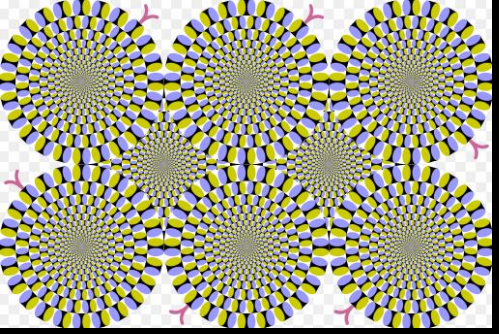
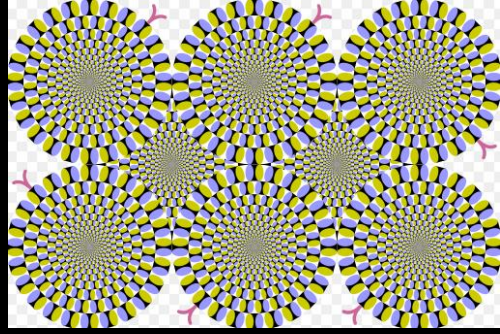
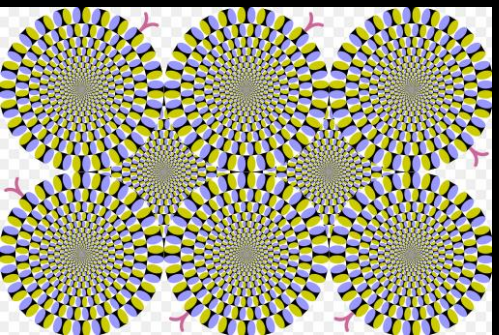
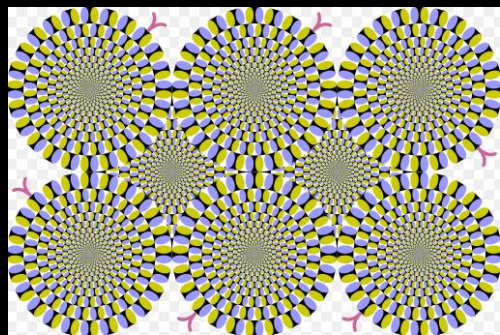
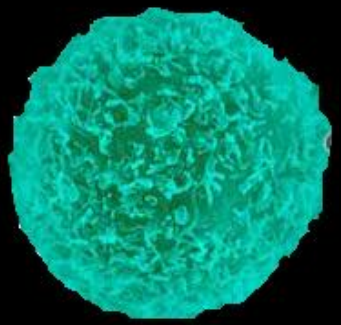



Tolerância Central
e
Periférica

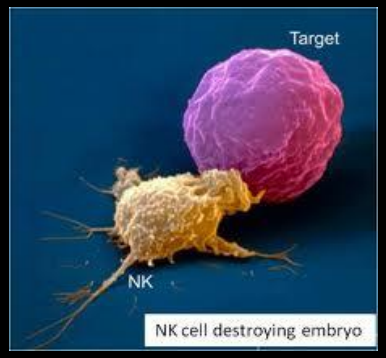
Prof. Dr. Jean Pierre Schatzmann Peron

Linfócitos T CD4 são os regentes da “orquestra” chamada Sistema Imune

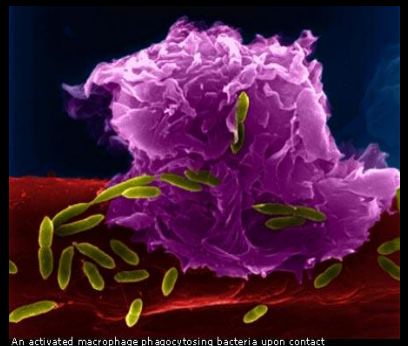


Linfócito T CD4



Linfócito T CD8

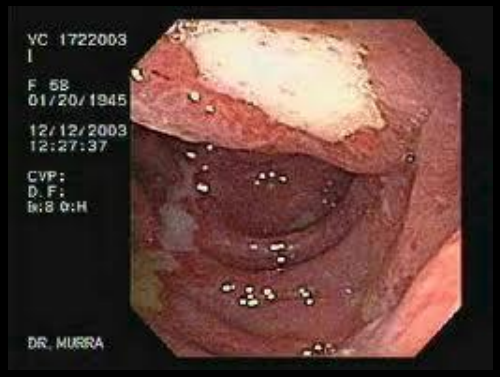
Neutrófilos



Macrófagos

Células Dendríticas

Imunodeficiências – Infecções de Repetição



Colite

Pneumonia

AIDS

Toxoplamose Ocular

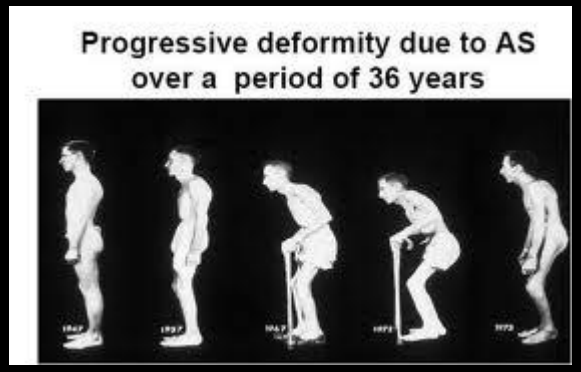
Candida

Staphilococcus

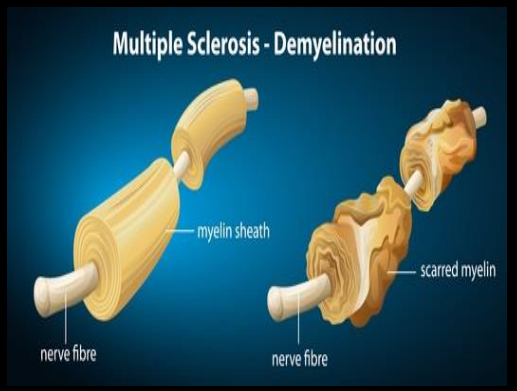
Infecções Múltiplas

Autoimunidade

LUPUS



Spondilite Anquilosante



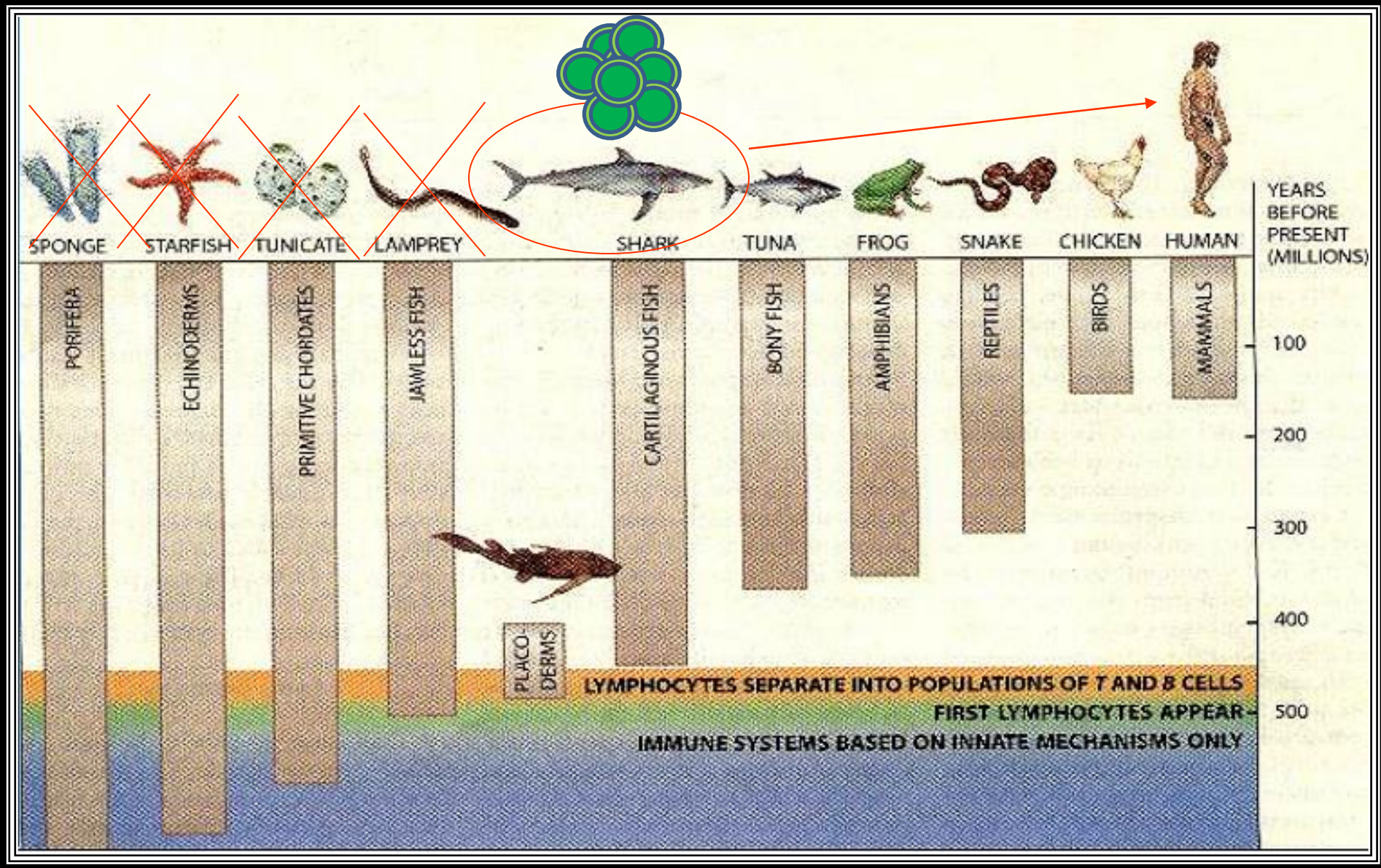
Esclerose Múltipla

Uveíte

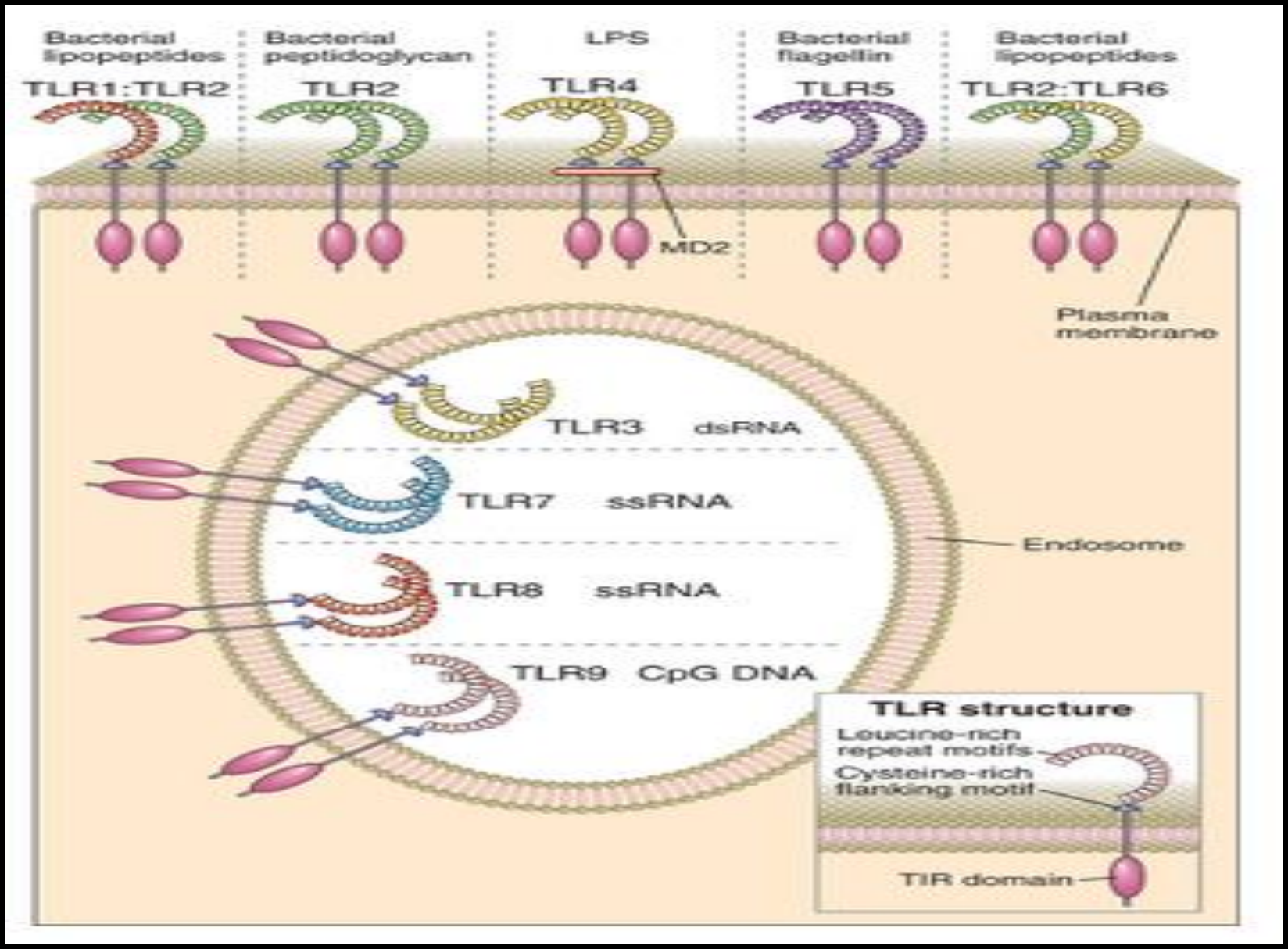
Doença de Graves

Psoríase

Linfócito é um Evento Recente

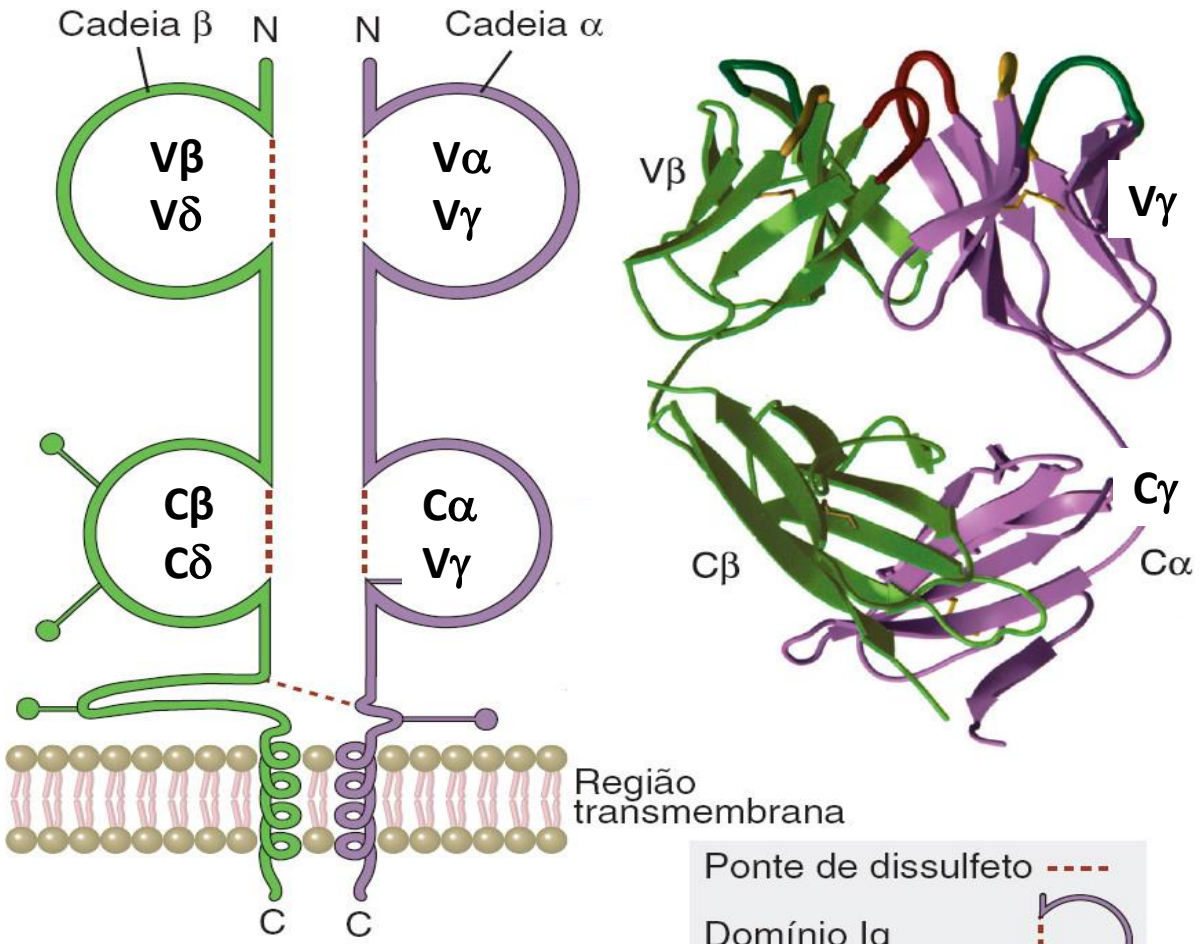


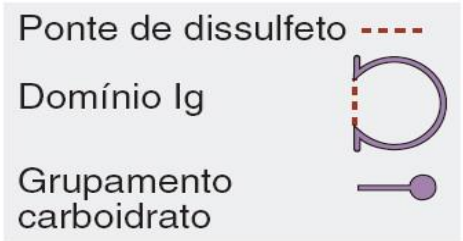
Natureza dos Antígenos



Linfócitos T também podem ser classificados quanto a seu TCR (αβ e γδ)

Imunologia Celular e Molecular
ISBN: 9788535222449
Elsevier Editora





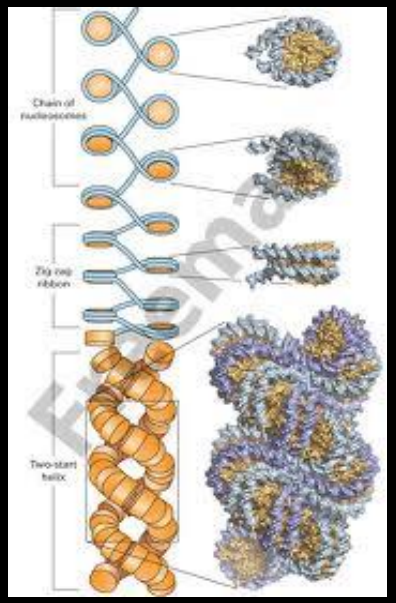
2 Regiões Constantes

2Regiões Variáveis

Alfa e gamma (V-J)

Beta e delta (V-D-J)

Loci do TCR



Imunologia Celular e Molecular
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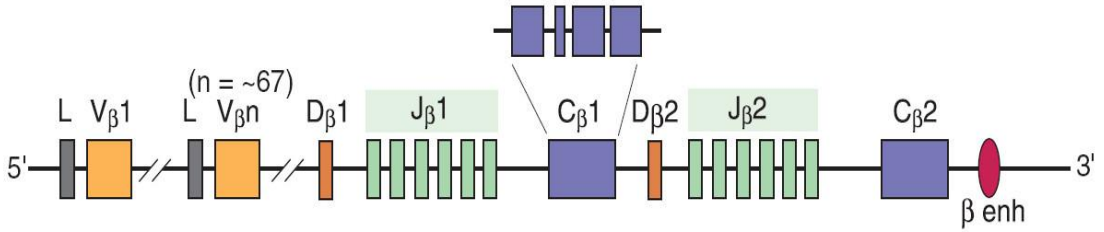
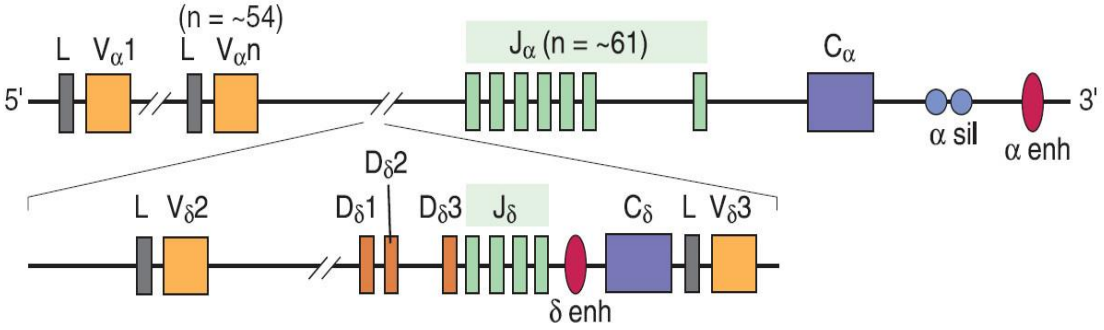
Elsevier Editora

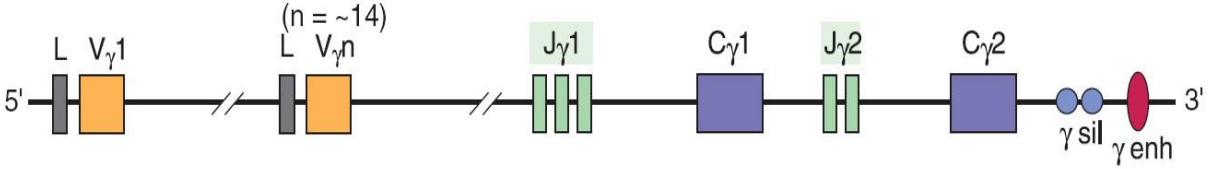
Locus da cadeia β do TCR humano (620 kb; cromossomo 7)



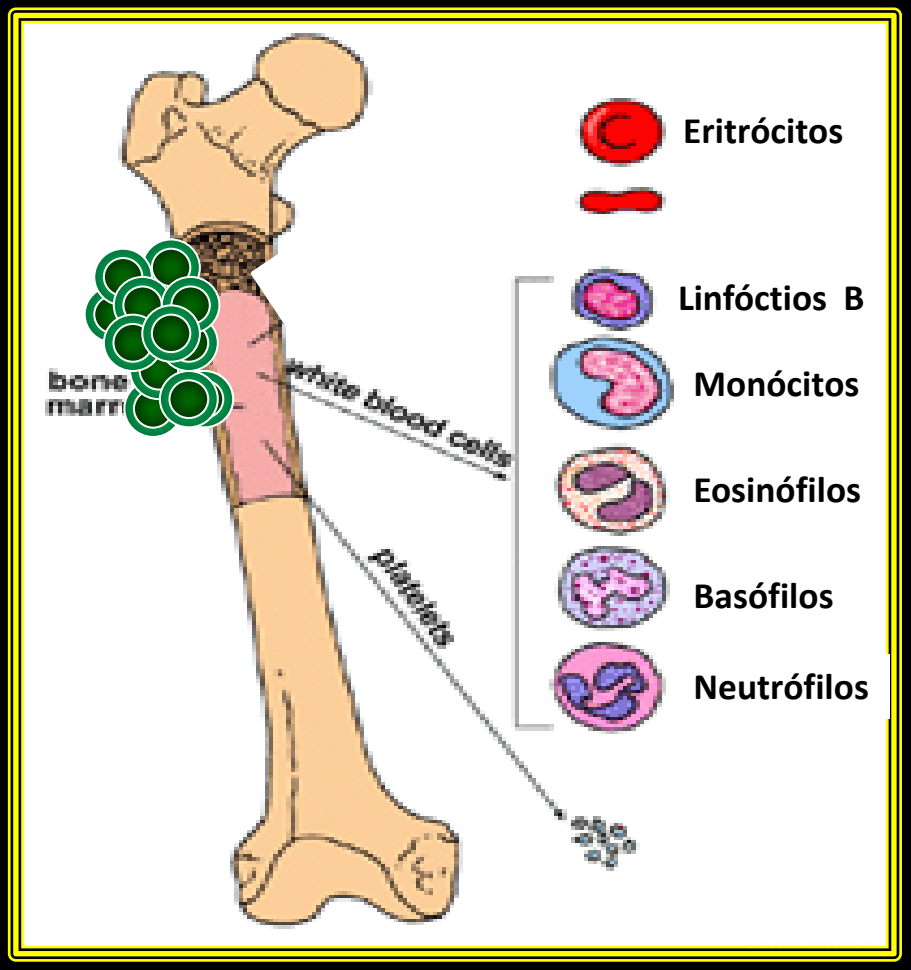
Locus das cadeias α e δ do TCR humano (1.000 kb; cromossomo 14)

