-2)

Indoleamine 2,3-dioxygenase (IDO)

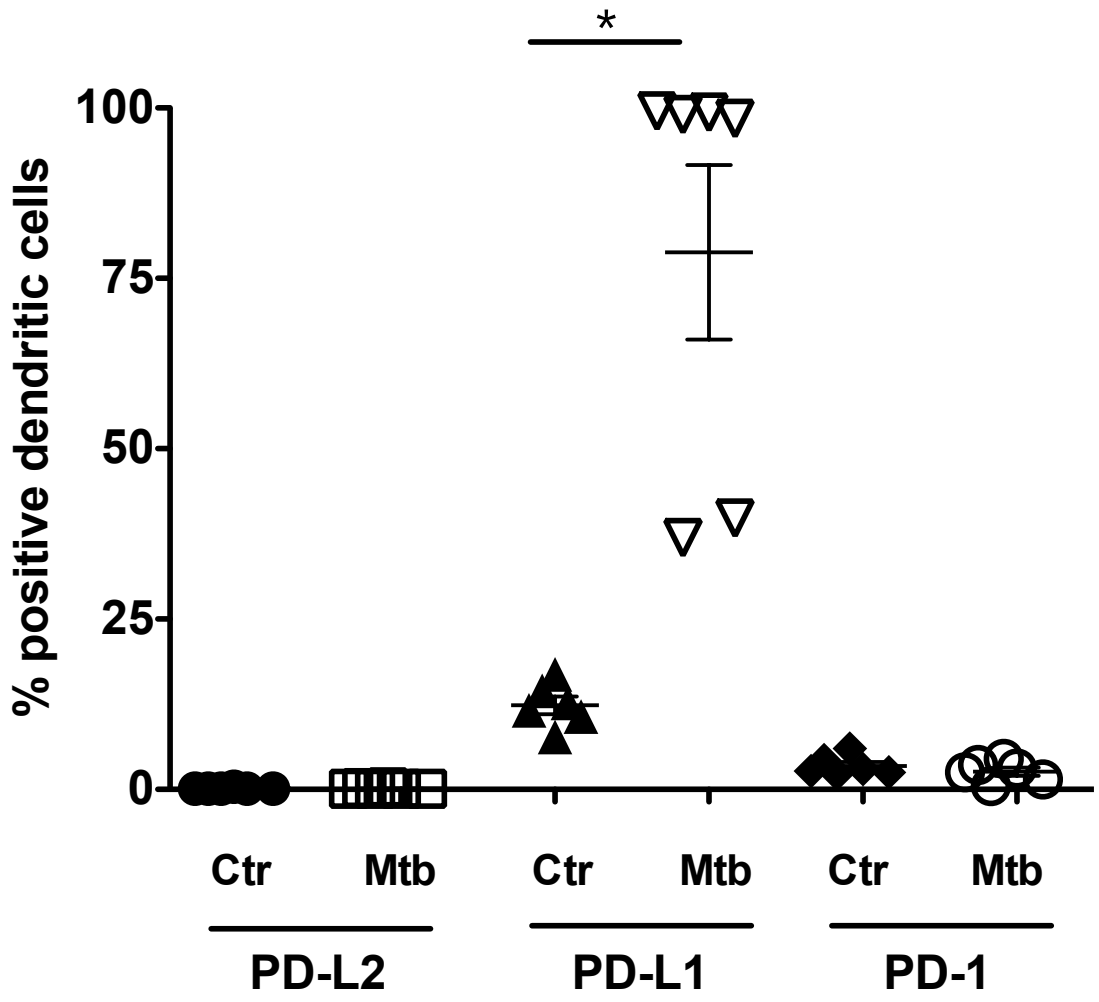
Inducible costimulatory ligand (ICOS-L)

CD252 (OX40L)

