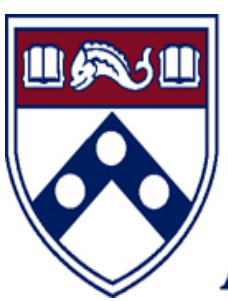
Primer on Tumor Immunology

International Society for Biological Therapy of Cancer

C. H. June, M.D.

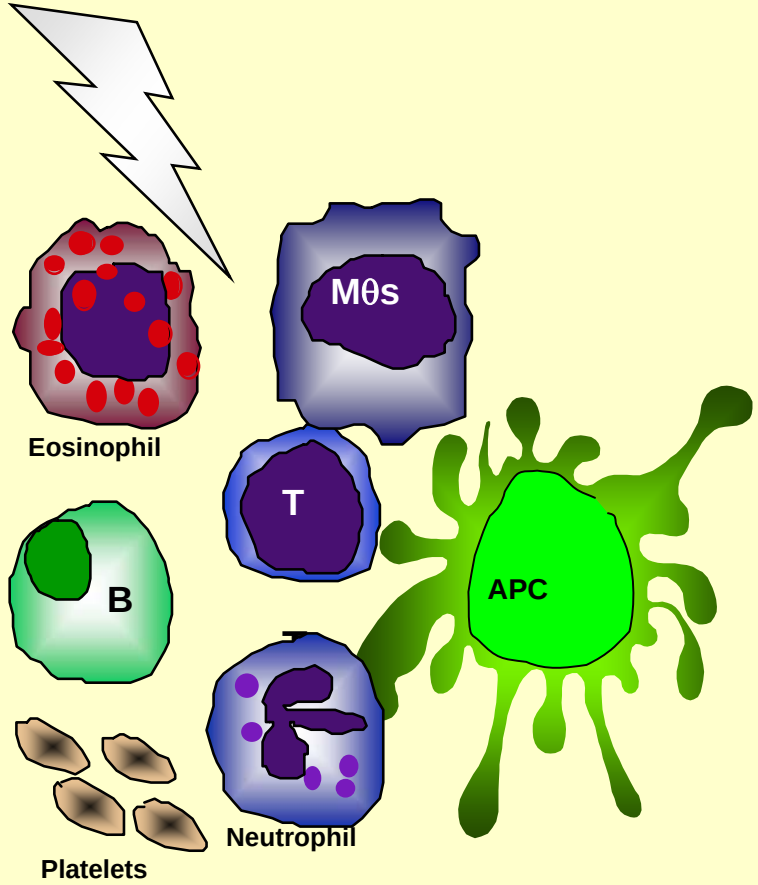
November 10, 2005



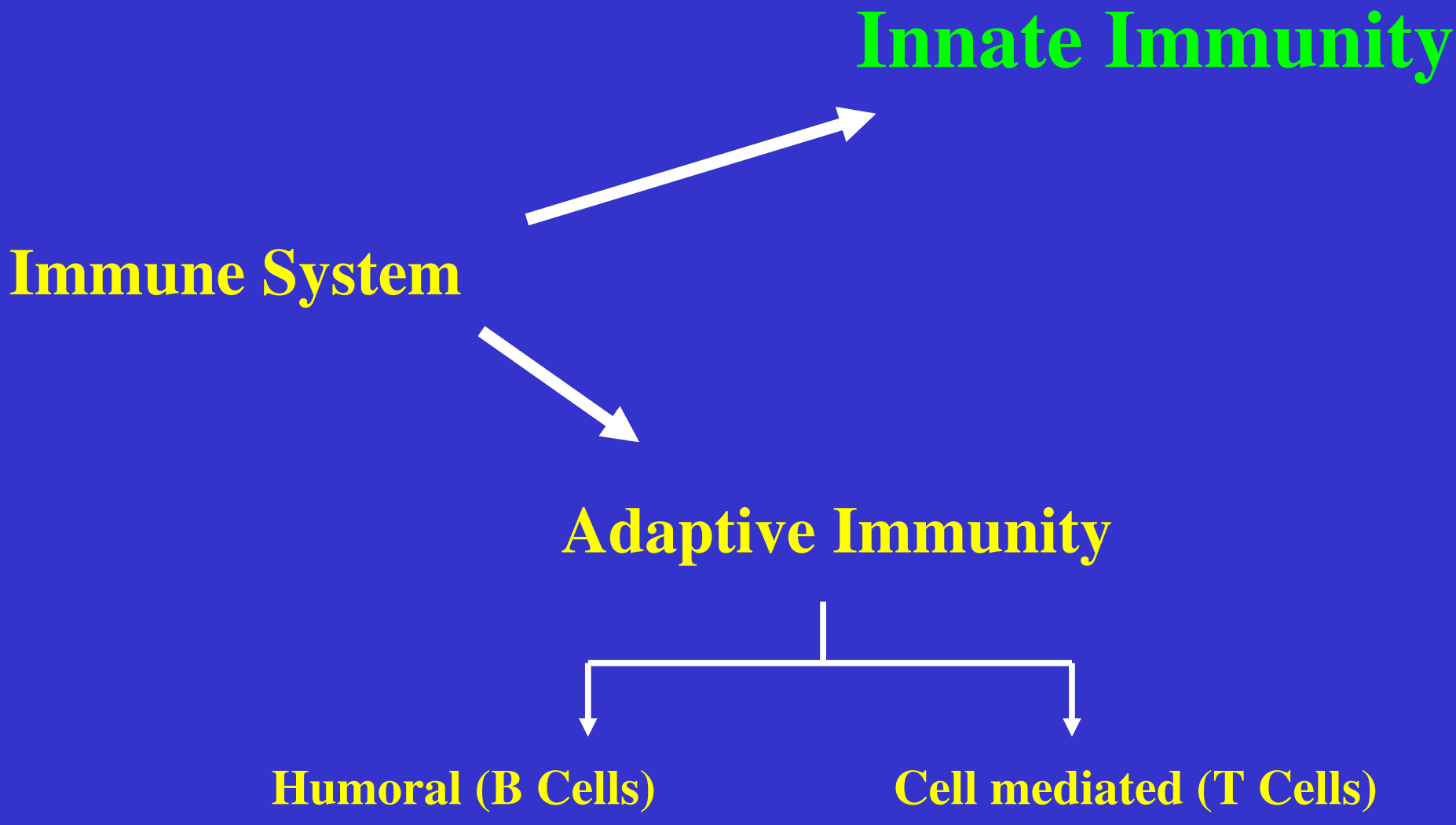
UNIVERSITY OF PENNSYLVANIA
Abramson Cancer Center

Outline: Primer on Tumor Immunology

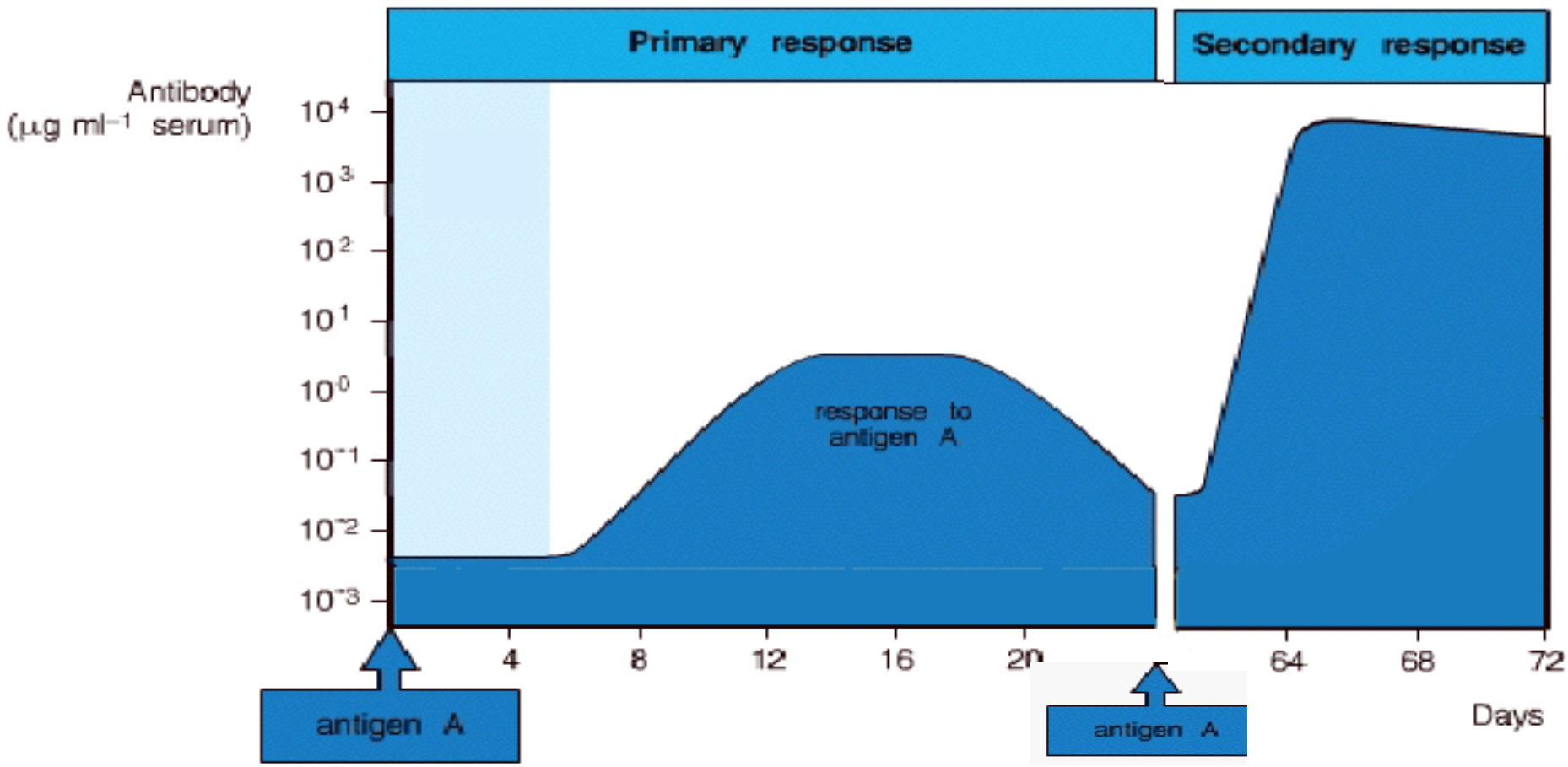
- T Cell Receptors
- T Cell Biology
- Tumor immunology



Organization of Immune System



The Course of Induction of Innate and Adaptive Immunity



Janeway, Immunobiology

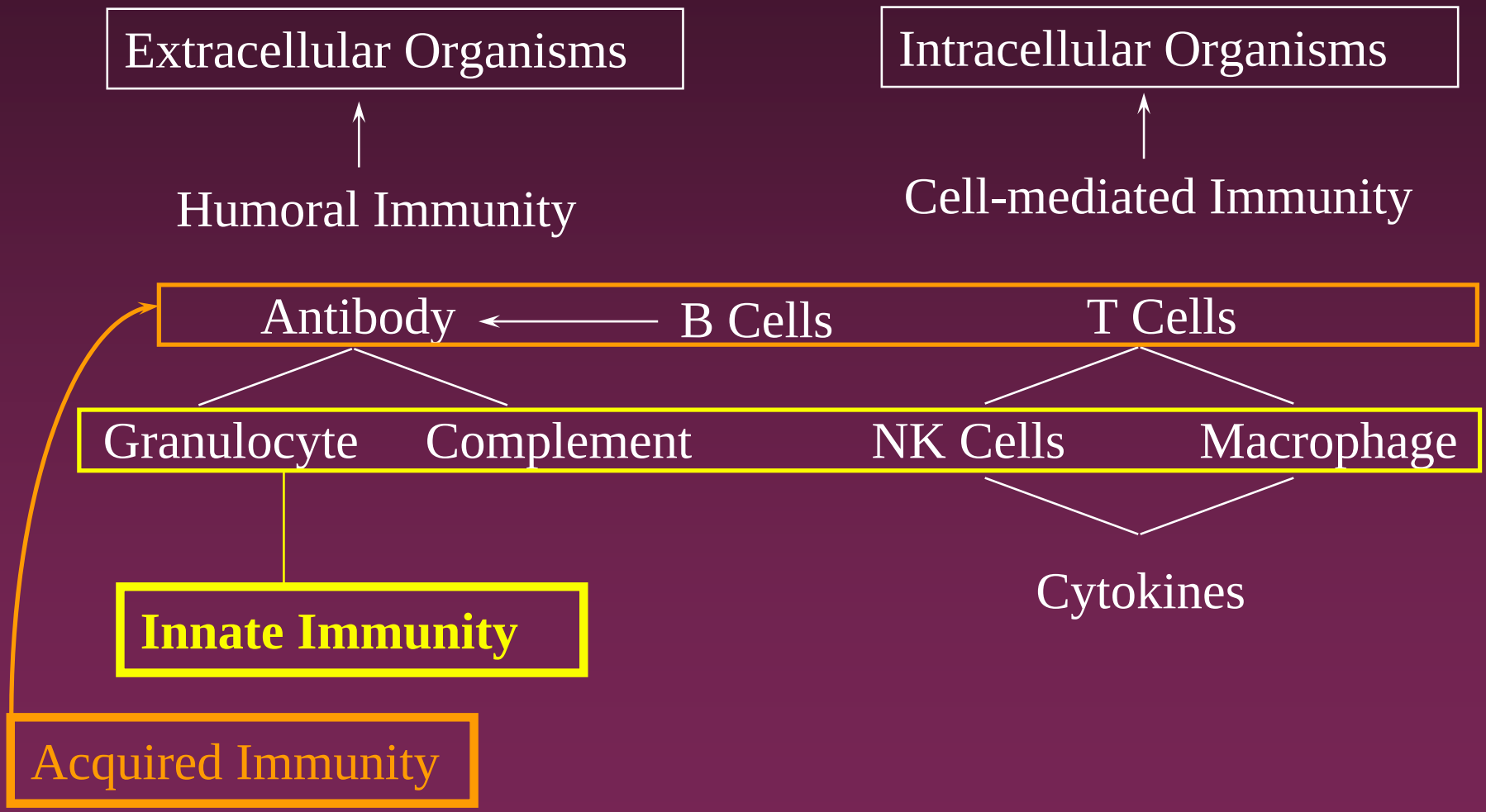
Hallmarks of the Adaptive Immune Response

