Immunology Basics Relevant to Cancer Immunotherapy:

Initiation of T Cell Reponses: Innate immunity, DCs, Antigen Presentation, MHC restriction

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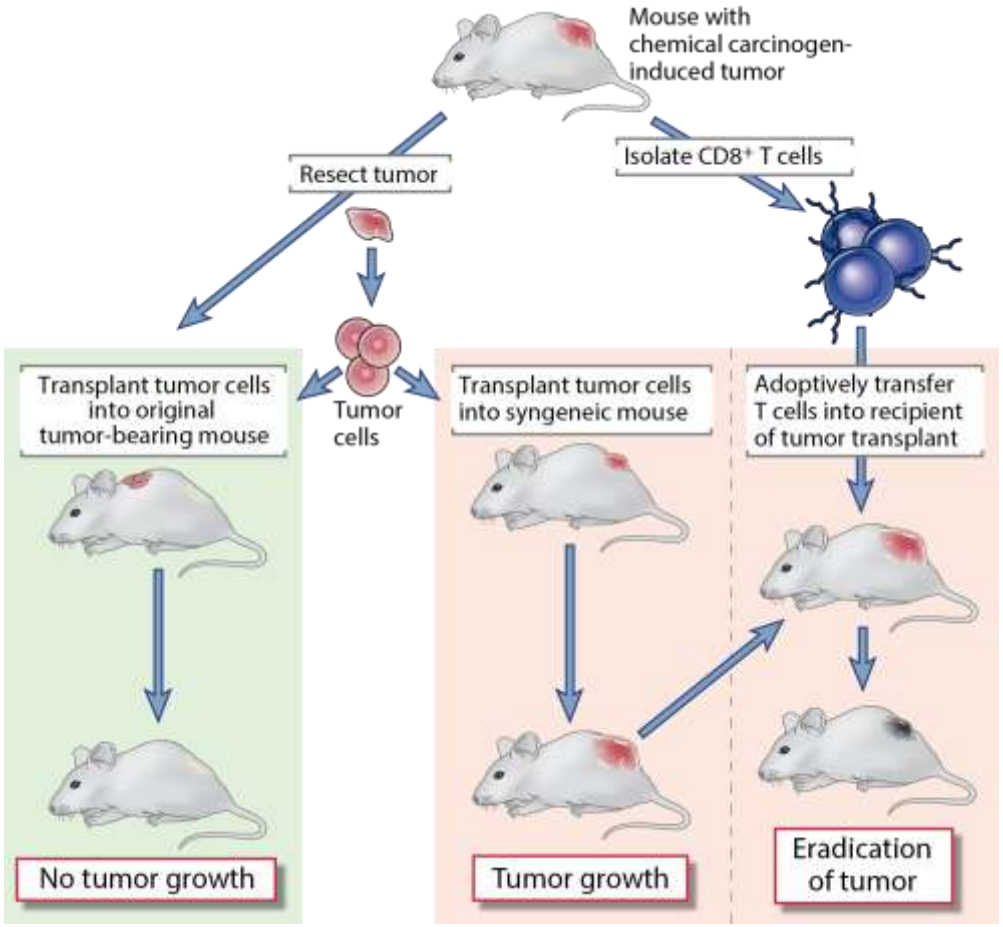
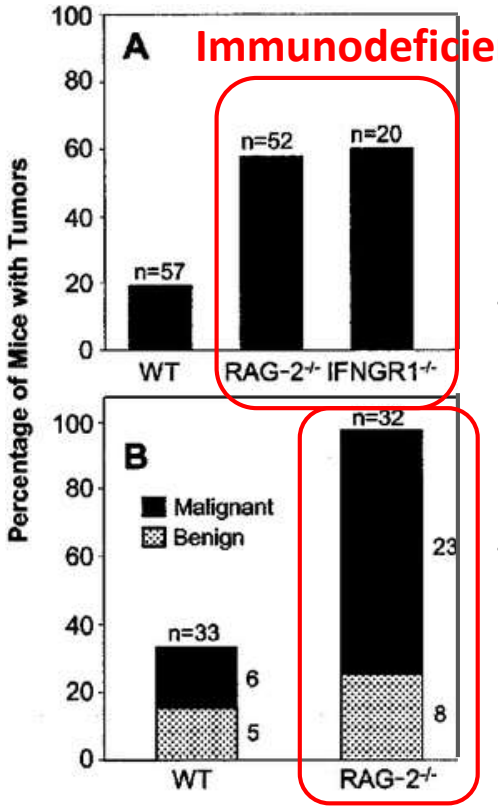
Lecture Outline

- Brief overview of tumor Immunity: why focus on T Cells?
- Innate immune activation of dendritic cells, by microbes and tumors
- T cell recognition of antigen
- Antigen processing and presentation pathways for CD8+ and CD4+ T cell responses
- Major histocompatibility complex molecules and MHC restriction
- Identifying tumor antigens that can be presented by MHC molecules

