Immune regulation and tolerance

Abul K. Abbas
UCSF

FOCiS

Lecture outline

- Self-tolerance; central and peripheral tolerance
- Inhibitory receptors of T cells
- Regulatory T cells

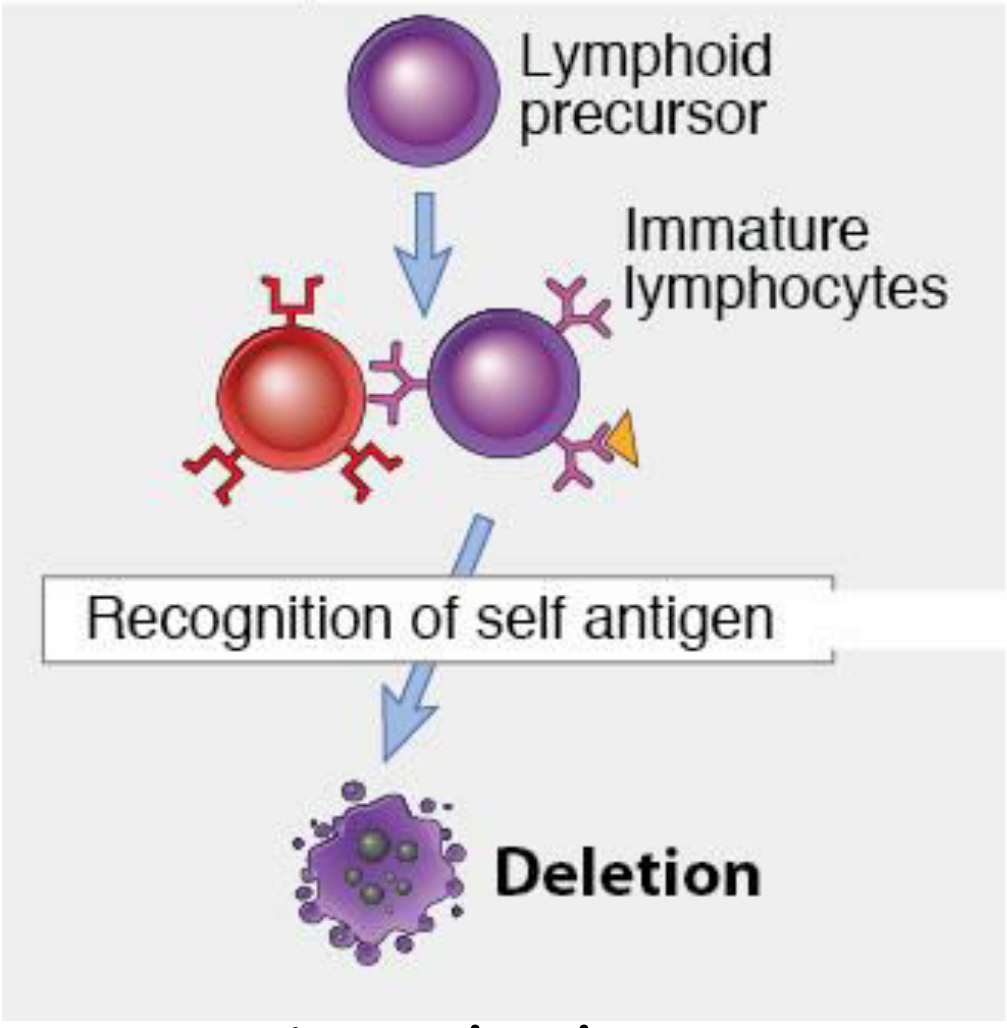
Immunological tolerance

- **Definition:**
  - unresponsiveness to an antigen induced by exposure of lymphocytes to that antigen; antigen-specific (unlike "immunosuppression")

- **Significance:**
  - All individuals are tolerant of their own antigens (self-tolerance); breakdown of self-tolerance results in autoimmunity
  - **Therapeutic potential:** Inducing tolerance may be exploited to treat autoimmune and allergic diseases

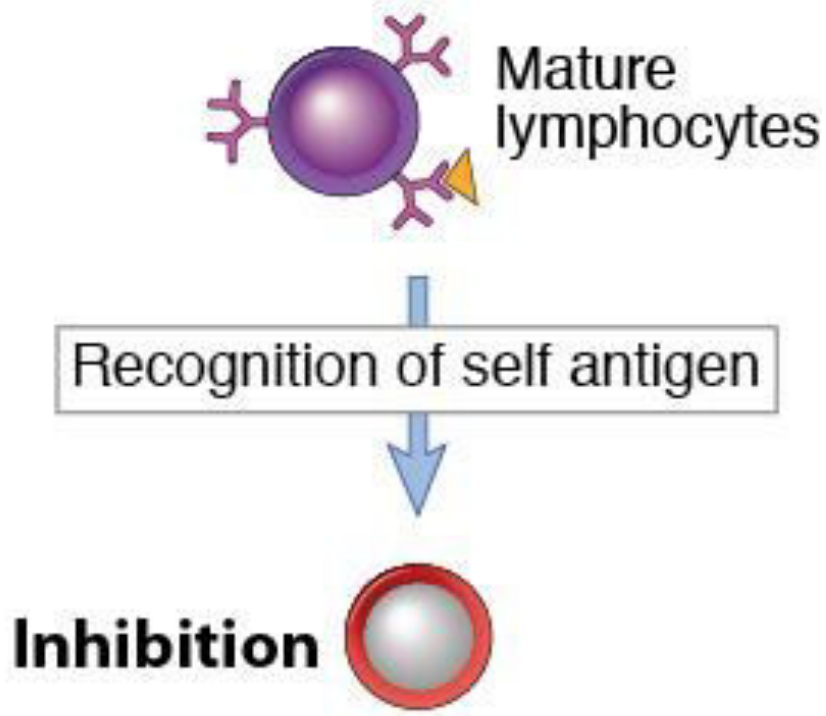
Where and when is self-tolerance induced?