Programmed death ligands

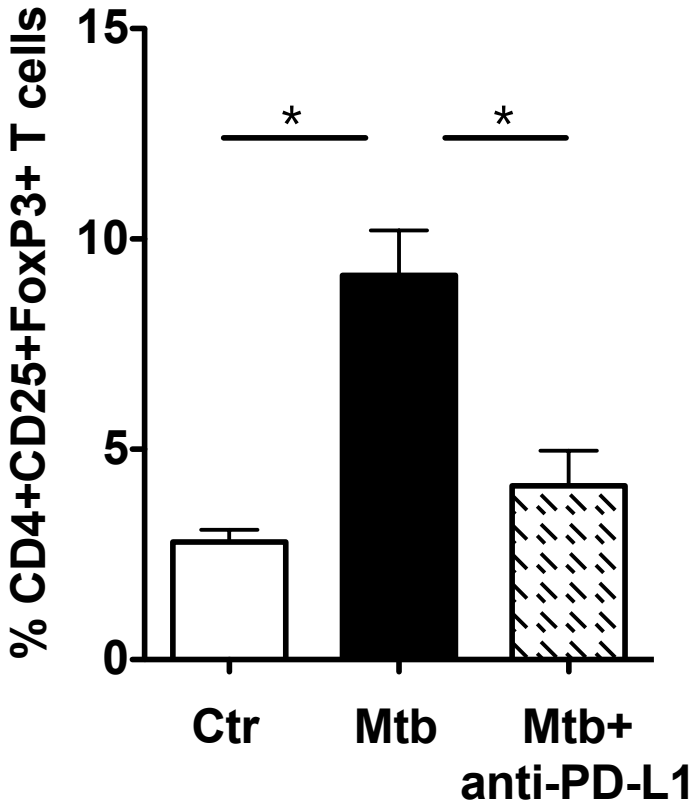
Pallotta et al., Nat Immunol. 2011
Trinath et al., Blood 2013
Gopisetty et al., Immunology 2013
Fantini et al., Immunology 2004
Sakaguchi et al., Cell, 2008
Dejaco et al., Immunology, 2007