Rodent Work in Tumor Immunology Established to Importance of T Cells





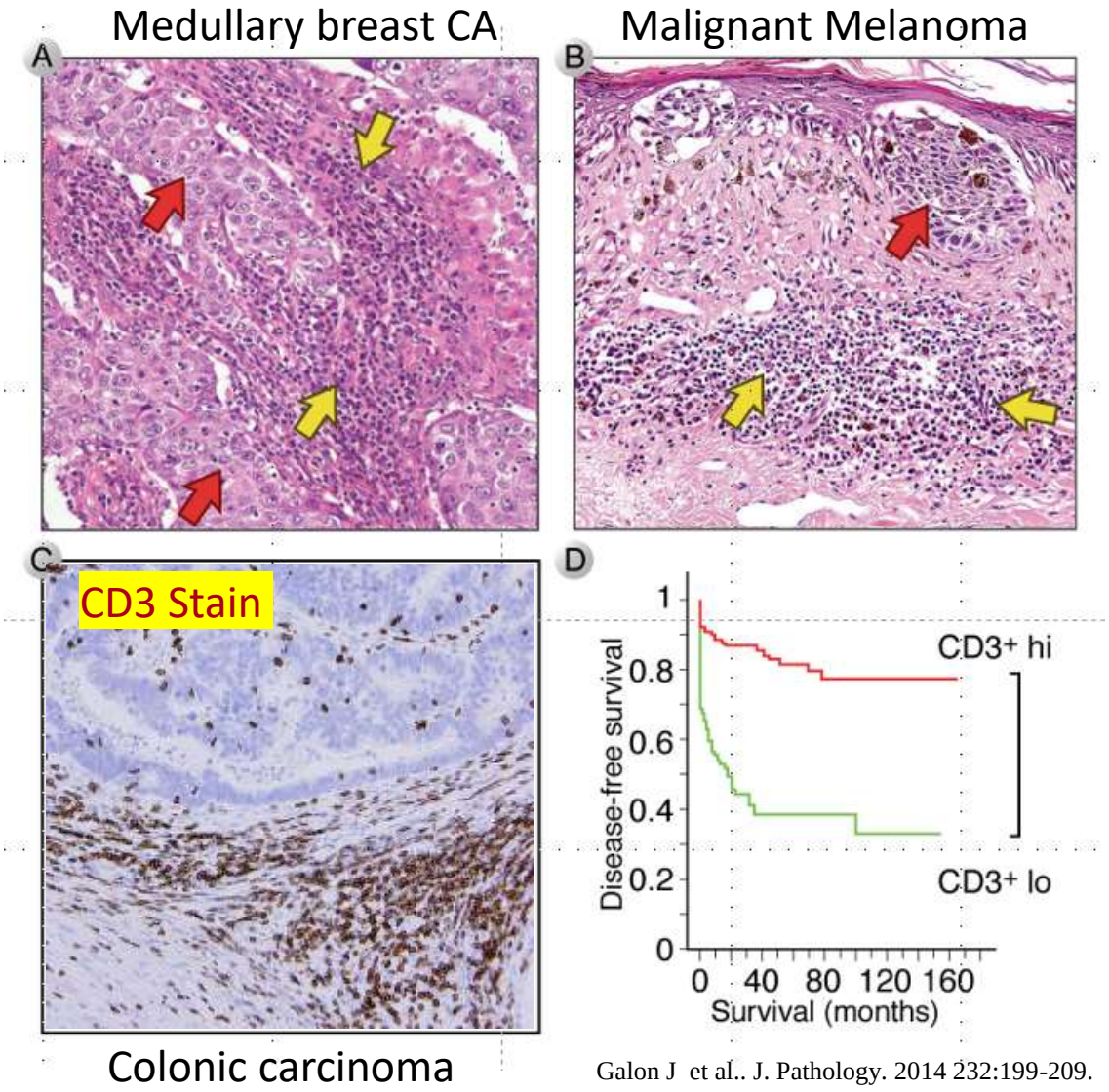
Chemical carcinogen induced tumors

Spontaneous tumors in aged mice

Dunn, Old and Schreiber· Ann Rev Imm 22:329. 2004

Abbas, Lichtman, Pillai. Cellular and Molecular Immunology. Elsevier. 2015

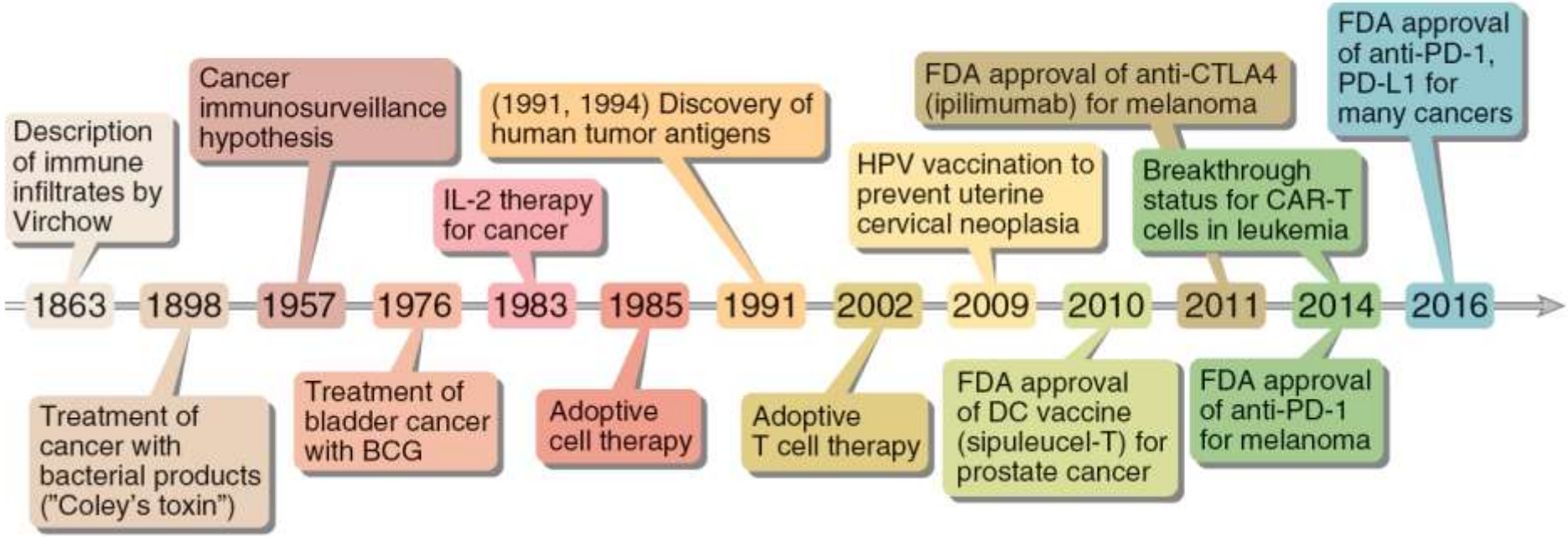
T lymphocytes infiltrate tumors and their presence improves prognosis



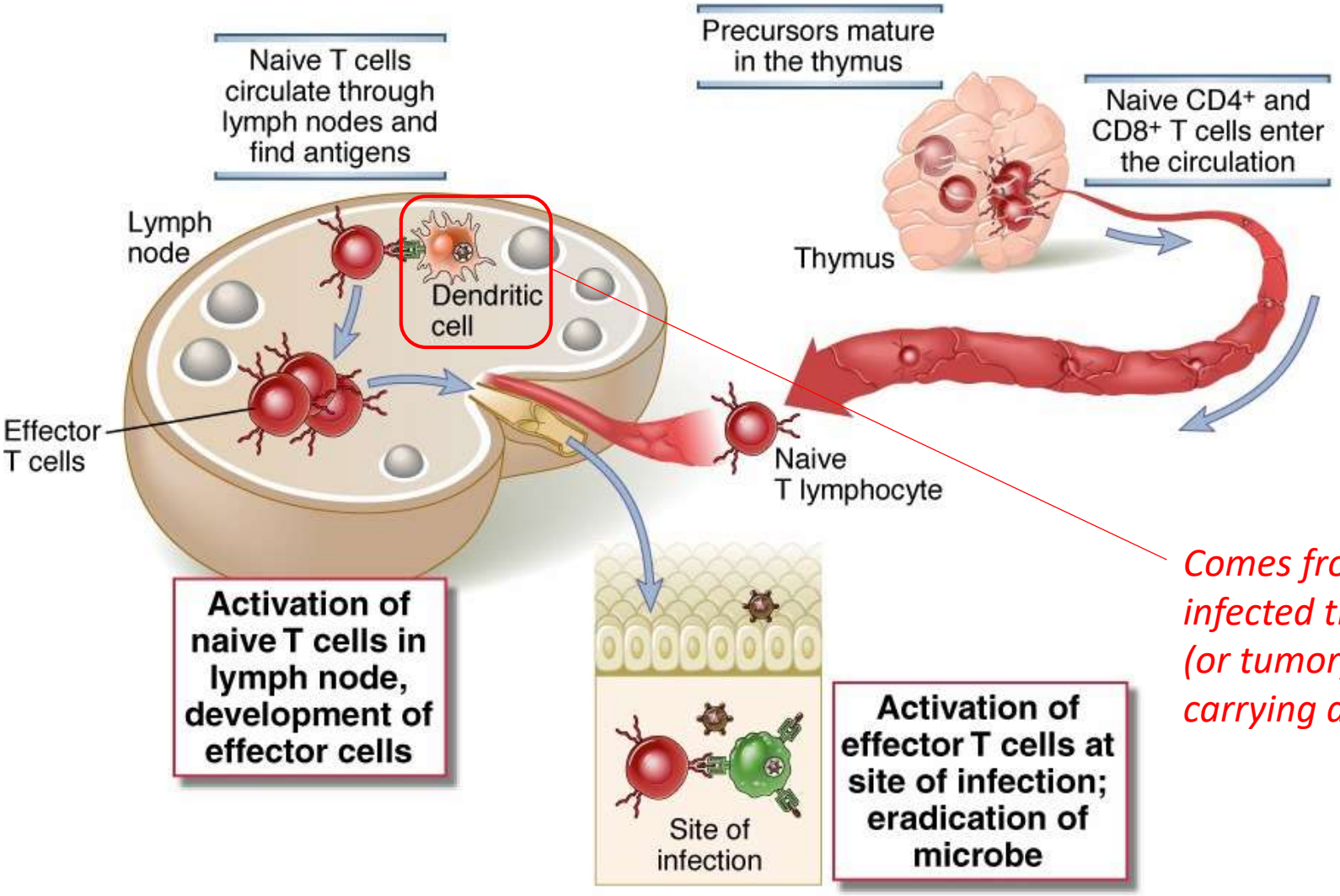
Galon J et al.. J. Pathology. 2014 232:199-209.