During lymphocyte maturation in thymus and bone marrow



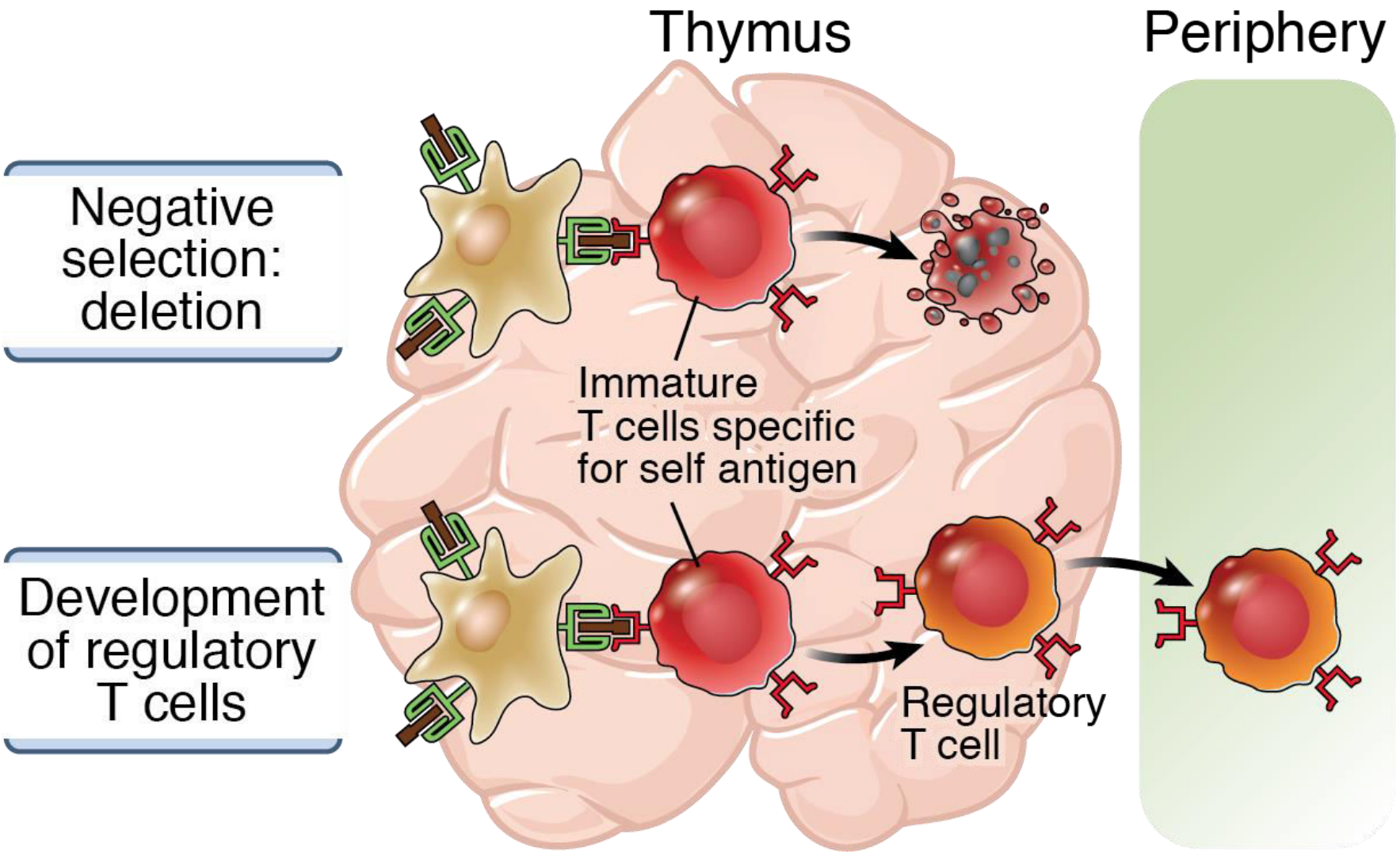
Central tolerance

After lymphocytes have matured, in peripheral tissues



Peripheral tolerance

Consequences of self antigen recognition in thymus



What self antigens are seen in the thymus?

- Ubiquitous cell-associated and circulating proteins

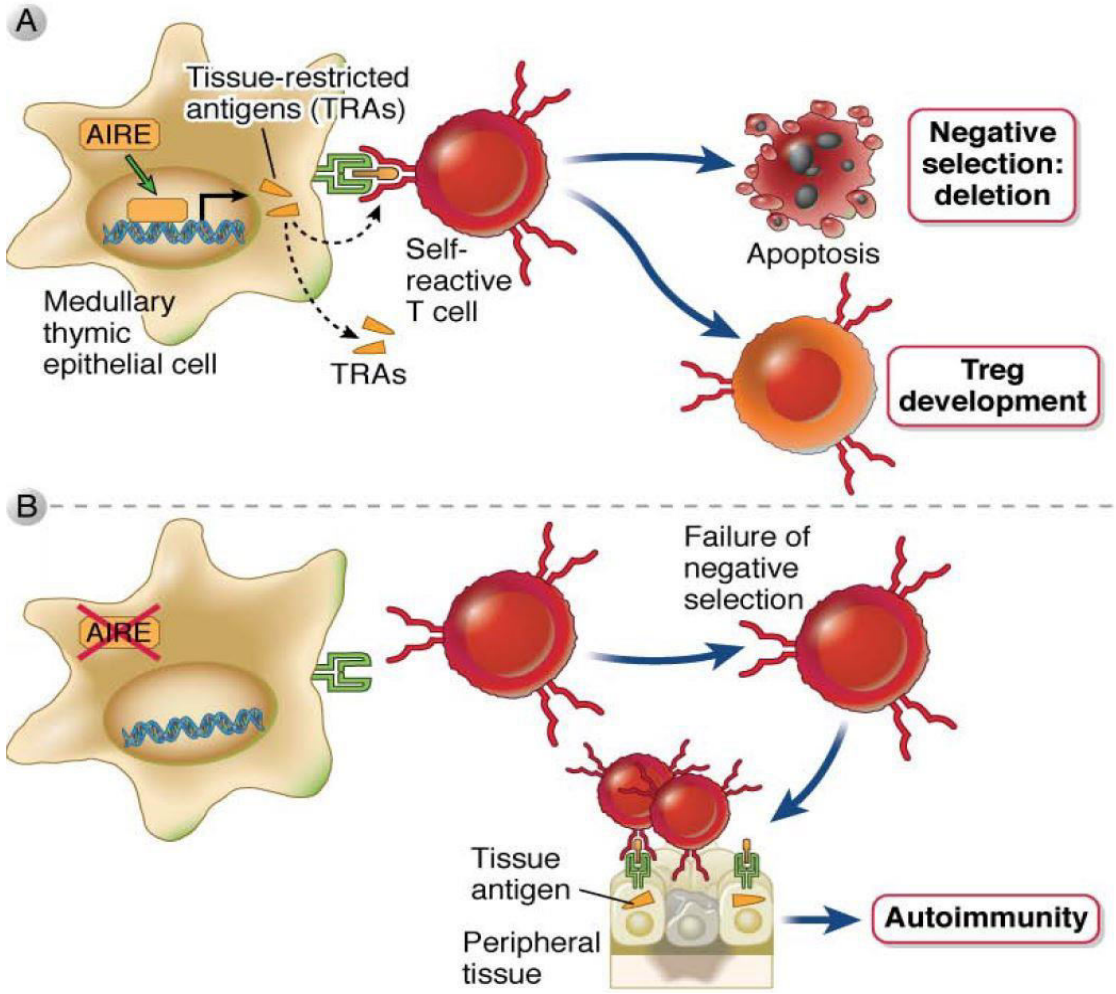
- The thymus has a special mechanism for displaying peripheral tissue antigens in medullary epithelial cells, where the antigens eliminate self-reactive lymphocytes (negative selection)

Consequences of AIRE mutation

- Human disease: autoimmune polyendocrinopathy with candidiasis and ectodermal dysplasia (APECED), also called autoimmune polyendocrine syndrome (APS-1)
  - Associated gene identified by positional cloning, named *AIRE* ("autoimmune regulator")