Regulatory T cell expansion by dendritic cells: Role of Programmed death ligands



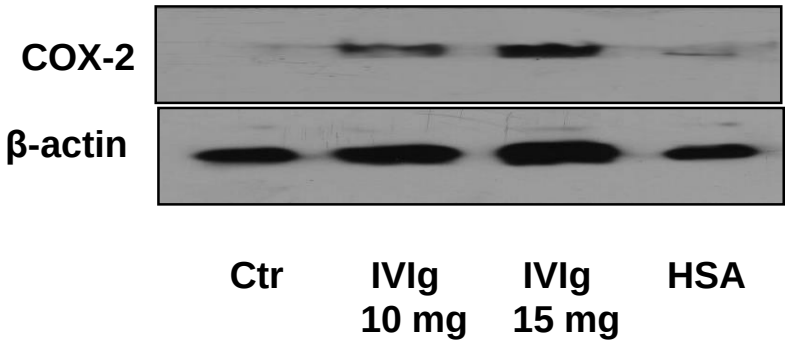
Trinath et al. J Infect Dis 2012

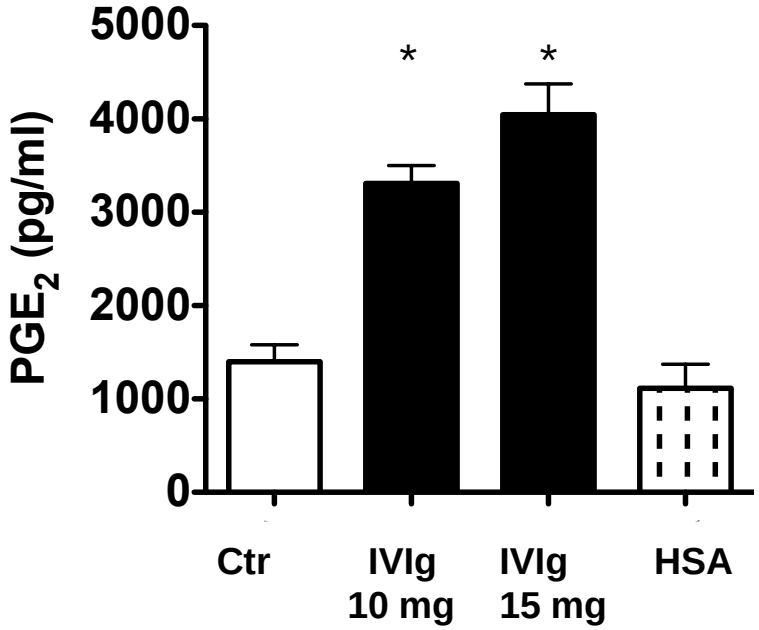
Regulatory T cell expansion by dendritic cells: Role of Programmed death ligands



Trinath et al. J Infect Dis 2012

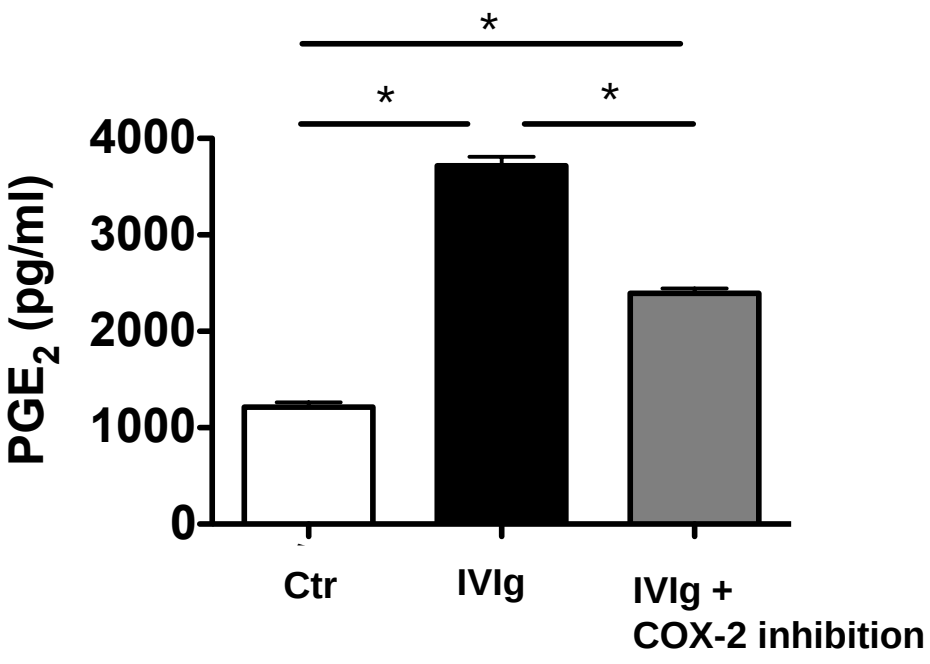
Regulatory T cell expansion by dendritic cells: Role of PGE2