History of Cancer Immunotherapy

It's all about T cells



The life history of T lymphocytes



Comes from infected tissues (or tumor), carrying antigen

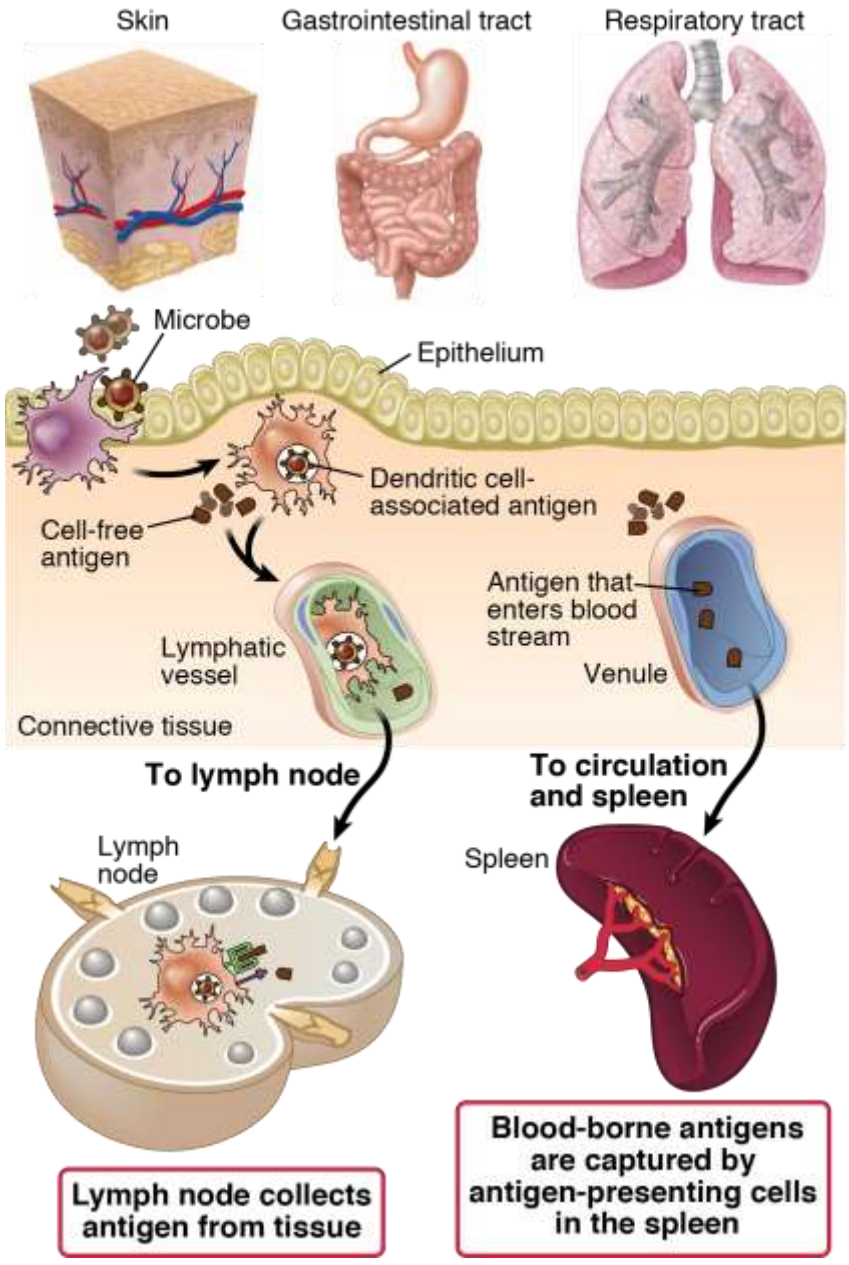
Abbas, Lichtman and Pillai. *Cellular and Molecular Immunology, 7th edition, 2011*

Capture of antigens

Sites of antigen entry

Sites of initial antigen capture

Sites of antigen collection and capture



Dendritic cell (DC) subsets

- Classical: CD11c+, located in epithelia (site of microbe entry), role in capture and presentation of most antigens
- Plasmacytoid: source of type I IFN; capture of blood-borne antigens, transport to the spleen

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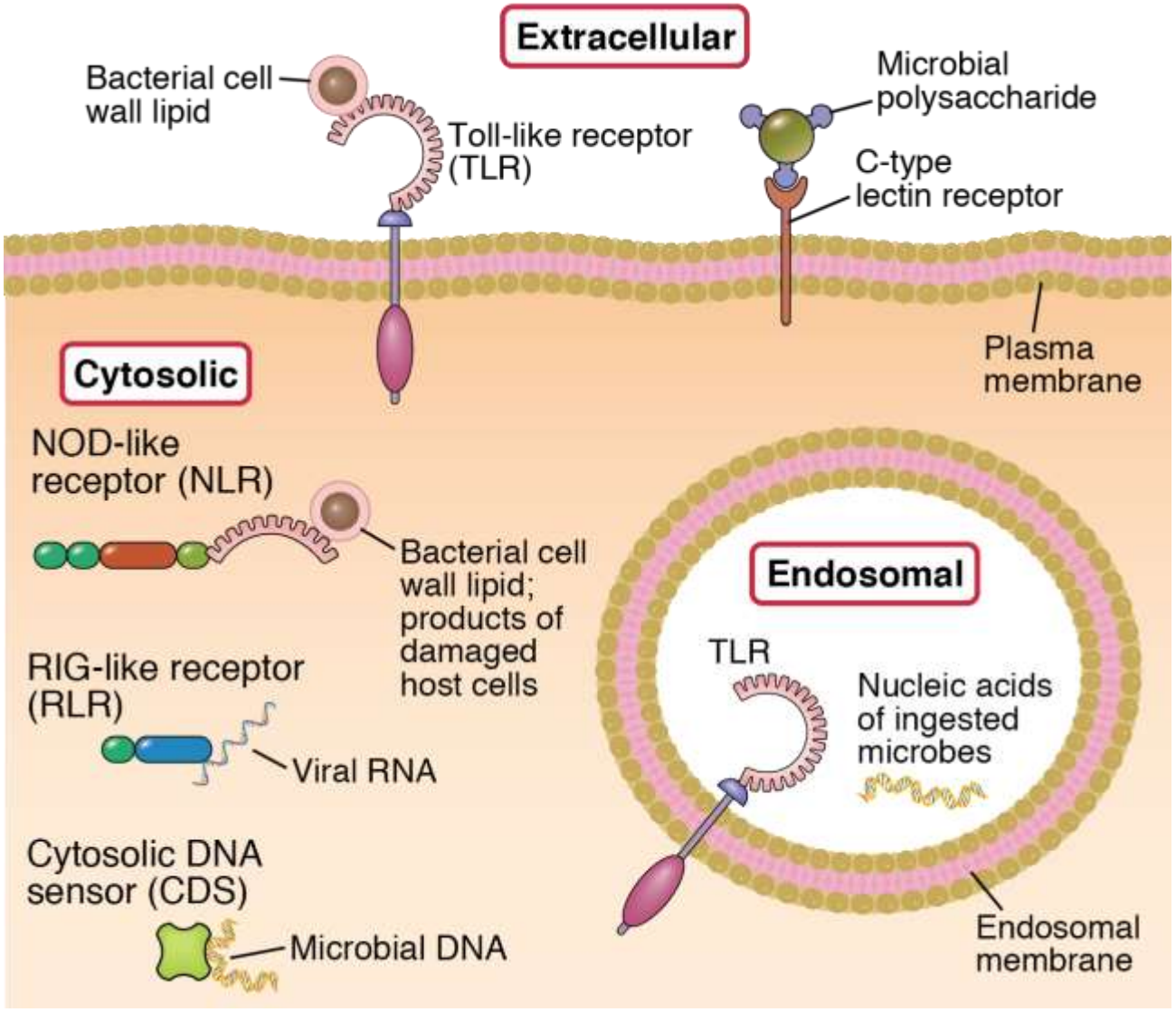
- Immature: in tissues; role in presentation of self antigens and maintenance of tolerance
- Mature: *activated by activated by innate immune responses:* TLR and other signals; role in T cell activation

Innate Immune System: What is recognized?

