Immunology of the GI Tract
a brief overview

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Objectives

- Understand the organization of the intestinal immune tissues
- Understand the immune cell populations and their distribution in the intestinal tract
- Understand the role of sIgA to immune barrier function and regulation
- Understand how oral tolerance develops
- Understand the the importance of the intestinal flora in immunity
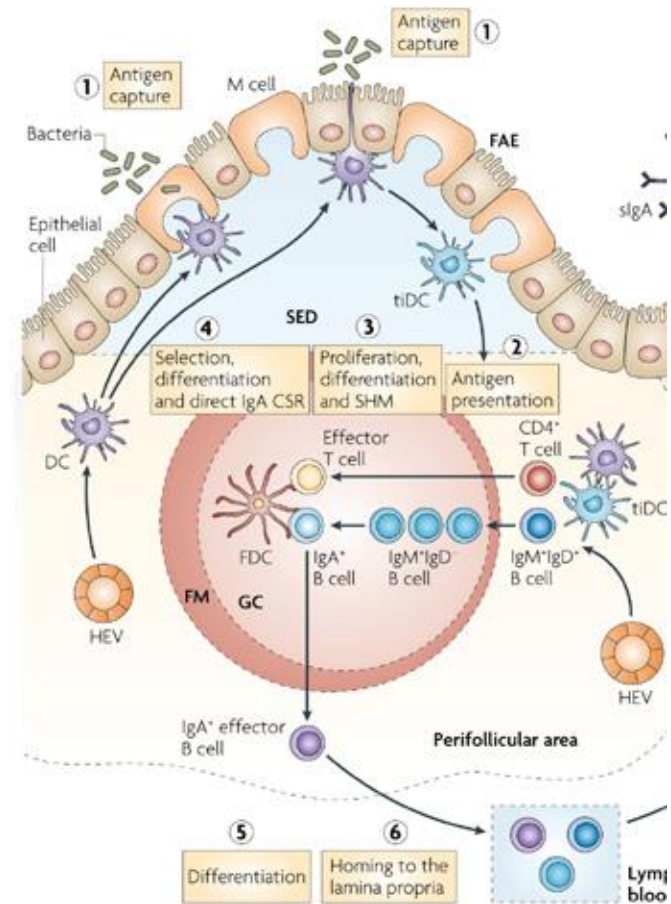
GALT

- The gastrointestinal associated lymphoid tissue (GALT) serves its function as
  - a barrier against pathogen entry and spread within the host,
  - an inductive site for innate and adaptive immune responses
  - site for immune cell expansion and survival

GALT sites of immune induction

Antigens transported by EC and captured by DC are presented to naïve T and B cells inducing their activation, proliferation, and differentiation in the:

- Peyers Patches
- Microfold cells (M cells)
- Mesenteric Lymph nodes
- Isolated lymphoid follicles (ILF)
- Cryptopatches

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GALT sites for effector immunity