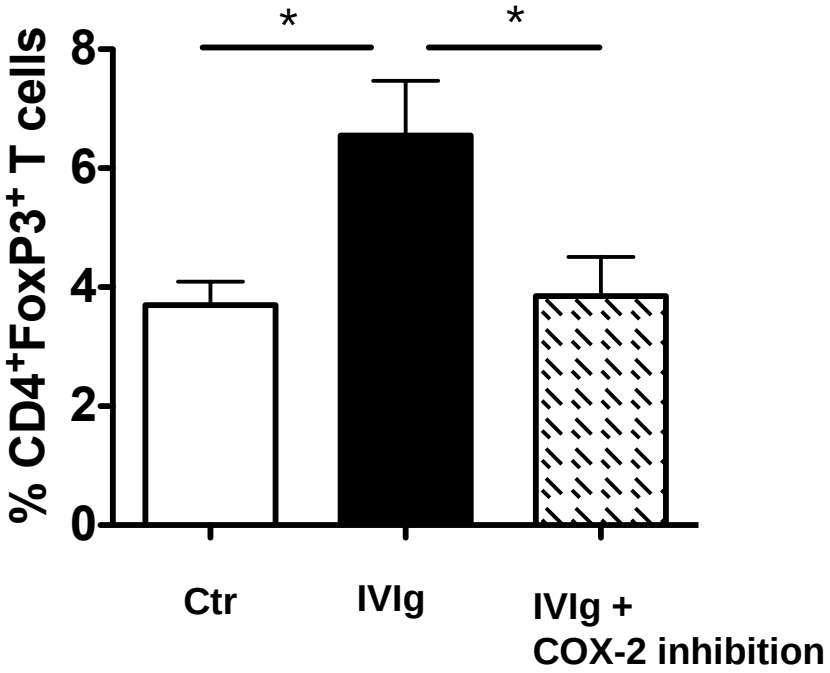




Trinath et al Blood 2013

Regulatory T cell expansion by dendritic cells: Role of PGE2

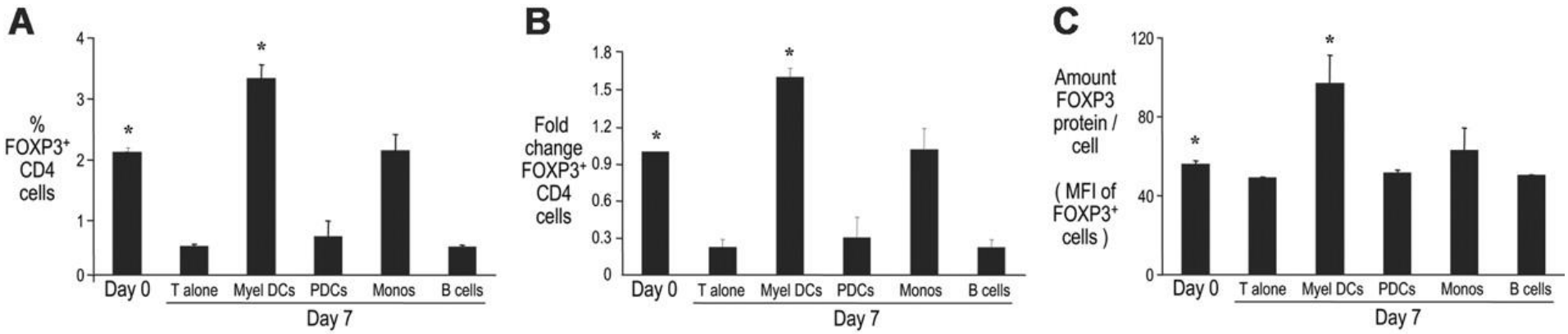




Trinath et al Blood 2013

Differentiation of iTregs by dendritic cells

Comparison of different cell populations for the induction of CD4+CD25+ FOXP3+ Treg's.