- Structures that are shared by various classes of microbes but are not present on host cells - Pathogen associated molecular patterns (PAMPs).
  - Innate immunity often targets microbial molecules that are essential for survival or infectivity of microbes (prevents escape mutants)
- Structures produced in damaged or necrotic host cells - Damage associated molecular patterns (DAMPs).
  - Cell injury and death associated with tumor growth may provide the DAMPs

Take home messages

Innate Pattern Recognition Receptors



Receptors are located where they can sample all cellular compartments

4 major classes of receptors:

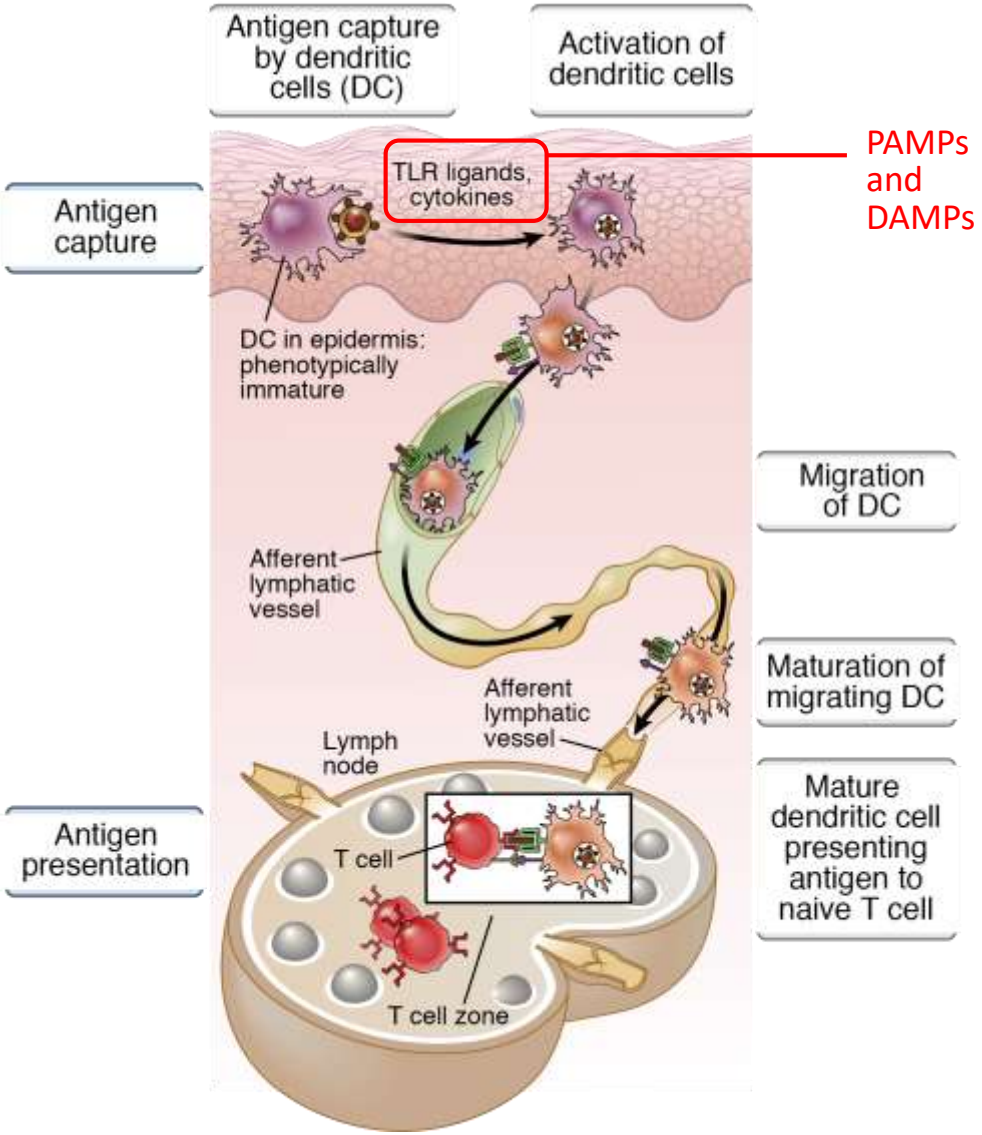
-TLRs: bacteria and viruses

-CLRs (C-type lectin receptors): fungi

-NLRs: bacteria and cell damage

-RNS/DNA sensors: viruses

Capture and Presentation of Antigens by DCs



Abbas, Lichtman and Pillai. *Basic Immunology, 5th edition, 2016, Elsevier*

***Sites of microbe entry**: skin, GI tract, airways (organs with continuous epithelia, populated with dendritic cells). Less often -- infected tissues, blood*

***Sites of lymphocyte activation**: secondary lymphoid organs (lymph nodes, spleen), mucosal and cutaneous lymphoid tissues*

Antigens and naive T cells come together in the same organs

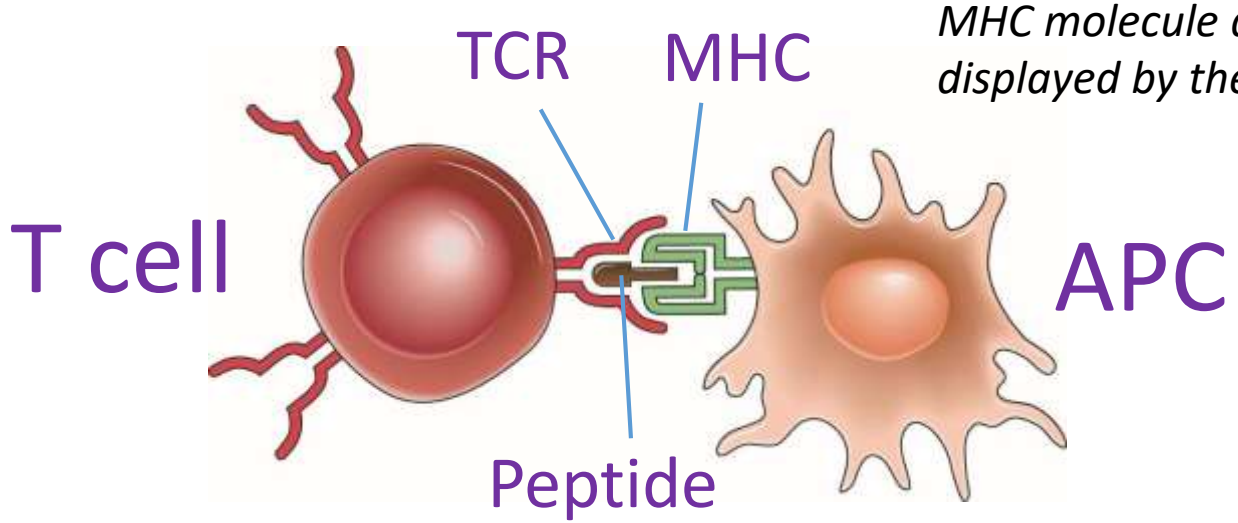
Why are dendritic cells the most efficient APCs for initiating immune responses?

- Location: at sites of microbe entry (epithelia), tissues
- Receptors for capturing and reacting to microbes: Toll-like receptors, other receptors
- Migration to T cell zones of lymphoid organs
  - Role of CCR7
  - Co-localize with naïve T cells
- Practical application: dendritic cell-based vaccines for tumors

Take home messages

What do T Cells Recognize?

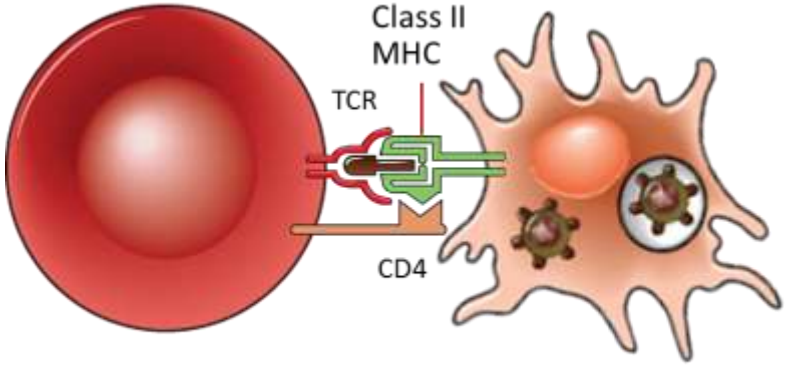
Most T cells only recognize peptides bound to **Major Histocompatibility complex (MHC)** molecules on the surface of other cells called **antigen presenting cells (APCs).**



The TCR binds simultaneously to an MHC molecule and a peptide displayed by the MHC molecules.