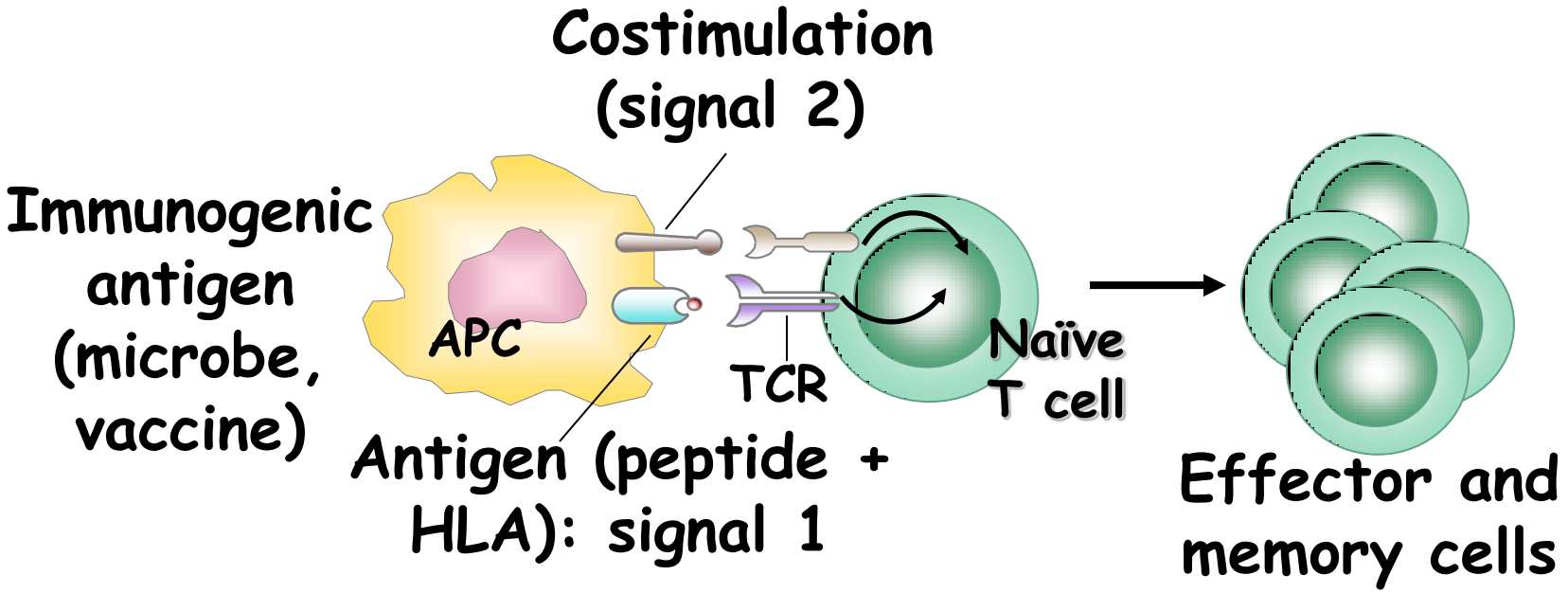
- Mouse knockout: autoantibodies against multiple endocrine organs, retina
  - Failure to express many self antigens in the thymus (revealed by transcriptome analysis of normal vs AIRE-/- thymic epithelial cells)

Deletion of self-reactive T cells in the thymus: how are self antigens expressed in the thymus?

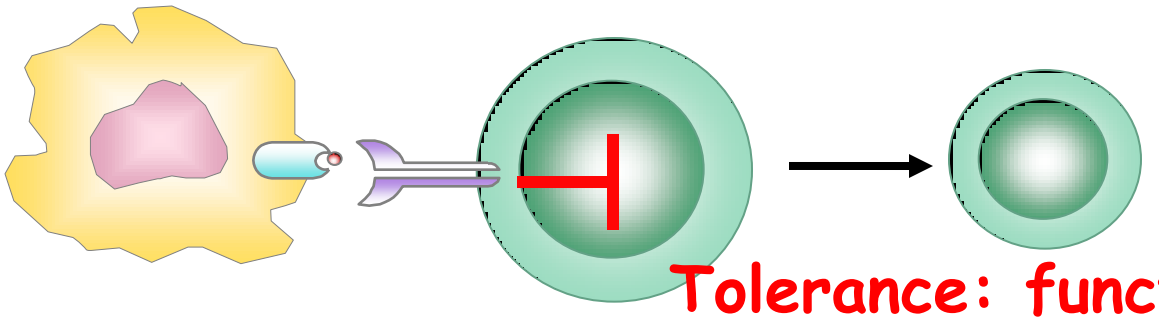


AIRE (autoimmune regulator) is a transcription factor that stimulates thymic expression of many self antigens which are largely restricted to peripheral tissues

Peripheral tolerance



Tolerogenic antigen (e.g. self)



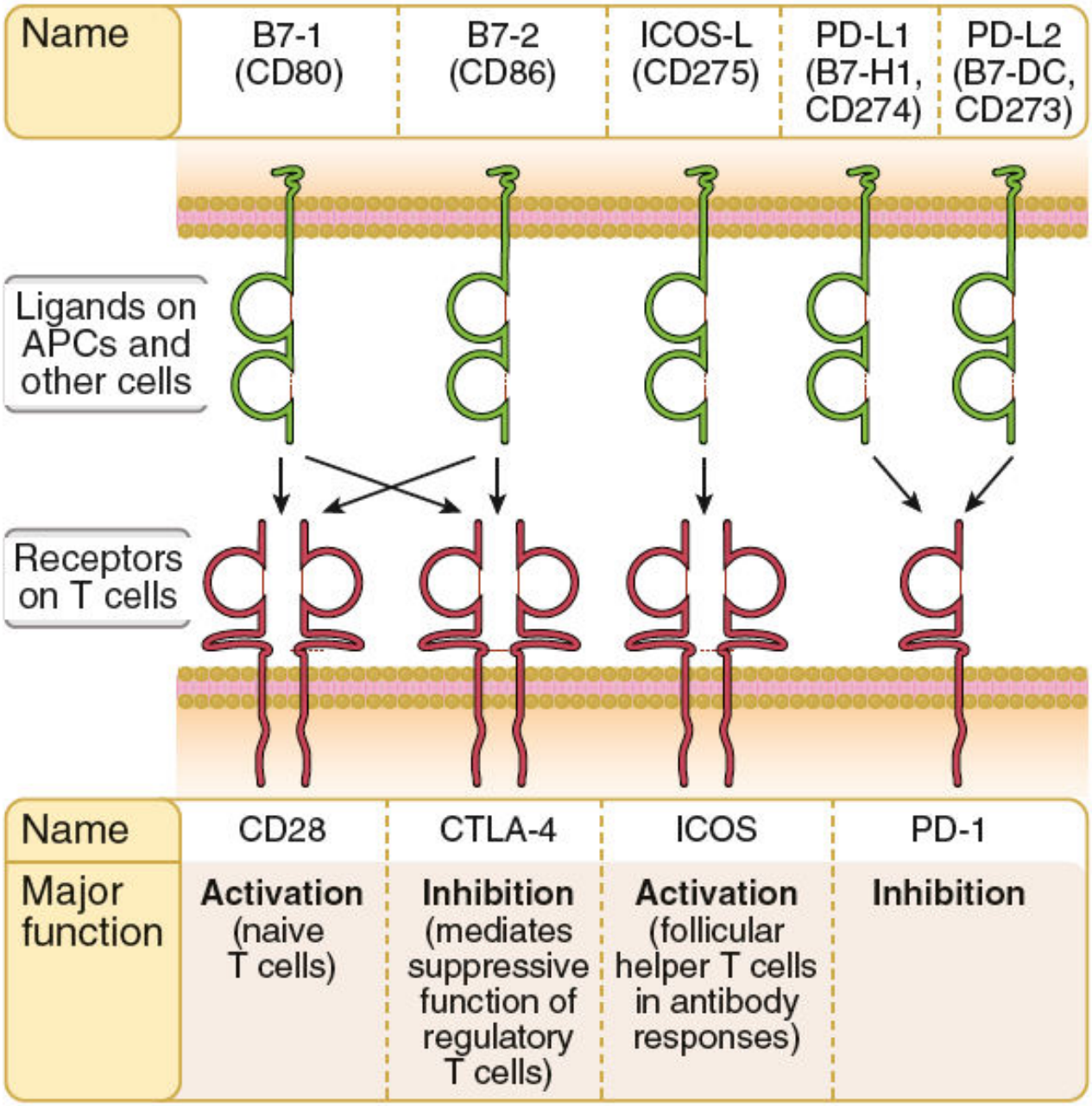
Tolerance: functional inactivation or cell death, or sensitive to suppression