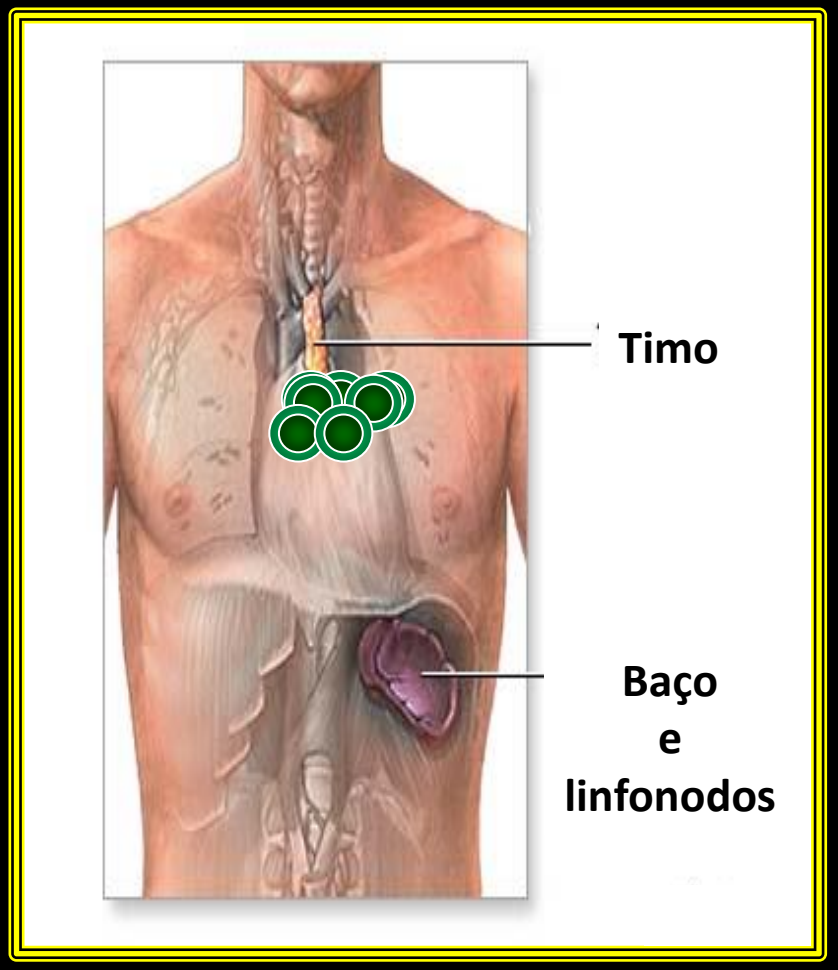


Locus da cadeia γ do TCR humano (200 kb; cromossomo 7)

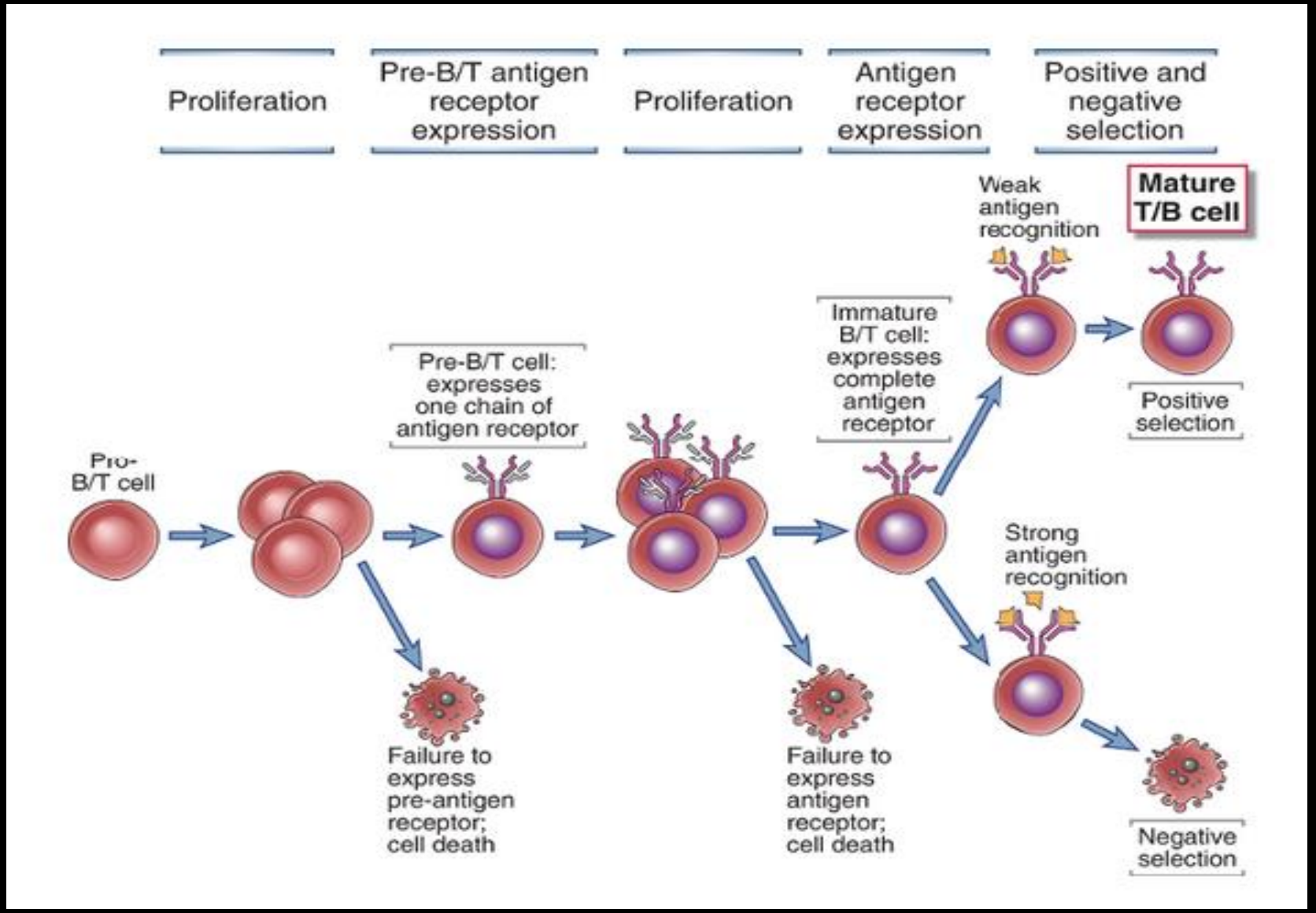


Precursores linfóides migram para o timo para sofrer maturação

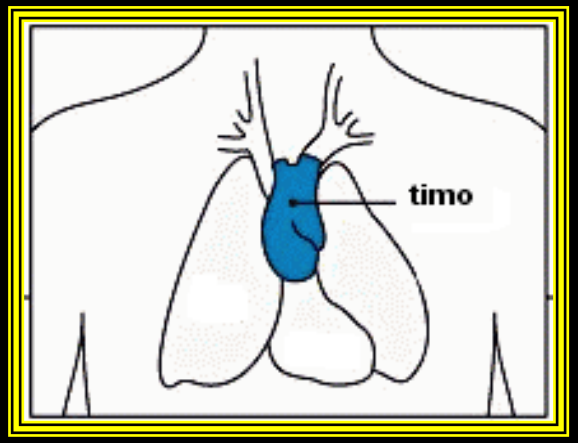




Tais Estágios Possuem Checkpoints que Garantem A Geração de Células Produtivas e não Deletérias

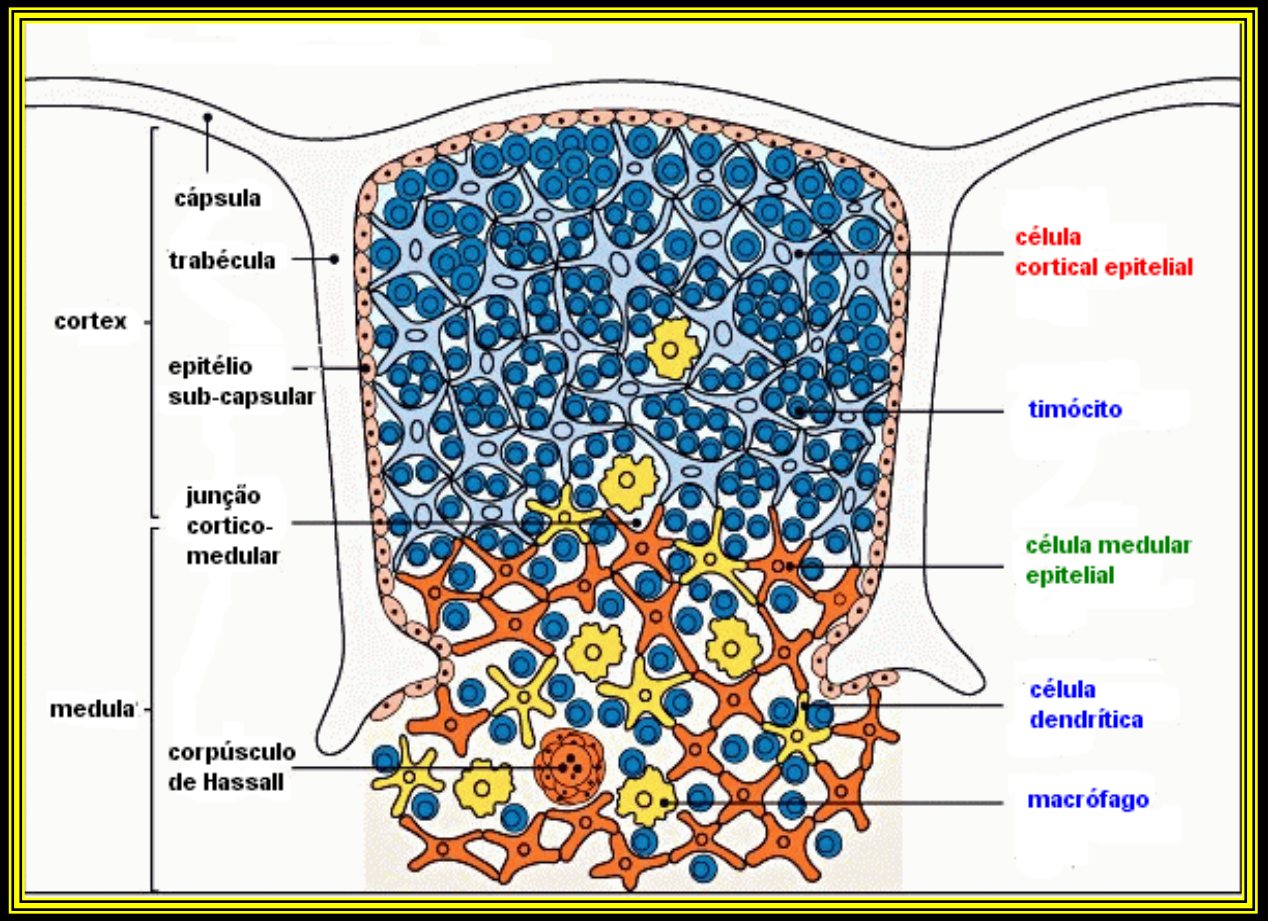


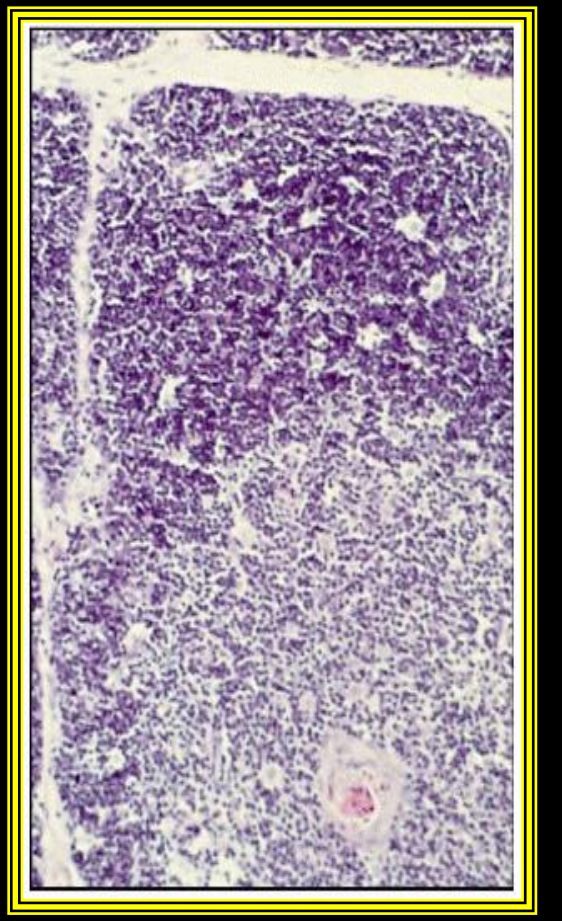
Timo



Idade em anos

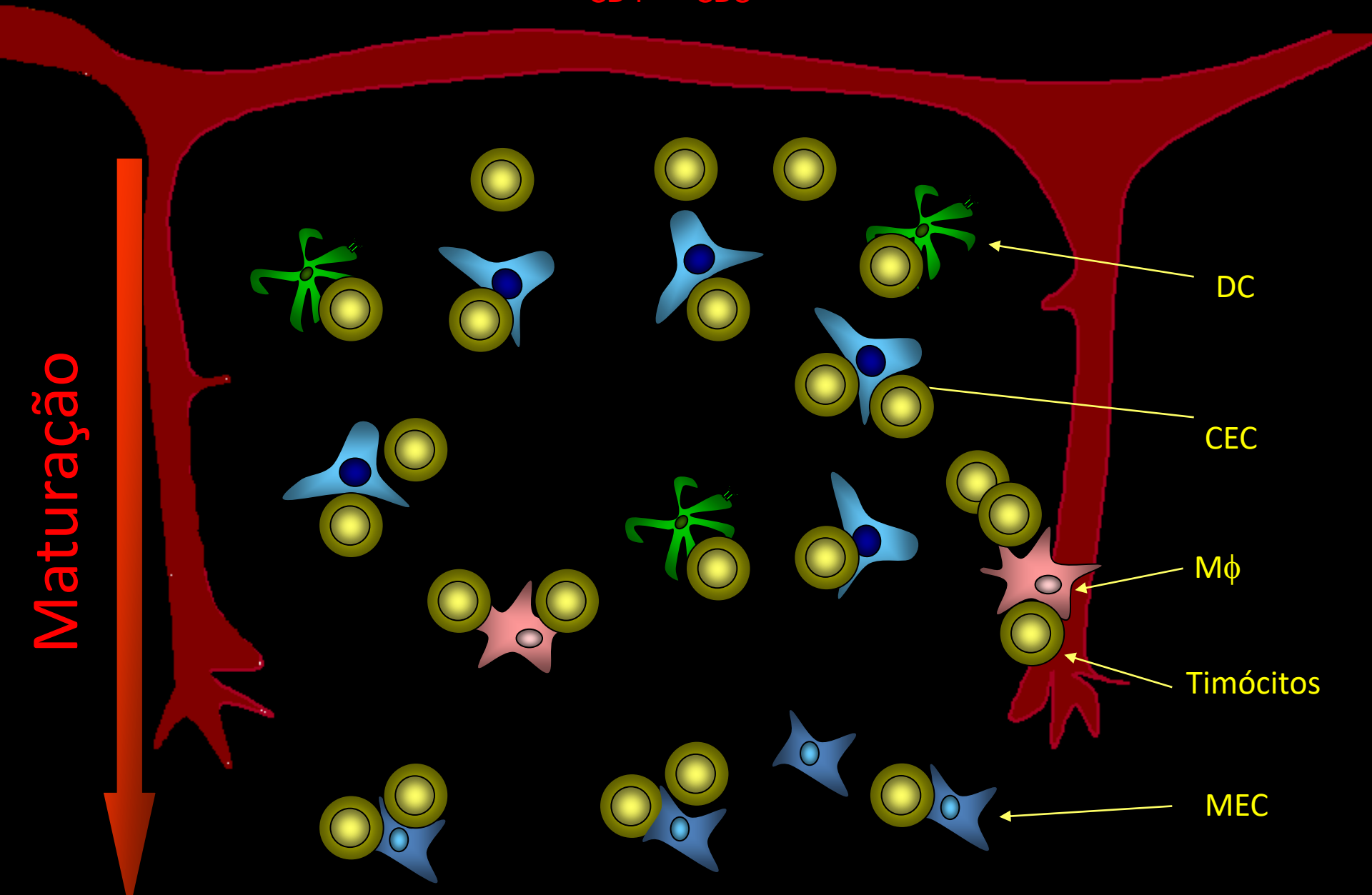
Estrutura Tímica



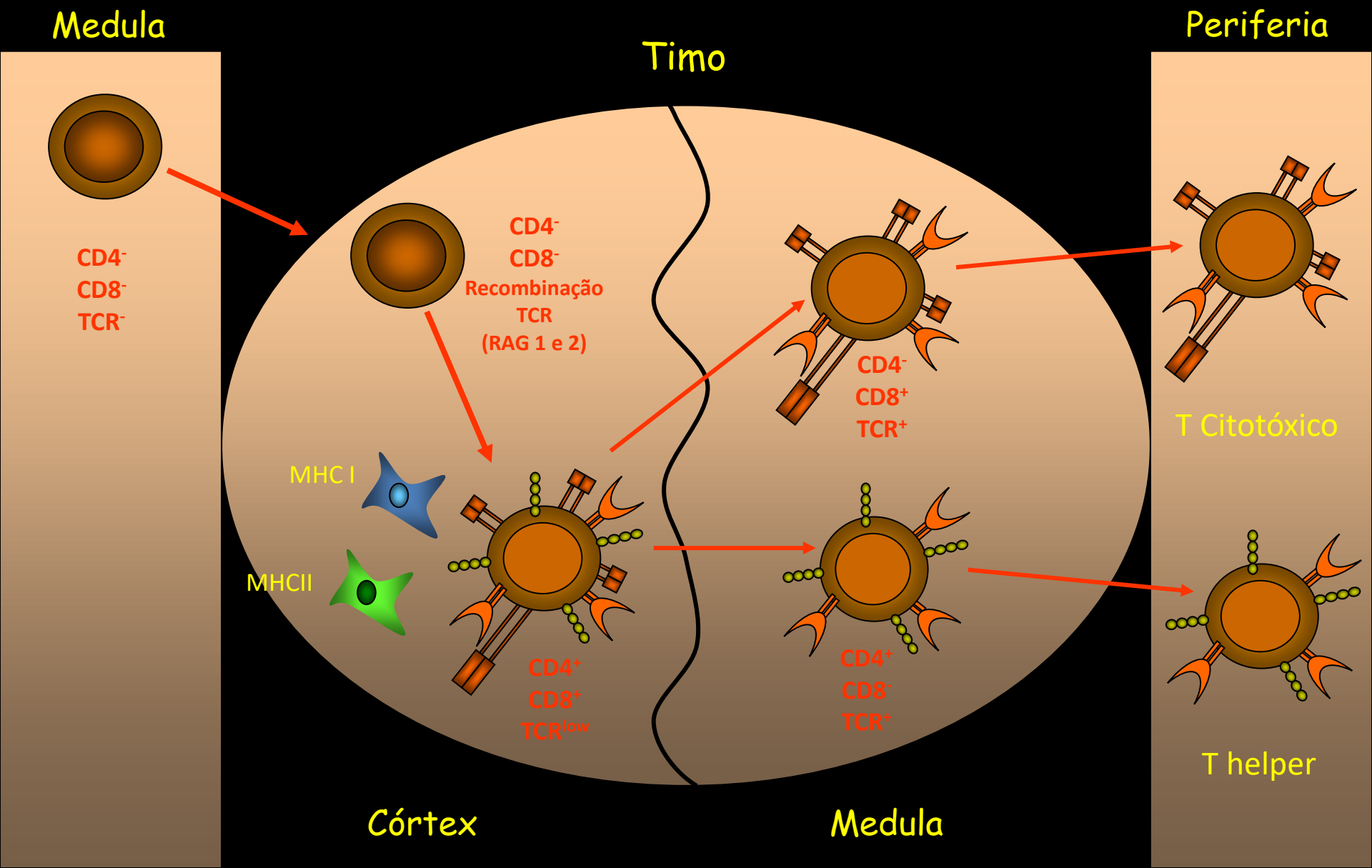


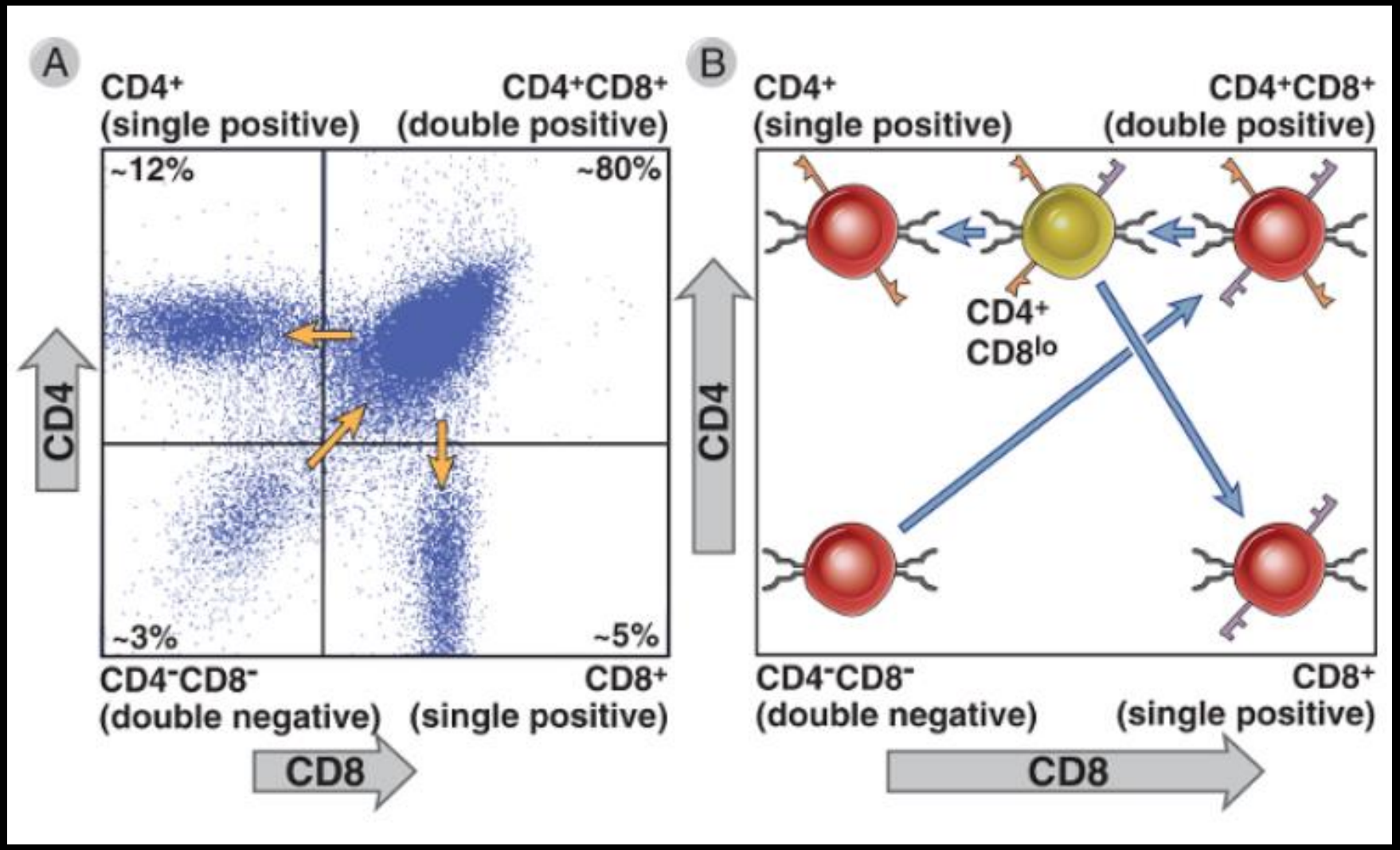
Precursores linfóides, agora chamados timócitos, migram da CÓRTEX PARA MEDULA e não expressam CD4 nem CD8