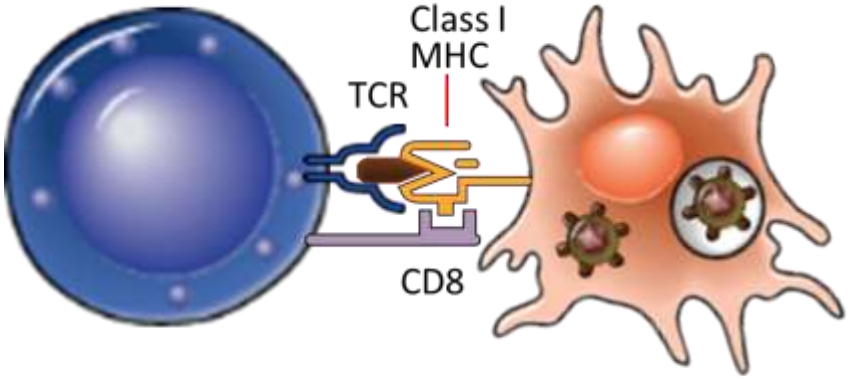
CD4+ and CD8+ T cells and MHC Class Restriction

CD4+ T cells (helper T cells)



CD4+ T cell recognition is class II MHC restricted

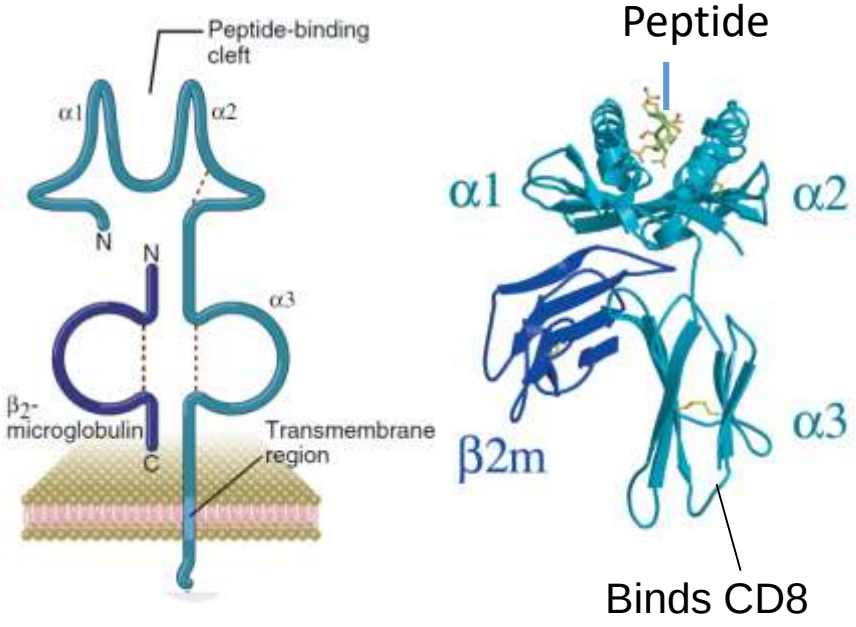
CD8+ T cells (cytotoxic T lymphocytes)



CD8+ T cell recognition is class I MHC restricted

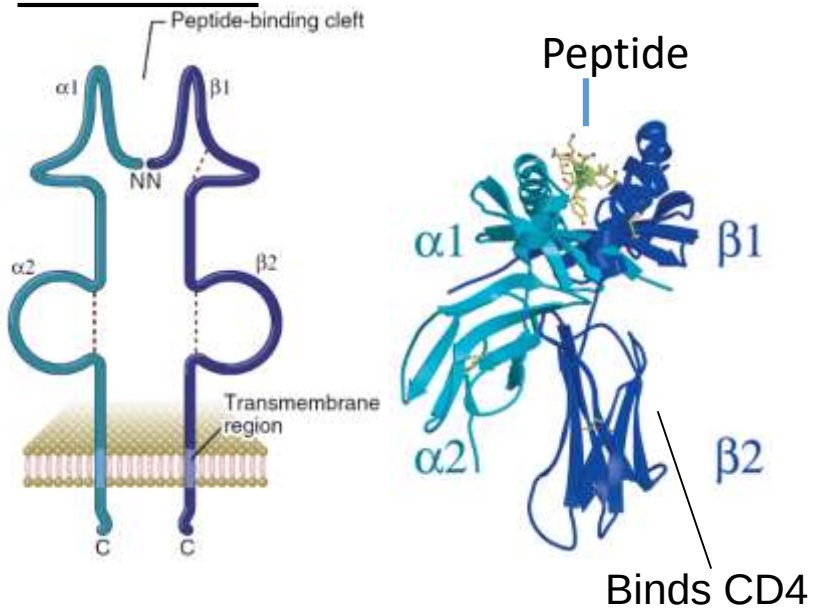
Human MHC (HLA) molecules

Class I



HLA-A
HLA-B
HLA-C

Class II



HLA-DP
HLA-DQ
HLA-DR

All MHC molecules have a similar basic structure: the cleft at the N-terminal region binds peptide antigens and is recognized by T cell receptors and the membrane-proximal domain binds CD4 or CD8 .

Abbas, Lichtman, Pillai. Cellular and Molecular Immunology. Elsevier. 2017

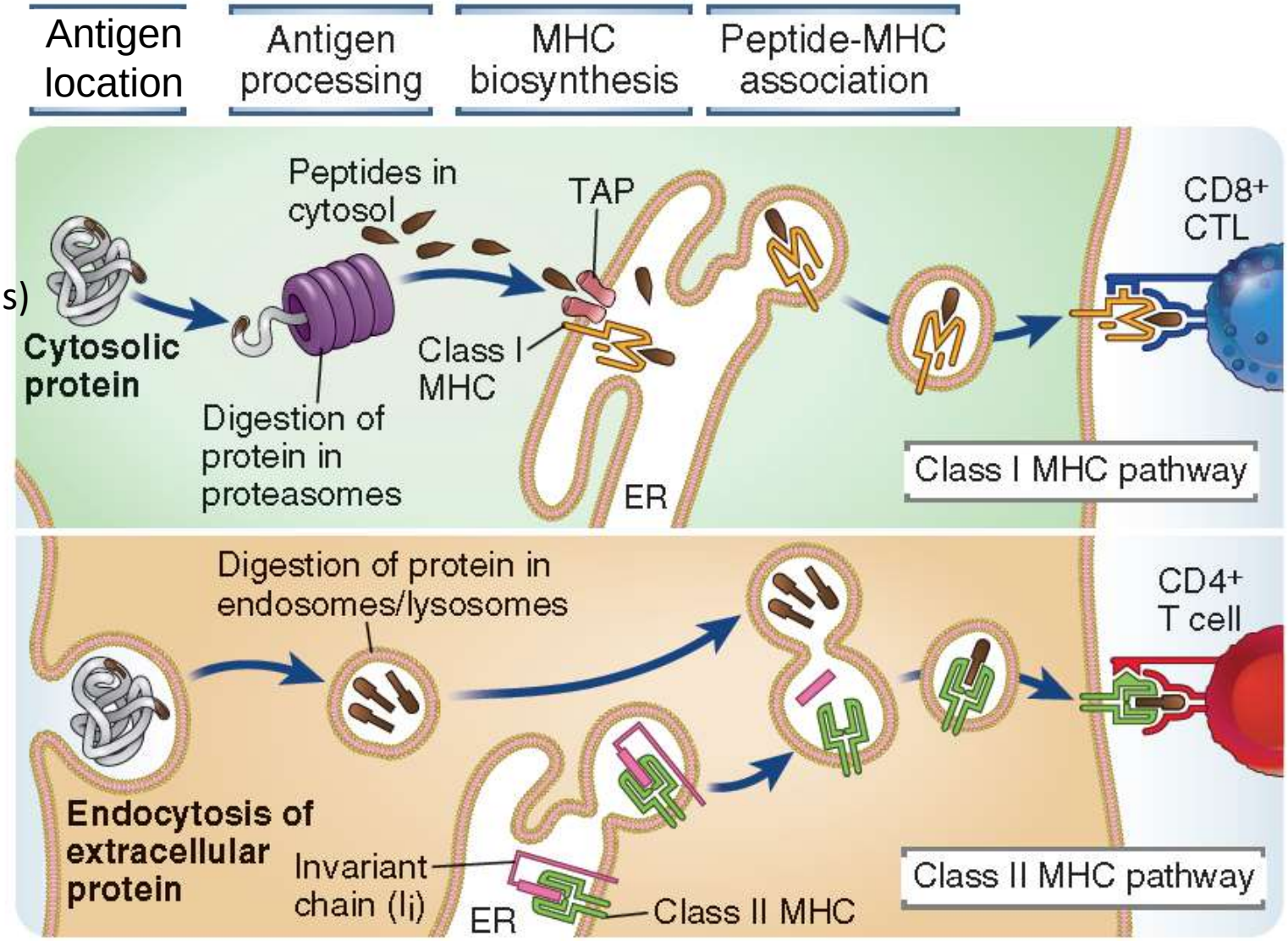
What is the Significance of Class II or Class I MHC Restriction of CD4+ and CD8+ T cells?