1. Increasing specificity

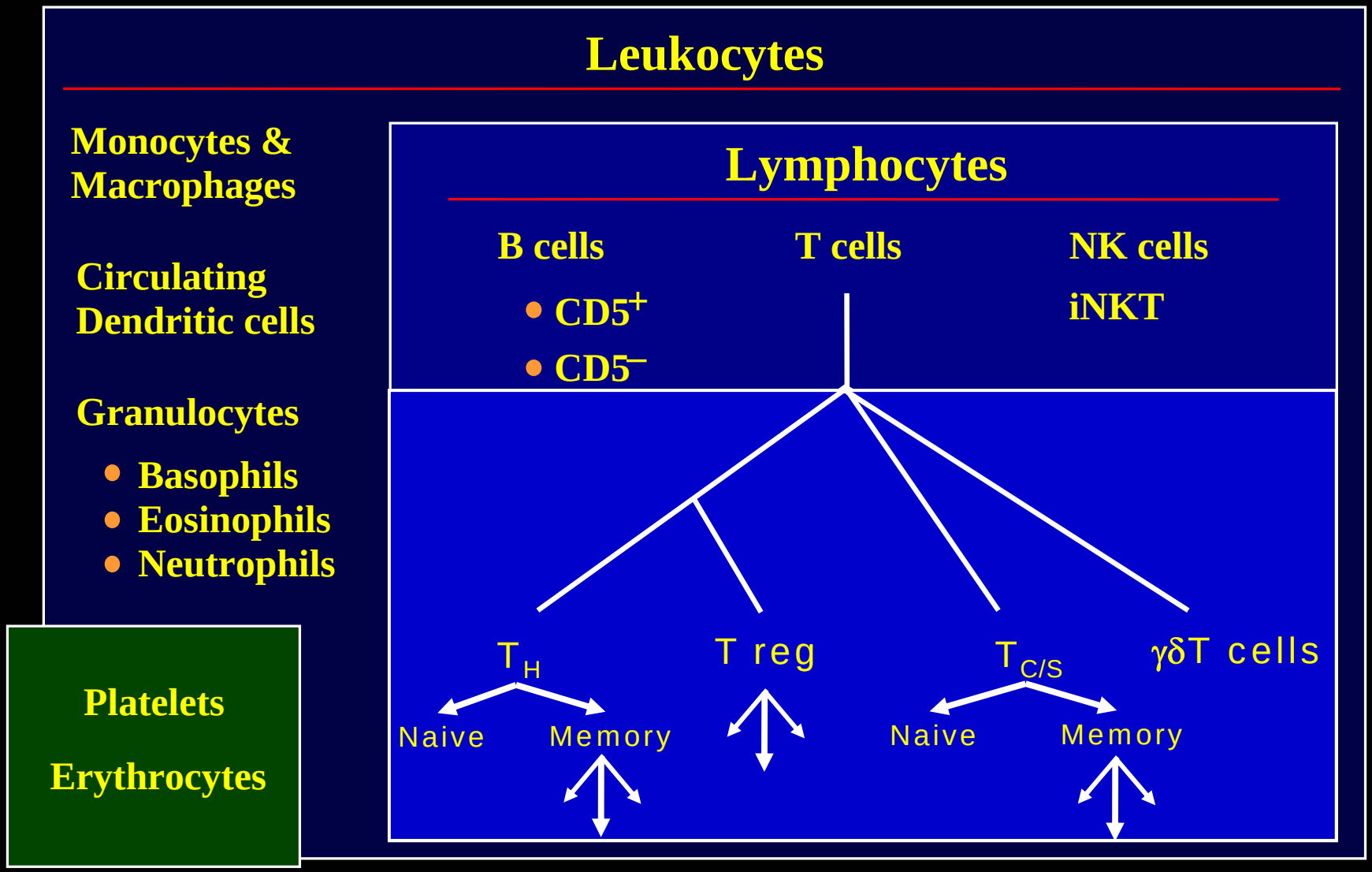
2. Rechallenge: response is more rapid and more robust

3. Memory: Selected T cells persist for years

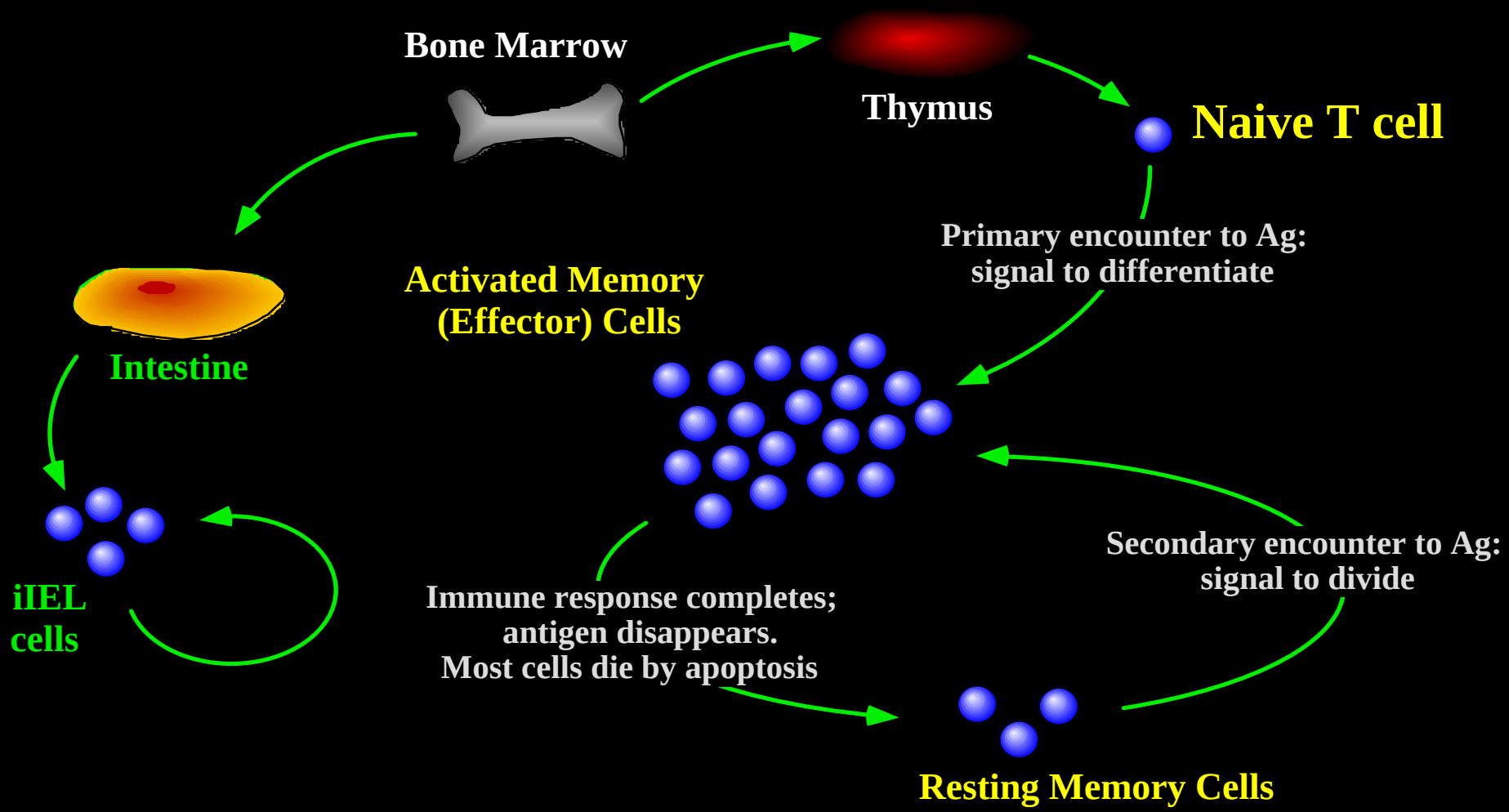
Innate and Acquired Immunity



T Cell Differentiation: Multiple Mature Subsets

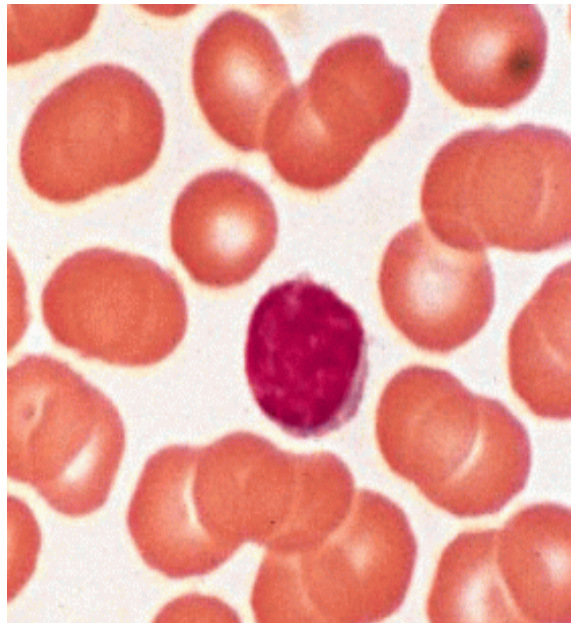


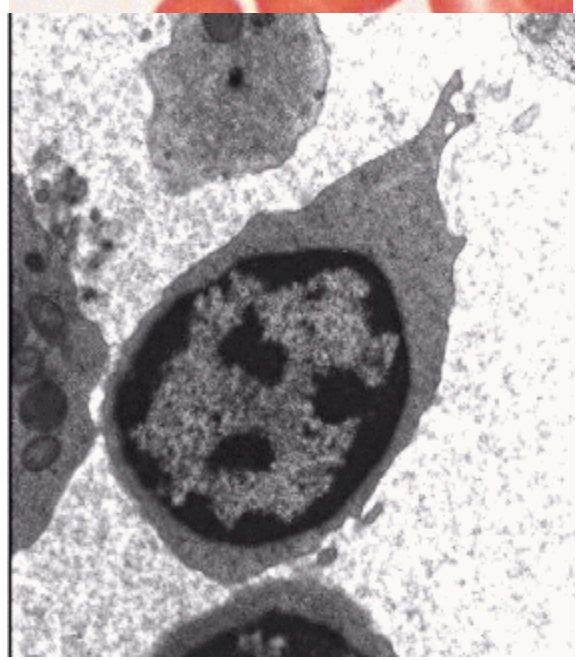
Generation of T cells



T Cell Biology

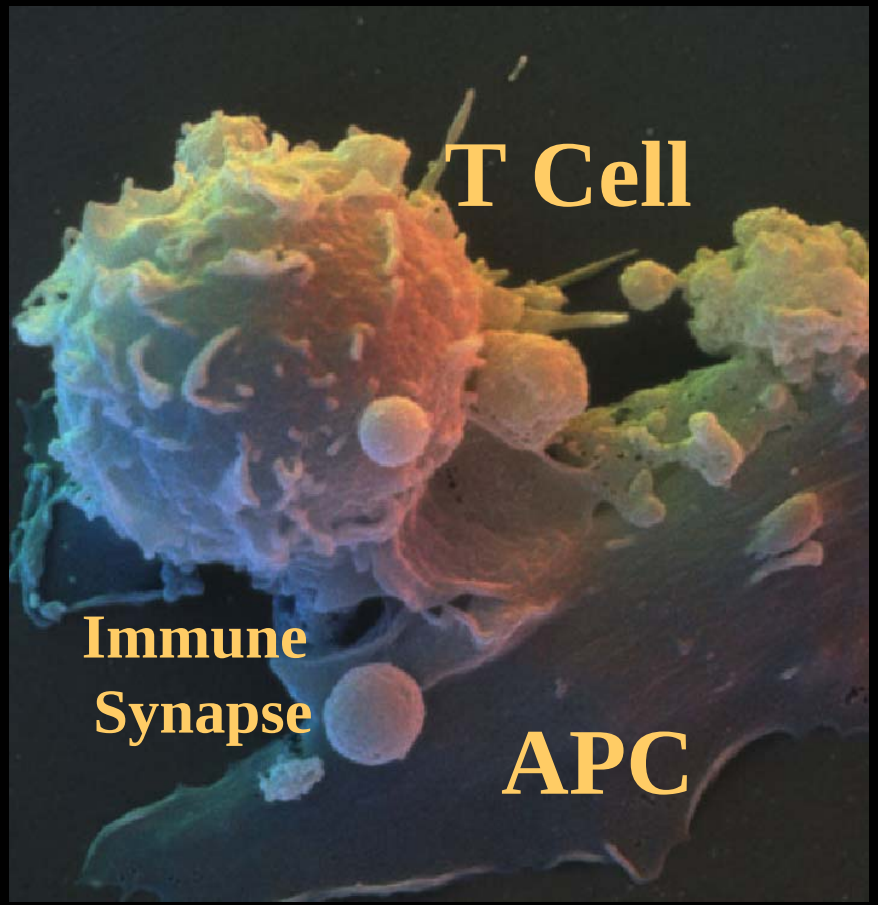
- T cells develop in the thymus
  - TCR rearrangement
- T cells belong to the class of cells that have capacity to enter and exit the cell cycle
- Mature T cells divide in secondary lymph tissue
  - Spleen, lymph nodes
  - Do not divide in peripheral blood