Inhibitory receptors of T cells

- Prevent reactions against self antigens (their physiologic function)

- Suppress immune responses to some tumors, chronic infections (HCV, HIV)
  - Therapeutic application: checkpoint blockade for cancer immunotherapy

The B7:CD28 families



Major functions of selected B7-CD28 family members

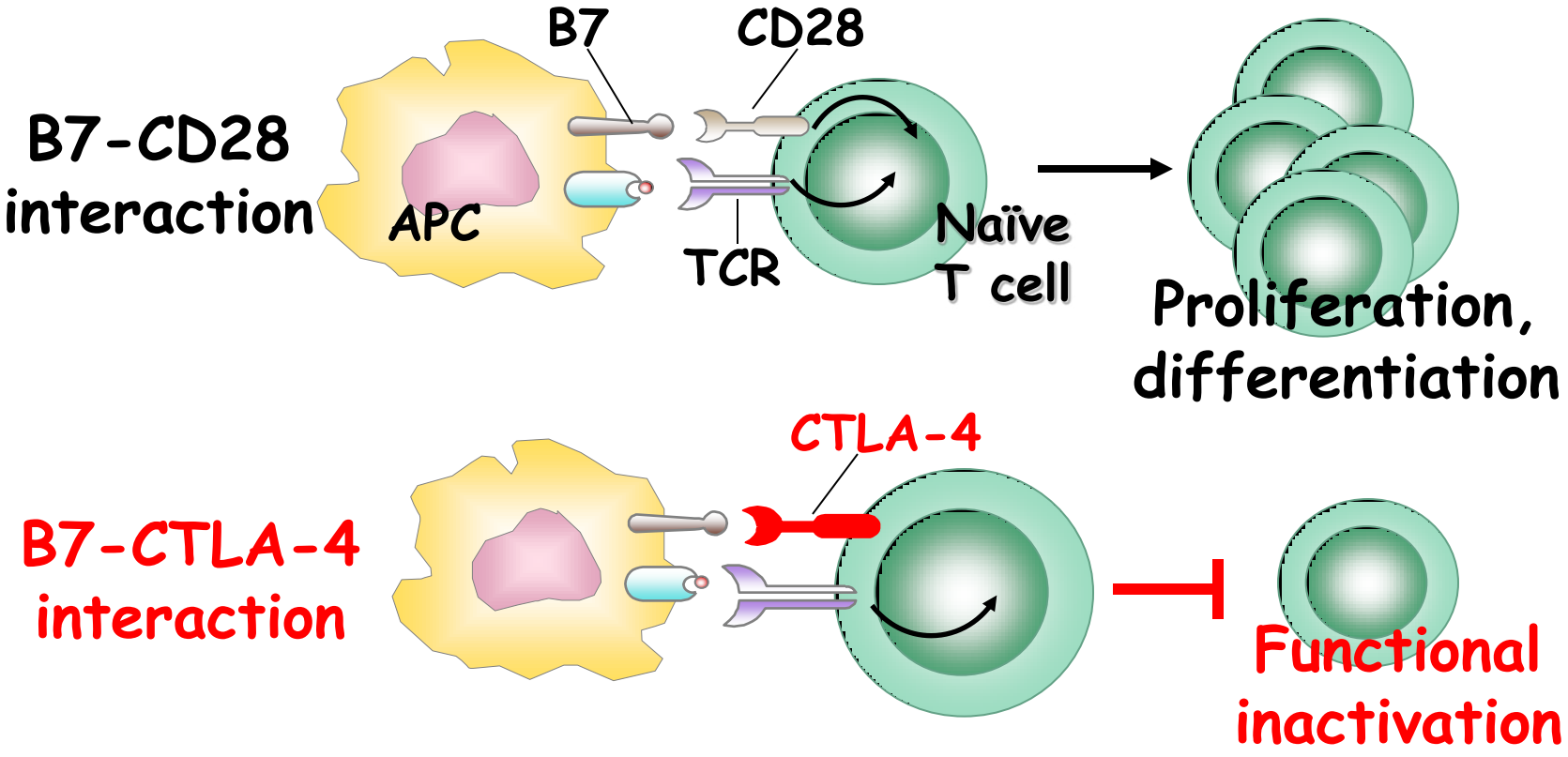
Activation

- CD28-B7: initiation of immune responses
- ICOS-ICOS-L: T cell help in germinal center reactions (antibody responses)

Inhibition

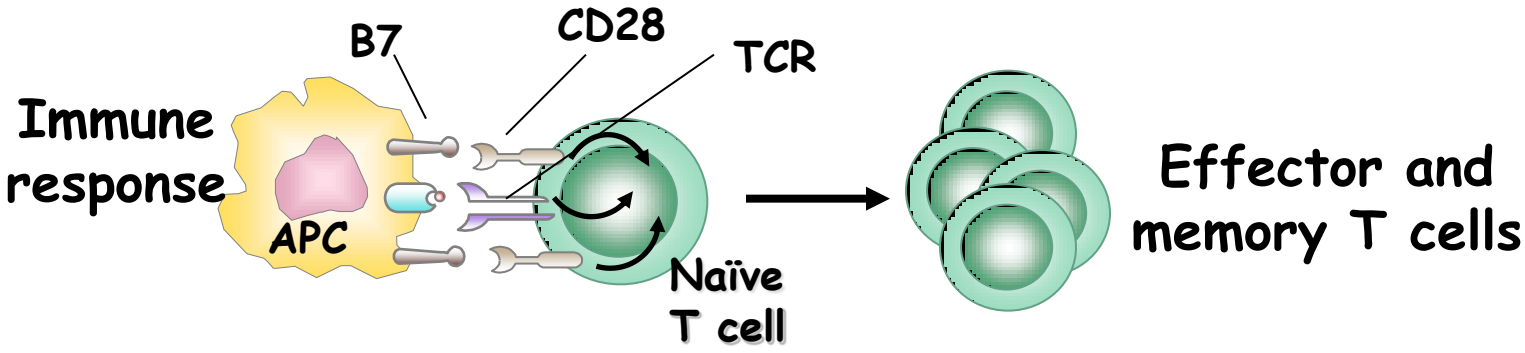
- CTLA-4-B7: inhibits early T cell responses in lymphoid organs
- PD-1:PD-L1,2: inhibits effector T cell responses in peripheral tissues