- **Lymphocytes must respond to each microbe in ways that are able to eradicate that microbe**
  - **Extracellular microbes: antibodies; destruction in phagocytes (need helper T cells)**
  - **Intracellular microbes (those that survive and reproduce inside our cells) : killing of infected cells (need CTLs)**
  - **T cells distinguish antigens in different cellular locations on the basis of class II vs. class I MHC**
- **Class II and Class I MHC molecules mainly present peptides from extracellular vs. intracellular microbes, respectively**
  - **This is based on antigen processing pathways**

Where do the MHC-binding Peptides Come From?

Viral proteins
(Tumor antigens)

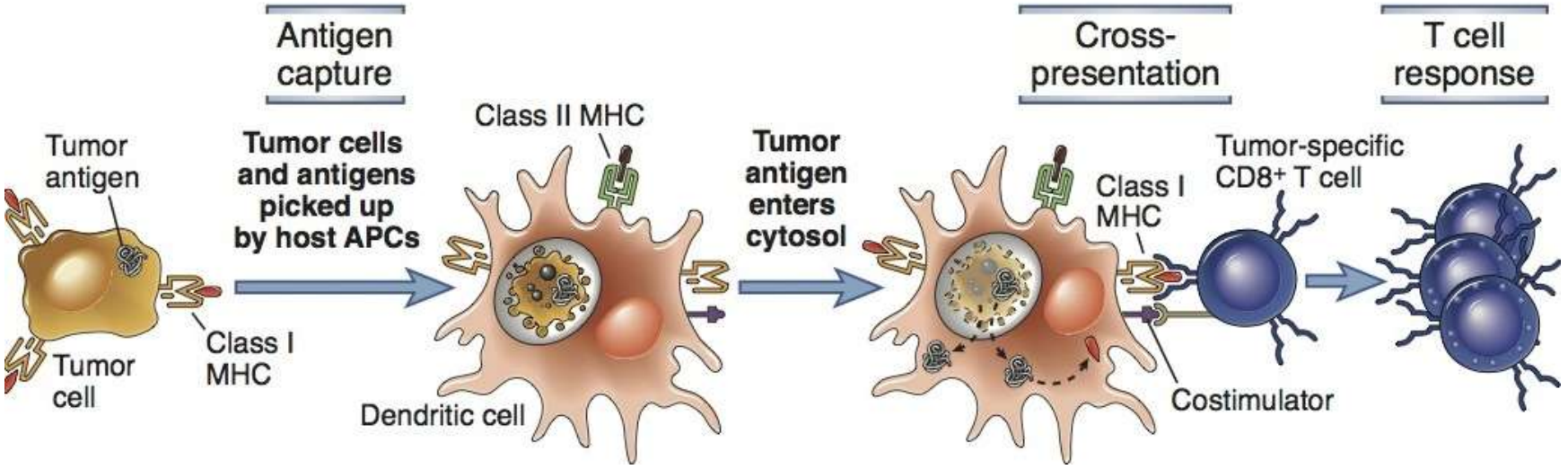
Extracellular
microbes



Abbas, Lichtman, Pillai. Cellular and Molecular Immunology. Elsevier. 2017

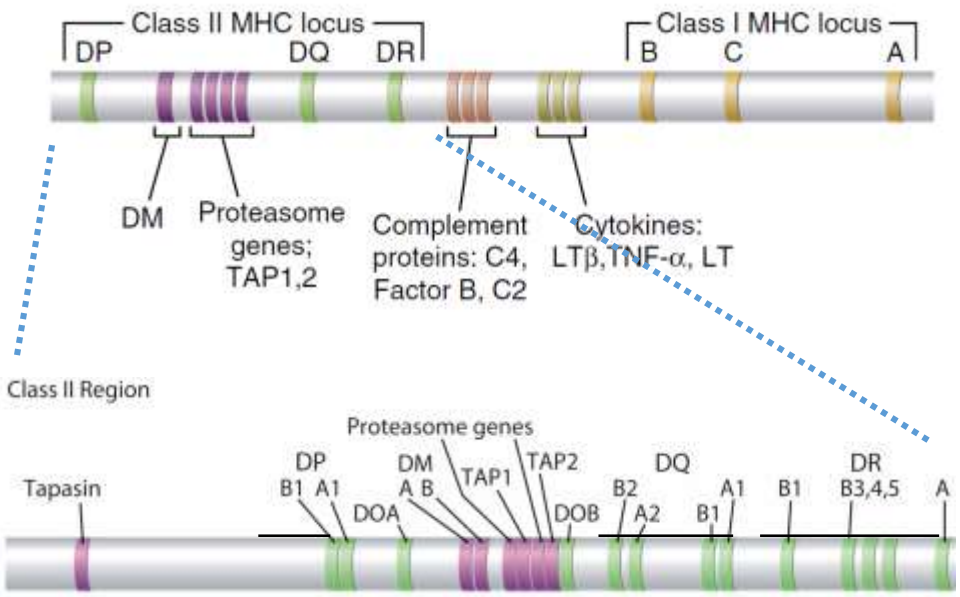
Cross-presentation

- Naive CD8+ T cells specific for tumor antigen need to be activated by DCs.
- Antigens taken into DCs by phagocytosis would typically be processed by the class II MHC pathways, for CD4+ T cell activation.
- But tumor antigens (also viral antigens) taken into phagosomes of DCs *can be* ***delivered to the cytosol*** for access to the class I MHC antigen processing pathways



Polymorphism of HLA genes

Human MHC : Chromosome 6

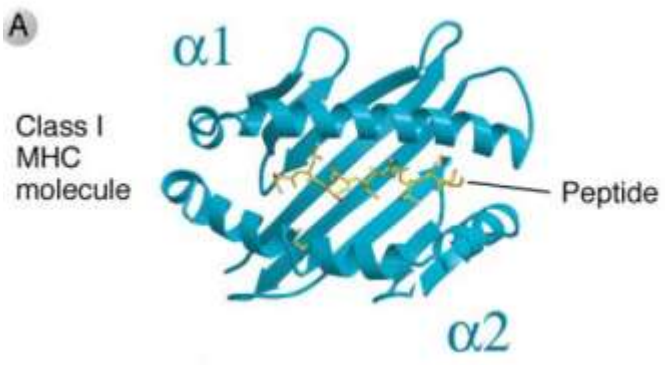


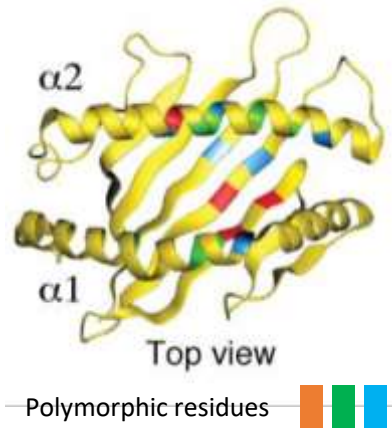
Locus	Number of alleles
A	2579
B	3285
C	2133
DRA	7
DRB	1512
DQA1	51
DQB1	509
DPA1	37
DPB1	248
Total	**10533 !!!**