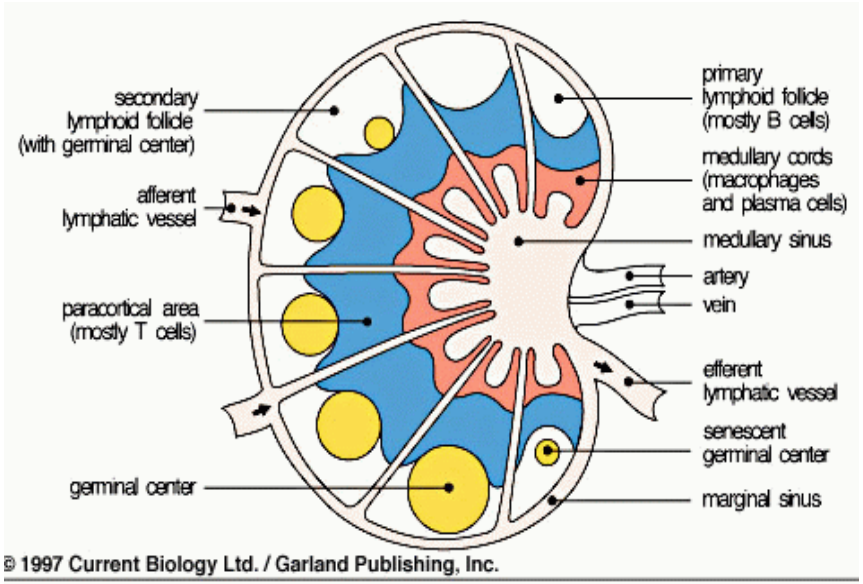
Principles of T Cell Activation

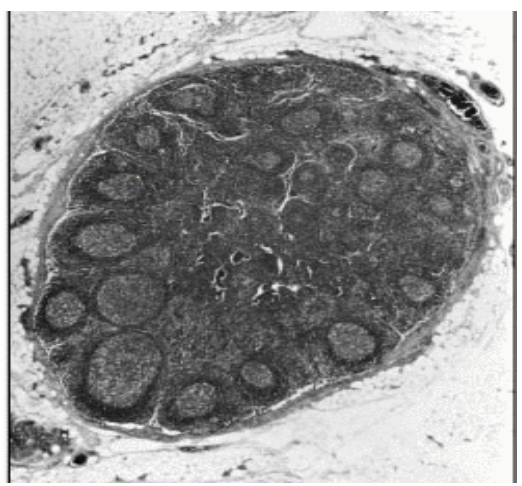
- T cells are activated by antigen presenting cells (APC)
- Any cell that expresses MHCI or MHCII can activate T cells
- Naïve T cells have more stringent requirements: only a DC can do the job



T Cell Activation

- **Immunosurveillance: DC / APC take up Ag in periphery**
- **T cell division and clonal expansion occurs in lymph nodes**
- **Geographic and temporal control**
  - **Naïve T cells reside in lymph nodes**
  - **DC bring antigen to the lymph node**





Janeway, Immunobiology

The Three Laws of Immunology

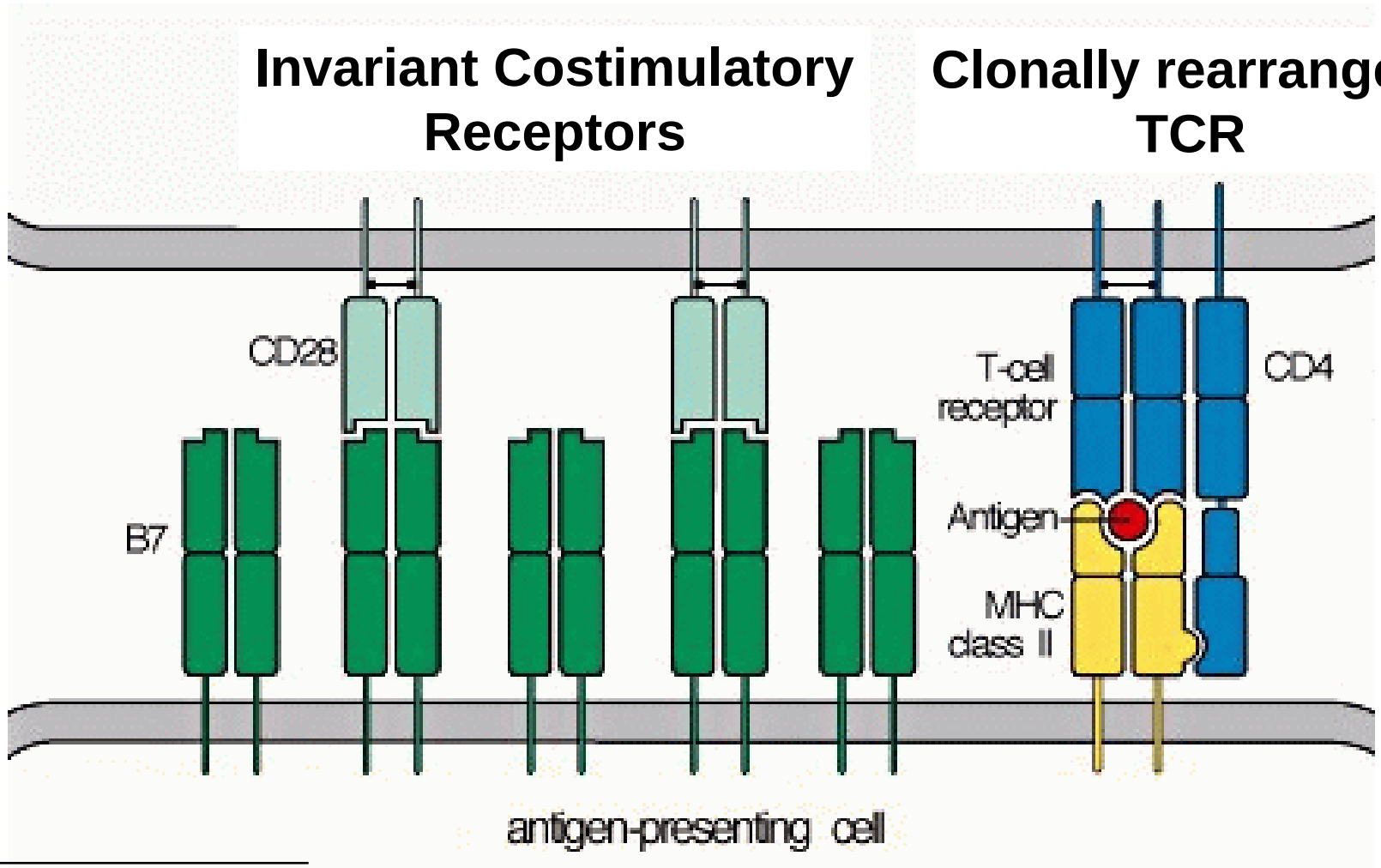
1. The Immune System is Capable of Recognizing a Virtually Unlimited Array of Specific Structures (*Universality*)

2. The Response to Self-Antigens is Eliminated or Controlled (*Tolerance*)

3. The Response is Appropriate to the Inducing Pathogen (*Appropriateness*)

Adapted from W. E. Paul, M.D., Editor, Fundamental Immunology

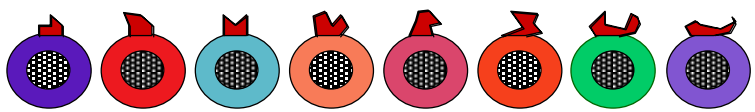
T Cell Activation



Janeway, Immunobiology

The First Law - *Universality*

Lymphocytes are the specific cells of the immune system.

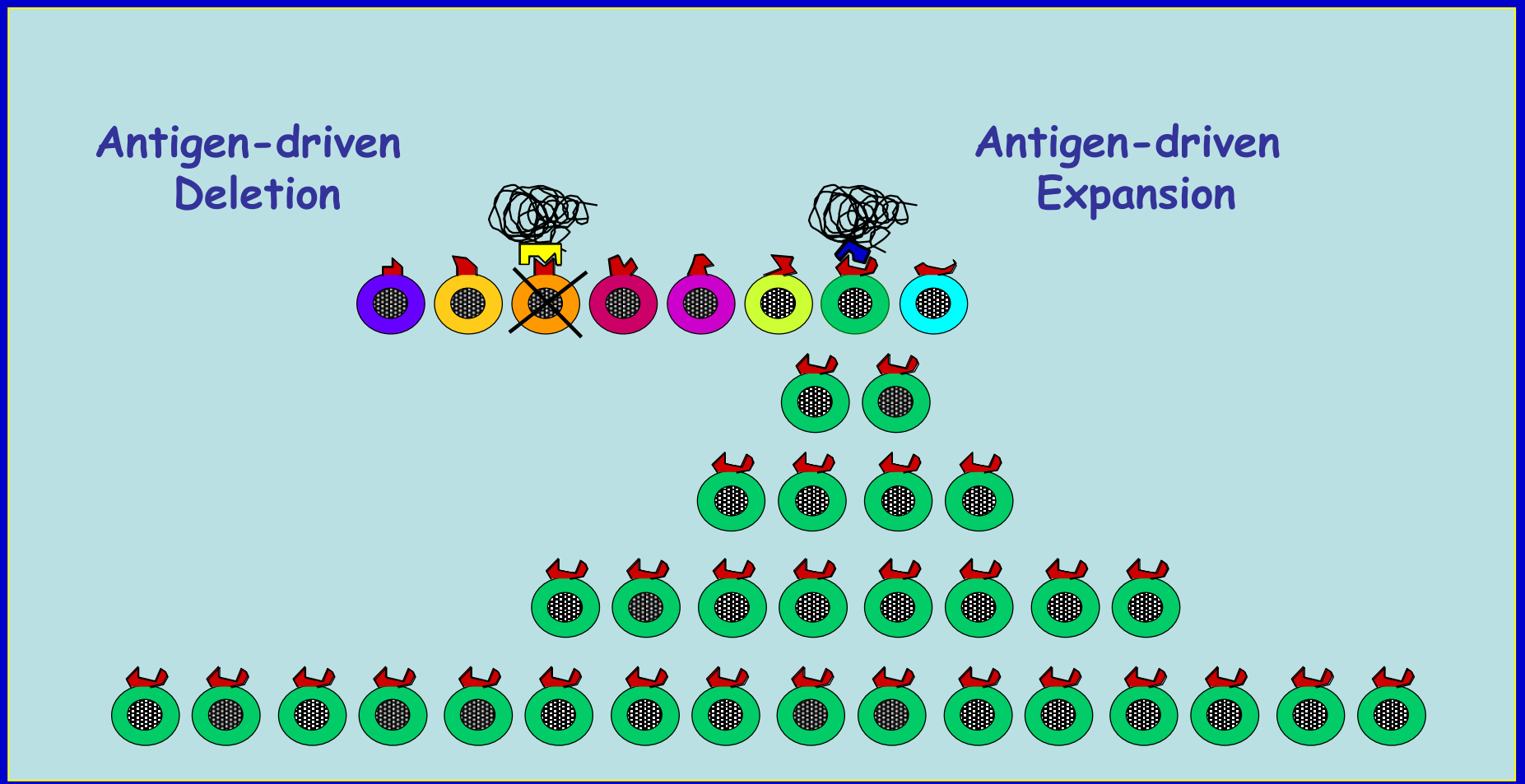


Each one is different in that it can recognize a different structure.

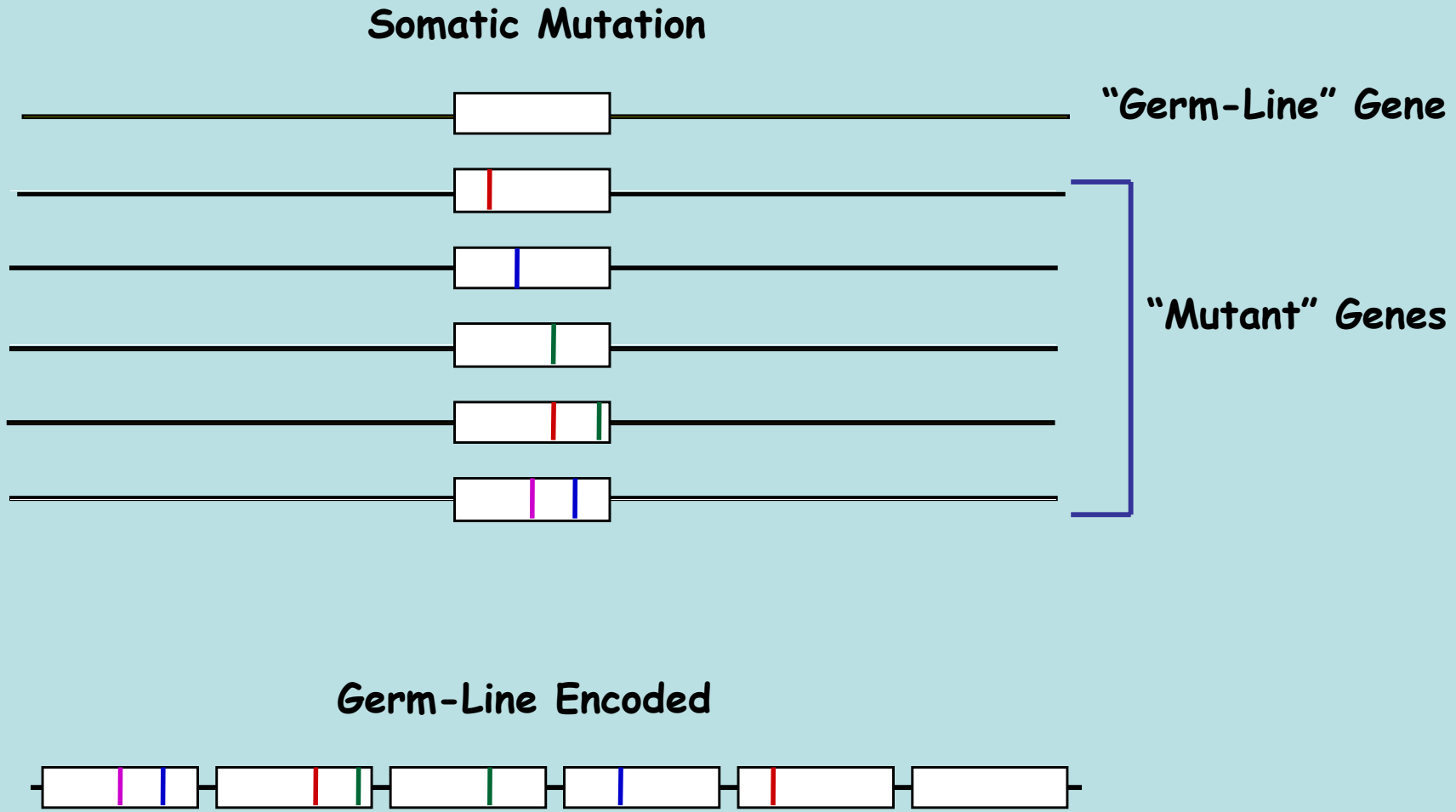
Lymphocytes use a cell surface receptor for this recognition.