$CD4^{neg}$ $CD8^{pos}$



Ao adentrar o timo, timócitos passam a expressar $CD4^{pos}$ $CD8^{pos}$

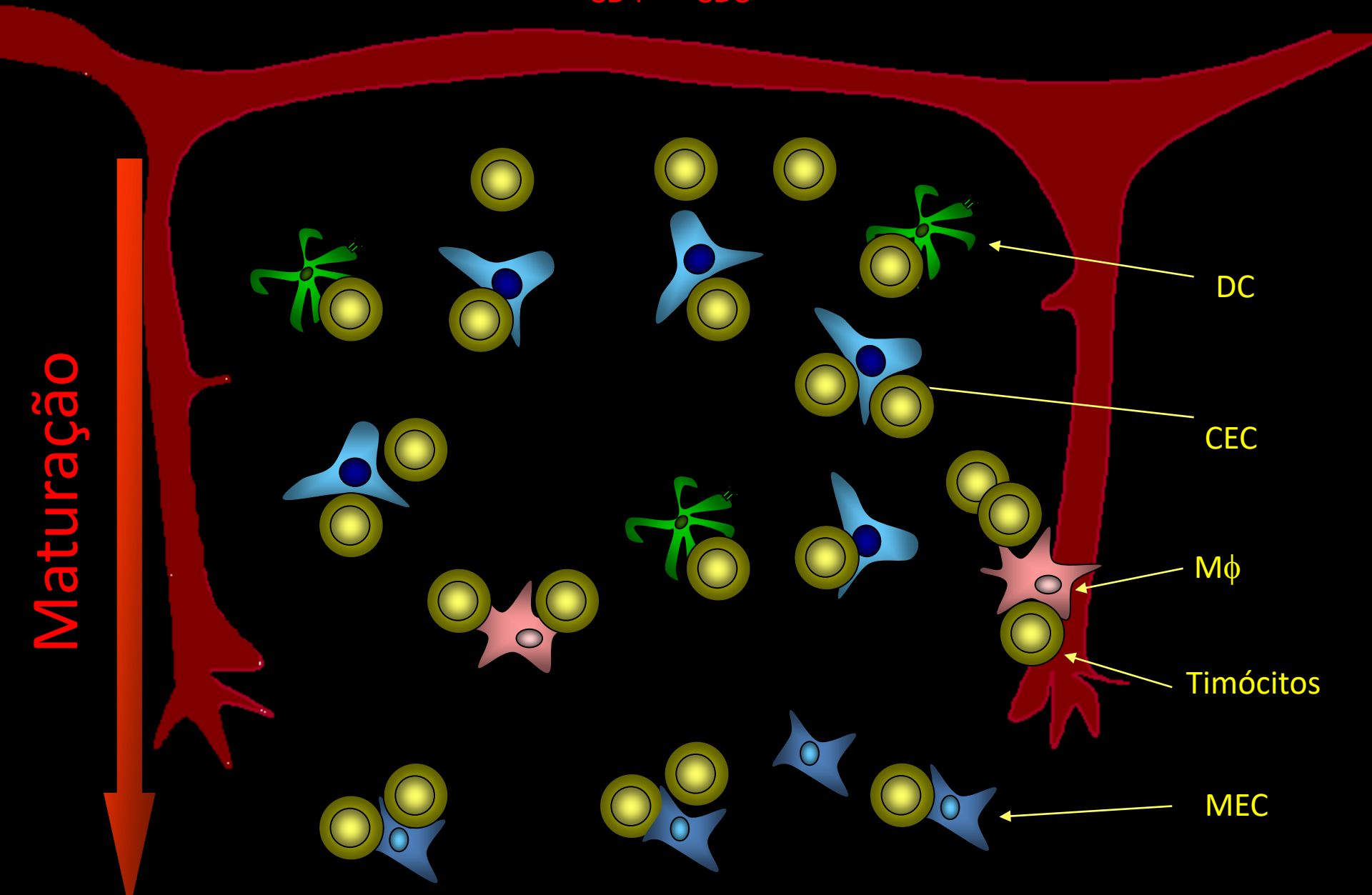



A
CD4+
(single positive)
CD4+CD8+
(double positive)
~12%
~80%
~3%
~5%
CD4
CD4-CD8-
(double negative)
CD8+
(single positive)
CD8
B
CD4+
(single positive)
CD4+CD8+
(double positive)
CD4+
CD8lo
CD4
CD4-CD8-
(double negative)
CD8+
(single positive)
CD8

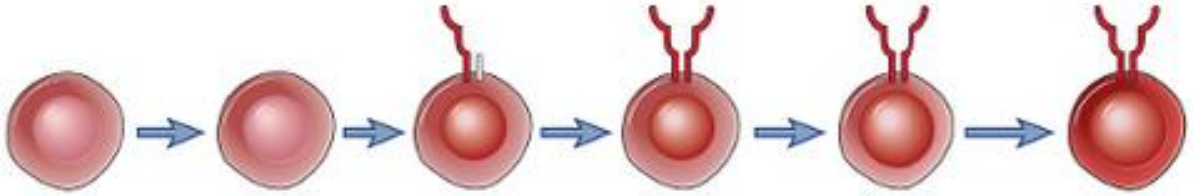
Limiar de Seleção
Regido pela Força de Interação Entre os Linfócitos e Antígenos Próprios

Precursores linfóides, agora chamados timócitos, migram da CÓRTEX PARA MEDULA e não expressam CD4 nem CD8

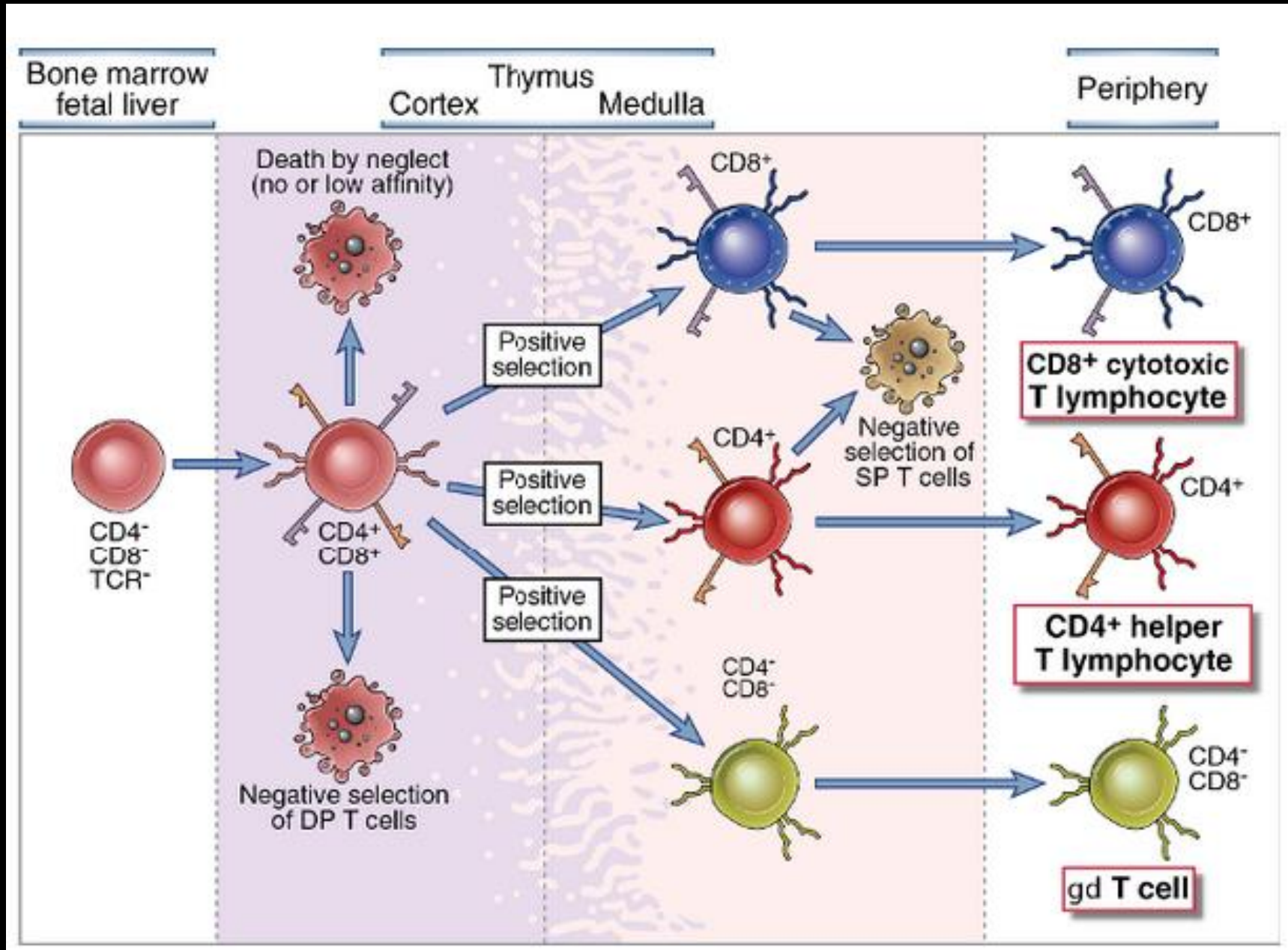
$CD4^{neg}$ $CD8^{pos}$



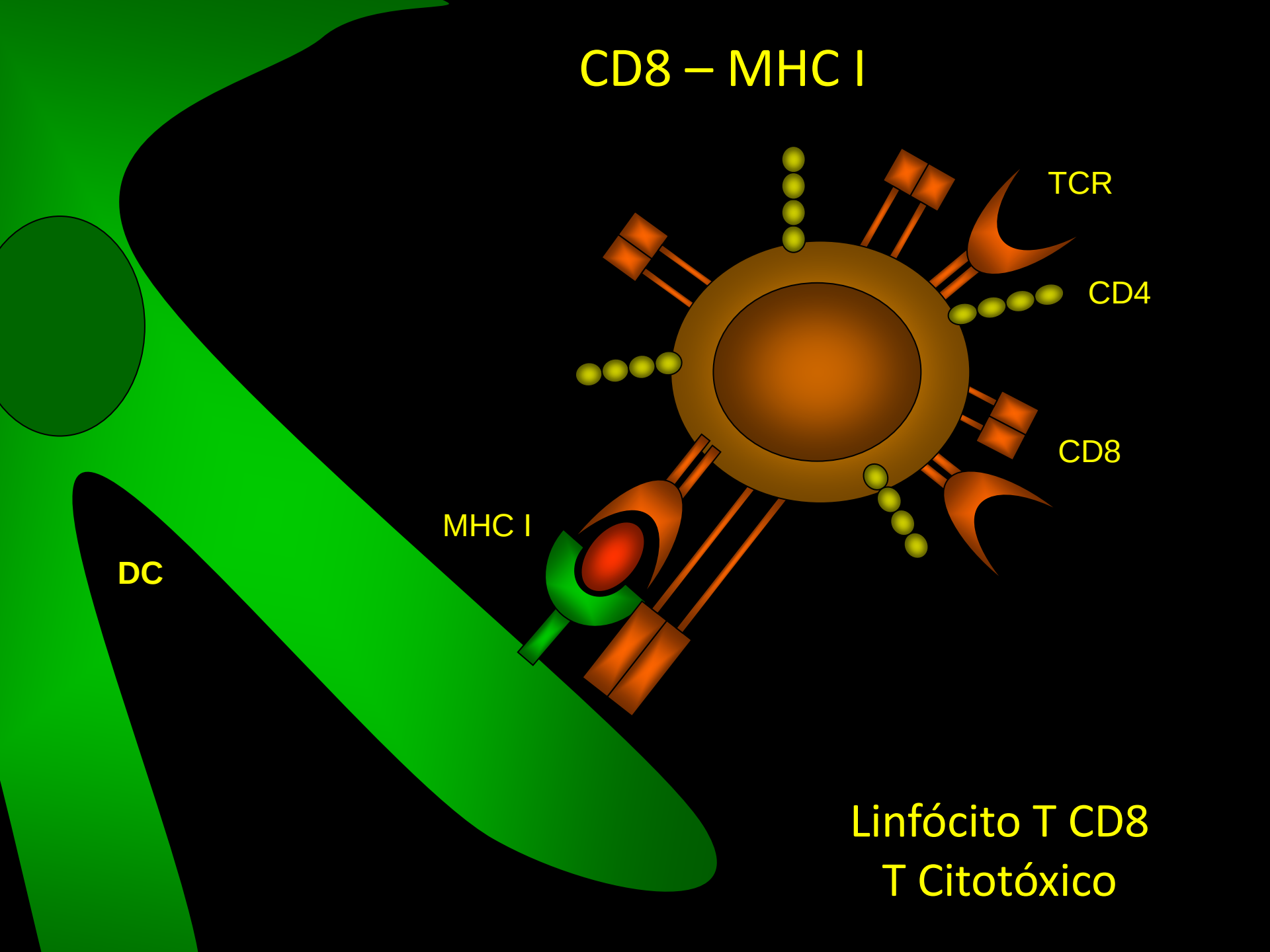
Estágios da Maturação de Linfócitos T



Stage of maturation	Stem cell	Pro-T	Pre-T	Double positive	Single positive (immature T cell)	Naive mature T cell
Proliferation						
RAG expression						
TdT expression						
TCR DNA, RNA	Unrecombined (germline) DNA	Unrecombined (germline) DNA	Recombined β chain gene [V(D)J-C]; β chain mRNA	Recombined β, α chain genes [V(D)J-C]; β and α chain mRNA	Recombined β, α chain genes [V(D)J-C]; β and α chain mRNA	Recombined β, α chain genes [V(D)J-C]; β and α chain mRNA
TCR expression	None	None	Pre-T receptor (β chain/pre-T α)	Membrane αβ TCR	Membrane αβ TCR	Membrane αβ TCR
Surface markers	c-*kit*$^+$ CD44$^+$ CD25$^-$	c-*kit*$^+$ CD44$^+$ CD25$^+$	c-*kit*$^+$ CD44$^-$ CD25$^+$	CD4$^+$CD8$^+$ TCR/CD3^{lo}	CD4$^+$CD8$^-$ or CD4$^-$CD8$^+$ TCR/CD3^{hi}	CD4$^+$CD8$^-$ or CD4$^-$CD8$^+$ TCR/CD3^{hi}
Anatomic site	Bone marrow	Thymus				Periphery
Response to antigen	None	None	None	Positive and negative selection		Activation (proliferation and differentiation)

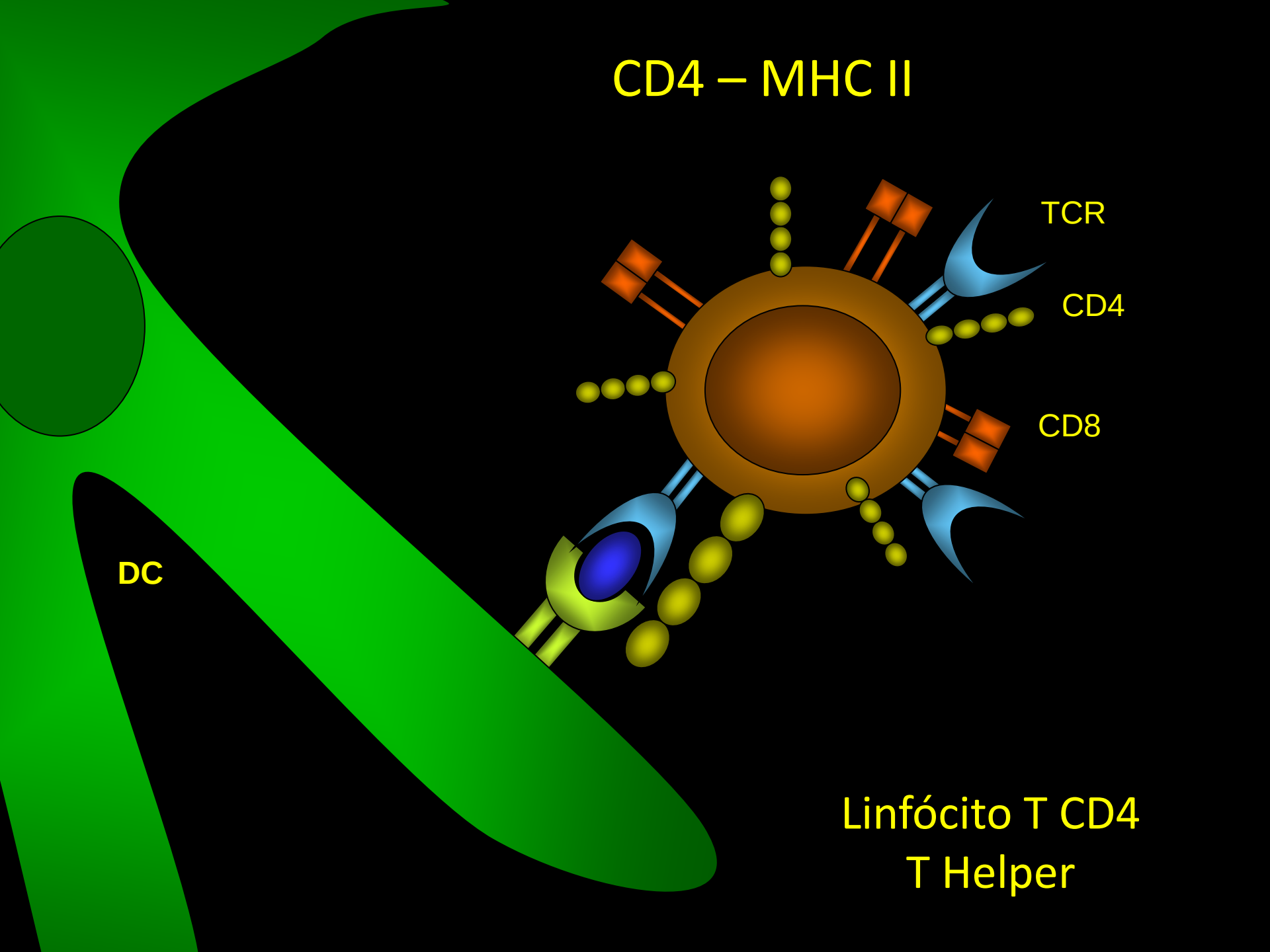

Bone marrow
fetal liver
Thymus
Cortex
Medulla
Periphery
Death by neglect
(no or low affinity)
CD4⁻
CD8⁻
TCR⁻
CD4⁺
CD8⁺
Positive selection
Positive selection
Positive selection
Negative selection
of DP T cells
CD8⁺
CD4⁺
CD4⁻
CD8⁻
Negative selection of SP T cells
CD8⁺
CD8⁺ cytotoxic
T lymphocyte
CD4⁺
CD4⁺ helper
T lymphocyte
CD4⁻
CD8⁻
gd T cell

CD8 – MHC I



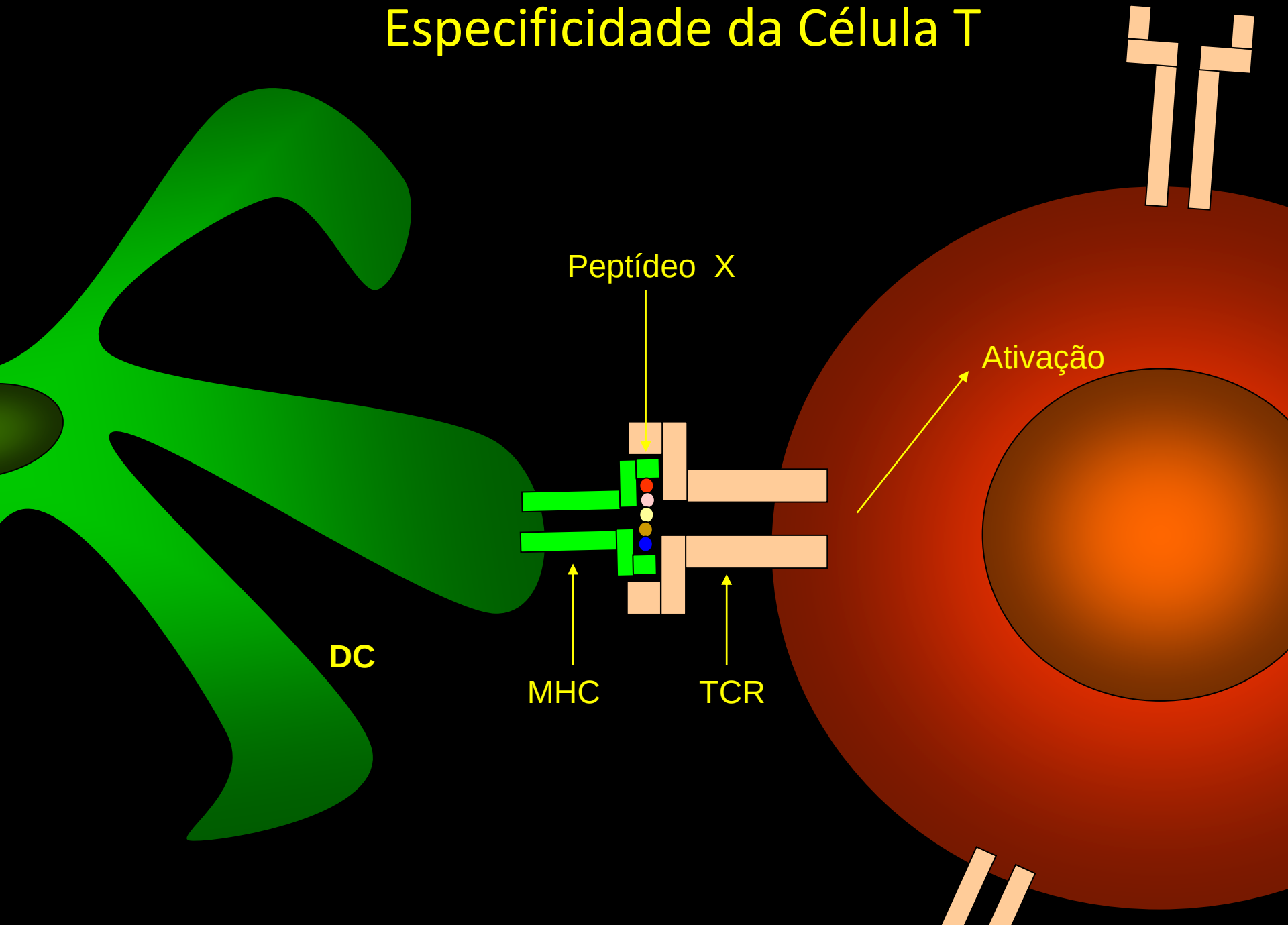
Linfócito T CD8
T Citotóxico

CD4 – MHC II

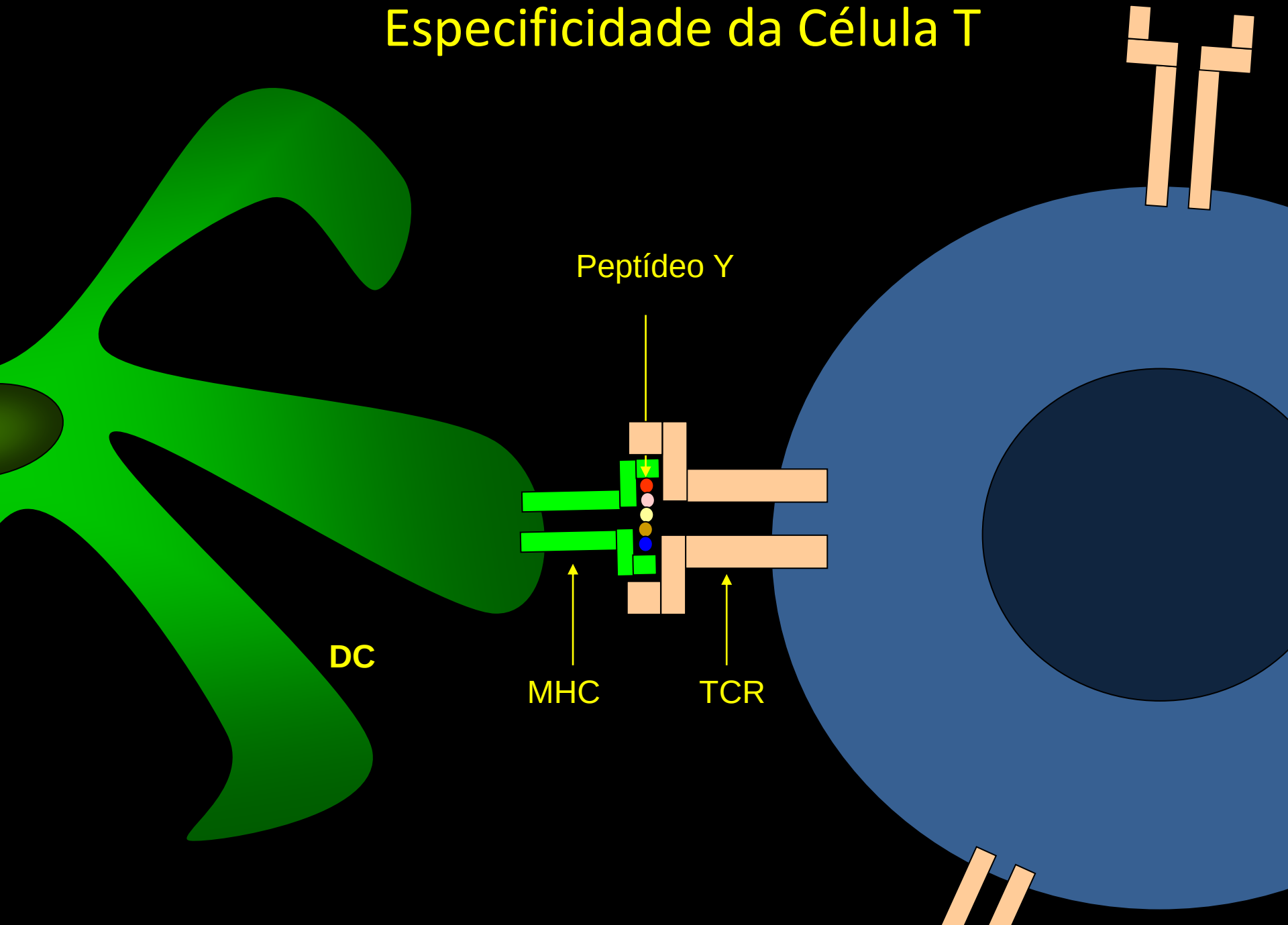


Linfócito T CD4
T Helper

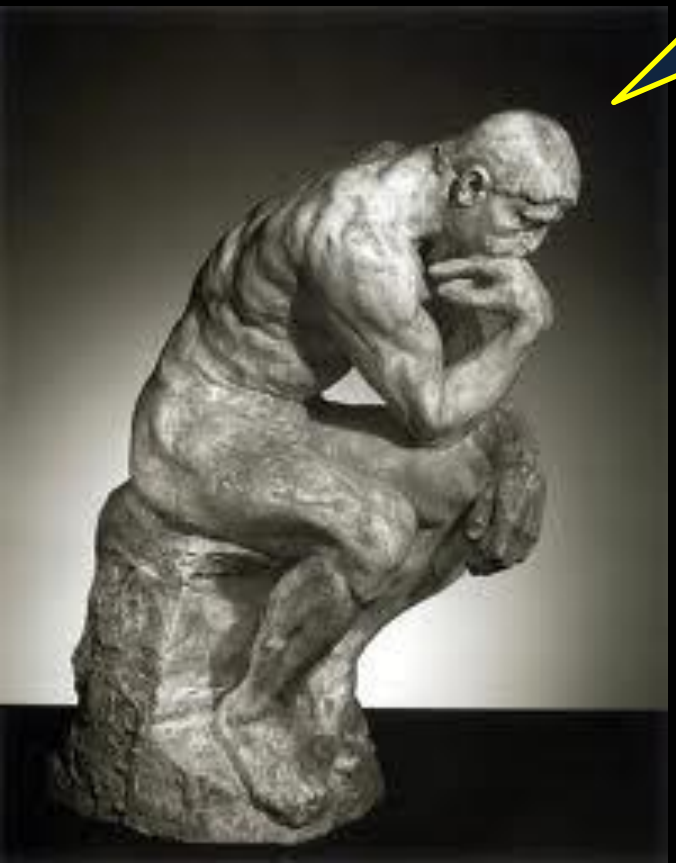
Especificidade da Célula T



Especificidade da Célula T



E se esse peptídeo for próprio ?