- Most polymorphic genes in biology
  - Large number of variants (alleles) in the population

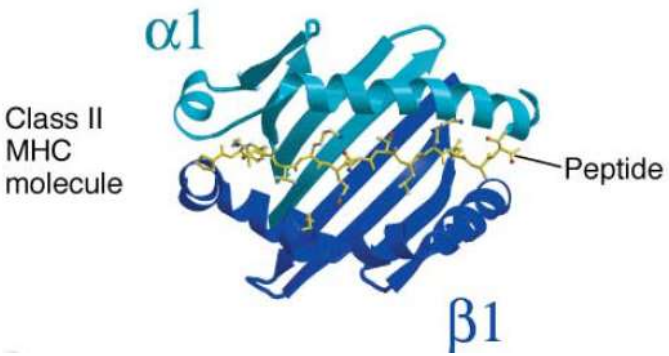
Human MHC (HLA) molecules

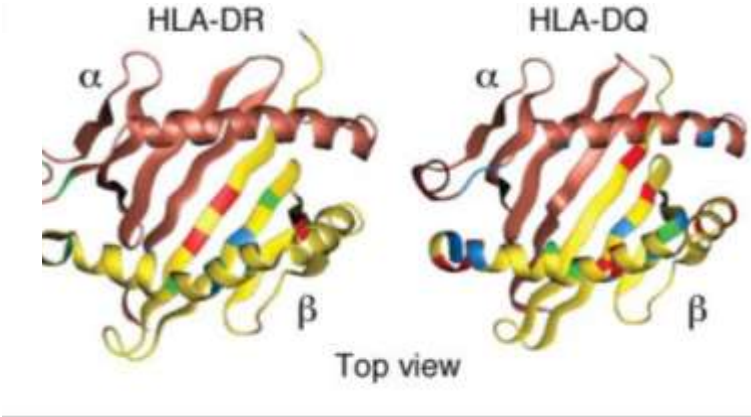
Class I





Class II

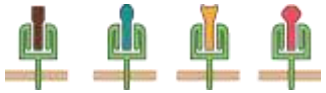
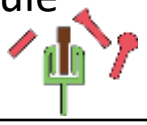




- *The polymorphic amino acid residues are all in the peptide binding grooves*
- *Different people will recognize different (but overlapping) sets of peptides*

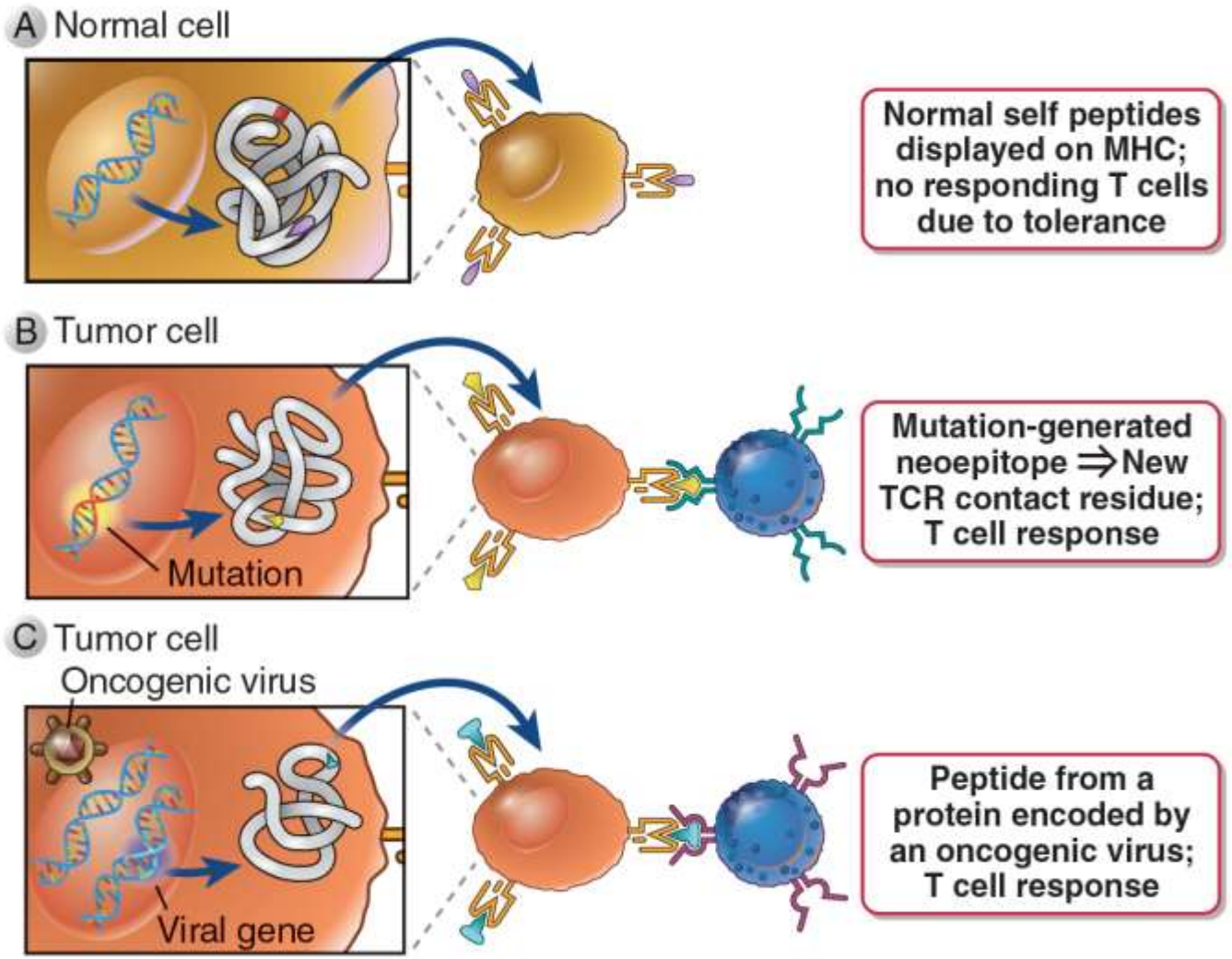
Abbas, Lichtman, Pillai. Cellular and Molecular Immunology. Elsevier. 2017

Peptide Binding Properties of MHC Molecules-1

Property	Significance
Broad specificity	Many different peptides can bind to the same MHC molecule 
Each MHC molecule displays one peptide at a time	Each T cell responds to a single peptide bound to an MHC molecule 
Class I vs. class II MHC bind different size peptides. Class I: **8-9** amino acids Class II: **10-30** amino acids	Class I and Class II MHC bind different peptides from same proteins. CD4+ and CD8+ T cells respond to different peptides from same protein
Peptides bind to MHC using 1 or 2 anchor residues, i.e. amino acid residues whose side chains fit into pockets in cleft floor	Only one or two amino acid residues determine if a peptide can bind to a particular MHC molecule; therefore many different peptides can bind any one MHC molecule