Banerjee D K et al. Blood 2006;108:2655-2661

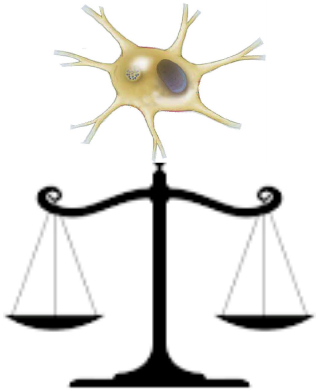
Take home message

Immature/tolerogenic DC

Immune tolerance

Prevention of autoimmunity

Induction of Tregs
Maintenance/homeostasis of Tregs



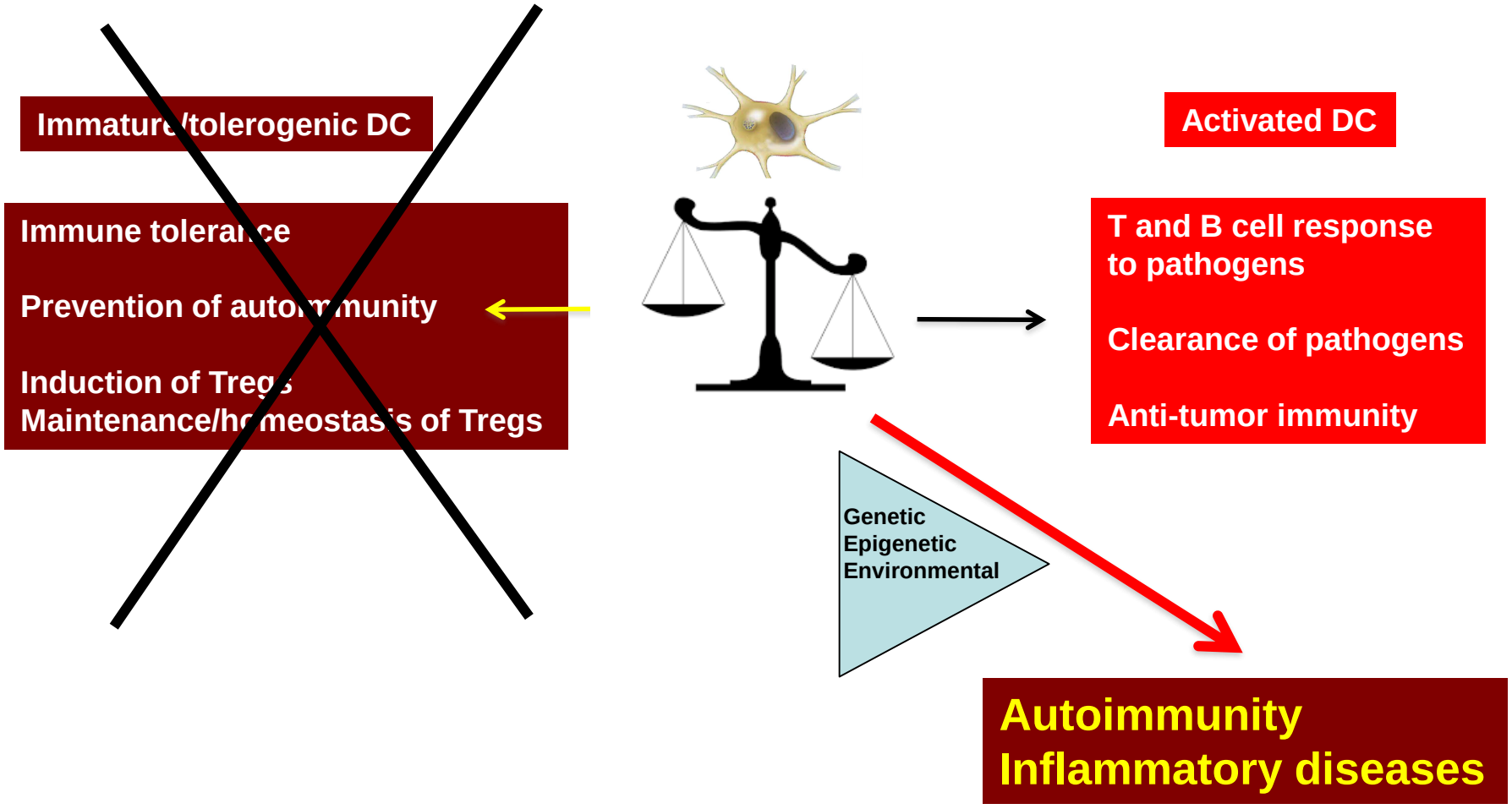
Activated DC

T and B cell response to pathogens

Clearance of pathogens

Anti-tumor immunity

Take home message



Thank you