- Following activation, effector cells selectively up-regulate chemokine and adhesion molecules for homing to the GIT mucosa
  - CCR9: binds CCL25 which is only expressed by EC in the GIT and thymus
  - A4β7: binds MADCAM-1 expressed by mucosal endothelial cells
- The tissues where effector immune function occurs are:
  - The lamina propria
  - The intra-epithelial cell compartment
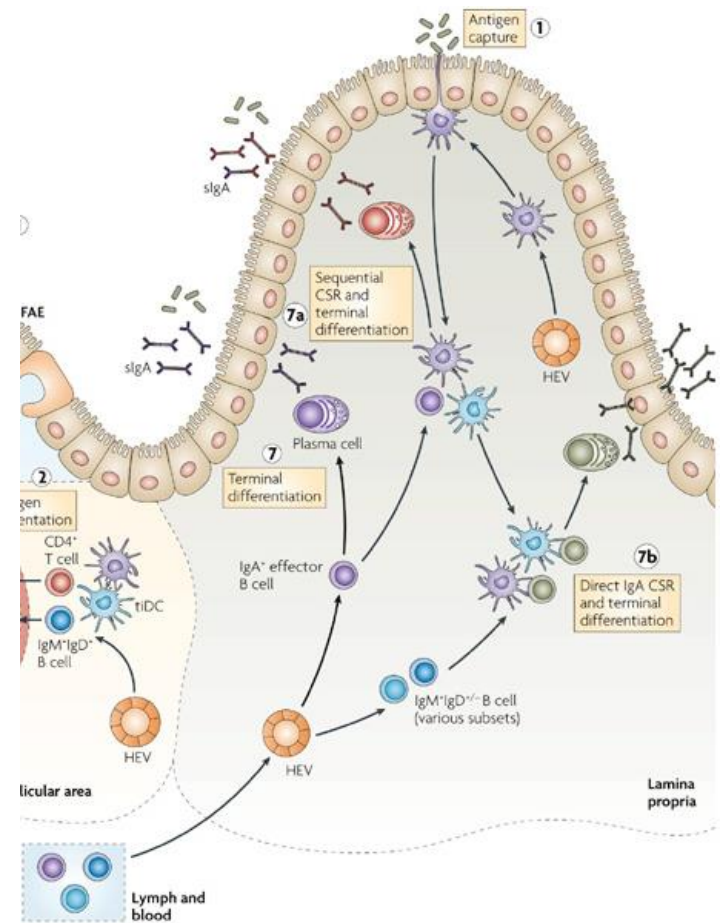
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GALT Effector Cells

Lamina Propria Cells

- Immunoglobulin A (IgA) secreting plasma cells
- Activated B cells
- T cells (CD4+>>CD8+)
- macrophages
- dendritic cells (DCs)
- innate lymphoid cells (ILCs)

Intraepithelial Cells

- Function based upon distribution along the length of the intestinal tract
- Exhibit an activated "antigen experienced" phenotype
- Majority are CD8$\alpha\alpha^+$ and TCR$\gamma\delta^+$ T cells

Epithelial Cells

- provide both a physical and immune barrier in the GIT

- at the "front line" of immune recognition in the GIT

- express extra and intracellular pattern recognition receptors (PRR) for the pathogen associated molecular patterns (PAMPs) expressed by bacteria and viral species