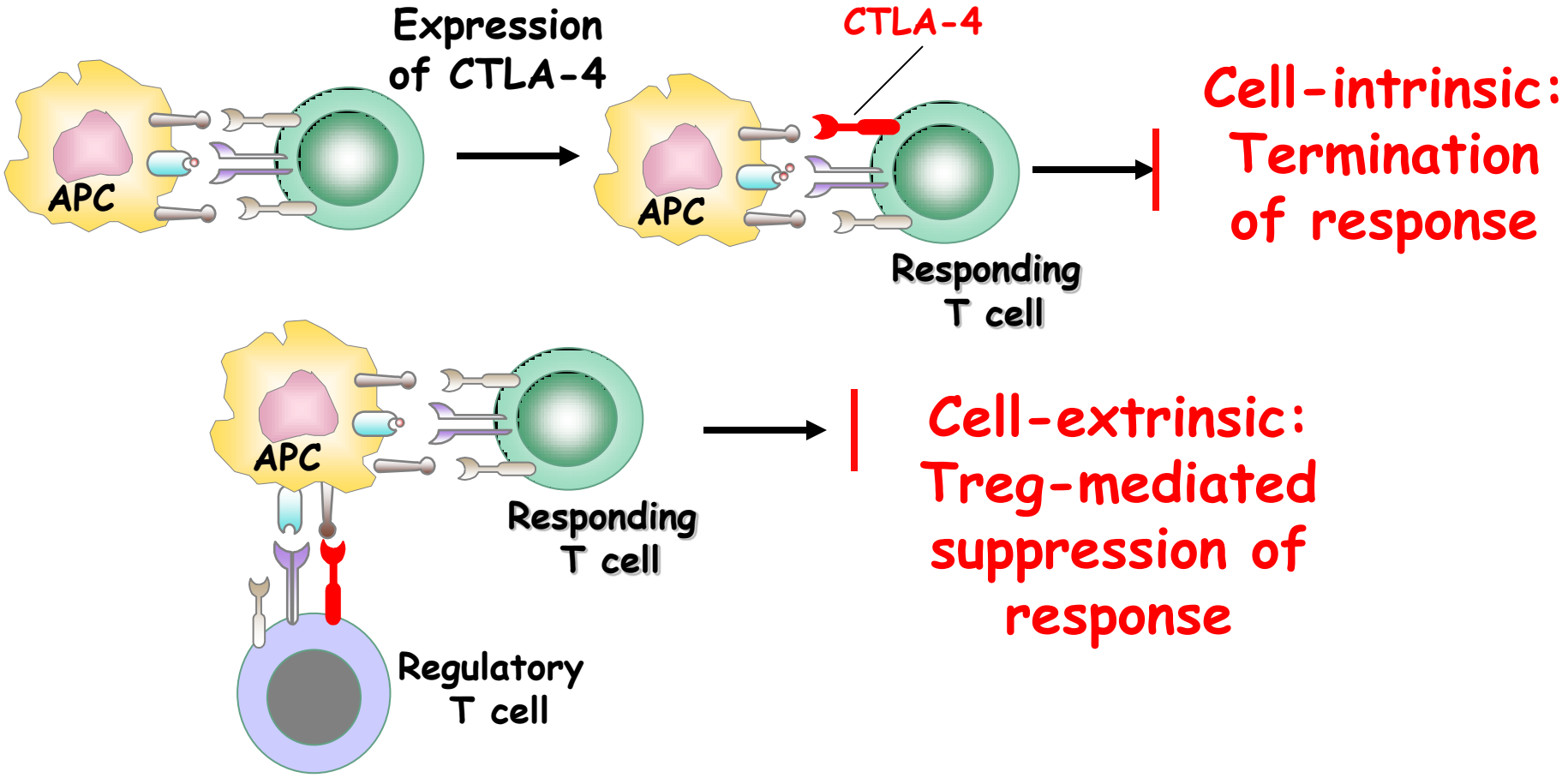
The opposing functions of CD28 and CTLA-4



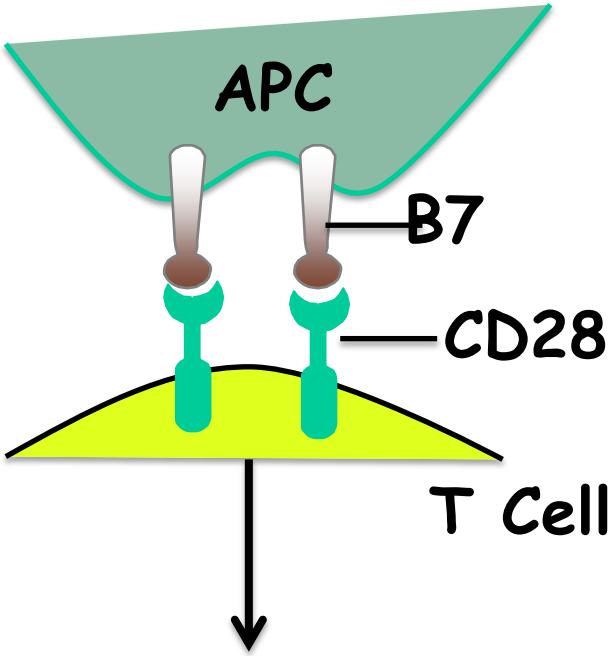
Knockout of CTLA-4 in mice and mutation in humans results in immune dysregulation (lymphoproliferation, multi-organ inflammation)