Tolerância Central

- Seleção Positiva
- Reconhecimento das porções polimórficas do MHC + Ag

- Restringe a resposta linfocitária aos MHCs do próprio indivíduo

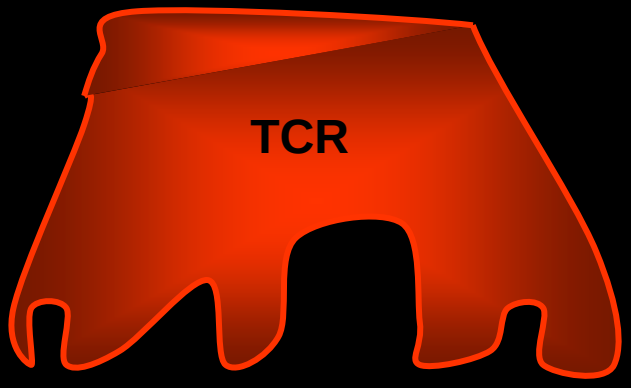
- Seleção Negativa

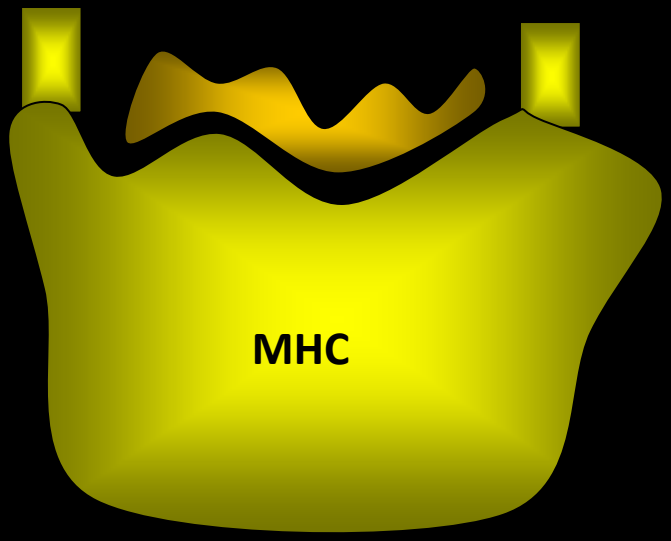
- Impede que linfócitos auto-reativos
- alcancem a periferia.

Seleção Positiva

Sinal de Vida

Negligência





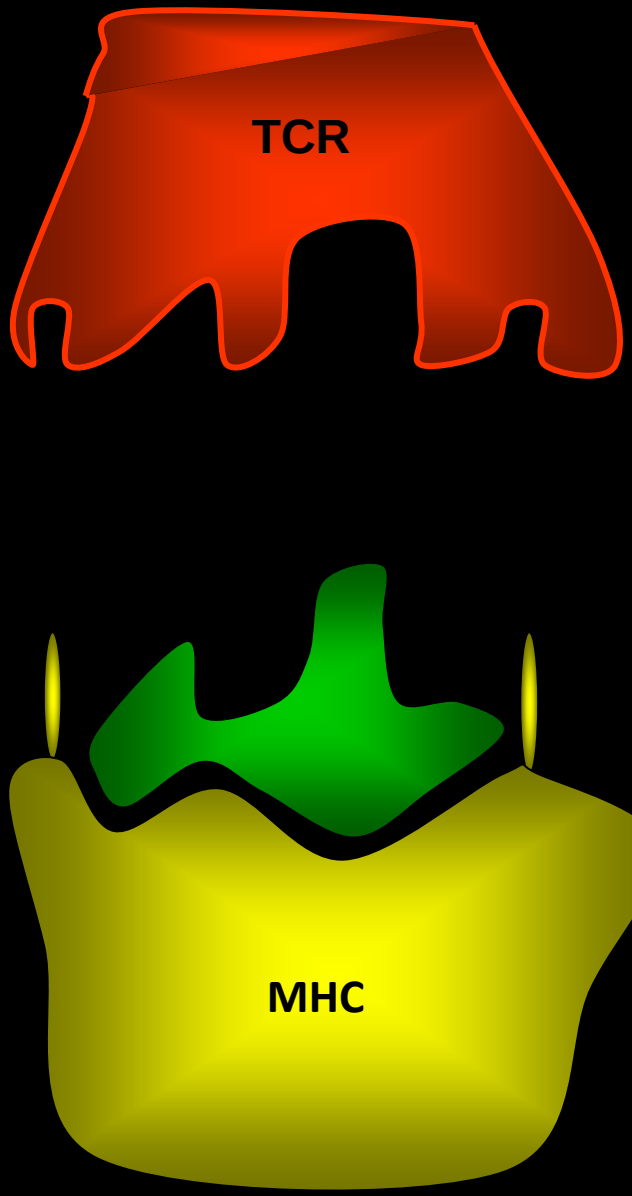
Nenhum Reconhecimento
Sinal de Morte
Morte por negligência

Seleção Negativa

Afinidade da interação
MCH-Peptídeo-TCR

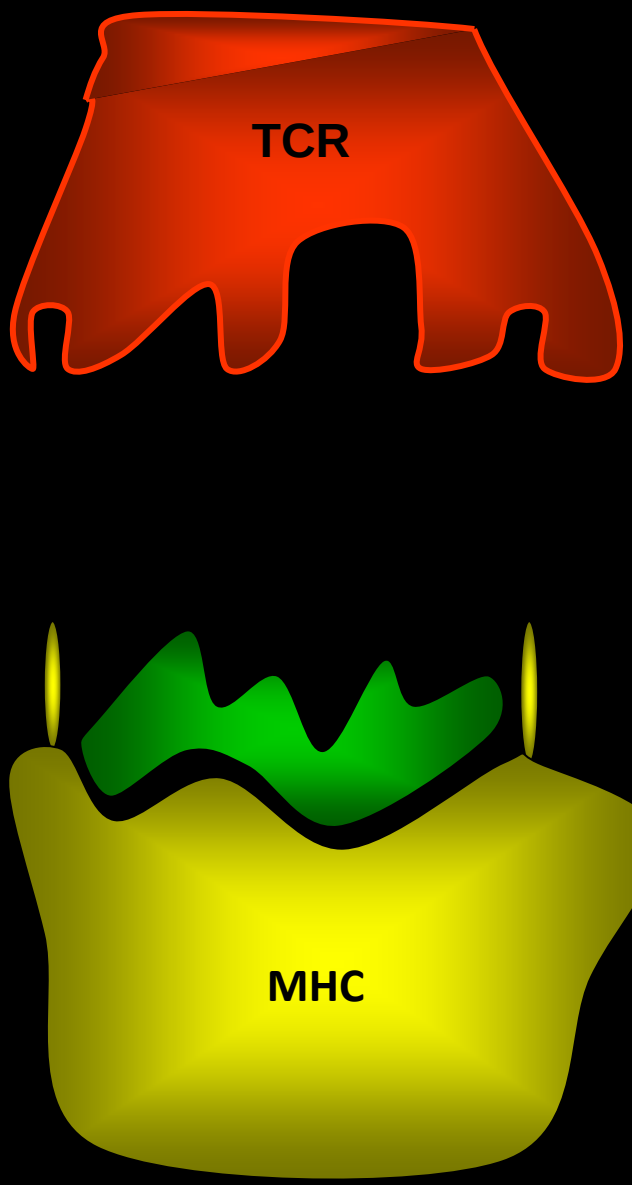
Impede que clones auto-
reativos alcancem a periferia

Seleção Negativa



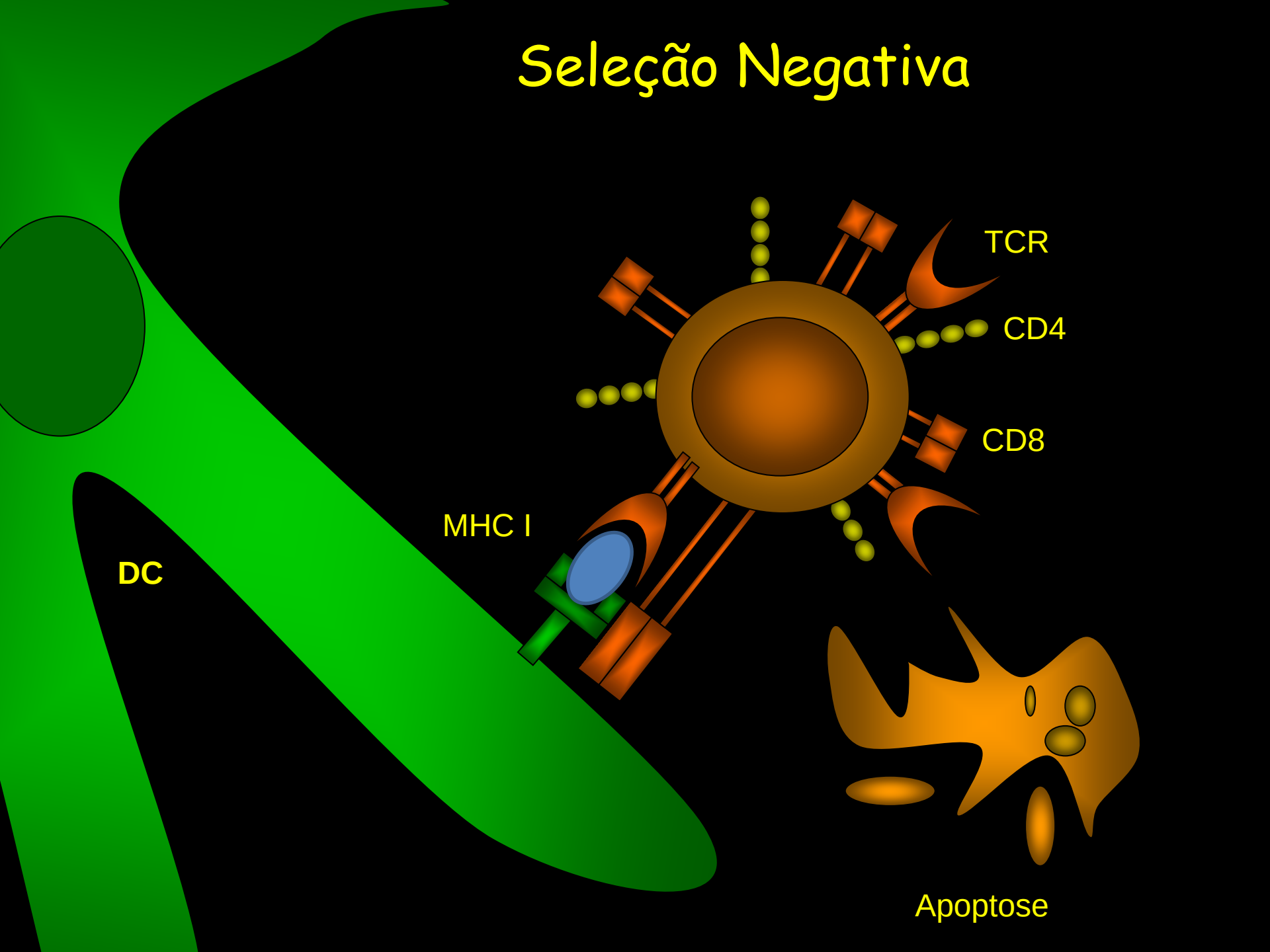
Sinal de Morte
Total
Complementaridade
Tregs

Seleção Positiva

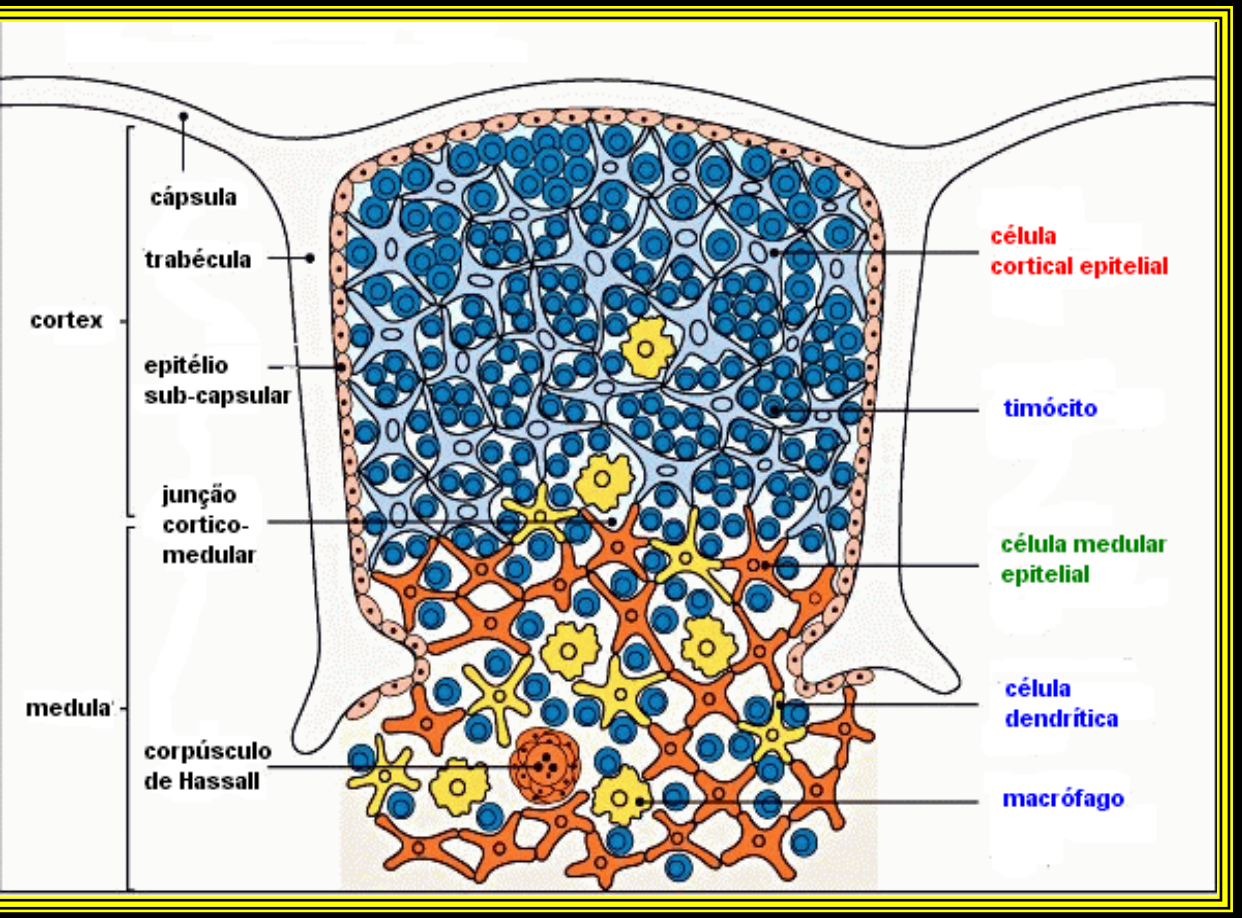


Sinal de vida

Seleção Negativa

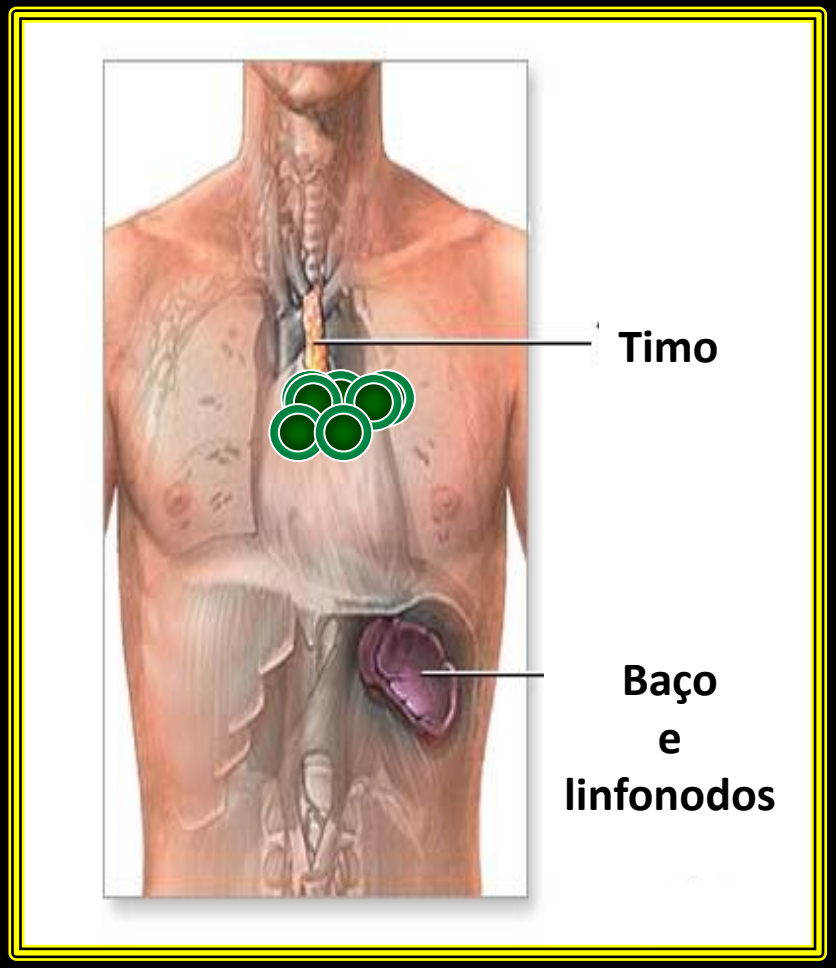


Antígenos não relacionados ao timo como insulina, mielina e antígenos oculares são expressos por células endoteliais da medula tímica (mTEC) por causa do fator de transcrição



AIRE

Regulador Auto-imune



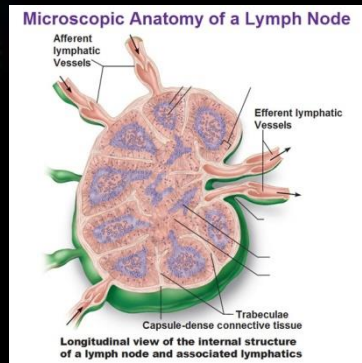
Linfócitos T CD4 e CD8 saem do timo para popular os órgãos linfóides secundários estão prontos para montar uma resposta imune

Mas Então Qual o Problema ?

ESCAPE

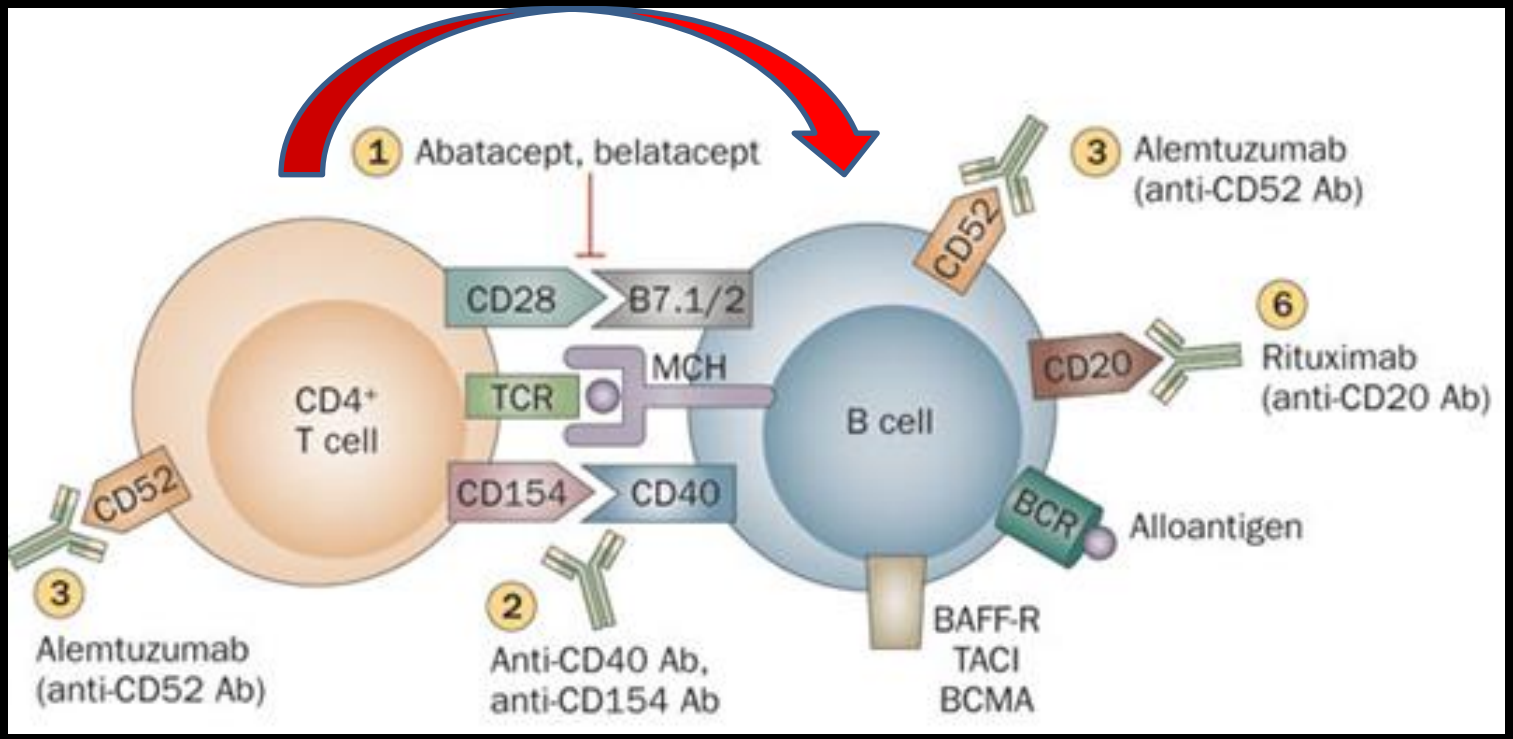
He finally did it!