PRRs and PAMPs

- MDP= muramyl dipeptide
- DAP= D-glutamyl-meso-diaminopimelic acid
- Not shown:
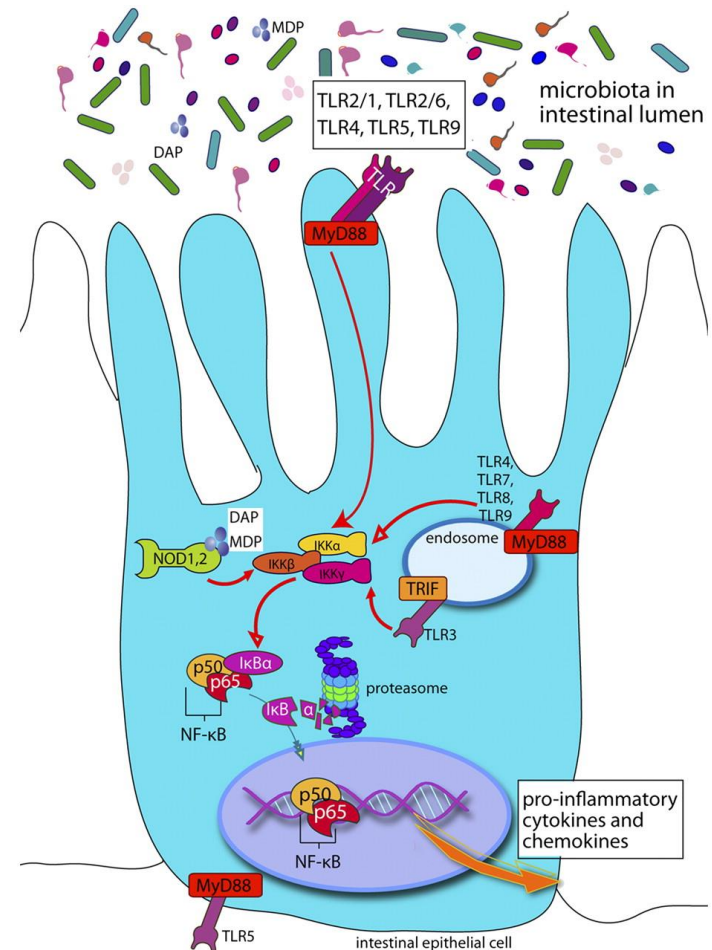 - The RIG-I intracellular receptors bind viral dsRNA
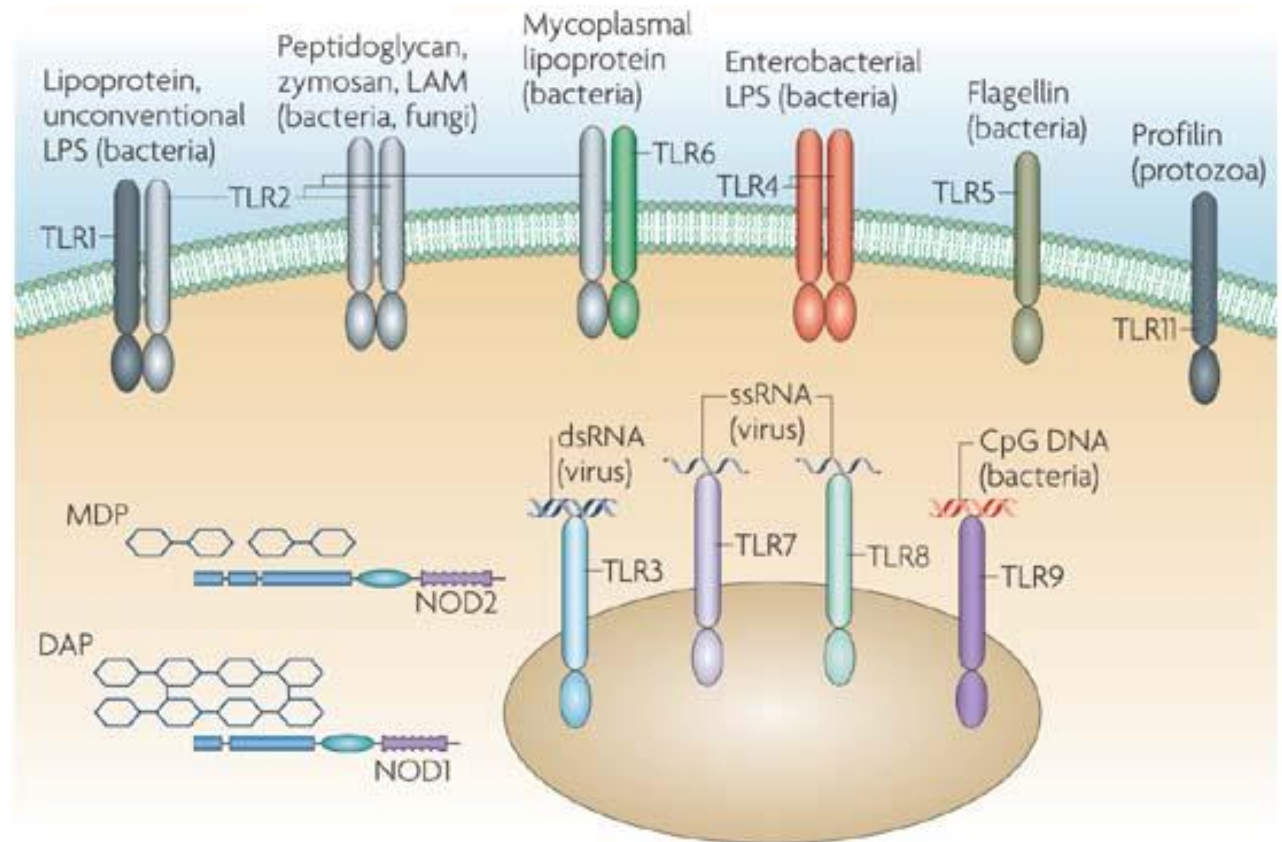
GIT epithelial cells recognize microbes binding to pattern recognition receptors