Abbas, Lichtman, Pillai. Cellular and Molecular Immunology. Elsevier. 2017

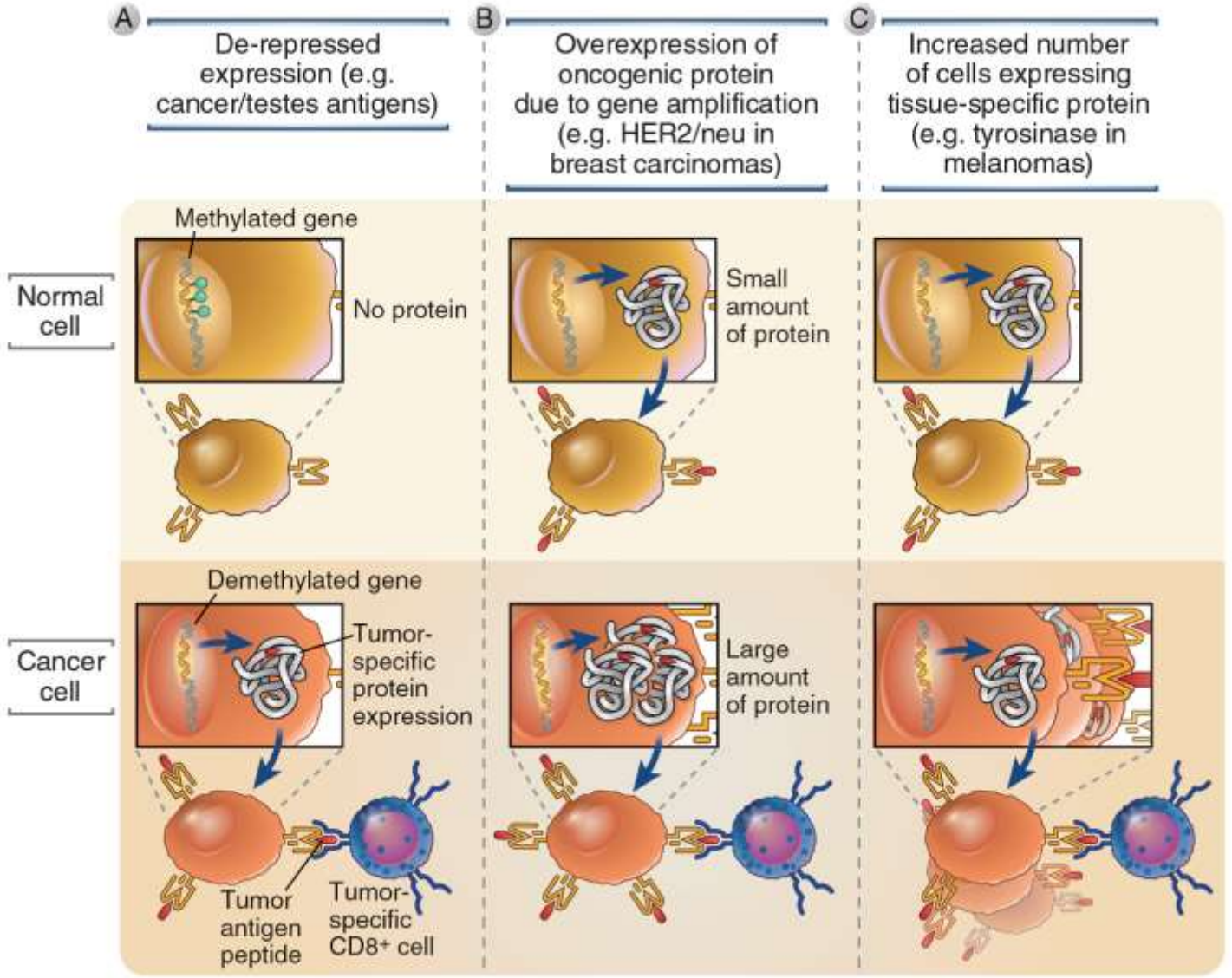
Cancer Patients' T cells Respond to Tumor Specific Antigens Derived from Mutated Proteins (neoantigens) and Oncogenic Viruses



Most cancer T cell antigens are generated by random mutations in genes whose function is unrelated to malignant phenotype. More mutations generates more neoantigens, and more T cell clones activated.

Abbas, Lichtman, Pillai. Cellular and Molecular Immunology. Elsevier. 2017

Cancer Patients' T cells Respond to Unmutated Protein Antigens



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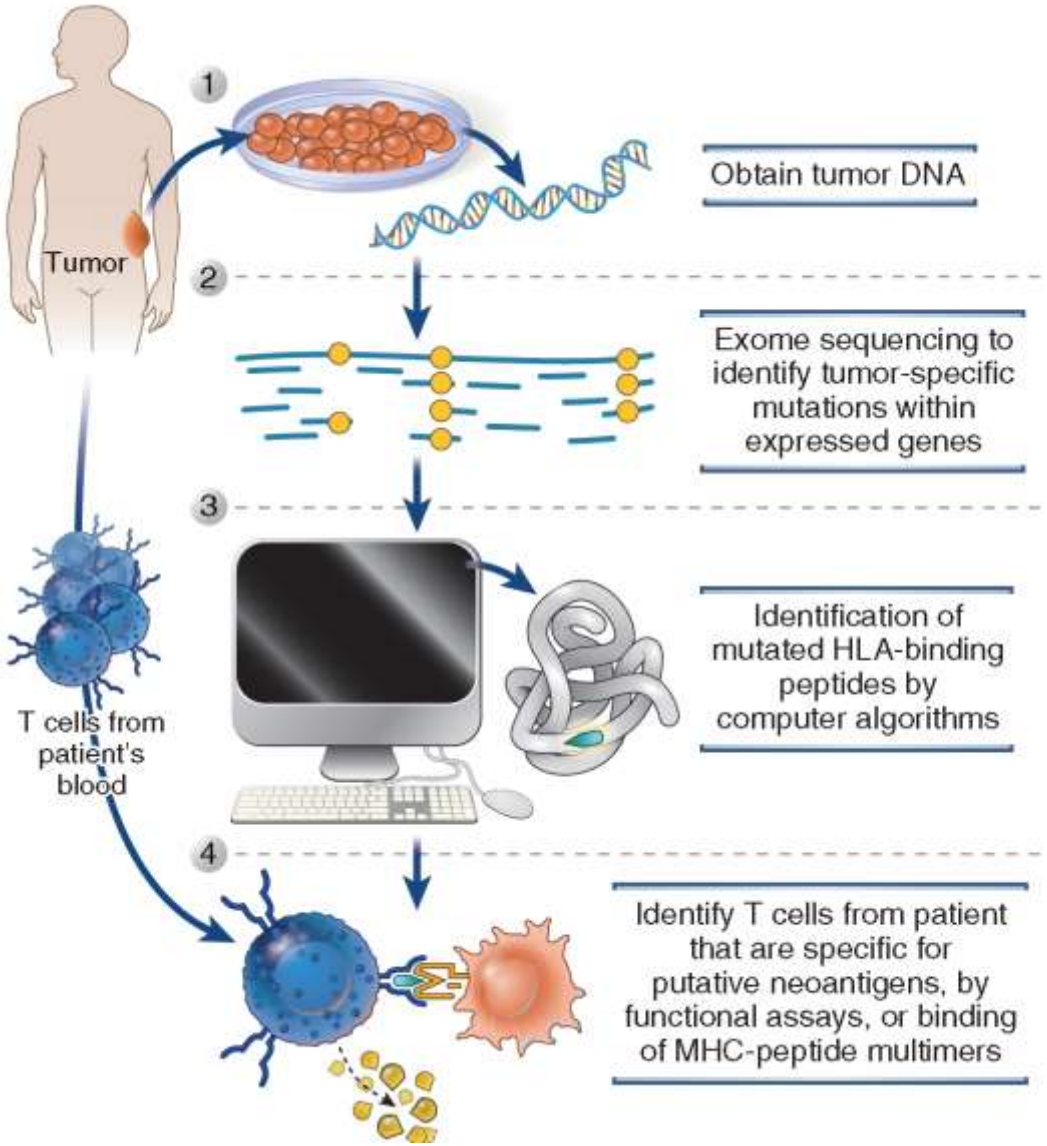
Identifying Mutant Tumor Peptides That Bind MHC Alleles for Personalized Tumor Vaccines



Personalize for HLA alleles of the patient

Relevance of MHC Polymorphism and T Cell MHC Restriction to Immunotherapy

- *"Intelligent design" of peptide vaccines against tumors*
- *Tumor vaccines composed of mutant tumor peptides will have to be personalized to ensure peptides bind to a particular patient's MHC alleles*
- *The tumor antigen receptors used in adoptive T cell approaches (e.g. CAR T cells) cannot be TCRs, in order to be widely applicable to many patients*

Take Home Points