Anti-Insulina
Anti-Mielina
anti-Colágeno
Anti Fator
Extrínseco



Impedir a Quebra da Tolerância
Impedir 1º. 2º. E 3º. Sinal

Citocinas



O Papel do Adjuvante

Louis Pasteur :

1885 Vacina anti-rábica : paralisia
Extratos de cérebro e medula espinhal de coelhos infectados

Thomas M. Rivers

1909 Esclerose Múltipla
1933 Virologista Rockfeller University

Por quê paralisia?
Qual o agente?

Extratos de cérebro e medula espinhal de coelhos normais e infectados

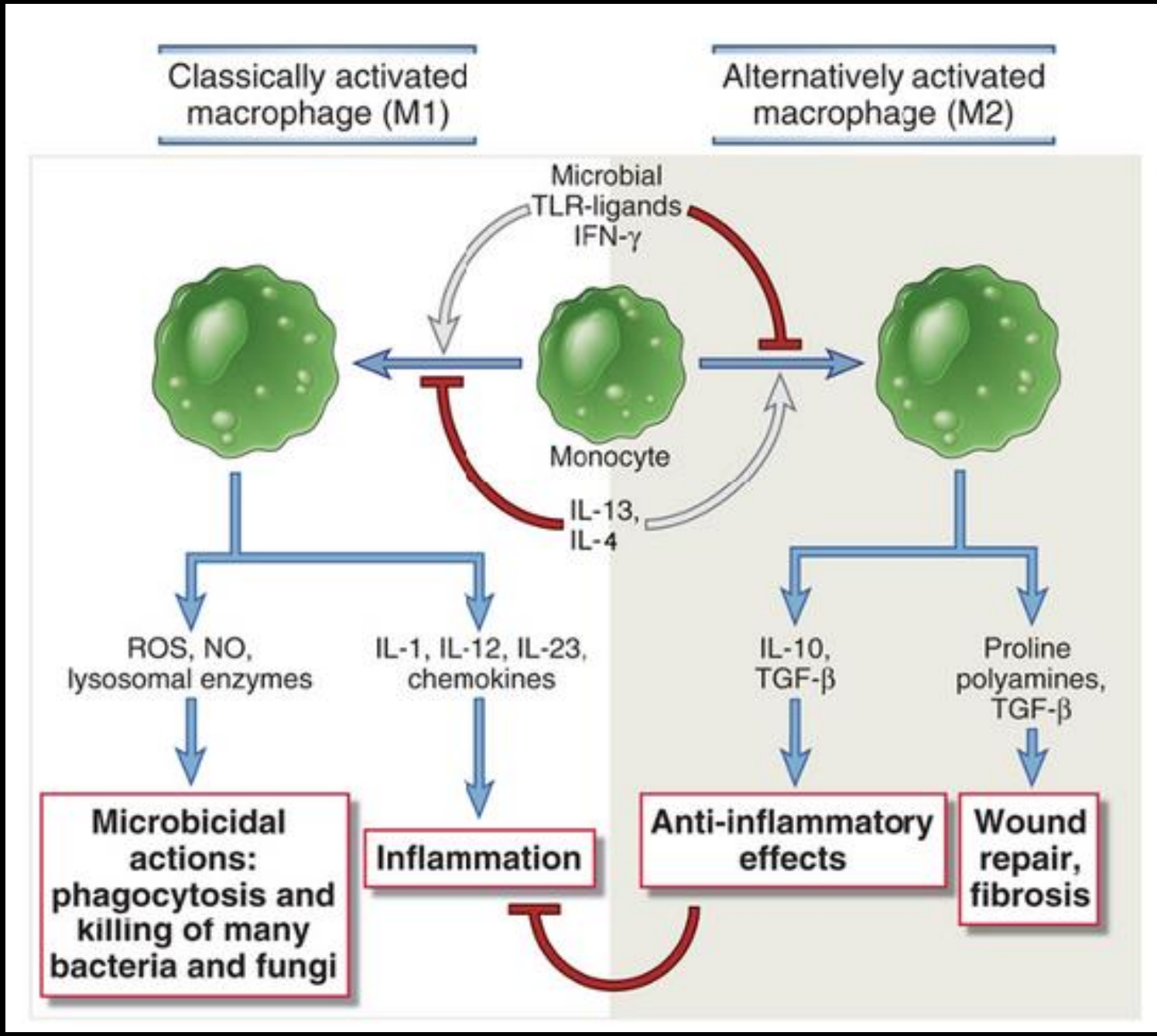
Macacos *Rhesus* desenvolviam paralisia também com extratos provindos de animais não infectados.
EAE

Adjuvante de Freund (1933)

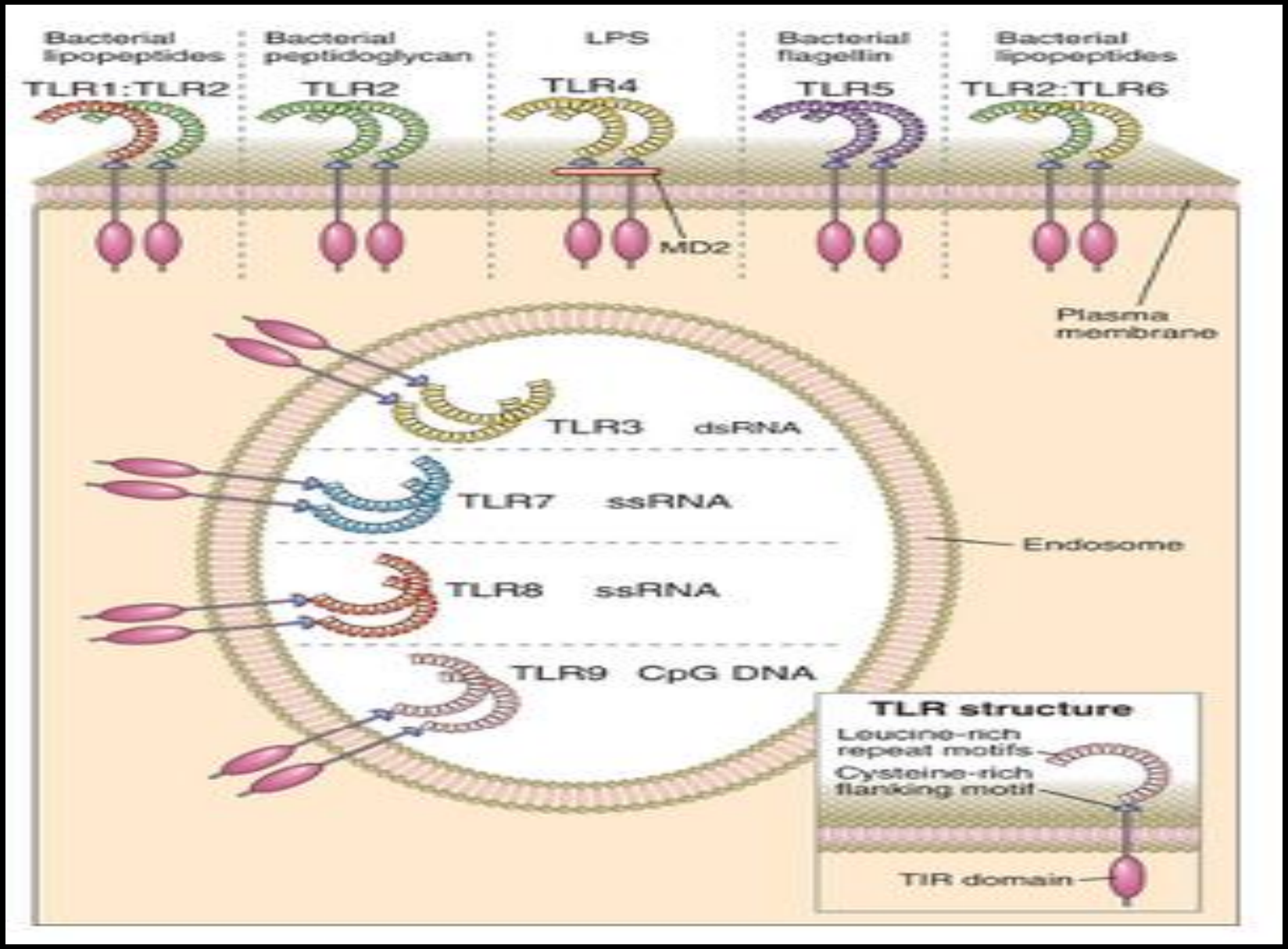
Rivers, T.M., D.H. Sprunt, and G.P. Berry. 1933. ***J. Exp. Med***. 58:39–56.

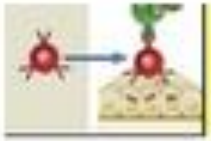
Como Evitar a Ação dos 3 Sinais ?

Mecanismos Supressores da Tolerância Periféria

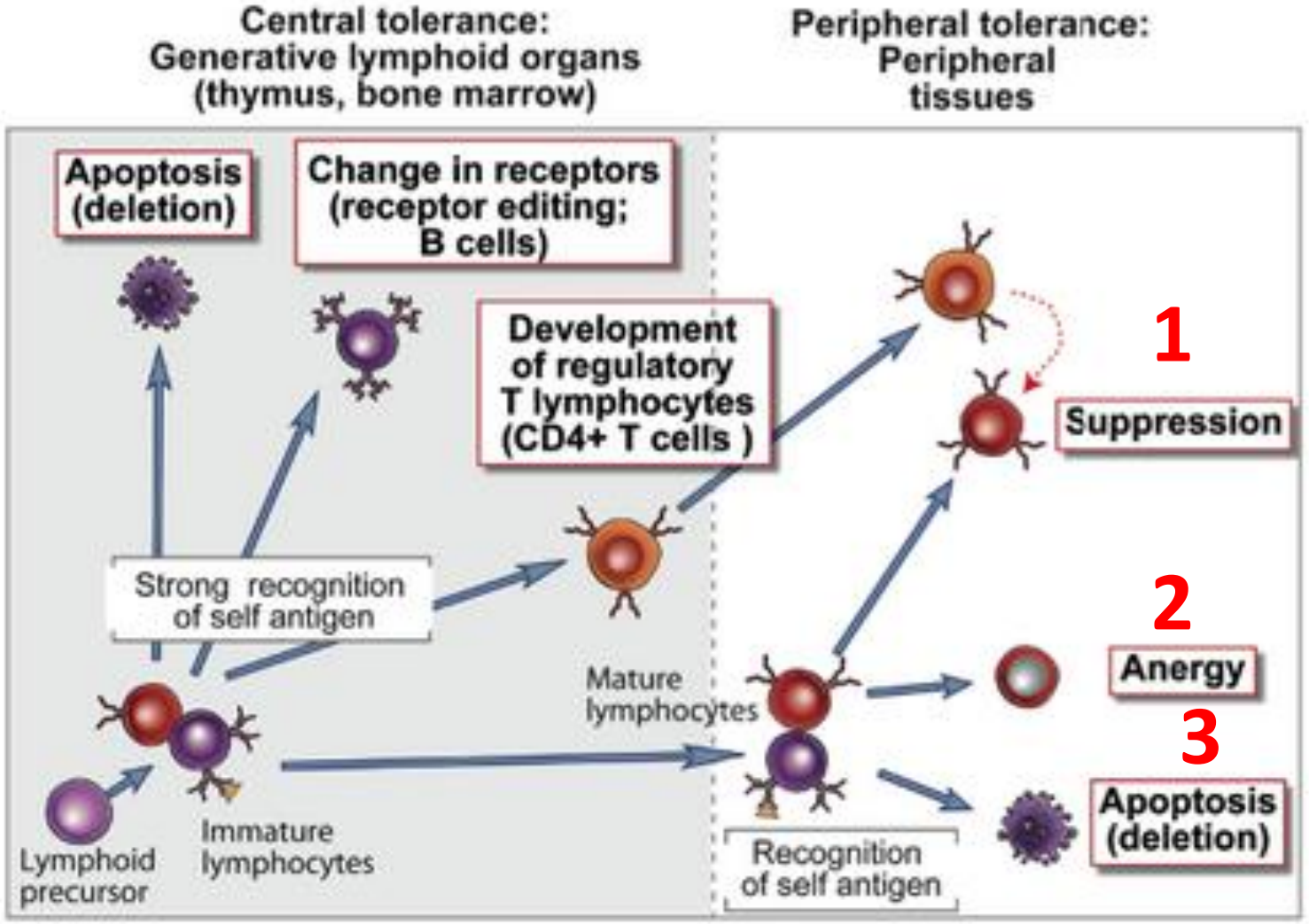

Classically activated macrophage (M1)
Alternatively activated macrophage (M2)
Microbial
TLR-ligands
IFN-γ
Monocyte
IL-13,
IL-4
ROS, NO,
lysosomal enzymes
IL-1, IL-12, IL-23,
chemokines
IL-10,
TGF-β
Proline
polyamines,
TGF-β
Microbicidal actions: phagocytosis and killing of many bacteria and fungi
Inflammation
Anti-inflammatory effects
Wound repair, fibrosis

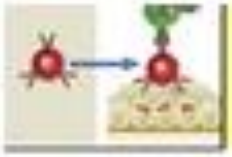
Natureza dos Antígenos





Central and Peripheral Self Tolerance

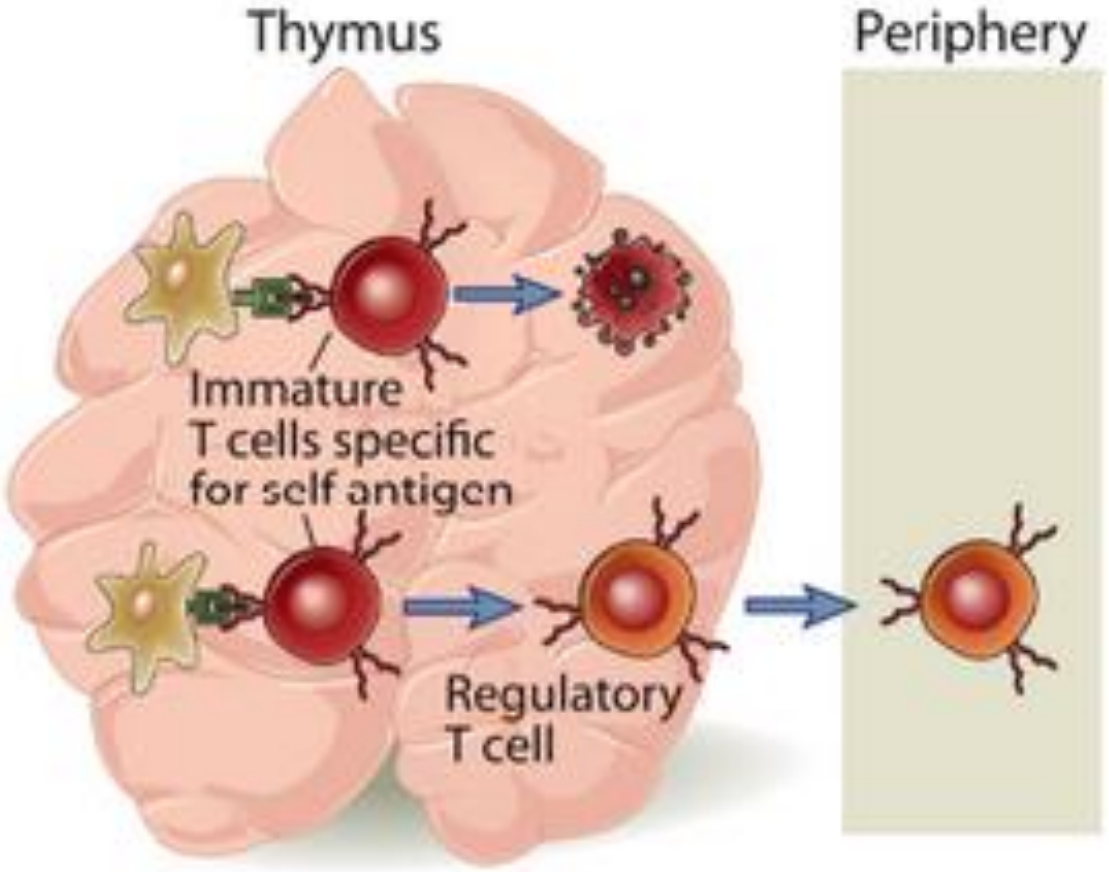


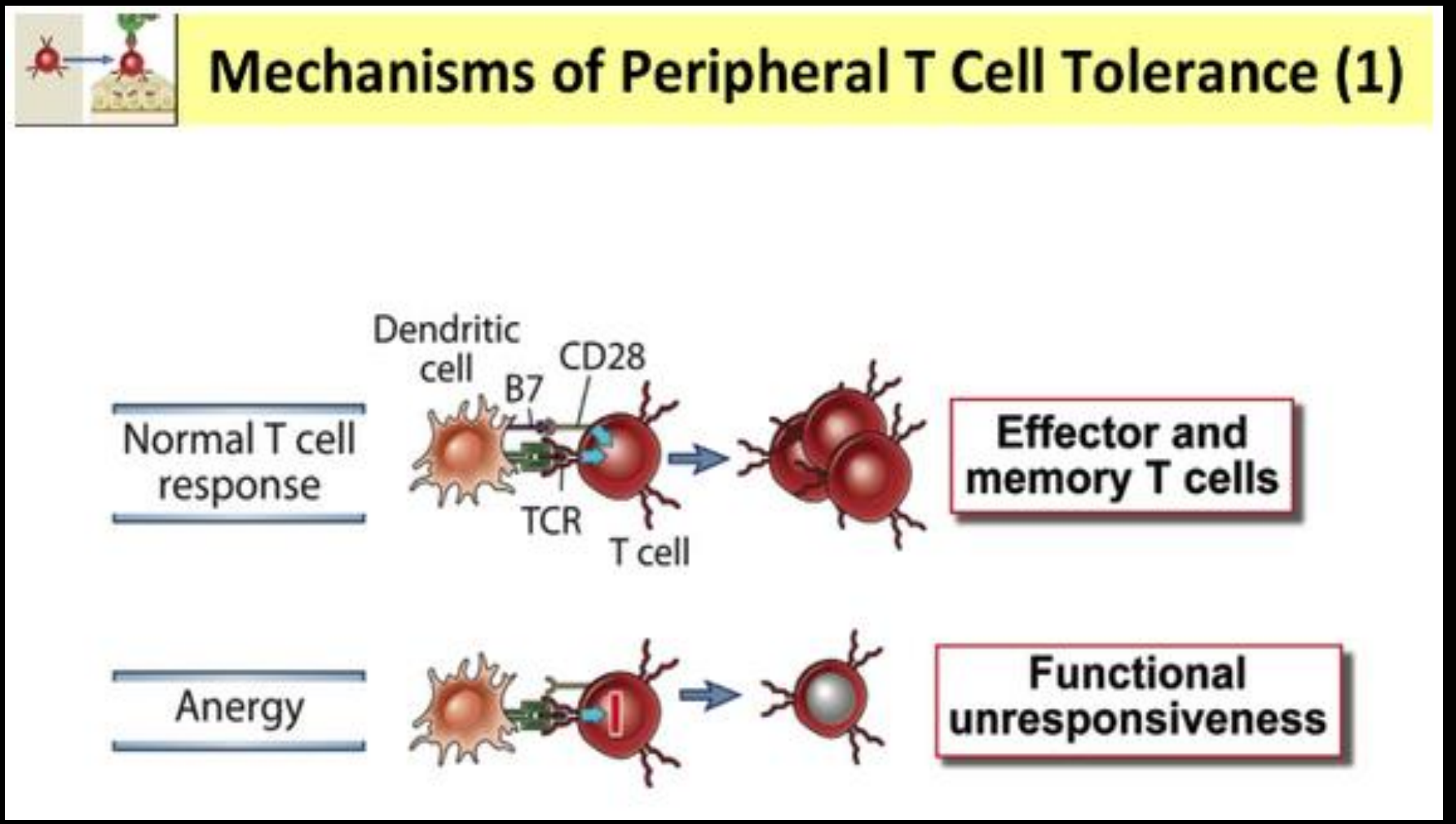


Central T Cell Tolerance

Negative selection: deletion

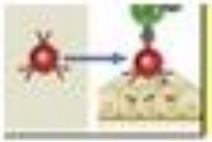
Development of regulatory T cells



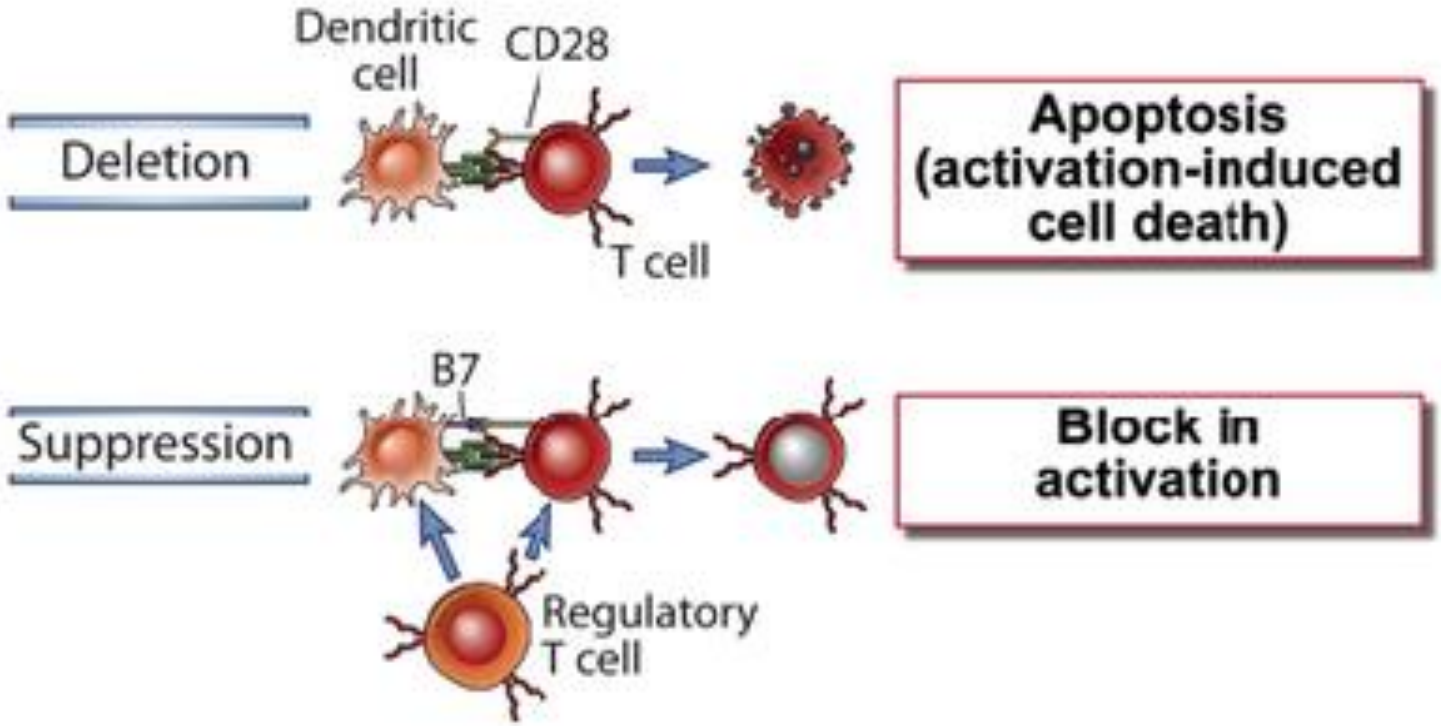


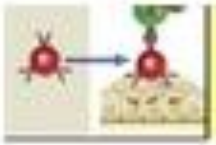
Ausência de Co-estimulação – Proteínas sem adjuvante

Não induzem resposta efetora – memória .