- Engagement by PAMPs triggers intracellular signaling cascade

- Subsequent gene transcription and production of cytokines & chemokines

- Recruit immune cells to fortify the EC response and limit microbial expansion or invasion

- Responding immune cells from the GALT include innate effectors (neutrophils, macrophages, dendritic cells, eosinophils, mast cells) and adaptive effectors (T cells and B cells)

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IgA

- IgA is the most abundant antibody isotype in the serum and in mucosal sites
  - 40 mg/kg or ~3 g is produced in an adult human per day!
- Contributes to maintenance of intestinal epithelial barrier function
- Requires epithelial pIgR expression and transcriptional regulation of J-chain production

J= Joining chain, required for EC transport of IgA dimers
SC= secretory chain, prevents proteolytic damage of IgA

http://www.buddycom.com/cells/immun/igxara/index.html
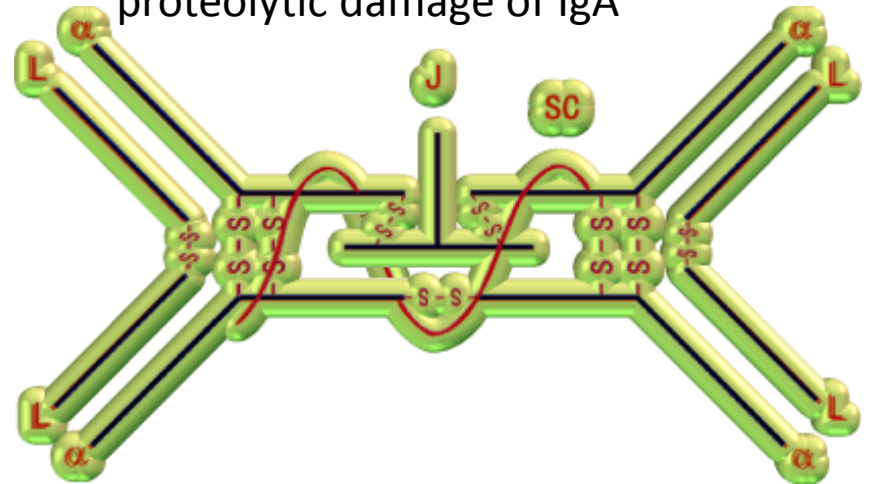
Functions of IgA
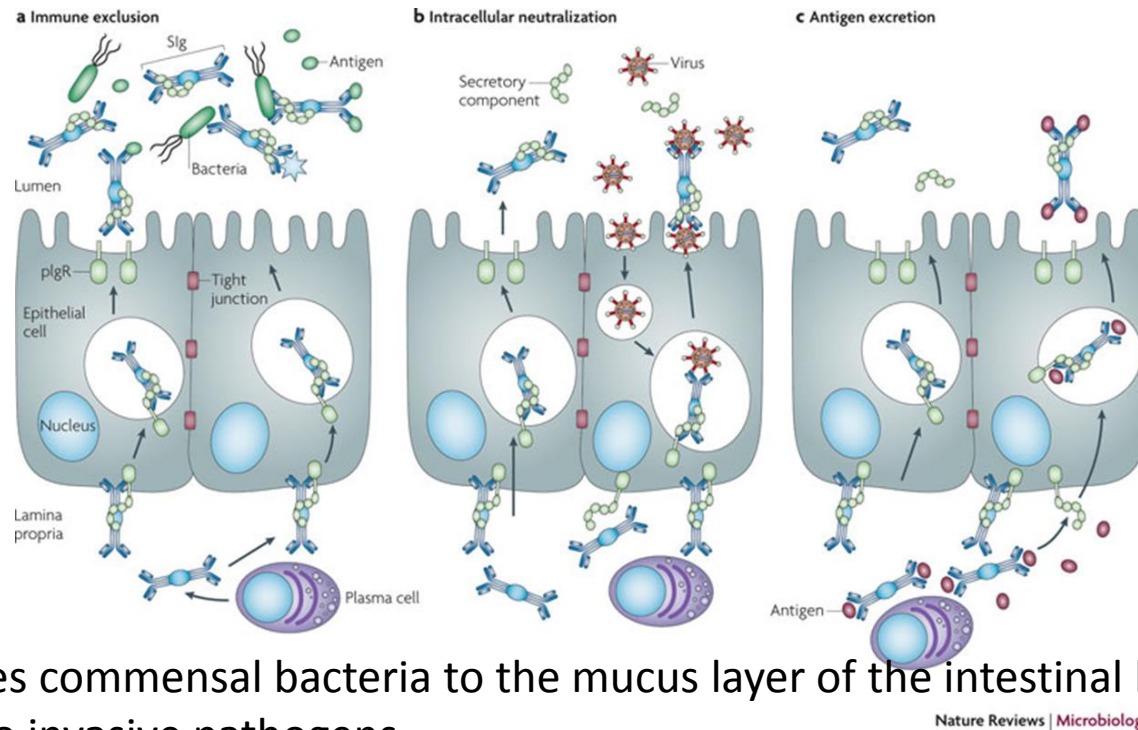
- Confines commensal bacteria to the mucus layer of the intestinal lumen
- Binds to invasive pathogens
- Neutralizes microbial toxins and other inflammatory microbial products
- Neutralization of antigens and pathogens in epithelial cell endosomes
- Uptake of luminal antigens
- Transport of antigens from the LP into the lumen