Actions of CTLA-4



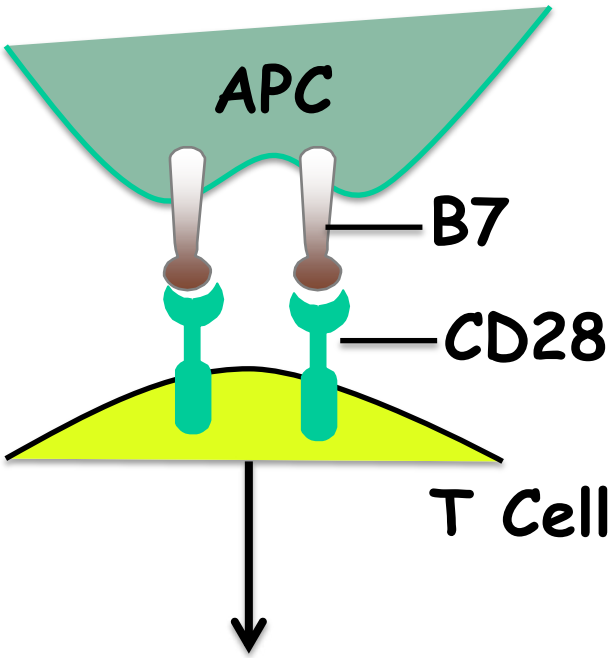


CTLA-4 competitively inhibits B7-CD28 engagement

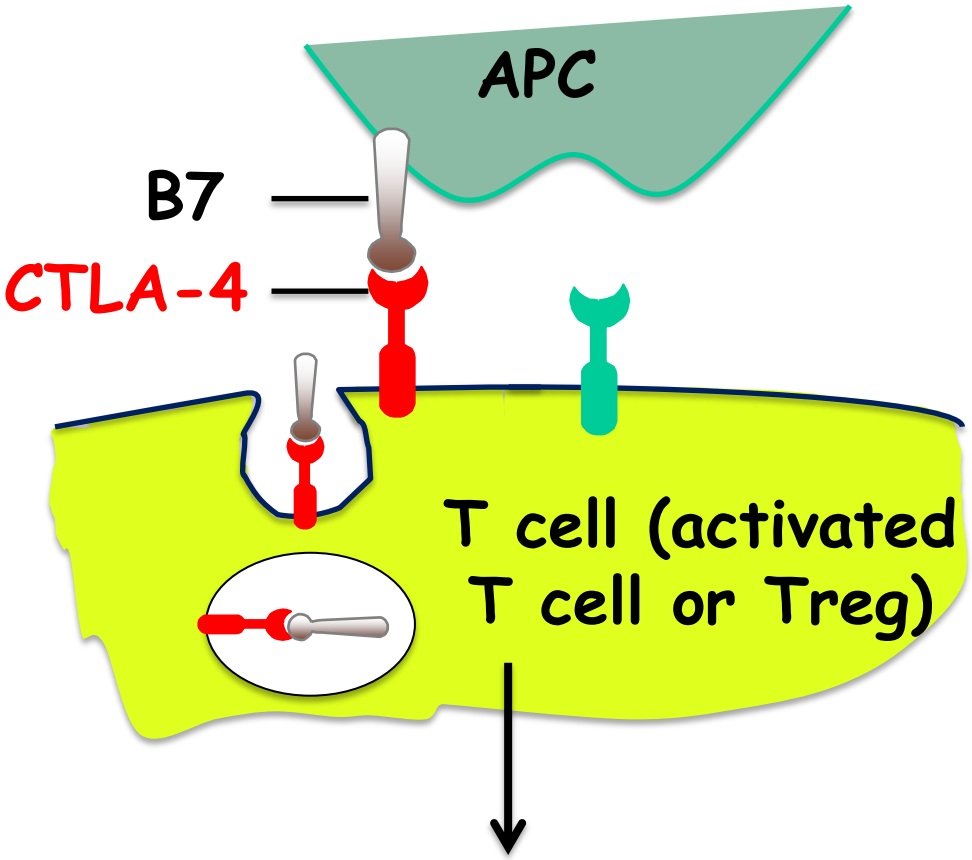


Costimulation →
T cell activation

CTLA-4 competitively inhibits B7-CD28 engagement



Costimulation →
T cell activation



CTLA-4 blocks and removes
B7 → lack of costimulation
→ T cell unresponsiveness

Functions of CTLA-4

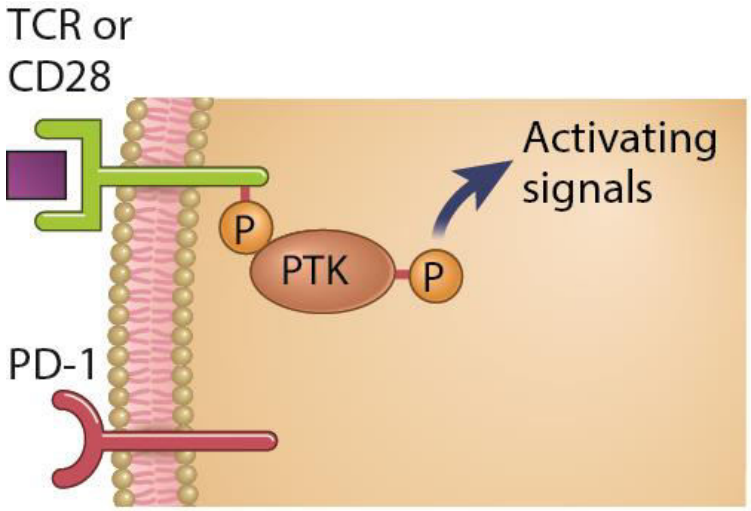
- Limits activation of responding T cells
- Mediates suppressive function of regulatory T cells (Tregs)
- How does the T cell choose to use CD28 to be activated (e.g. with microbes) or CTLA-4 to shut down (e.g. with self Ag)?
  - Level of B7 expression and affinity of receptors: Low B7 (e.g. when DC is displaying self or tumor antigen) --> engagement of high-affinity CTLA-4
  - High B7 (e.g. after microbe encounter) --> engagement of lower affinity CD28

The PD-1 inhibitory pathway

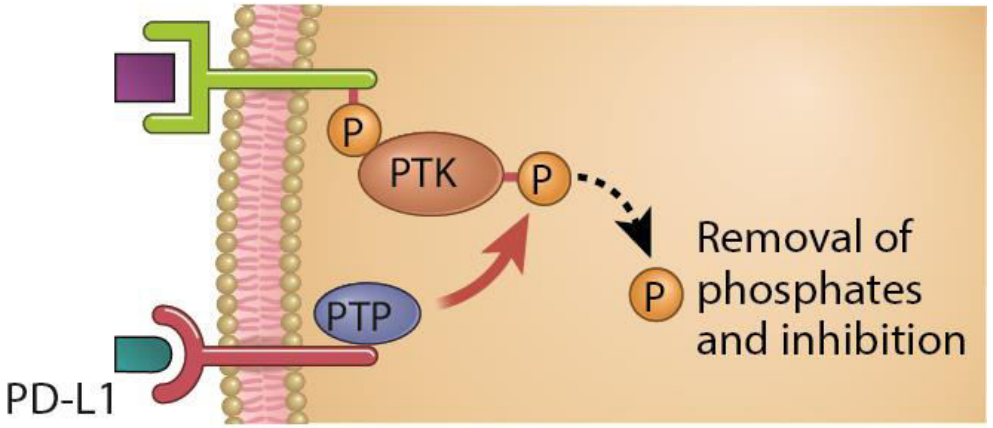
- PD-1 recognizes two widely expressed ligands (PD-L1, PD-L2)
- Knockout of PD-1 leads to autoimmune disease (less severe than CTLA-4-KO)
- Role of PD-1 in T cell suppression in chronic infections, tumors?