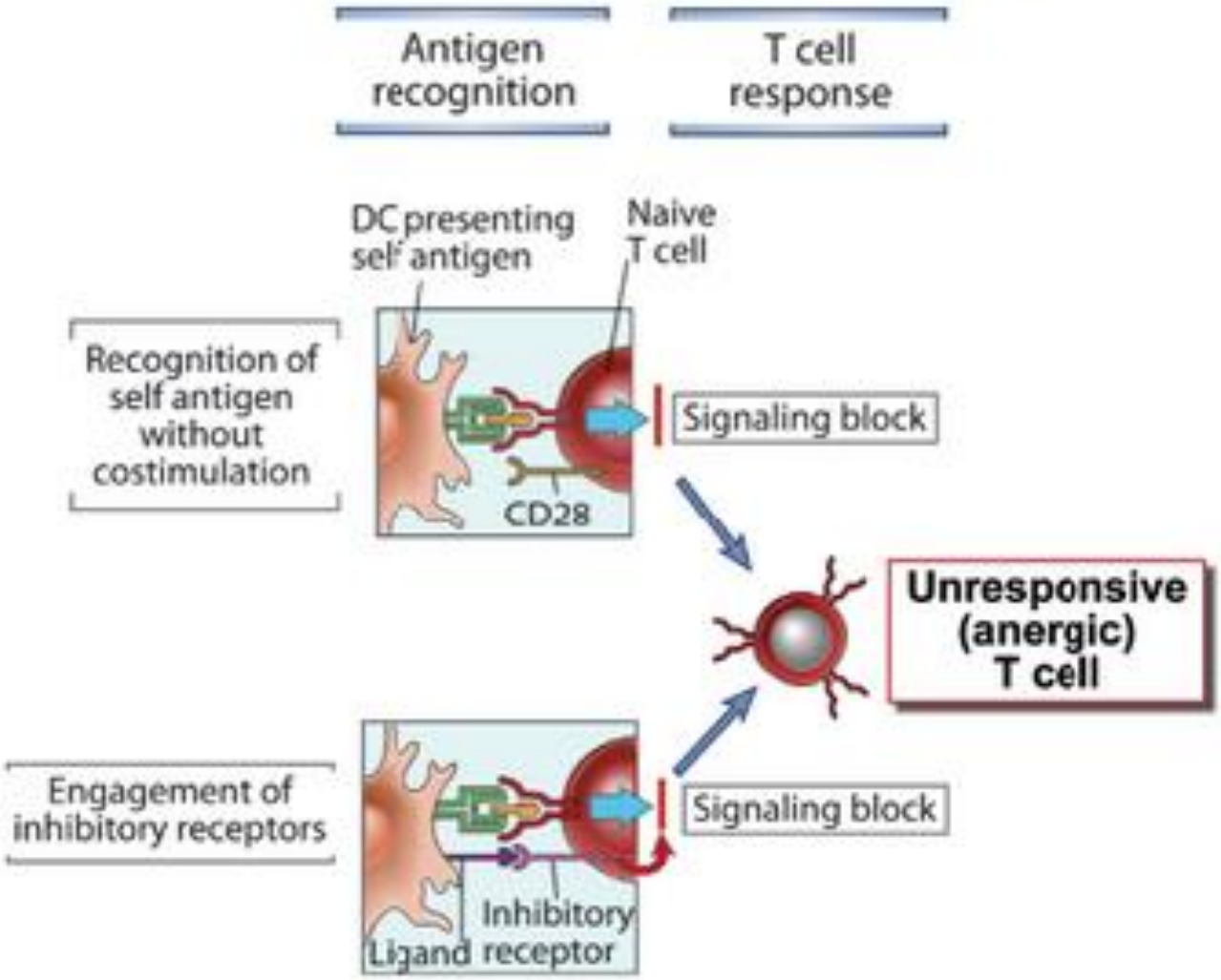


Central and Peripheral Self Tolerance (2)

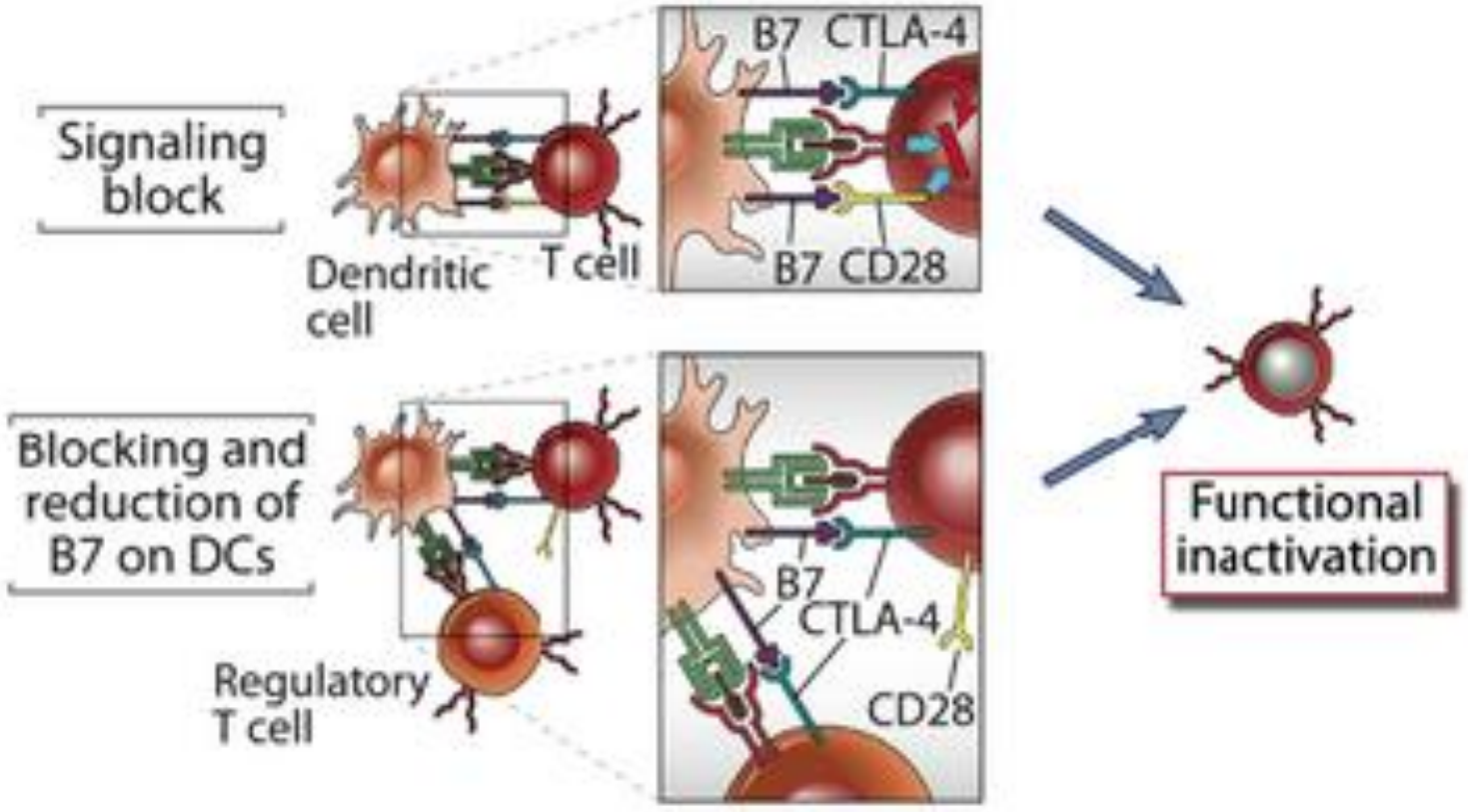




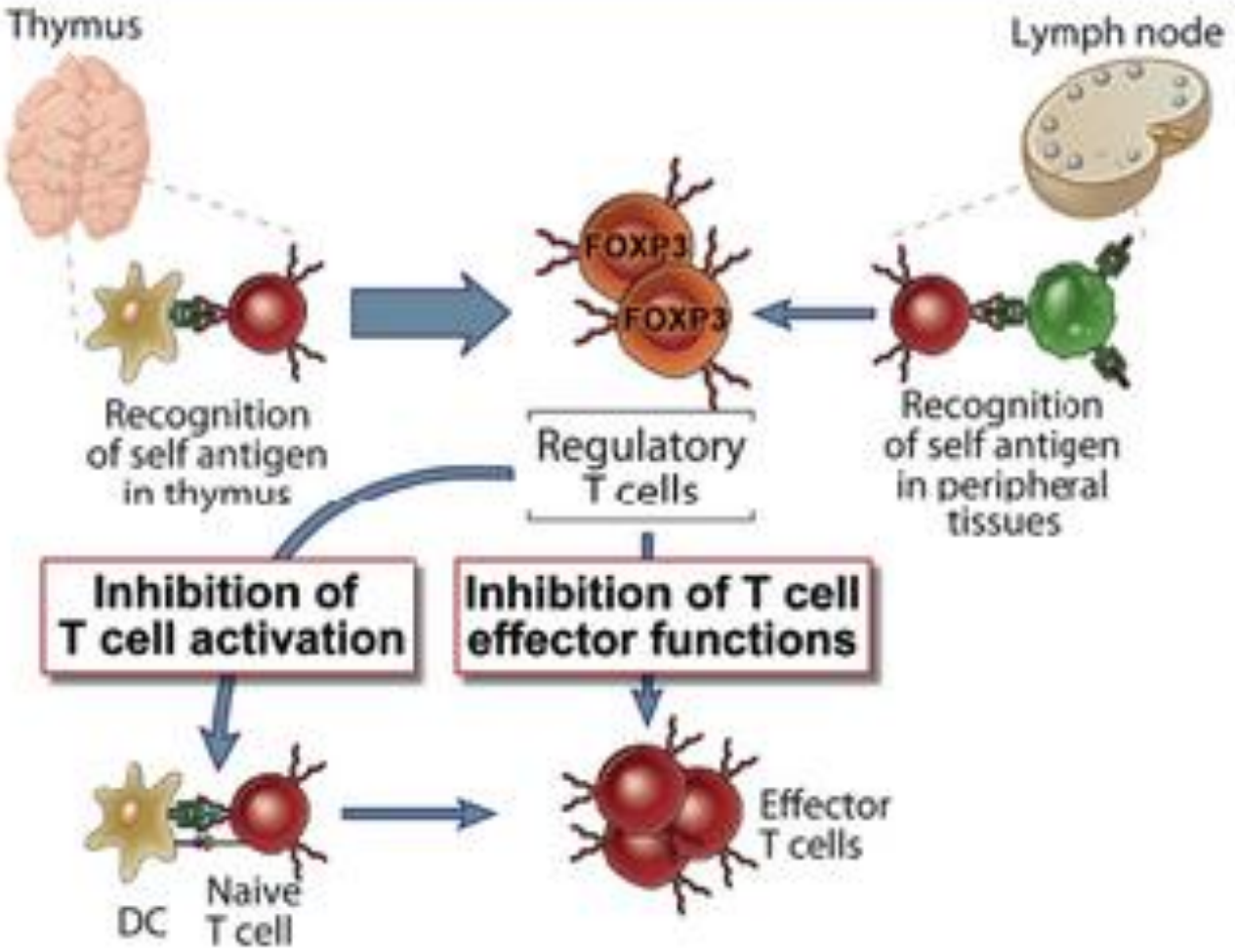
Mechanisms of T cell Anergy

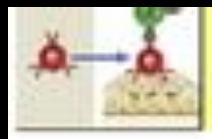


Mechanisms of Action of CTLA-4

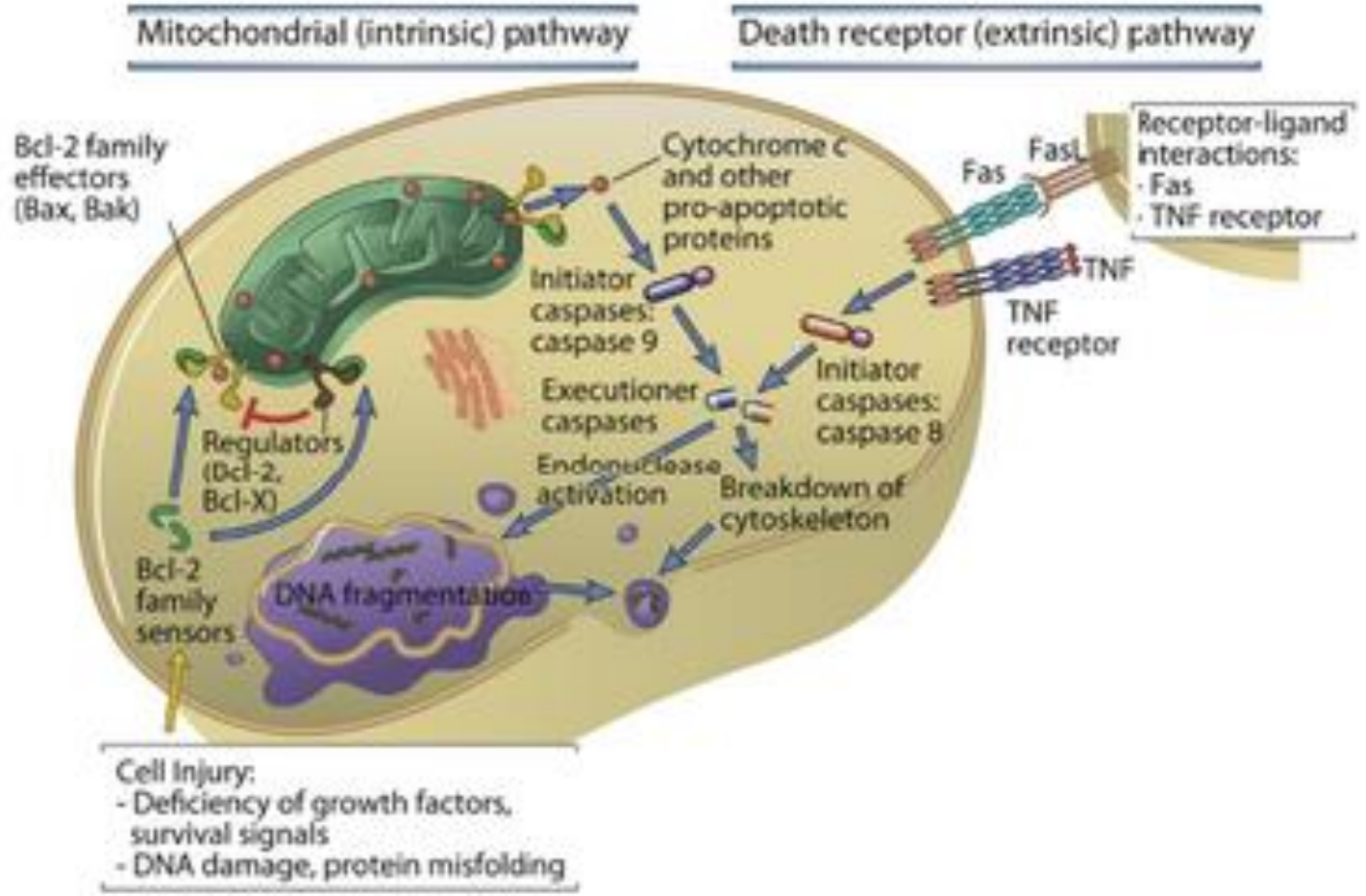


Regulatory T Cells

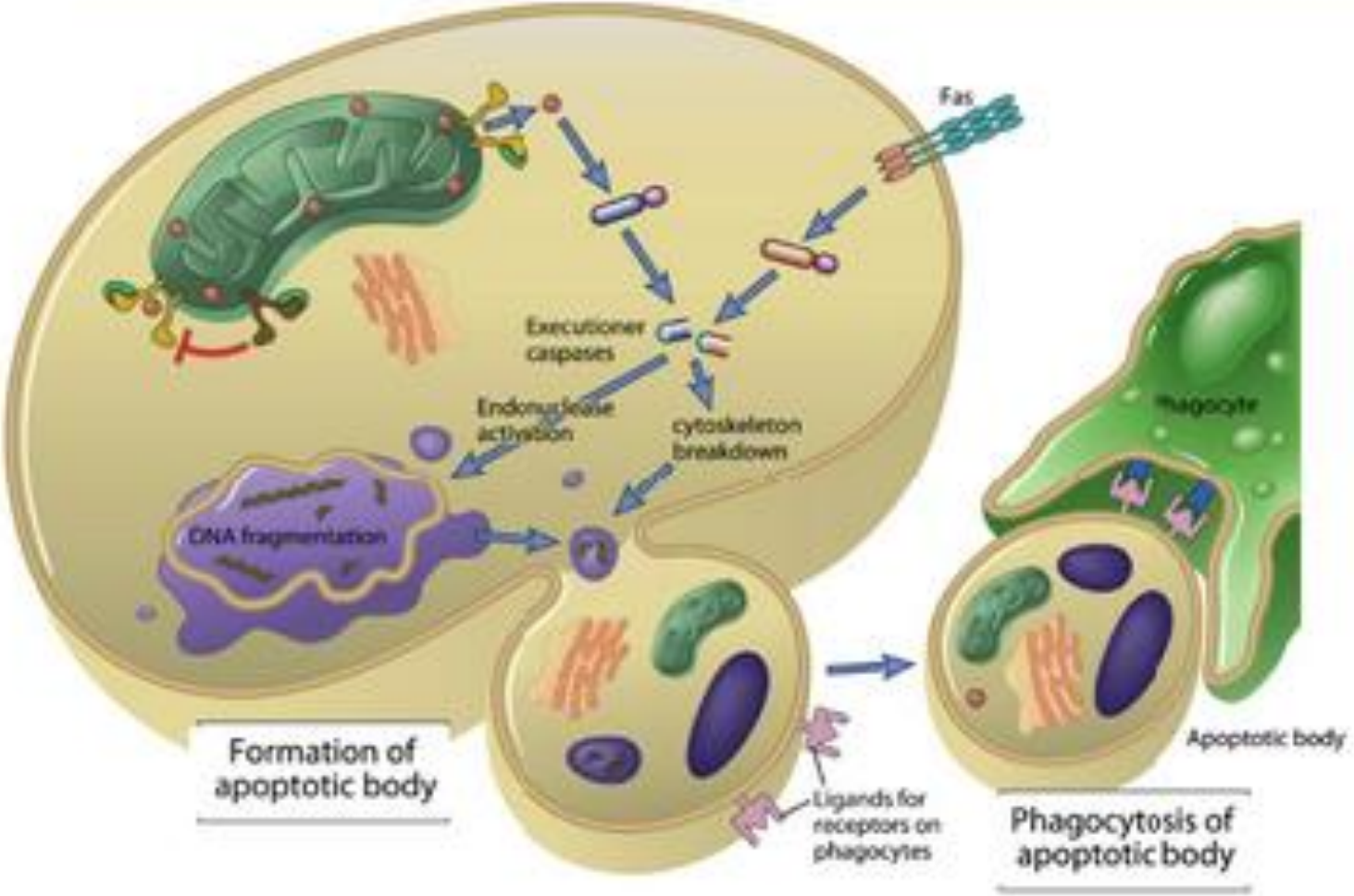




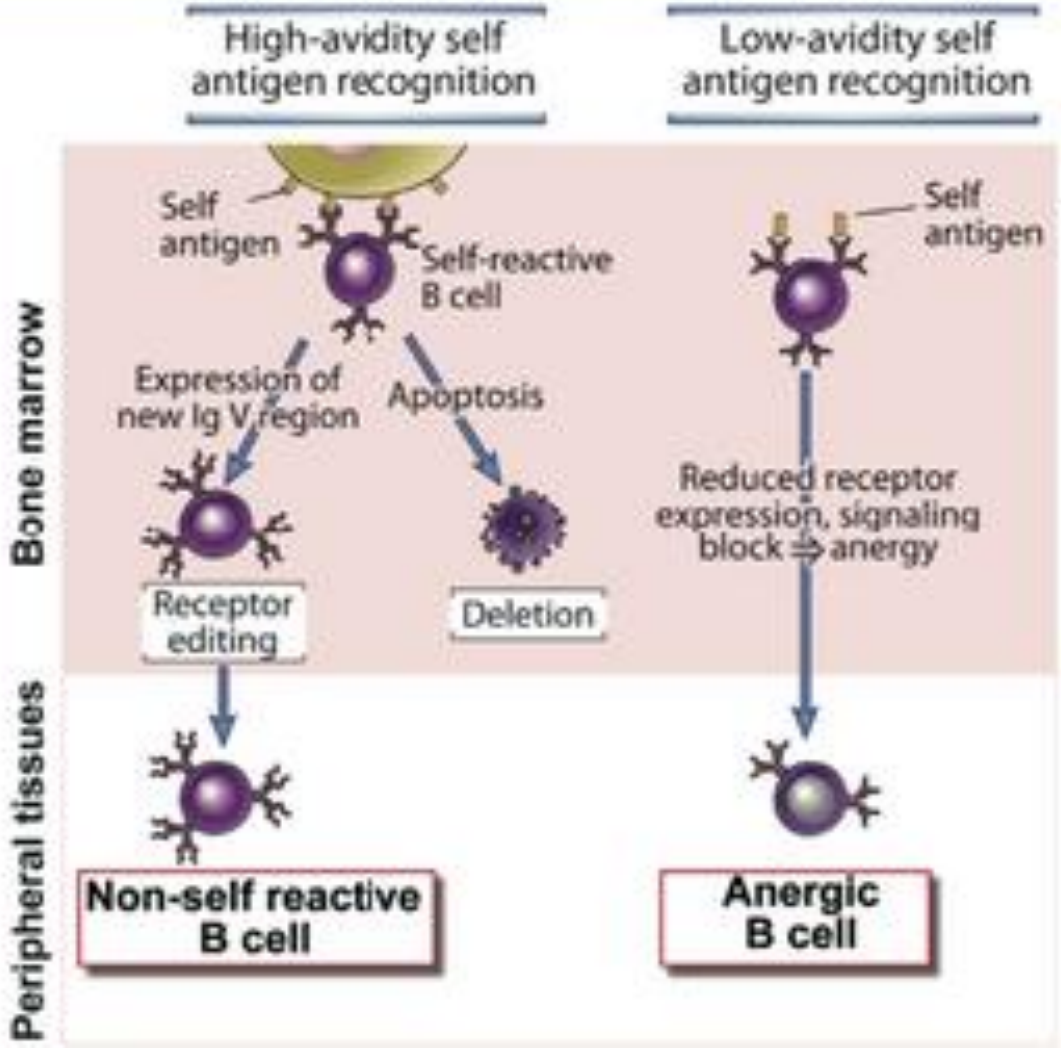
Pathways of Apoptosis (1)

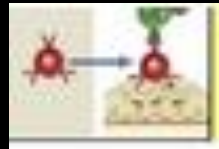


Pathways of Apoptosis (2)