Humans have ~10^{12} lymphocytes

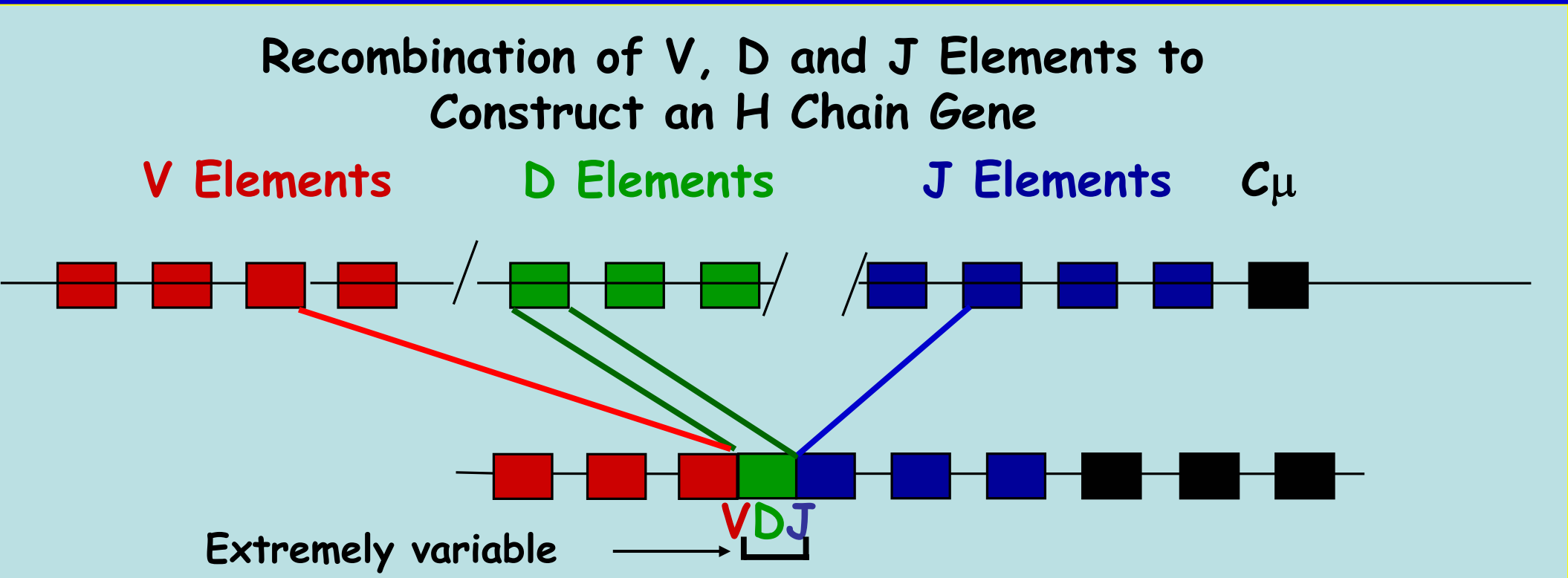
Clonal Selection



Genetic Basis of Specificity



Genetic Basis of Specificity



At V & D and D & J junctions there is joining imprecision, deletion of nucleotides and addition of untemplated nucleotides; the result is a virtual *random peptide generator* creating an enormous potential repertoire of H chains

T Cells Resemble Unicellular Organisms

- Individual T cells are capable of expanding or contracting in number depending on whether they have a selection advantage (recognition of antigen).
- The immune system evolves *somatically* and thus provides a mechanism to deal with the rapidly evolving microbial world.
- The TCR repertoire is continually evolving

T Lymphocyte Homeostasis

T Cell Production
Thymic
Extrathymic

Repertoire ~10^9
T lymphocyte mass ~10^{12}
Life-span: months to years

T Cell Pools

Pathogen or Tumor Induced Demise

Natural Demise apoptosis

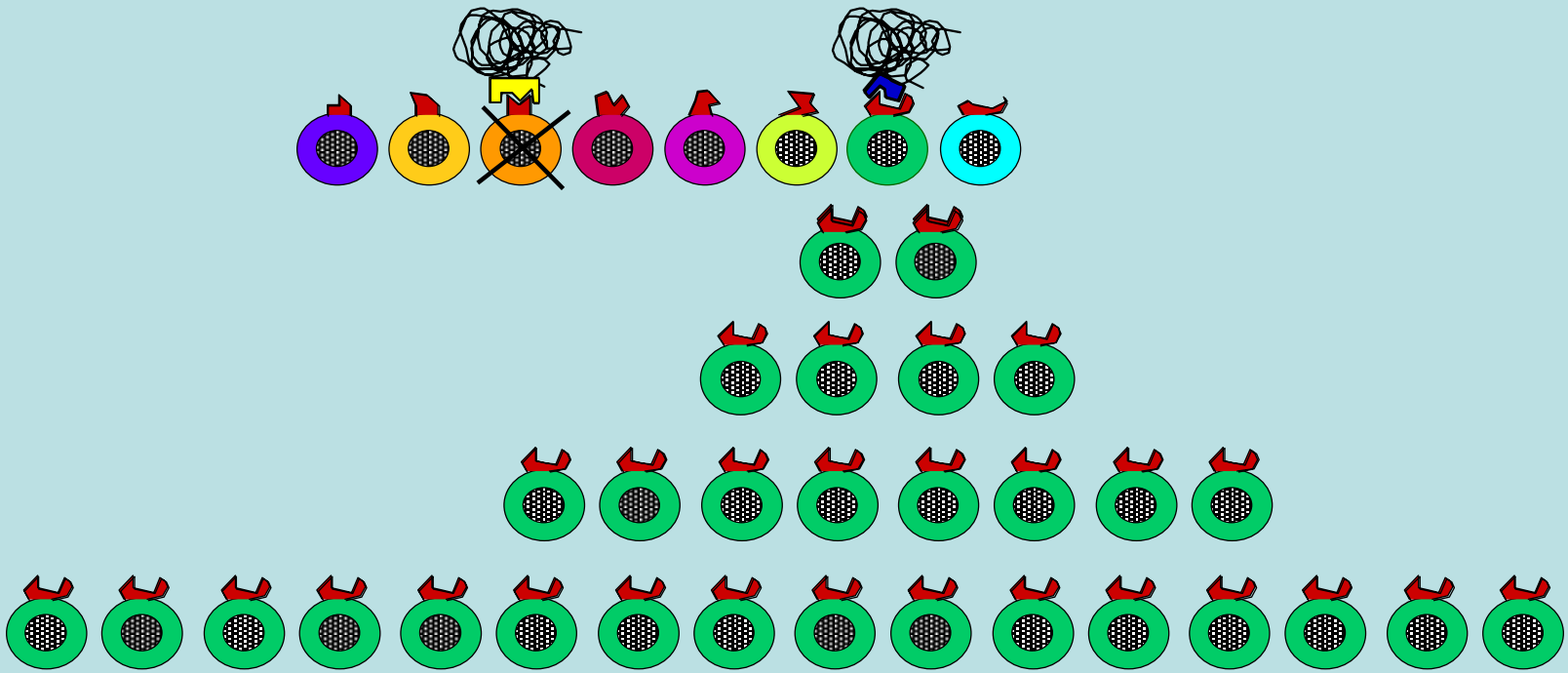
The Second Law - *Tolerance*
T Cells

- Negative selection in the thymus
- Clonal elimination in the periphery
  - ?Encounter with cognate antigens on immature dendritic cells
- Clonal anergy
  - Encounter with cognate antigens in the absence of co-stimulation inactivates T cells
- Regulatory/suppressor T cells
  - Is this a tolerance mechanism or a mechanism to control the magnitude of immune responses

Clonal Elimination; "Classical View"

Antigen-driven Deletion;
Occurs during T Cell Development

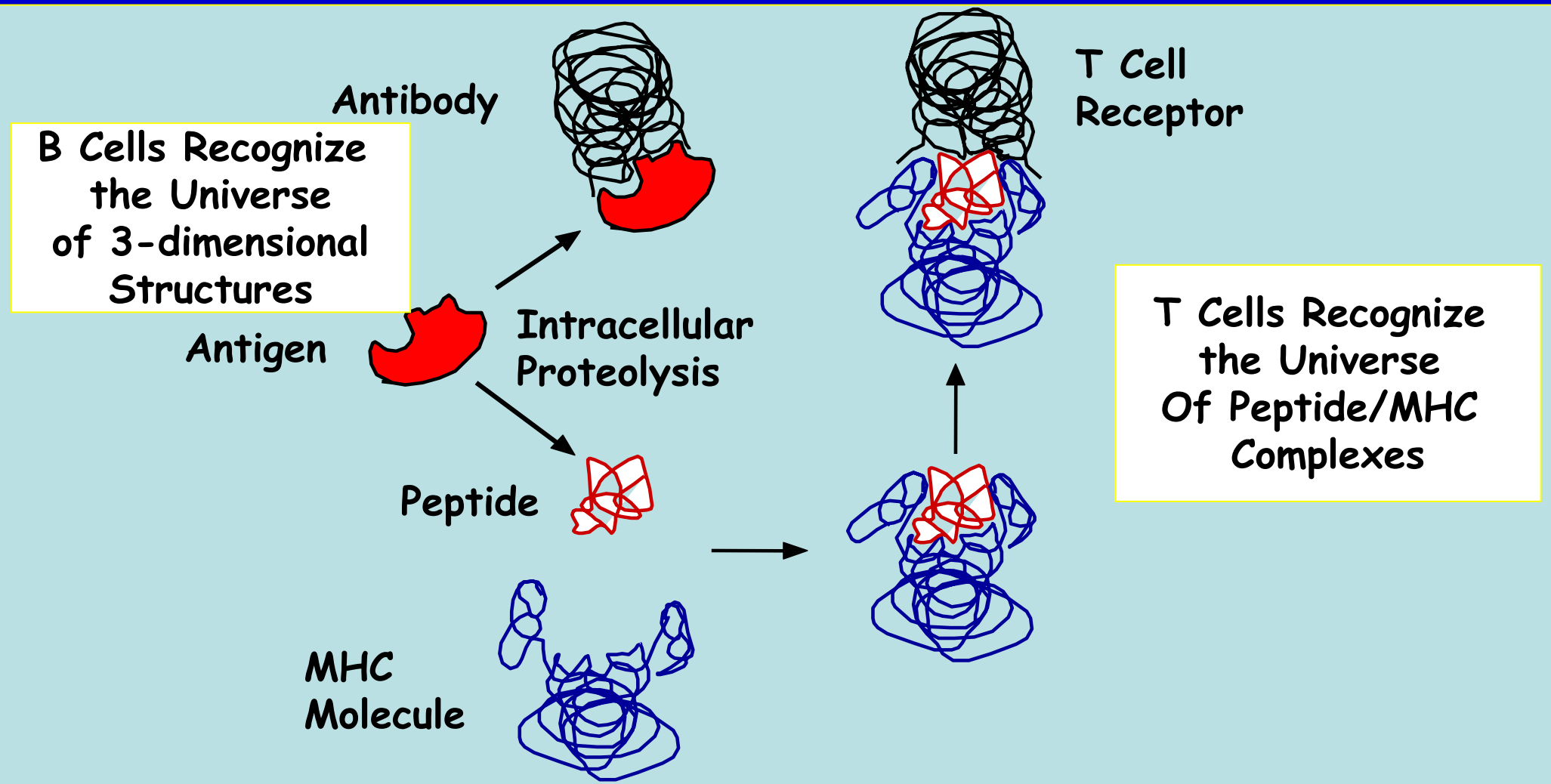
Antigen-driven Expansion;
Occurs when T cell is Mature