Action of PD-1

Normal response



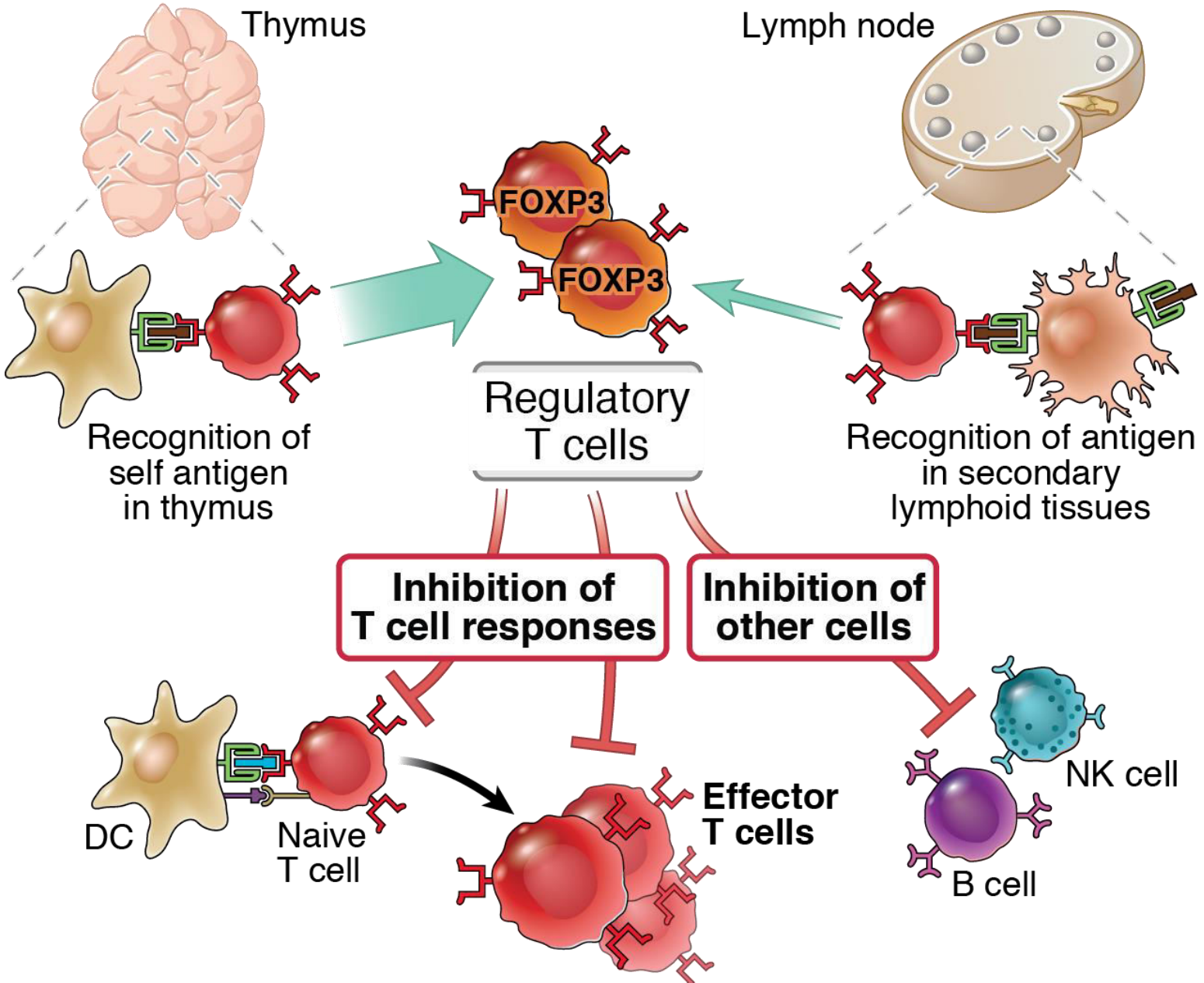
PD-1 engagement



Functions of CTLA-4 and PD-1

	CTLA-4	PD-1
Major site of action	Lymphoid organs	Peripheral tissues
Stage of immune response suppressed	Induction	Effector phase
Main signals inhibited	CD28 costimulation (by reducing B7)	Chronic antigen receptor stimulation
Cell type suppressed	CD4+ > CD8+	CD8+ > CD4+
Inflammatory reactions following antibody treatment	More severe	Milder

Regulatory T cells



Properties of regulatory T cells

- **Phenotype:** CD4+, high IL-2 receptor (CD25), Foxp3 transcription factor; other markers

- **Essential features** of stable Tregs:
  - Foxp3 expression: requires demethylated non-coding CNS2 sequence in promoter
  - CD25 (IL-2Rα) expression: IL-2 is a necessary survival factor
  - CTLA-4 expression: required for suppressive function of most Tregs
  - (Inability to produce IL-2)

The significance of Foxp3+ Tregs

- Genetic evidence: Foxp3 mutations --> autoimmune disease (IPEX); in mice, disease can be corrected by providing normal Foxp3+ cells

- Do defects in Foxp3+ Tregs or resistance to Treg-mediated suppression contribute to common autoimmune diseases?
  - Inconsistent and variable data

Mechanisms of action of Foxp3+ Tregs

- CTLA-4 on Tregs removes B7 on APCs, reduces CD28 engagement and T cell activation
  - Genetic deletion of CTLA-4 in Foxp3+ cells results in severe systemic autoimmunity and lymphoproliferation
  - Commonly used assay for Treg function in humans bypasses the need for B7 and cannot measure this activity

Mechanisms of action of Foxp3+ Tregs

- CTLA-4 on Tregs removes B7 on APCs, reduces CD28 engagement and T cell activation

- **Inhibitory cytokines produced by Tregs (IL-10, others?) suppress immune responses (DCs, Macs, T cells)**
  - **IL-10 is especially important for regulating mucosal immune responses (deletion of IL10 in Foxp3+ cells results in colitis)**

- **Consumption of IL-2**
- **Many others reported**

Role of Tregs in fetal tolerance

- In evolution, placentation developed at the same time as the ability to generate FoxP3+ peripheral Tregs
- Paternal antigens expressed in the fetus induce long-lived antigen-specific Tregs
- Replacement of fetal antigen-specific Tregs with polyclonal Tregs in mice results in fetal resorption

- Anatomic restriction of immune regulation?
- Role in humans? Are defects in regulatory memory the basis of recurrent fetal loss?

Regulatory T cells

- Explosion of information about the generation, properties, functions and significance of these cells
- Will cellular therapy with ex vivo expanded Treg become a reality?
- Therapeutic goal: induction or activation of Treg in immune diseases

The therapeutic potential of regulatory T lymphocytes

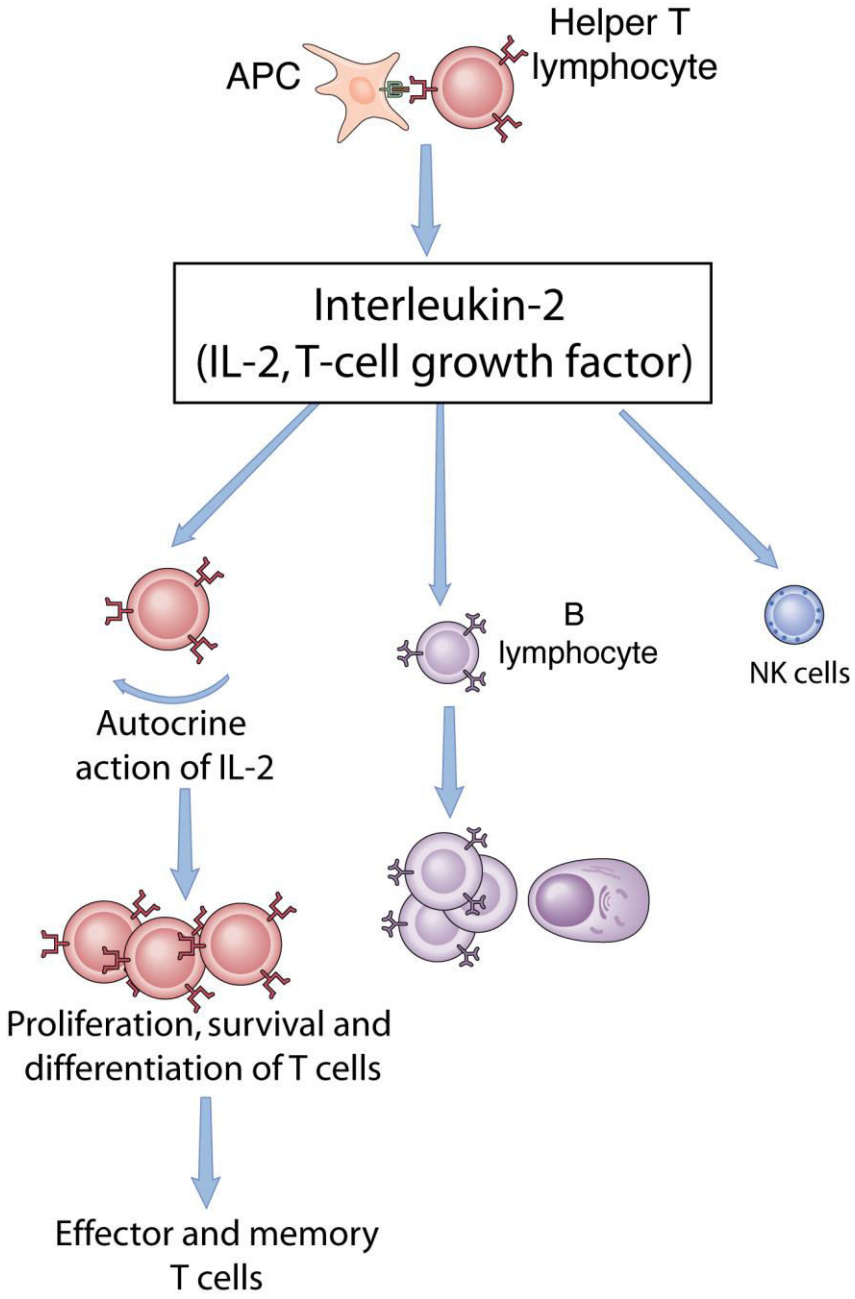
- Cell transfer of autologous Tregs to suppress immune responses
  - Grow up patient's Tregs ex vivo
  - Ongoing clinical trials in graft rejection, T1D show it is safe
  - Very little efficacy data
  - Technically difficult, individualized