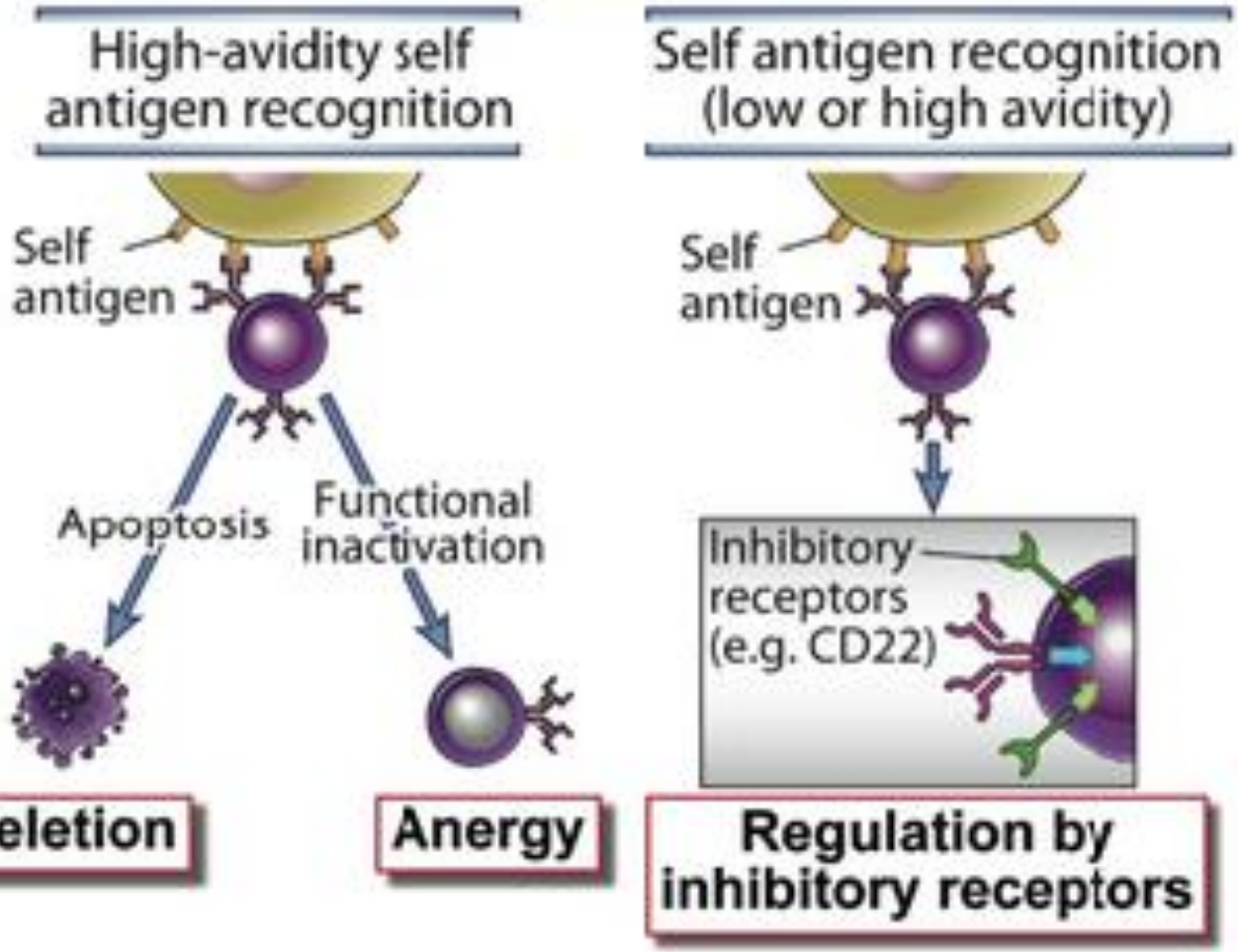


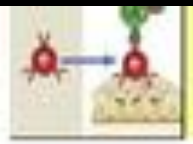
Central Tolerance in B cells



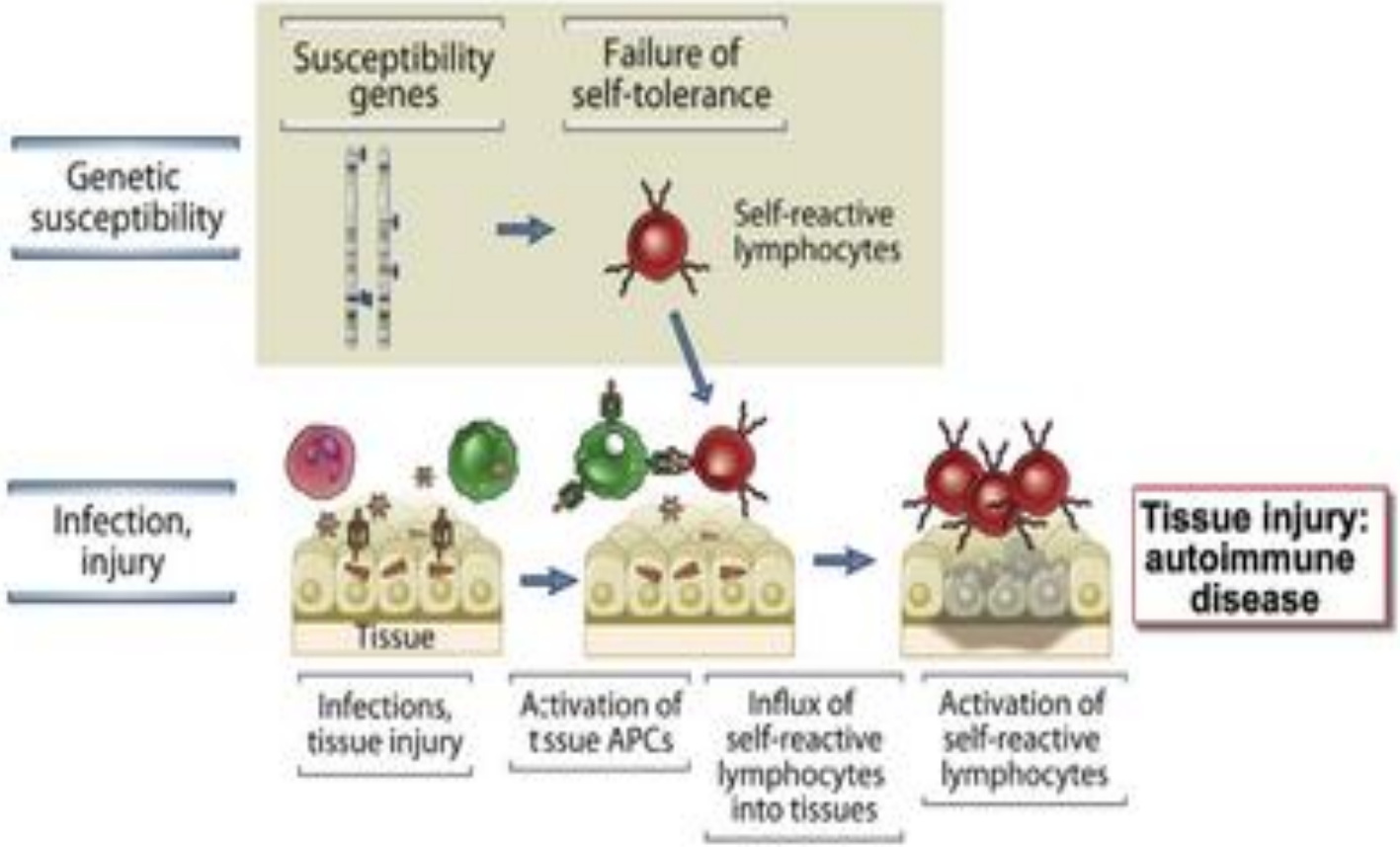


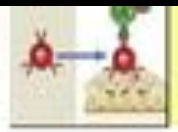
Peripheral Tolerance in B cells



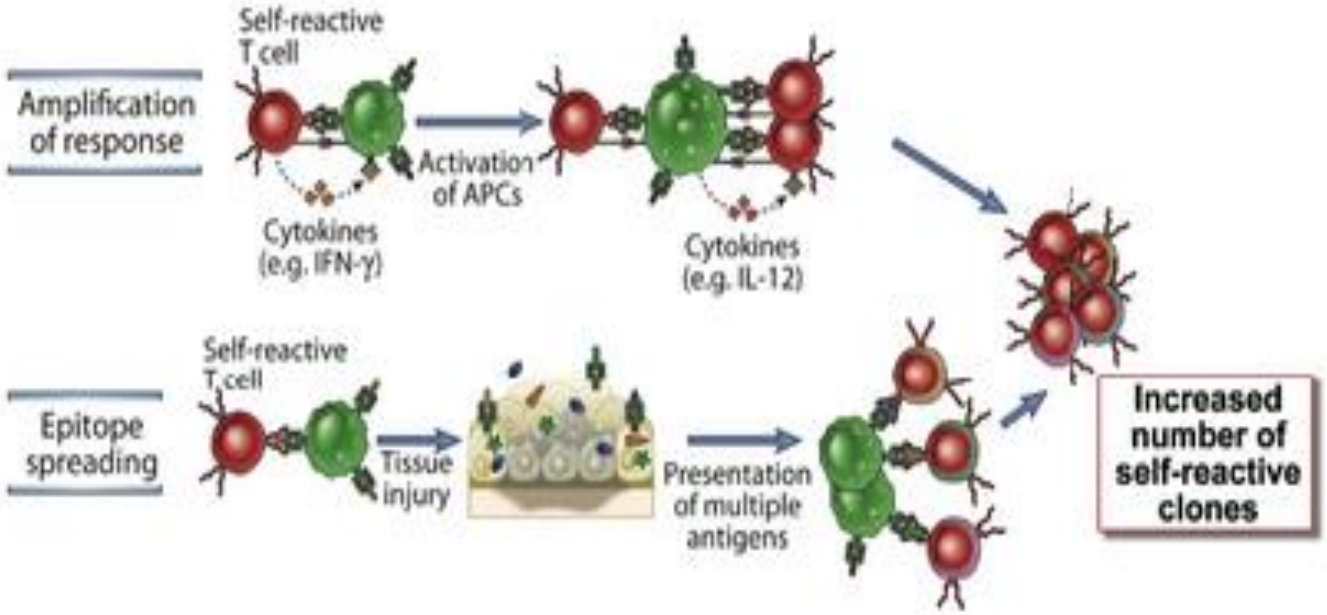


Postulated Mechanisms of Autoimmunity

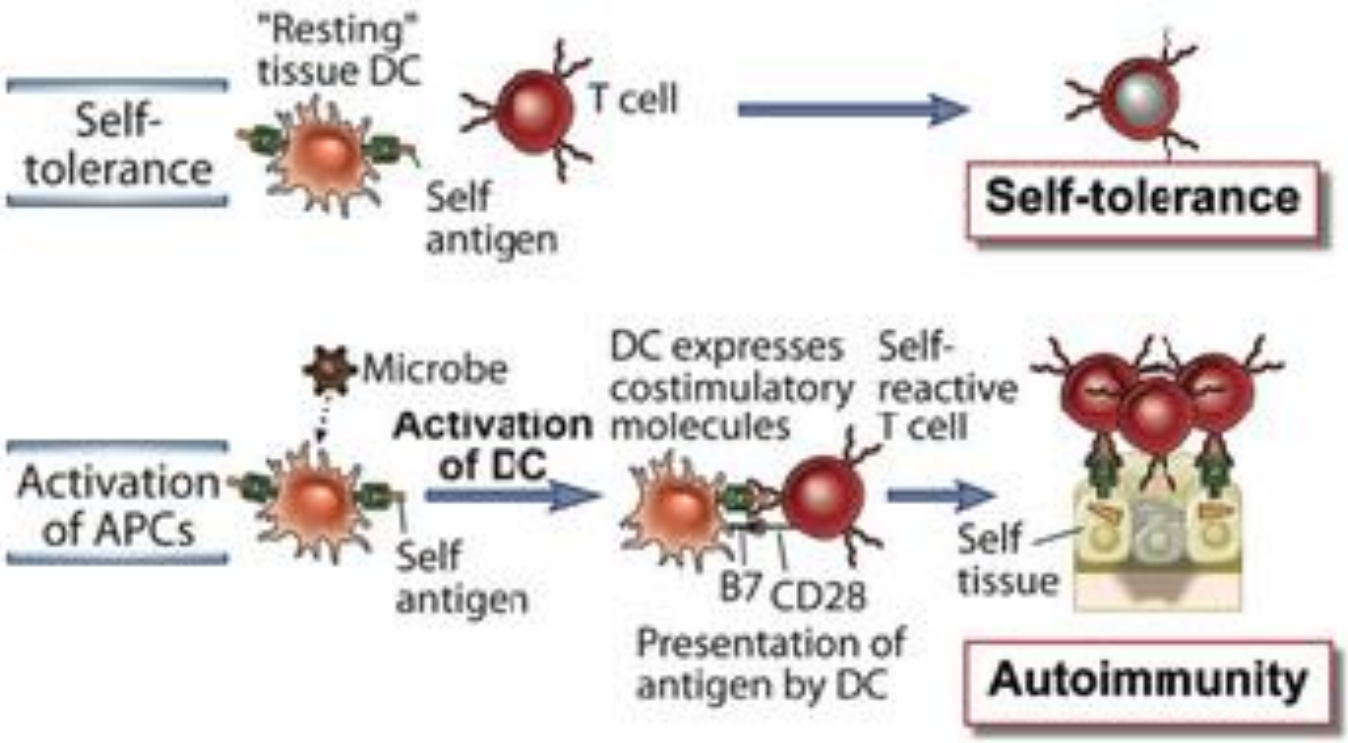


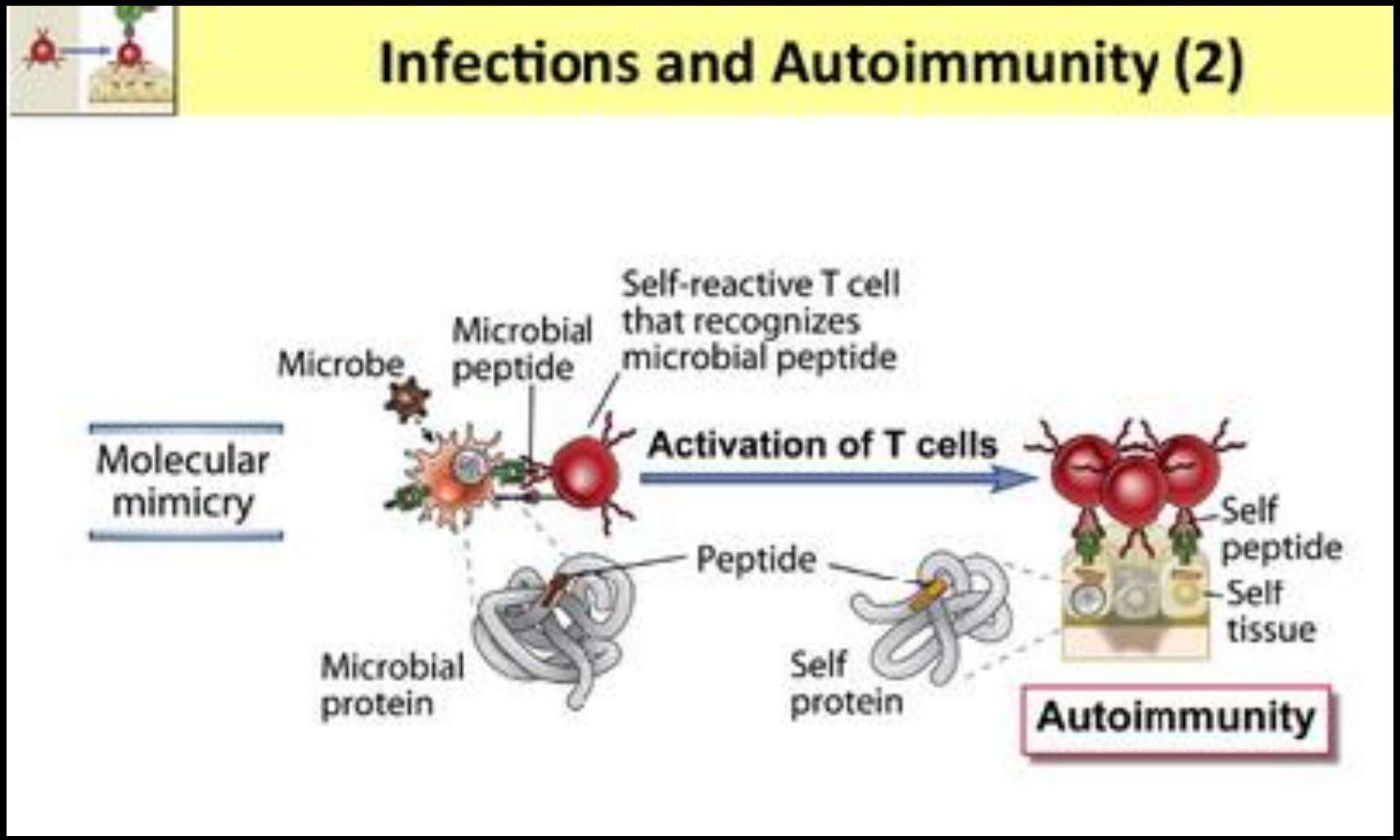


Chronicity of Autoimmune Diseases



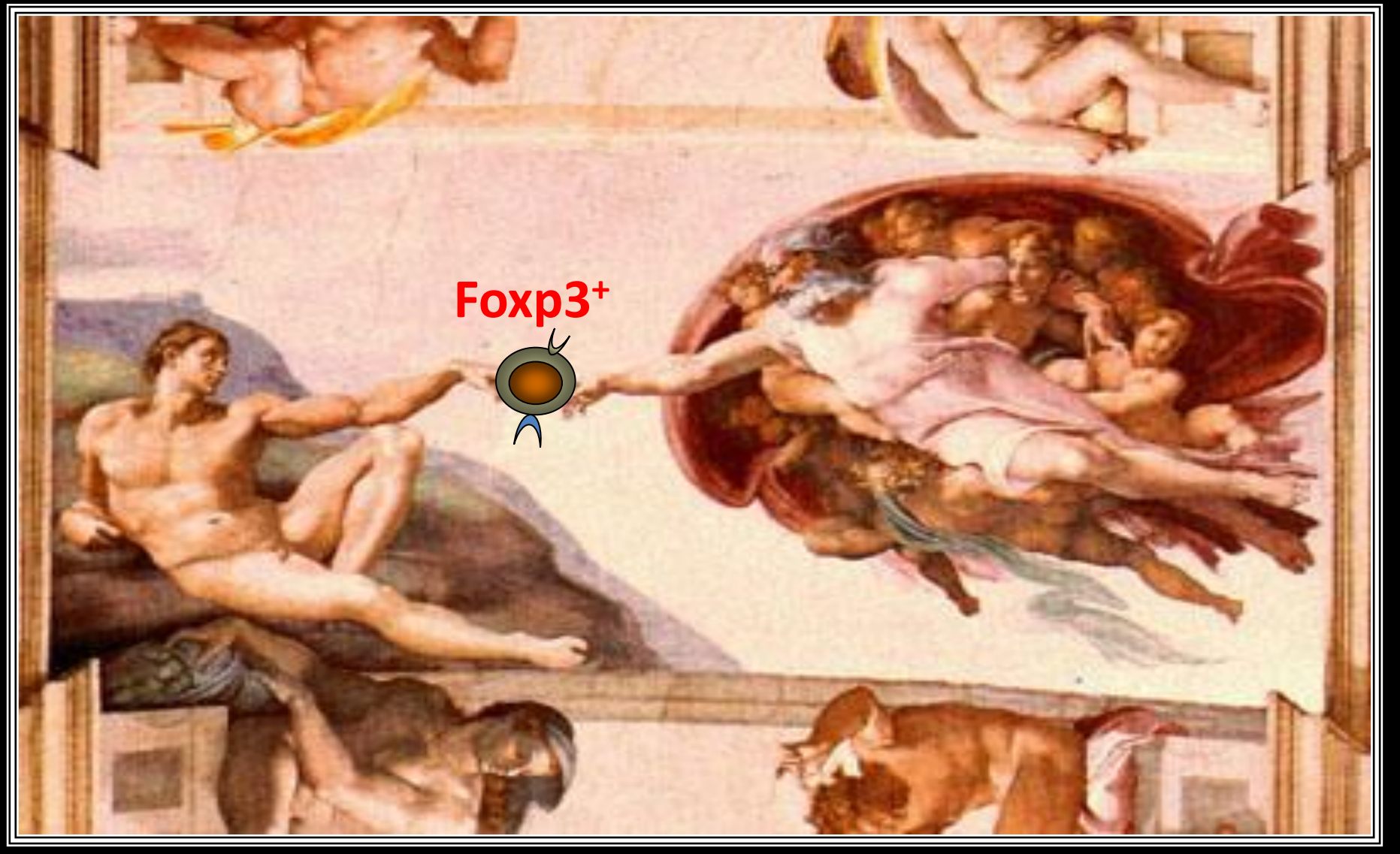
Infections and Autoimmunity (1)



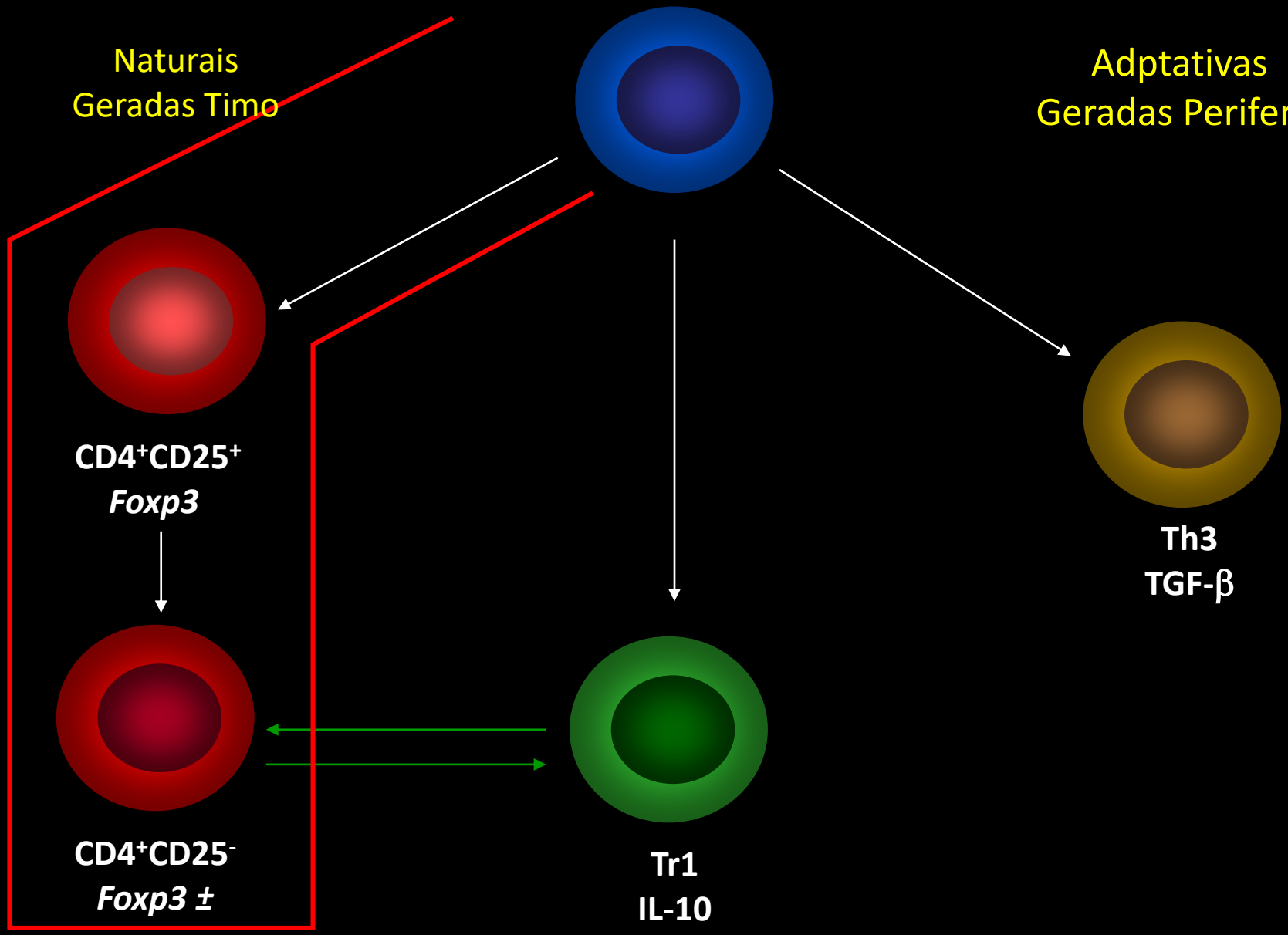


Mimetismo Molecular

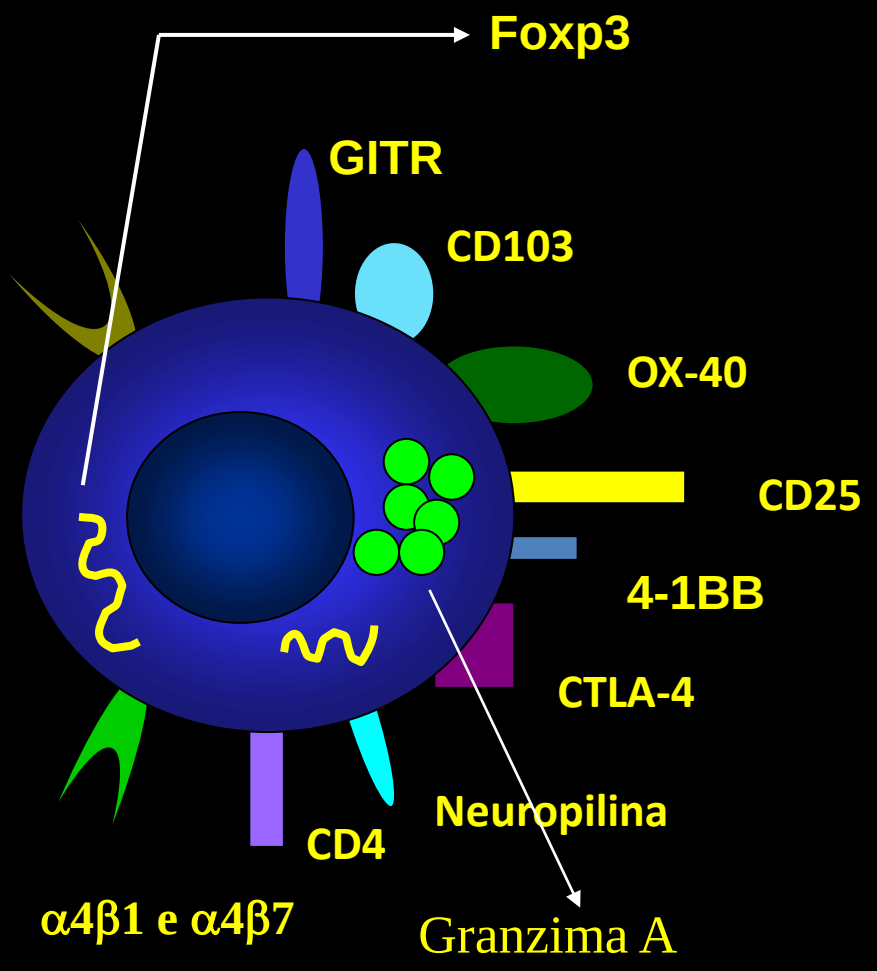
1995 Renascimento das células T reguladoras