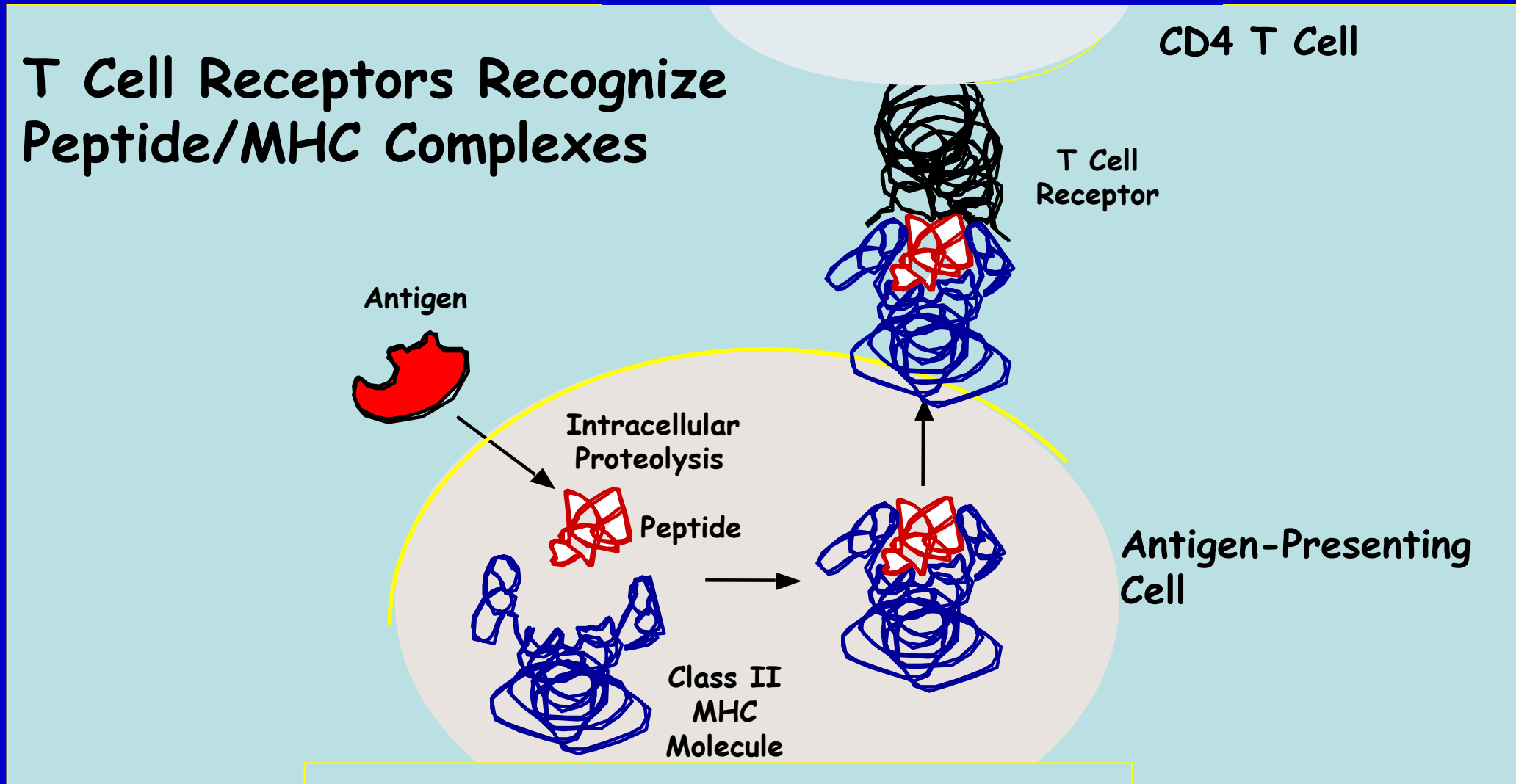
Self Recognition: Both Essential and Dangerous

- The immune system plays a "dangerous" game.
- It relies on self-recognition both to create the repertoire of T cells that are allowed to populate the periphery and for the survival of these cells and yet it needs to control self recognition to avoid a dangerous attack on self-tissues.
- A current view is that the border between these is quantitative – that "good" self recognition is low affinity and results only in survival signals while dangerous self-recognition is high affinity and results in expansion and differentiation into effector cells.

B Cells & T Cells Deal with Different Antigenic Universes

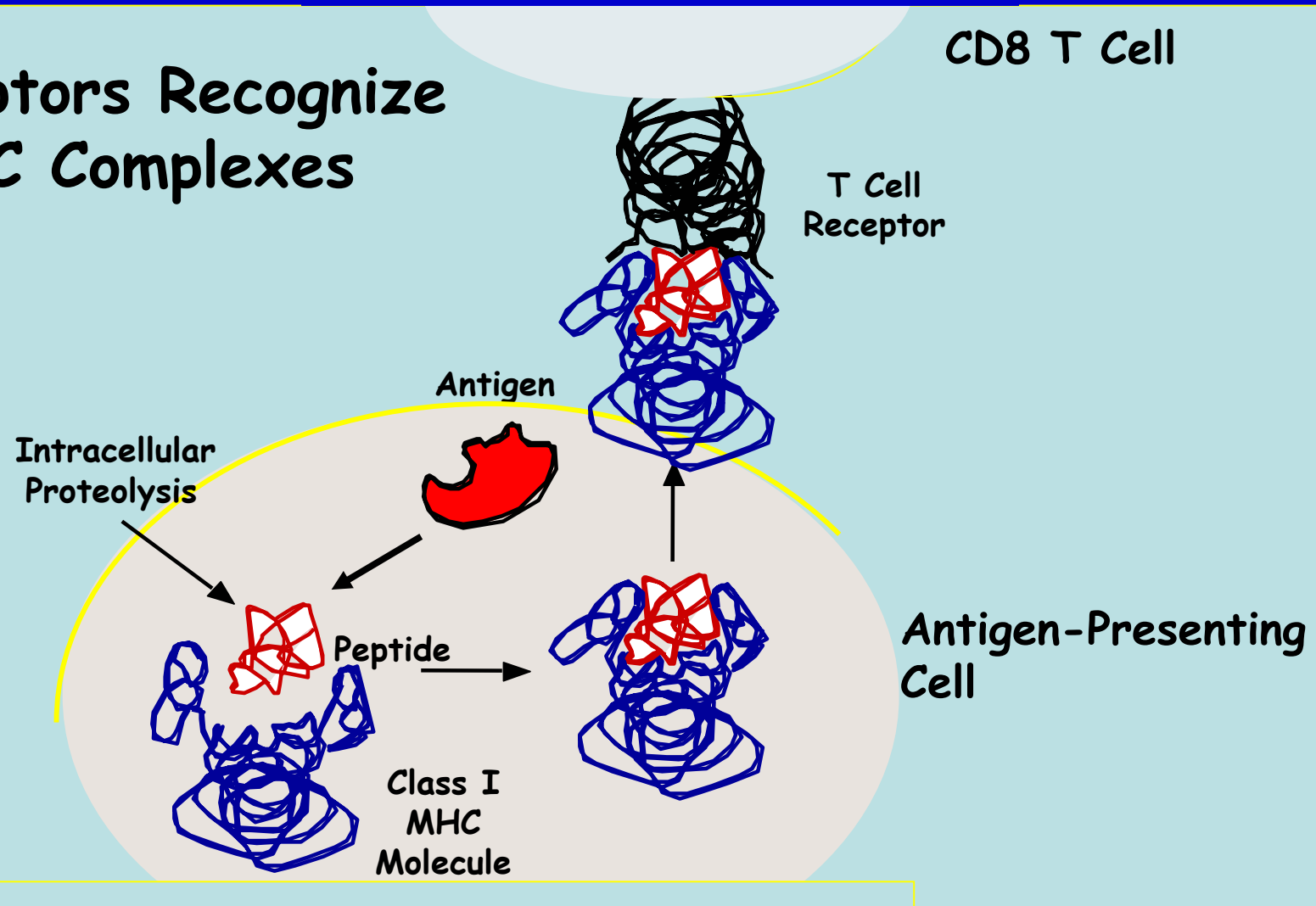


The CD4 T Cell Antigenic Universe

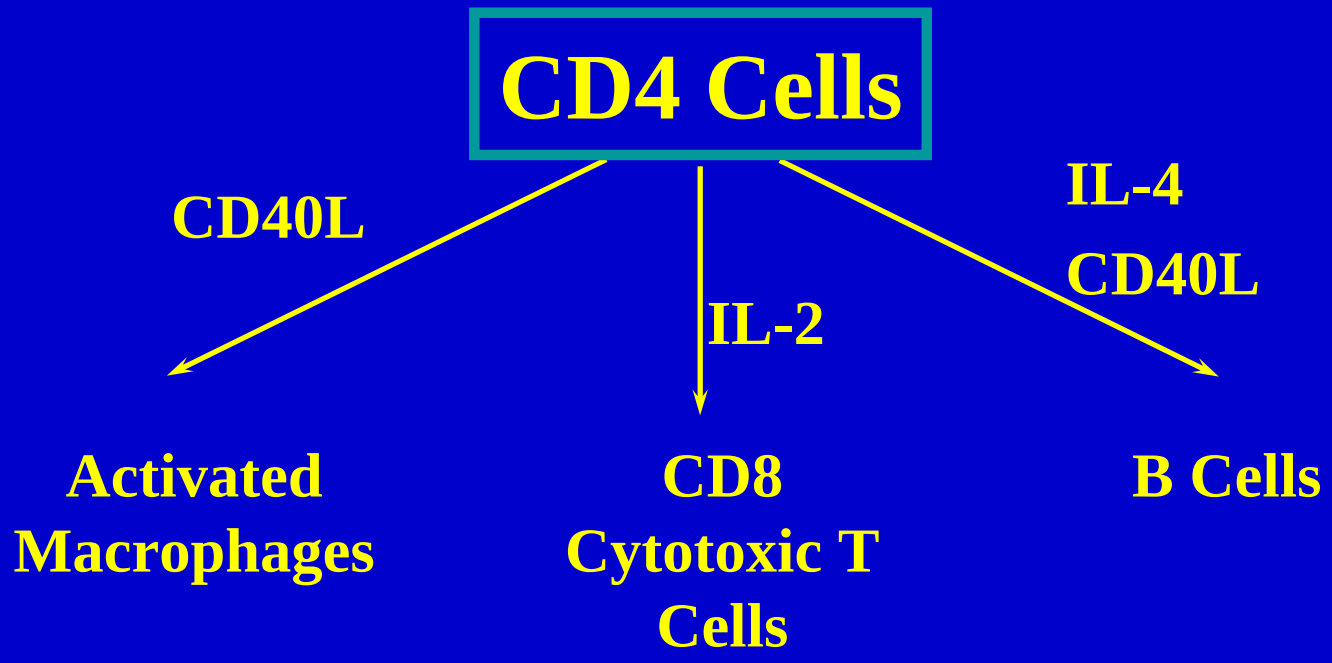


The CD8 T Cell Antigenic Universe

T Cell Receptors Recognize Peptide/MHC Complexes

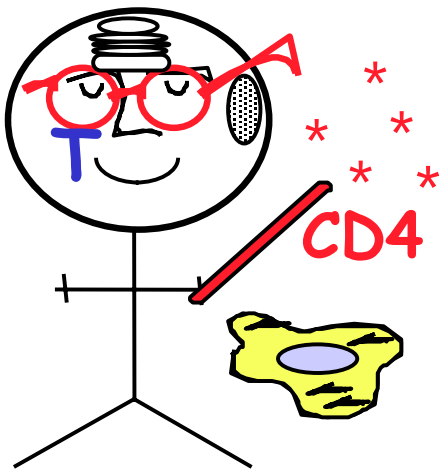


Pivotal Roles of CD4 Cells



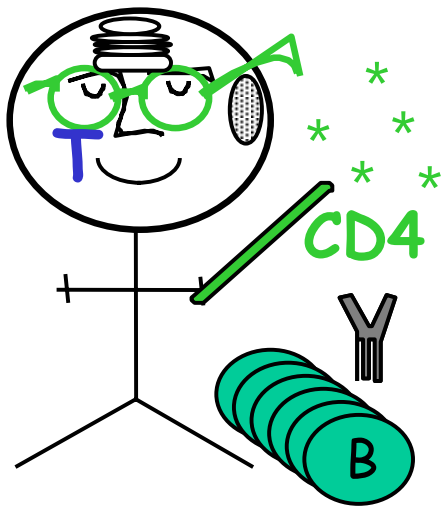
Tolerance is Controlled by CD4 Cells
Three Faces of CD4 T Cells