The therapeutic potential of regulatory T lymphocytes

- Cell transfer of autologous Tregs to suppress immune responses

- **Administer antigen or cytokine in ways that preferentially induce Tregs?**
  - **Antigen without adjuvant?**
  - **IL-2**

Functions of Interleukin-2: the dogma



The unexpected biology of IL-2

- Interleukin-2 is the prototypic T cell growth factor (TCGF), required for initiating clonal expansion of T cells in response to antigen

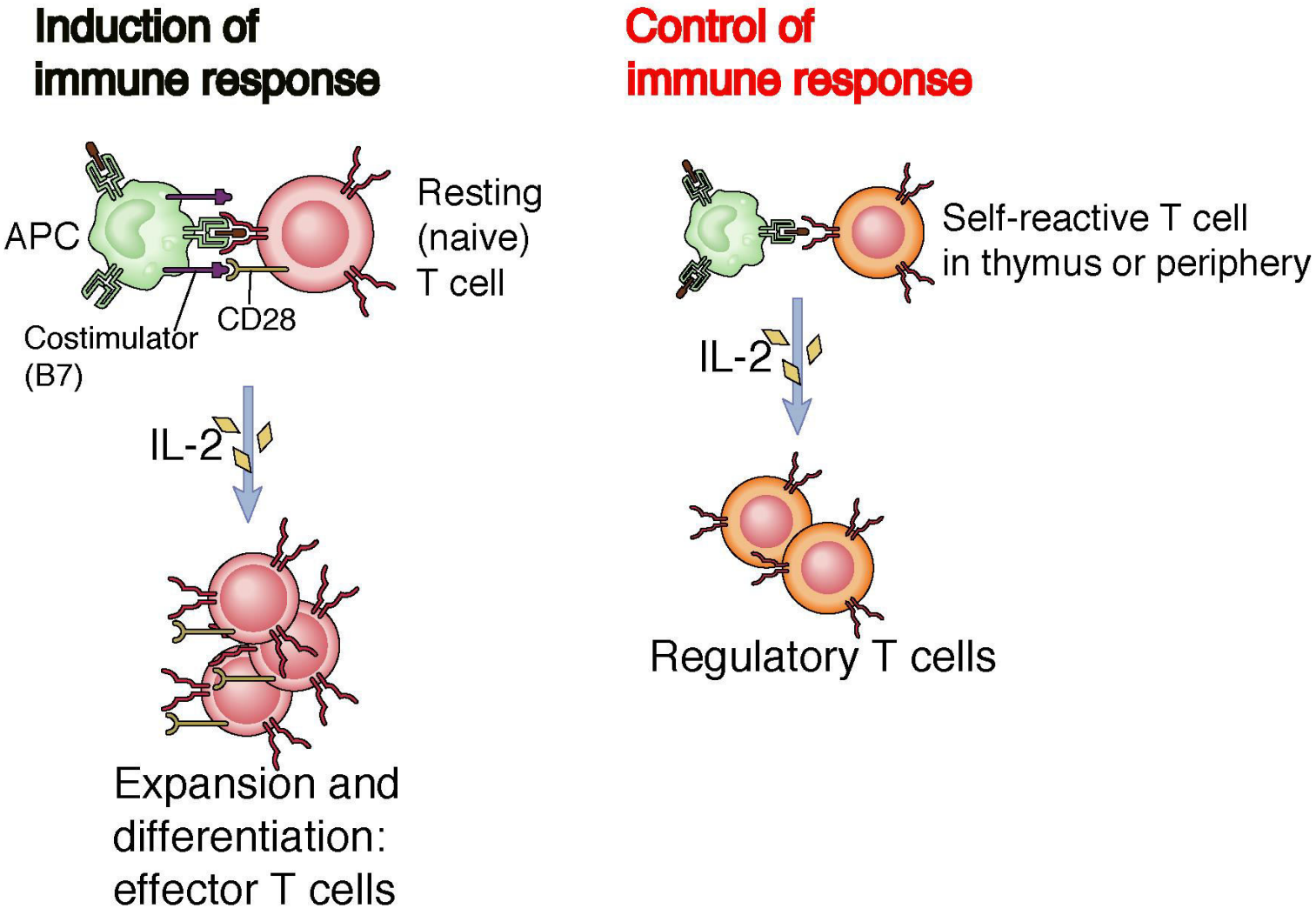
- **Prediction**: what will be the consequence of eliminating IL-2 or the IL-2 receptor?

The unexpected biology of IL-2

- Interleukin-2 is the prototypic T cell growth factor (TCGF), required for initiating clonal expansion of T cells in response to antigen

- BUT: knockout of IL-2 or the α or β chain of the IL-2R results not in immune deficiency but in systemic autoimmunity and lymphoproliferation

Dual roles of IL-2 in T cell responses



Surprising conclusion from knockout mice: the non-redundant function of IL-2 is in <u>controlling</u> immune responses

Therapeutic potential of IL-2: a revision

- IL-2 was originally used to boost immune responses in cancer, HIV infection (enhancing effector and memory T cells)
  - IL-2 treatment can increase number and functional activity if Tregs
- Use of IL-2 to boost Tregs: design IL-2 to bind to high-affinity CD25
  - Low-dose IL-2
  - Mutant IL-2 that binds preferentially to CD25

Regulating immune responses: where are we?

- Elucidating the mechanisms of immune regulation is one of the dominant themes of modern Immunology; obvious relevance to immune-mediated inflammatory diseases, therapeutics, vaccines

- Already leading to new therapeutic strategies

- Continuing challenge is to establish the importance of control mechanisms in the development of inflammatory diseases

Pathogenesis of autoimmunity