Tipos de Tregs



Tregs Naturais



Ação por contato;
IL-2;
Geradas no timo;
GITR
Foxp3
Granzima A

Mecanismos Regulatórios

- Anergia clonal
- Citocinas supressoras
- Down-regulation de moléculas apresentadoras
- Indoleamina, 2,3-dioxigenase
- Citotoxicidade
- Fosfatases, ubiquitina ligases, SOCS
- miRNA

Células T Reguladoras

Timectomia

BALB/c adulto

$CD4^+CD25^+$
ou
$CD4^+CD25^-$

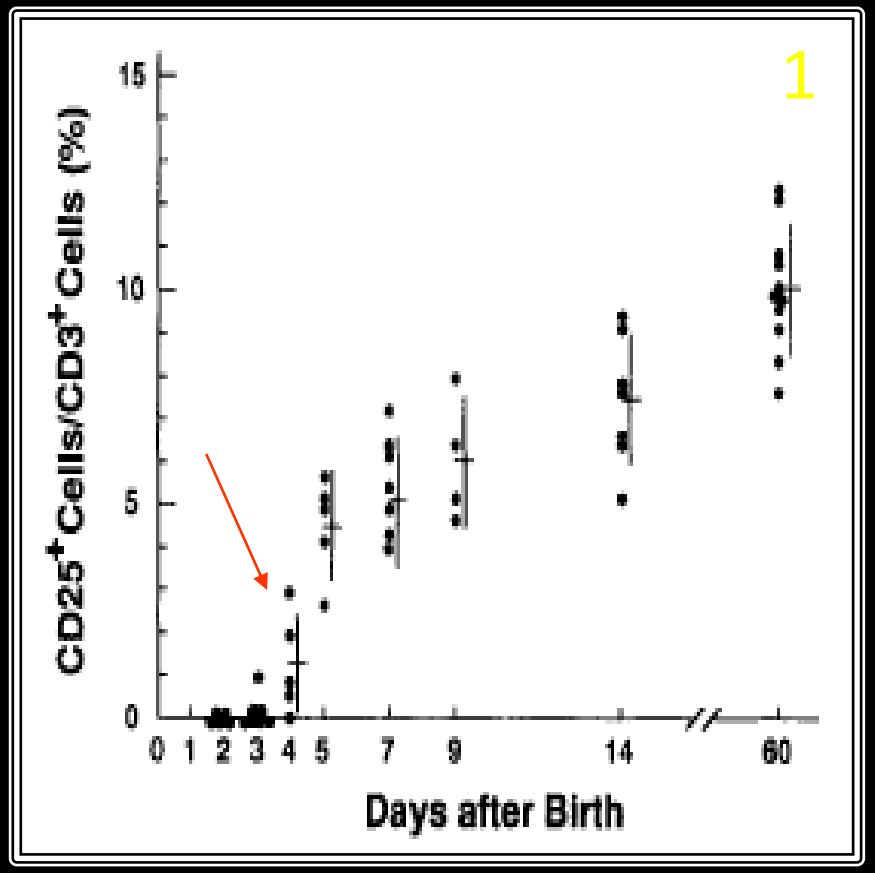
Timectomia dia 3

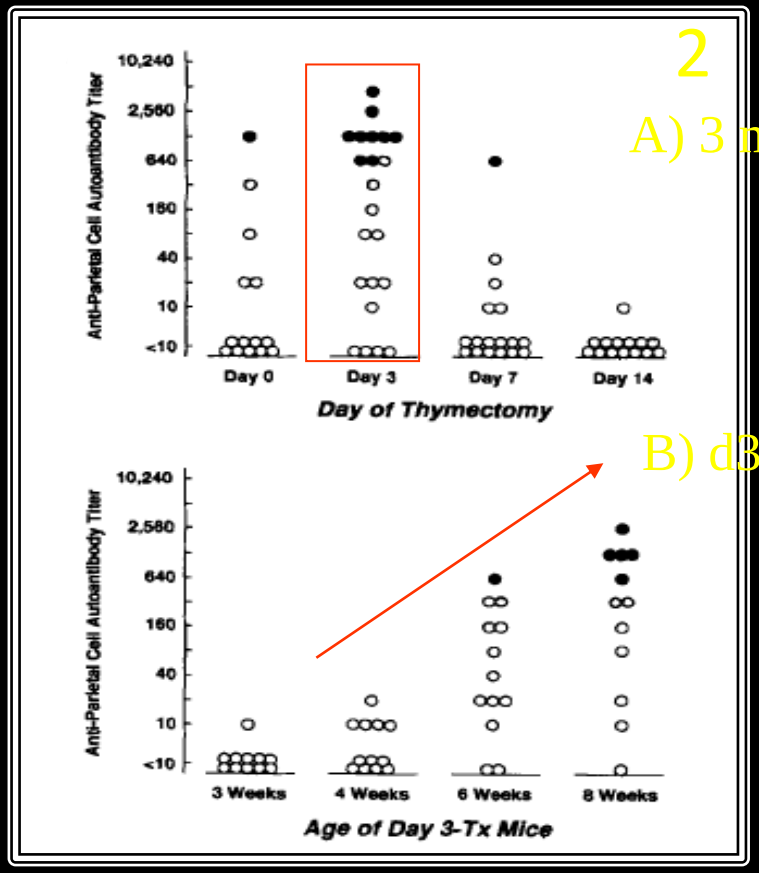
7 dias

3 meses

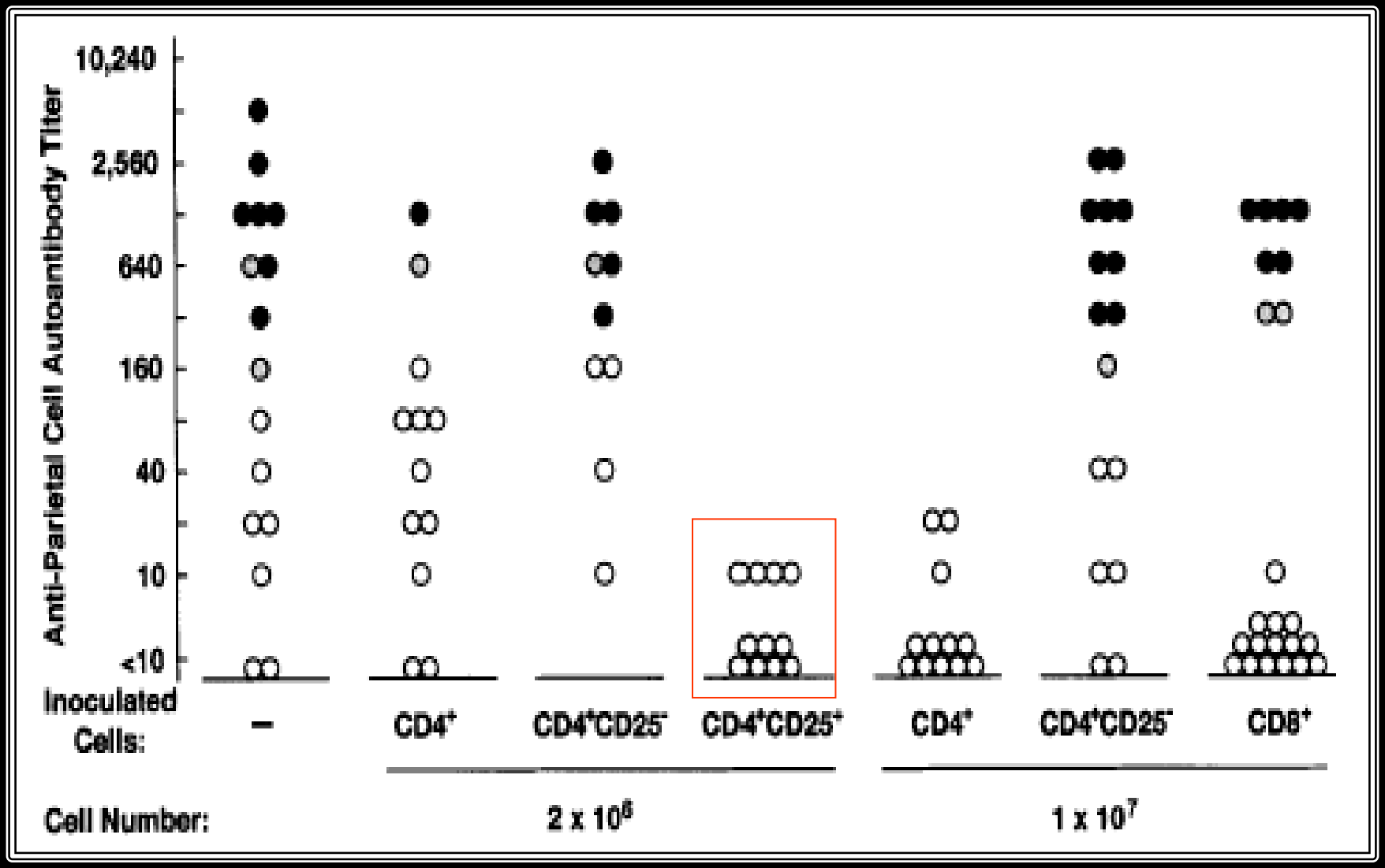
Asano, M.; Toda, M.; Sakaguchi, N. and Sakaguchi, S. Autoimmune disease as a consequence of developmental abnormality of a T cell subpopulation. ***J Exp Med*** 184: 387, 1996.

Resultados





1) Número de células T $CD4^+CD25^+$ no baço de camundongos BALB/c nos dias 1-60.
2) Auto-anticorpos em camundongos que receberam células $CD4^+CD25^+$ ou $CD4^+CD25^-$.


Anti-Parietal Cell Autoantibody Titer
10,240
2,560
640
160
40
10
<10
Inoculated Cells:
–
CD4+
CD4+CD25-
CD4+CD25+
CD4+
CD4+CD25-
CD8+
Cell Number:
2 x 10^6
1 x 10^7

Conclusão

Aparentemente as células T reguladoras $CD4^+CD25^+$ são geradas no timo a partir do 3o. dia de vida.

Periferia

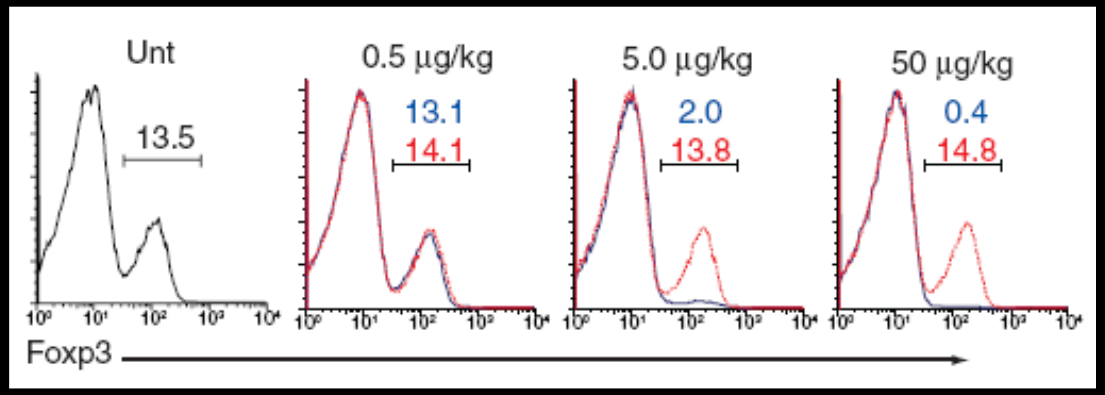
nature immunology

Regulatory T cells prevent catastrophic autoimmunity throughout the lifespan of mice

Jeong M Kim[1], Jeffrey P Rasmussen[1] & Alexander Y Rudensky[1,2]

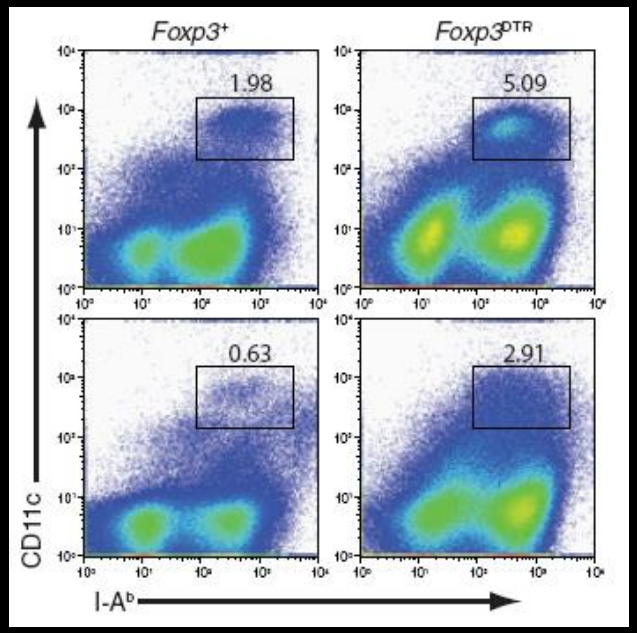
Mice lacking the transcription factor Foxp3 (*Foxp3*$^{-}$) lack regulatory T (T_{reg}) cells and develop fatal autoimmune pathology. In *Foxp3*$^{-}$ mice, many activated effector T cells express self-reactive T cell receptors that are expressed in T_{reg} cells in wild-type mice. Thus, in wild-type mice, most self-reactive thymocytes escaping negative selection are diverted into the T_{reg} lineage, and whether T_{reg} cells are critical in self-tolerance in wild-type mice remains unknown. Here, acute *in vivo* ablation of T_{reg} cells demonstrated a vital function for T_{reg} cells in neonatal and adult mice. We suggest that self-reactive T cells are continuously suppressed by T_{reg} cells and that when suppression is relieved, self-reactive T cells become activated and facilitate accelerated maturation of dendritic cells.

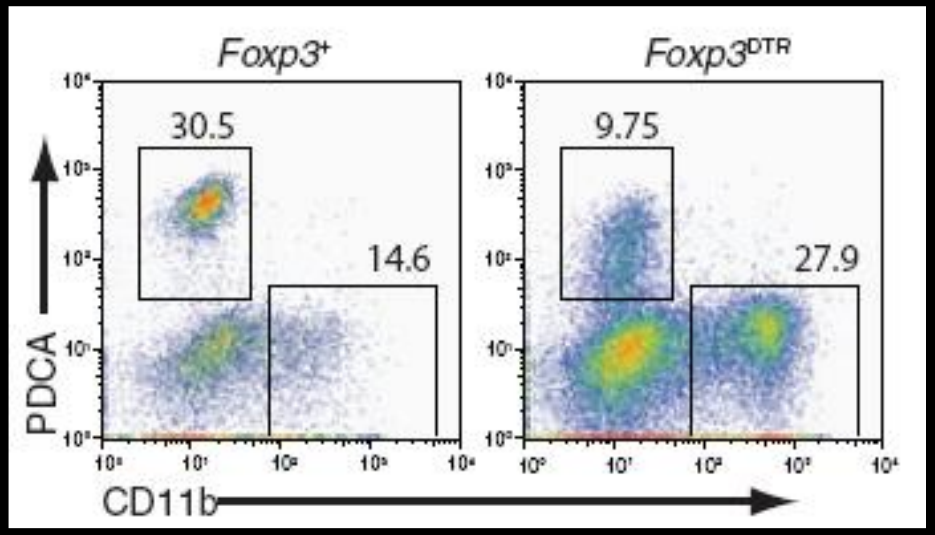
Depleção de Tregs



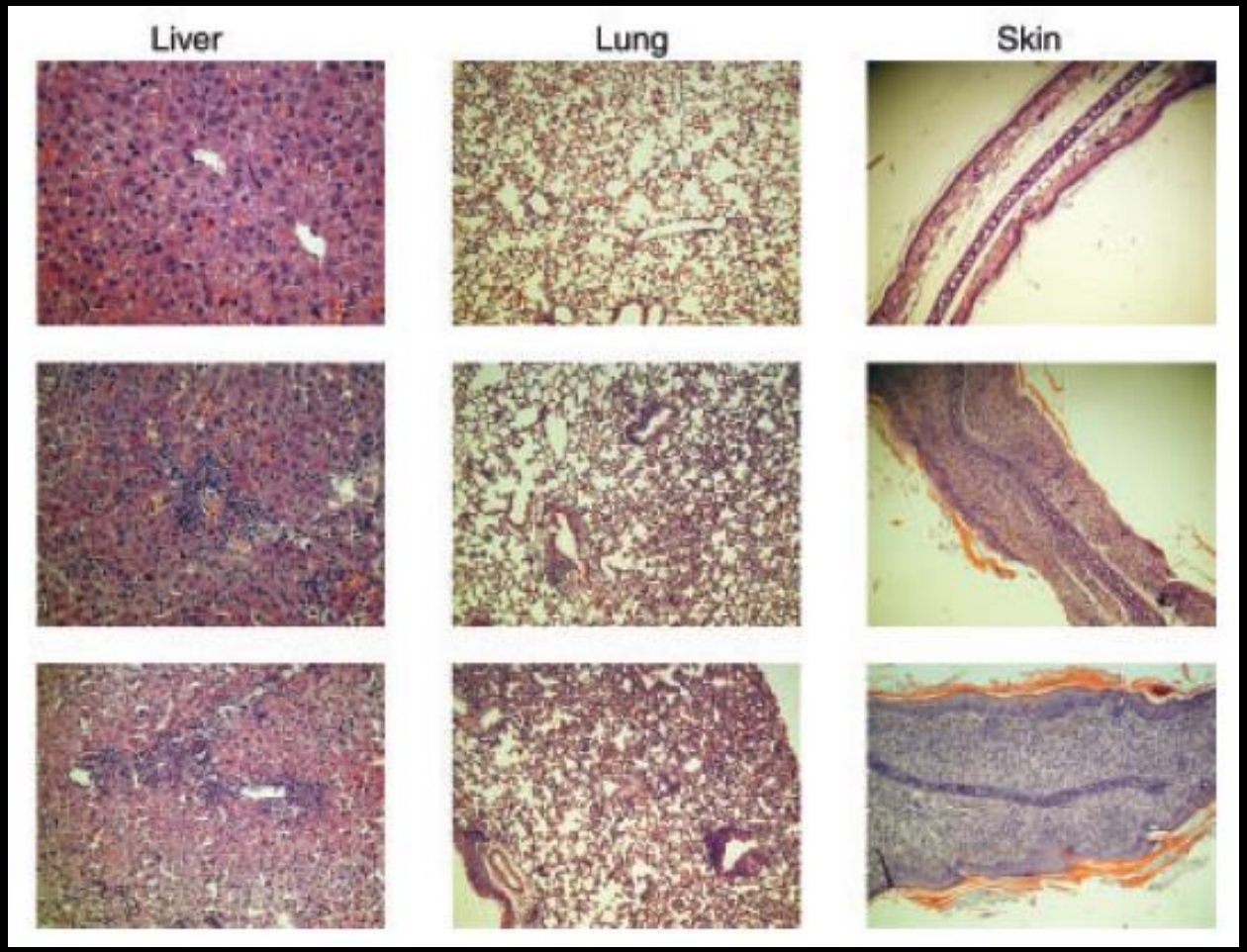
Baço

LN

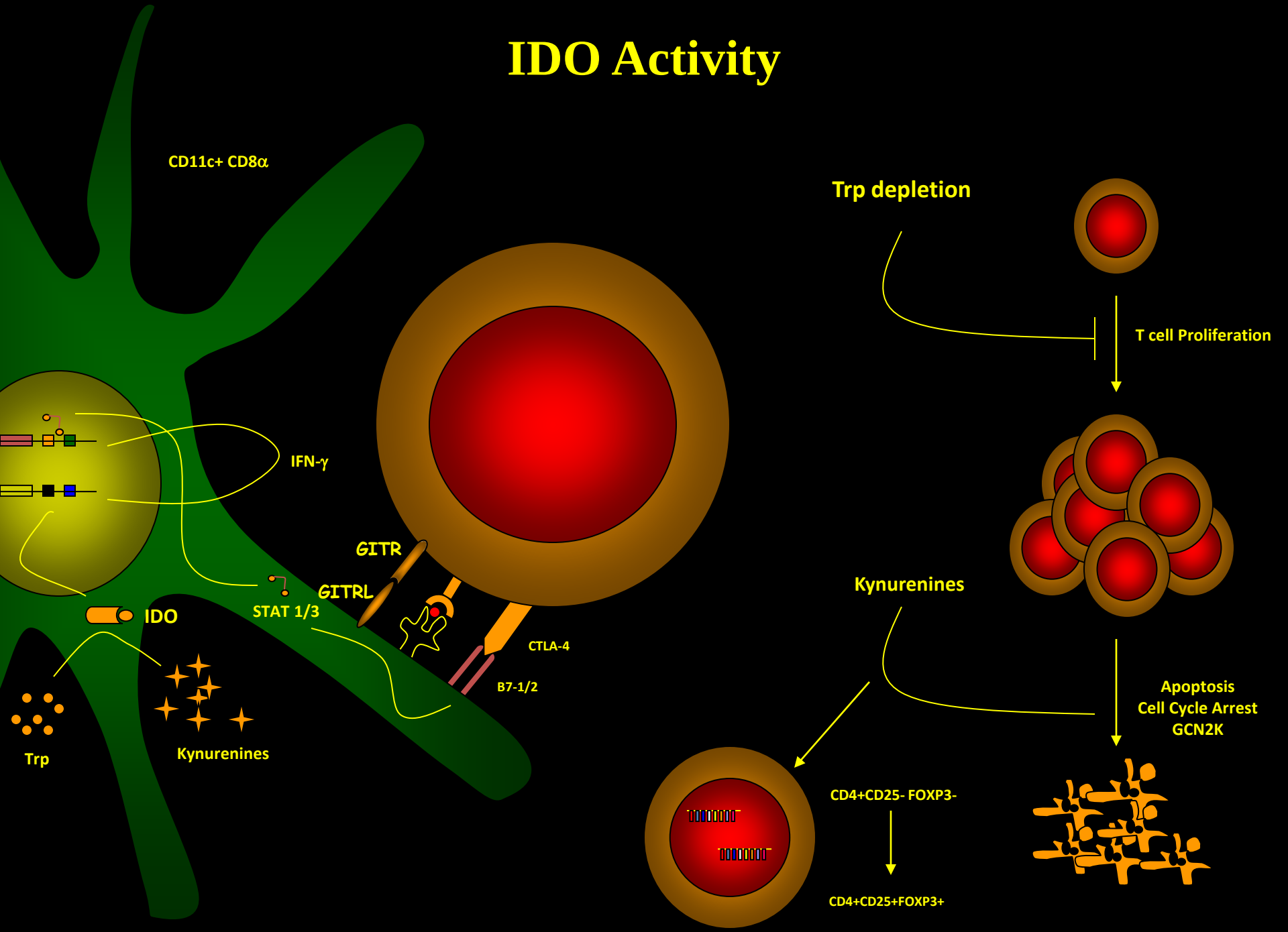




Depleção de Tregs leva a infiltrado inflamatório expontâneo



IDO Activity



Impedir a Quebra da Tolerância
Impedir 1º. 2º. E 3º. Sinal

Citocinas