T-bet



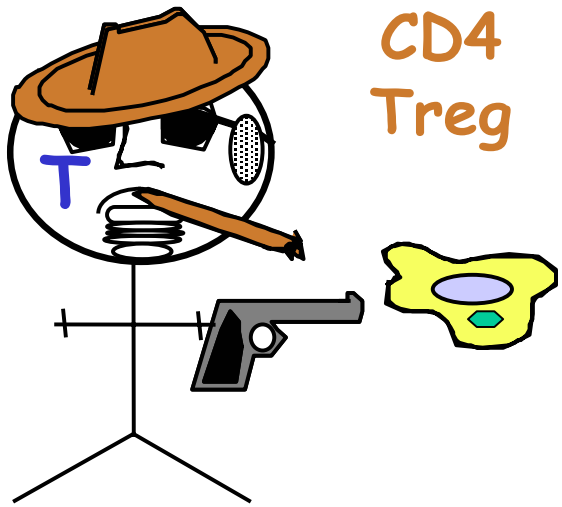
Th1: Fight
Intra-cellular
Pathogens

Gata-3



Th2: Fight
Extra-cellular
Pathogens

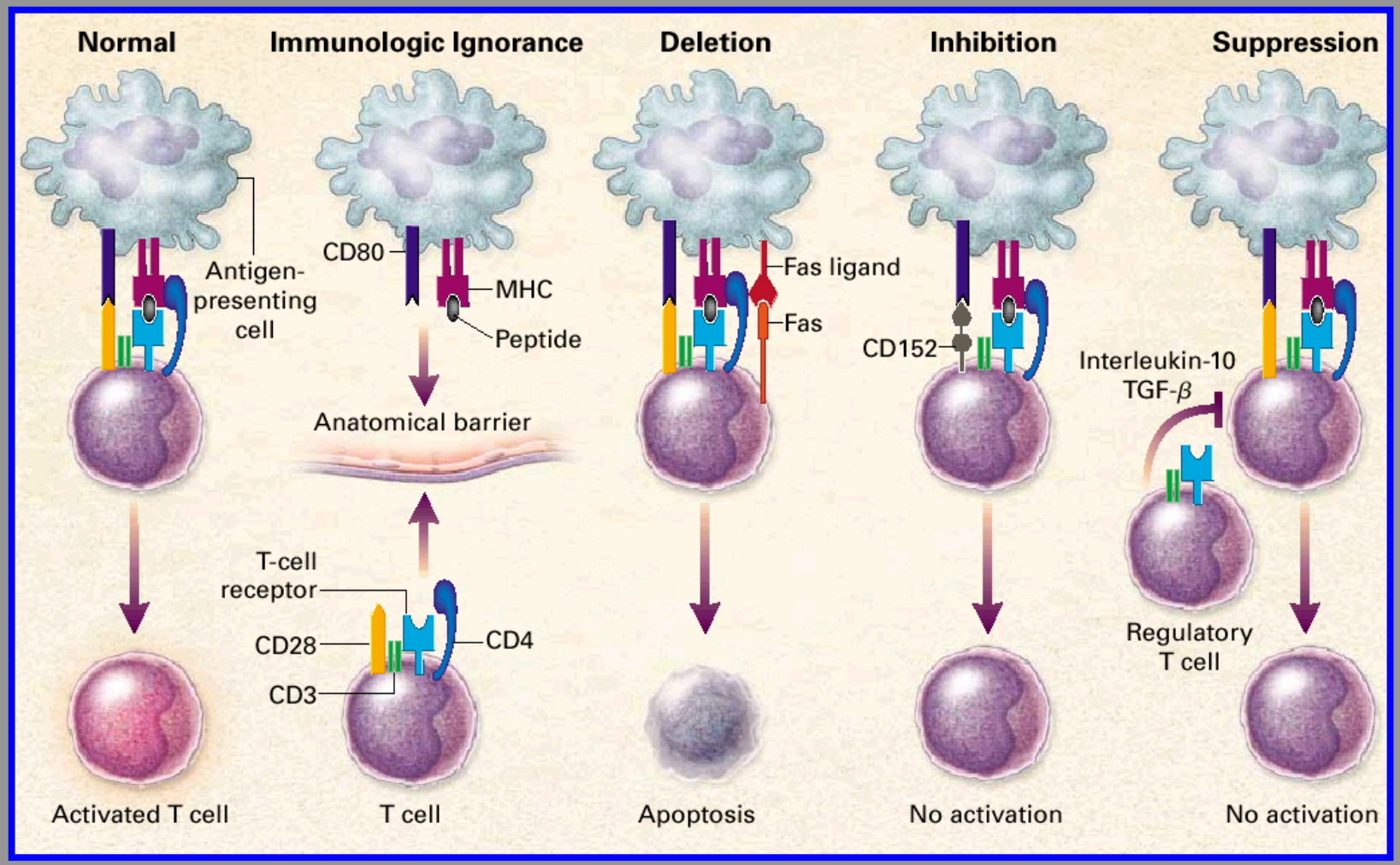
FoxP3



Treg: Silences
Effector T Cells

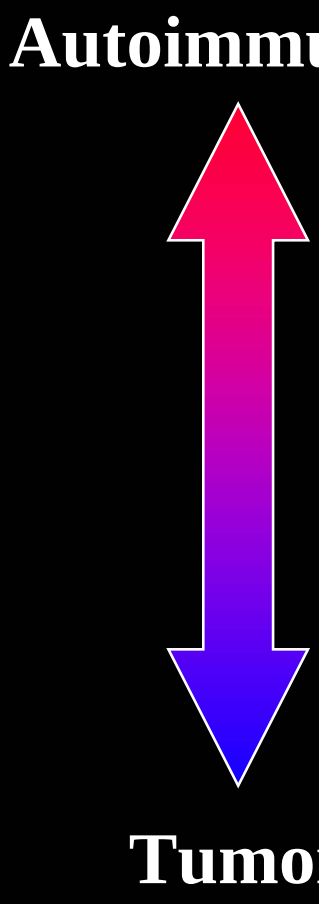
Accumulate at
tumor microenvironment

Mechanisms of Tolerance Induction



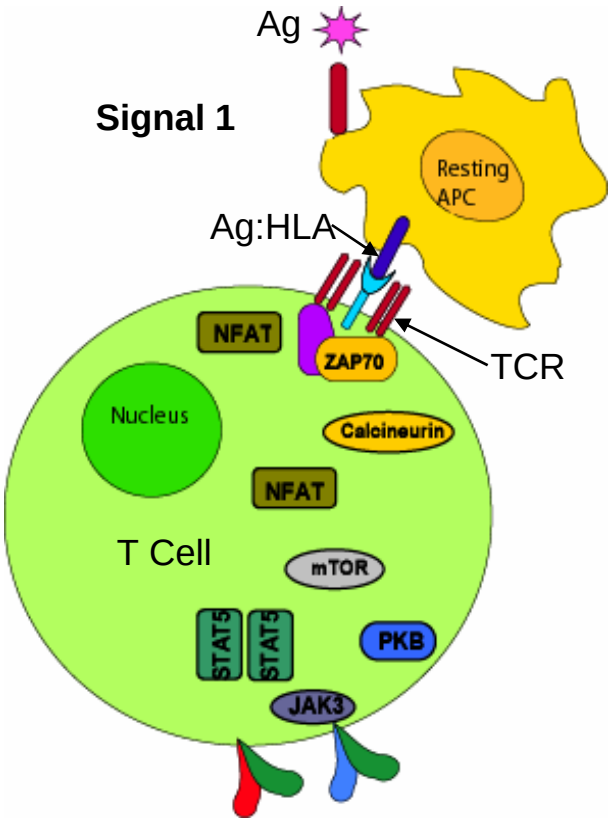
NEJM 344: 655, 2001

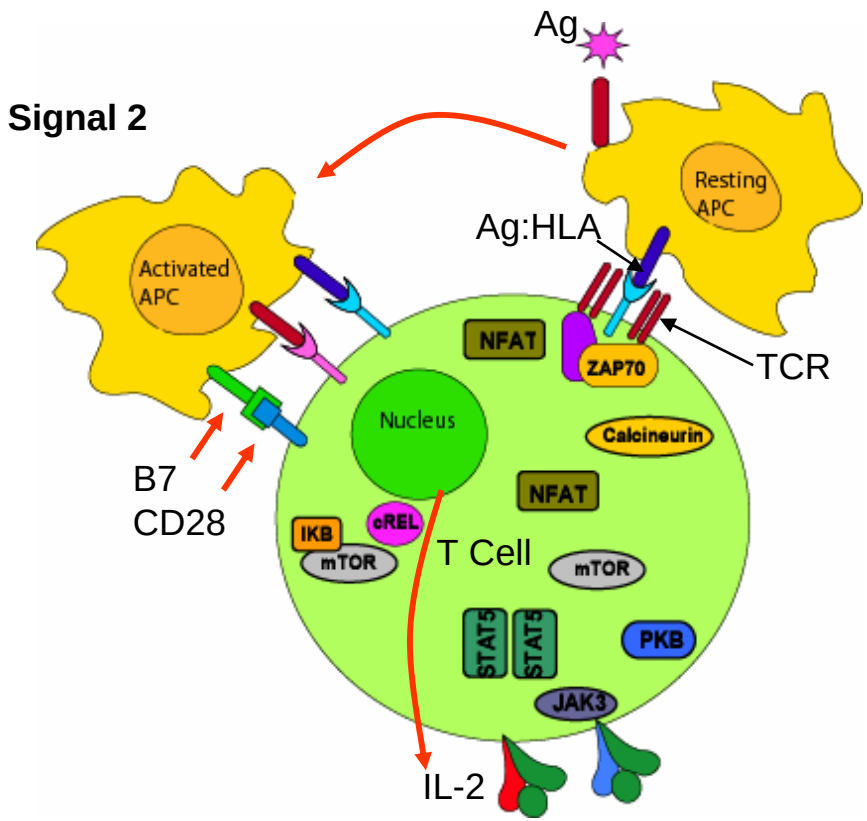
Tumor Immunity and Autoimmunity Paradigm



- **Autoimmunity is the loss of tolerance to self antigens**
- **Many approaches to tumor therapy attempt to *break* tolerance to self antigens expressed on tumors**
- **Expected consequences: tissue specific autoimmunity**

T Cell Activation: antigenicity vs immunogenicity
The concept of costimulation





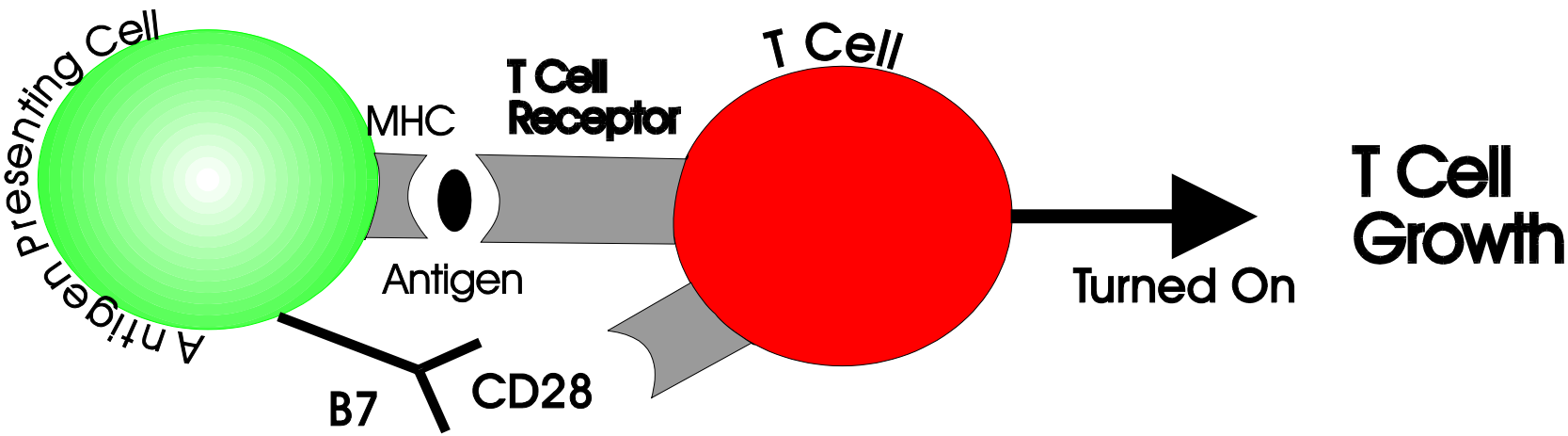
<u>Antigen:APC T cell interaction: no costimulation:</u>

Result: T cell anergy, apoptosis or suppression (Treg cells)

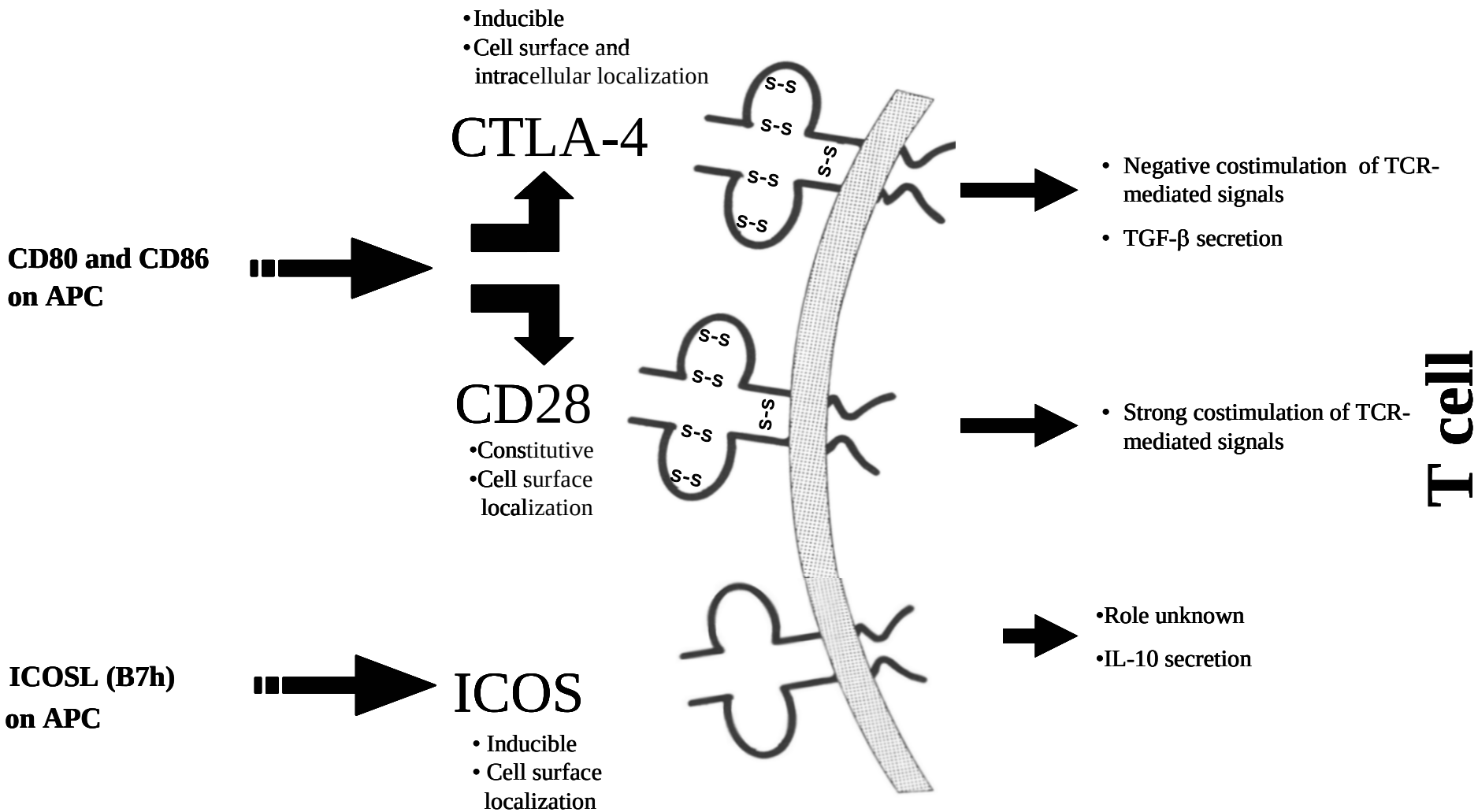
<u>Antigen:APC T cell interaction: with costimulation:</u>

Result: T cell activation, clonal expansion, effector functions

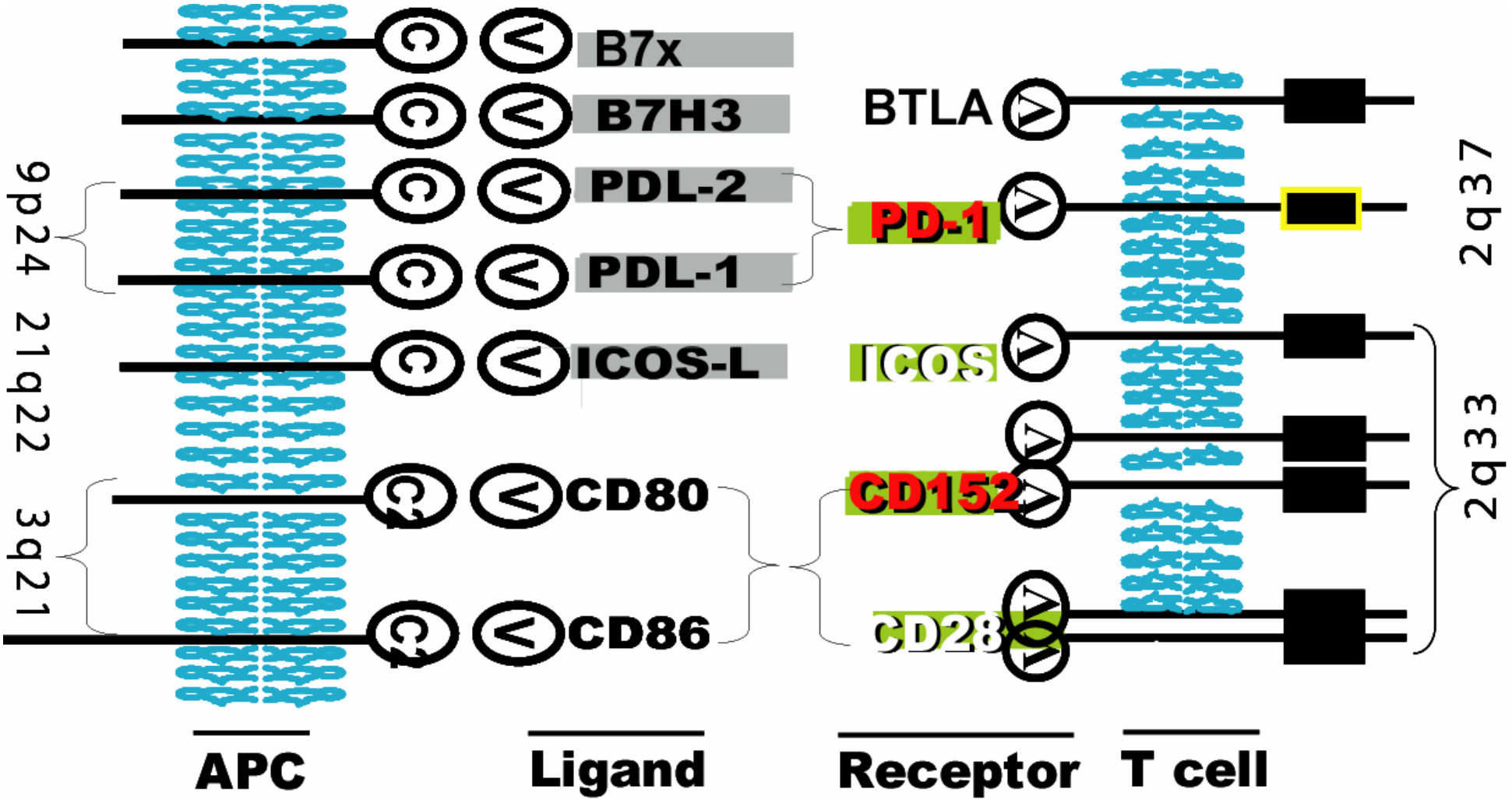
The CD28 and B7 Receptor Families



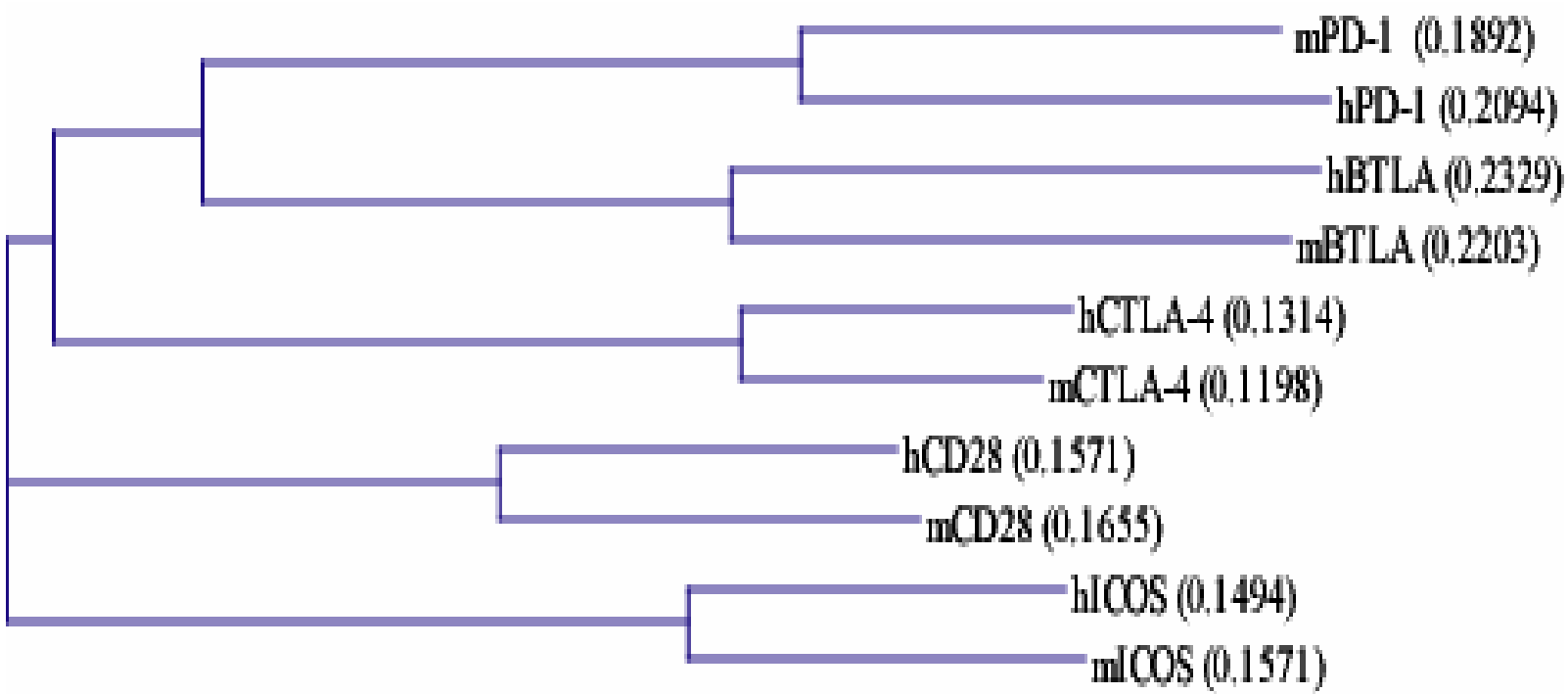
CD28 Receptor Family - 1999



The CD28 and B7 Receptor Families - 2005



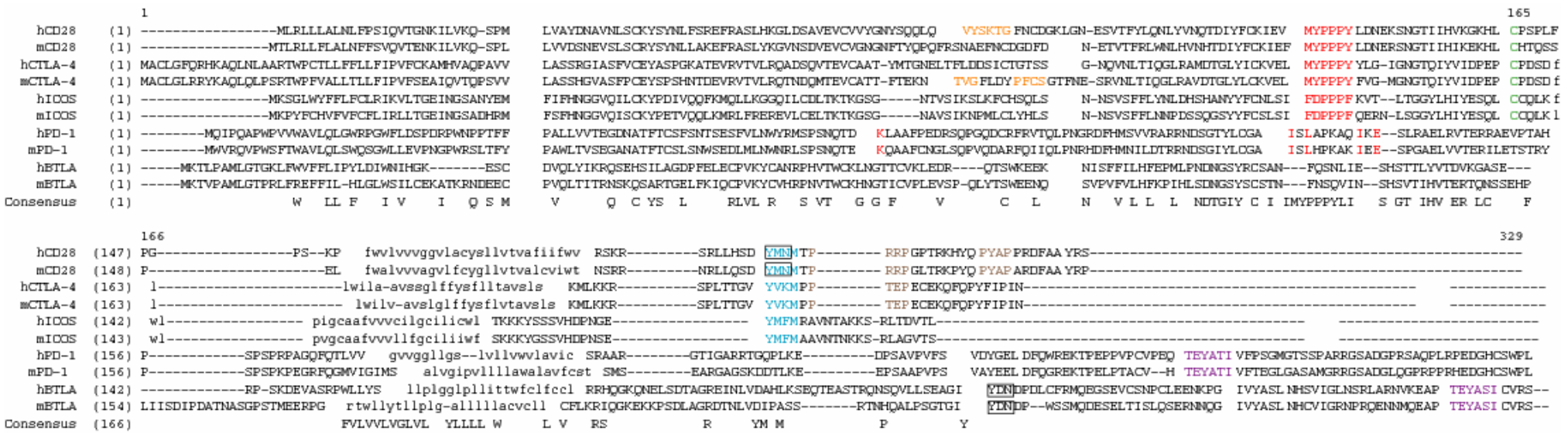
The CD28 Family Homologies



Roles of CD28

- Induction and maintenance of cytokine and chemokine secretion
- Cell survival: bcl-X induction and promotes clonal expansion
- Enhanced telomerase activity
- Required for T cells to increase their glycolytic rate (PI3K and Akt)
- Down regulation of beta chemokine receptor expression
- Costimulation and “superagonists”

The CD28 Family



- Residues important for natural ligand binding
- Residues important for superagonist binding
- Unpaired Cysteine
- SH2 Binding Domain
- SH3 Binding Domain
- ITIM
- ITSM
- YXN GRB2 Binding Domain

CTLA4 Regulates T Cell Numbers

Negative roles

- **CTLA-4-deficient mice die within 4 wk after birth due to a lymphoproliferative disorder of CD4 T cells**
- **Lymphoproliferative disease in the absence of CTLA-4 is not T cell autonomous: bone marrow chimeras producing CTLA-4-/- and normal T cells are healthy**
- **CTLA-4 recruits phosphatases to the TCR complex**

Positive roles

- **Increases beta chemokine receptor expression**

Science 1995; 270: 985

CTLA4 Regulates T Cell Mass

CTLA4 -/- **CTLA4 +/+**

***Science* 1995; 270: 985**

***Immunity* 1995; *3*: 541**

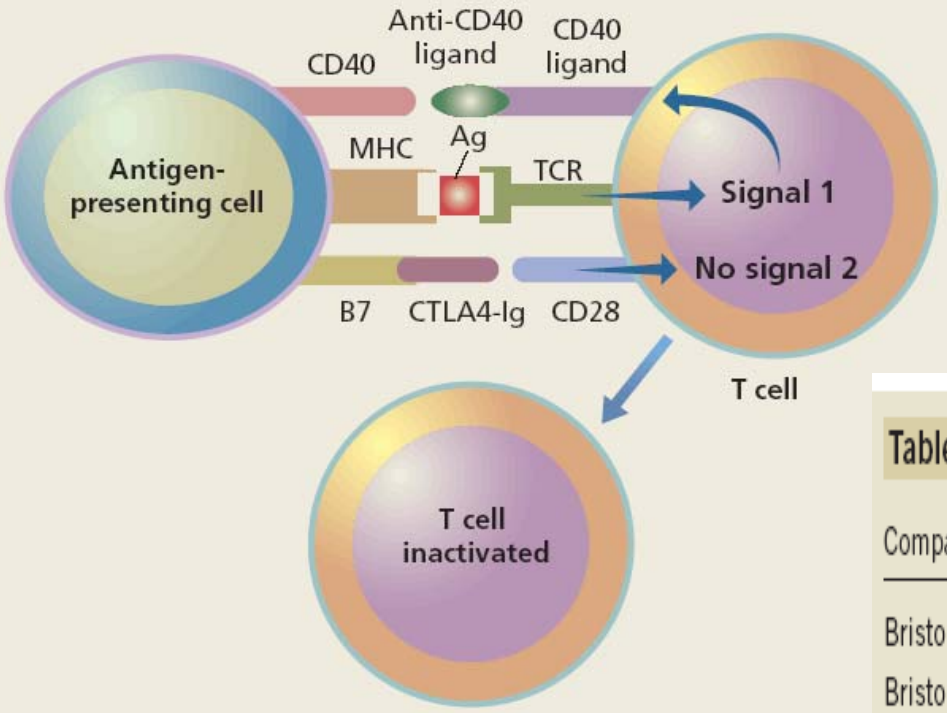
Dysregulated Negative Costimulatory Receptor Expression Can Lead to Autoimmunity

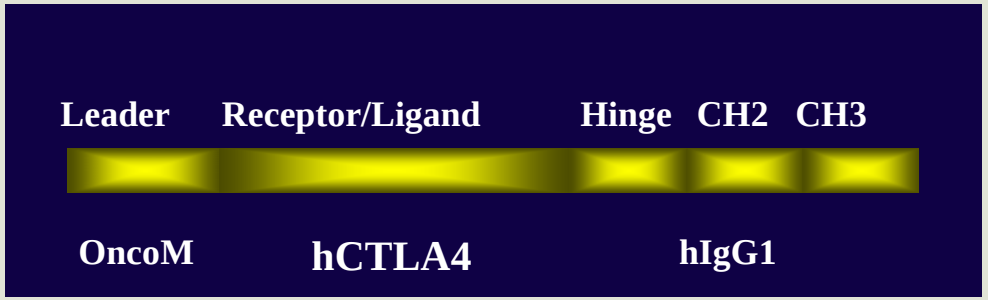
- **Polymorphisms of CTLA4 associated with multiple autoimmune disorders**
  - **Mouse and human reports: T1 DM, Graves' Disease (Ueda et al, Nature 2003: 423: 506)**
  - **Polymorphism in the 3' UTR of CTLA4**
  - **Reduced production of a splice form encoding a molecule lacking the CD80/CD86 ligand-binding domain.**
  - **Mechanism: effects on CTLA4 signals or Treg ?**

Blocking Negative Costimulatory Receptor Function Can Lead to Autoimmunity

- **Antagonistic CTLA4 antibody therapy in cancer patients leads to autoimmunity**
  - **Phan et al, PNAS 2003; 100: 8372**
  - **Multiple organs affected**
  - **Prominent target organ is GI tract: IBD like lesion**
  - **Symptoms resolve with corticosteroids**
  - **Augmented anti-tumor effects**

Soluble Fusion Proteins of CTLA4 Can Treat Autoimmune Disorders





- **Rationale: soluble ligand for B7 that prevents B7:CD28 interaction.**

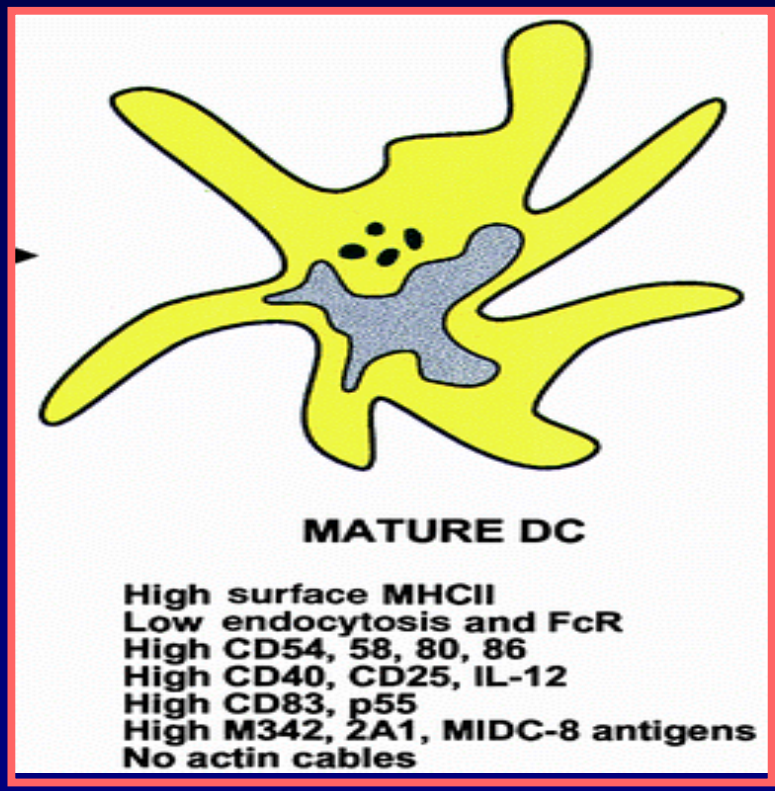
Table 1 Selected CTLA4-Ig drugs in clinical trials

Company	Indication	Drug	Stage of development
Bristol-Myers Squibb	Rheumatoid arthritis	CTLA4-Ig	Phase 3
Bristol-Myers Squibb	Multiple sclerosis	CTLA4-Ig	Phase 2
Bristol-Myers Squibb	Kidney transplantation	LEA29Y	Phase 2
Repligen	Multiple sclerosis	CTLA4-Ig (C.gamma4)	Phase 1/2
Bristol-Myers Squibb	Lupus	CTLA4-Ig	Phase 2 (pending)
Repligen	Lupus	CTLA4-Ig (C.gamma4)	Phase 1/2 (pending)

Why are there so many costimulatory molecules?

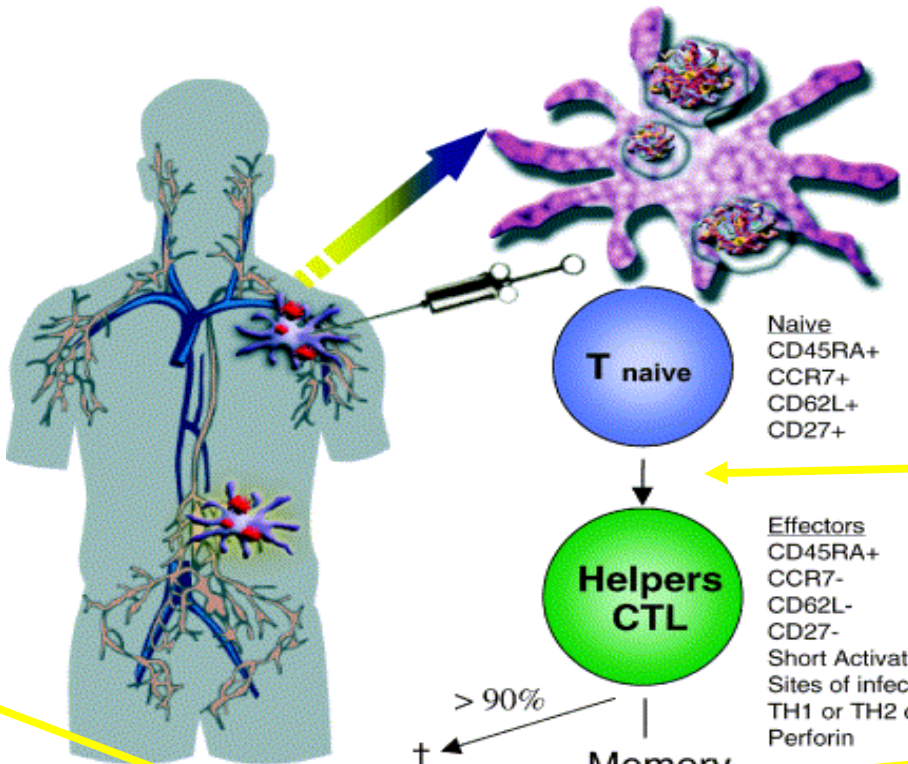
Hypotheses:

1. **Fine Tuning of the Immune Response**
2. **Tissue Specificity**
3. **Distinct signal requirements to trigger differentiation and drive activation**
4. **Instructional Role of DC in Generation of T cell Subsets**



Banchereau J and Steinman RM.
Nature. 1998 (392) 245-252.

Generation of CD8 Memory T Cells



T naive

Naive
CD45RA+
CCR7+
CD62L+
CD27+

CD28

Helpers CTL

Effectors
CD45RA+
CCR7-
CD62L-
CD27-
Short Activation
Sites of infection
TH1 or TH2 cytokines
Perforin

CD107
CD83

> 90%

†

IL-15
IL-21?

Memory

T Effector Memory
CD45RA+ or CD45RO-
CCR7-
CD62L-
CD27-
Short Term Activation
Periphery & Mucosae
TH1 or TH2 cytokines
Perforin

T EM

T CM

T Central Memory
CD45RA+ or CD45RO+
CCR7+
CD62L+
CD27 +
Delayed Activation
Skin & Lymph Nodes
IL-2 + IL-10
DC help (CD40L)

4-1BB

Esser MT, et.al.

Tumor Immunology: Basic Concepts

- Current Dogma: "Tumor cells are antigenic but not immunogenic"
- T cells (T for "tumor and thymus") mediate specific tumor rejection. Natural killer cells also have a role in tumor elimination.
- B cells and antibody appear to have little or no physiologic role in immune tumor elimination since these immune effector systems are best suited for extracellular antigens.
- "Tumor Darwinism": Tumors have evolved sophisticated means to avoid immune detection.

Cancer Immunosurveillance Theory

- The T cell immune system almost certainly evolved as a defense mechanism to control viral infections; it definitely did not evolve to control tumors!
- Given the demonstration of tumor specific antigens, during the 1960s and 1970s, there was wide acceptance of the "immunosurveillance" model put forth by Lewis Thomas and MacFarlane Burnet.
- 1980s, SCID mice (T-NK+): had *normal* incidence of tumors: toss the theory!
- 2000, mice with deficient IFNgamma signaling have large increase in spontaneous tumors (Schreiber et al, Nature, 2001): resurrects theory.

Tumor Immunosurveillance

- **Immunosurveillance: Compelling data from human patients indicates that cancer immunosurveillance acts as an extrinsic tumor suppressor**
- **Immunoediting: immune surveillance facilitates the outgrowth of tumors with reduced immunogenicity (Schreiber and Old)**

Enhanced susceptibility of immunodeficient mice to spontaneous and chemically induced tumors in immunosuppressed animals

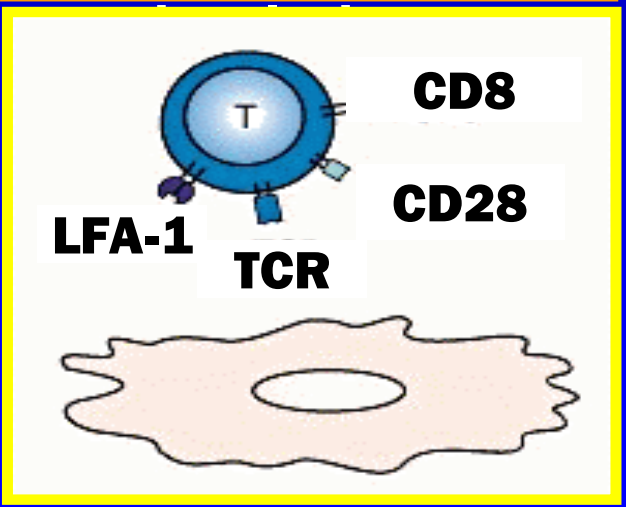
Technology	Immune status	Tumor susceptibility relative to wild type
RAG-2 $^{-/-}$	Lacks T, B, NKT cells	↑ MCA-induced sarcomas;
		↑ spontaneous intestinal neoplasia
RAG-2 $^{-/-}$ ×STAT1 $^{-/-}$	Lacks T, B, NKT cells;	↑ MCA-induced sarcomas; ↑ spontaneous intestinal and mammary neoplasia
	IFNγ-, α/β-insensitive	
LMP2 $^{-/-}$	Lacks LMP2 subunit	↑ Spontaneous uterine neoplasms

Annu Rev Immunol 2004; 22: 329-360

Mechanisms of Tumor Immune Evasion

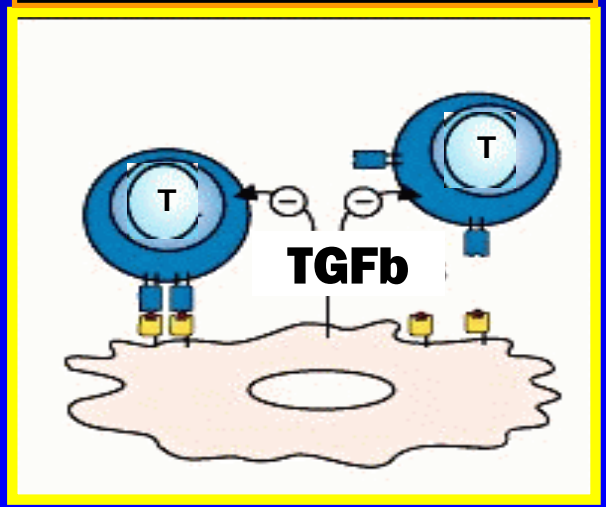
Immunogenicity

- decreased peptide:MHC complexes
- decreased



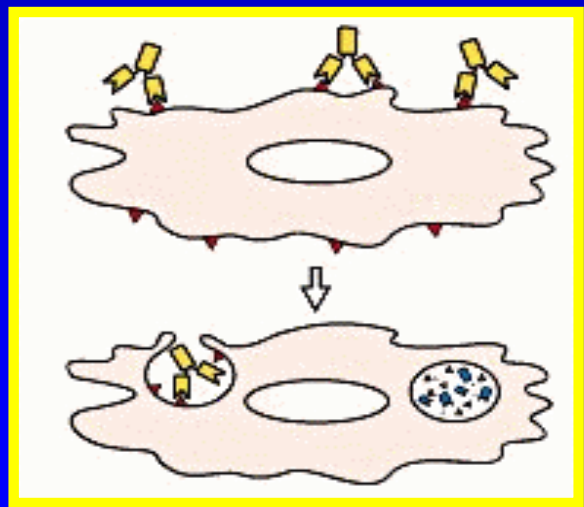
Tumor Induced Immune Suppression

- e.g. TGFβ
- e.g. IL-10



Antigenic Modulation

- Selection for "loss variants"



Summary: Tumor Immunology

- Basic principles of tumor immunology
  - Know the three laws!
- Cancer antigens: generally "self" antigens
  - Implies immune system will be biased to *tolerogenic* responses
- Tumor immunosurveillance and immunoediting
- Tumor induced immune suppression

Outline: Primer on Tumor Immunology

- T Cell Receptors
- T Cell Biology
- Tumor immunology