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IgA Deficiency
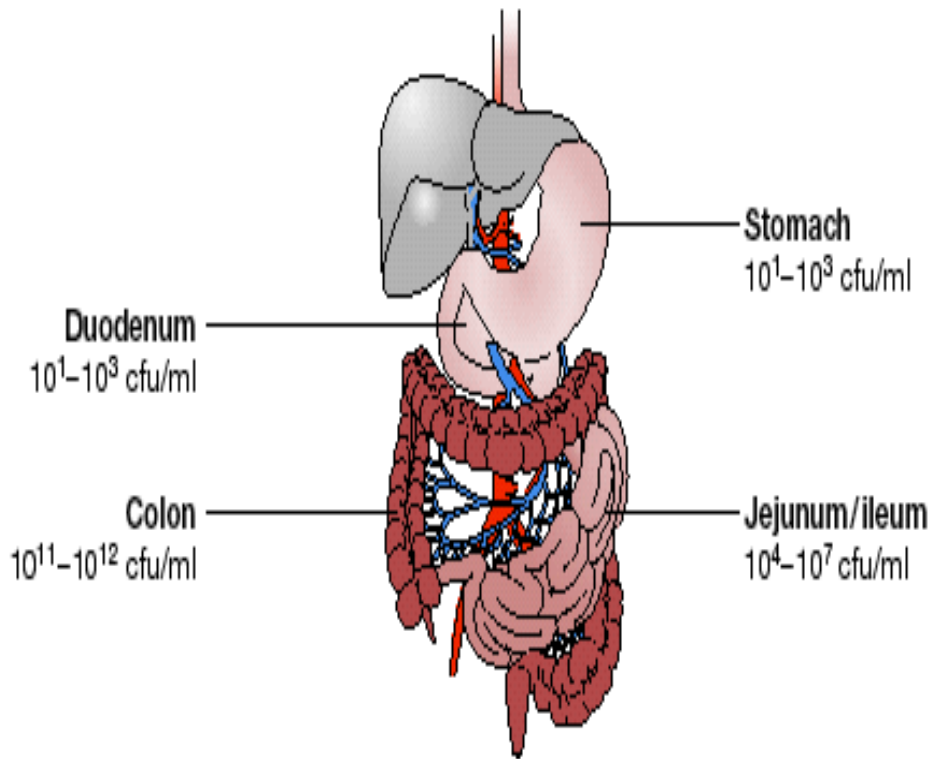
- Deficiency is not severely detrimental
- Compensation by other antibody isotypes
  - intestinal sIgM also binds pIgR for translocation into the intestinal lumen and other innate pathways
- However, sIgA deficiency may influence the development of autoimmunity and allergy
  - limits innate immune responses,
  - modulates intestinal regulatory T cells,
  - regulates the commensal flora
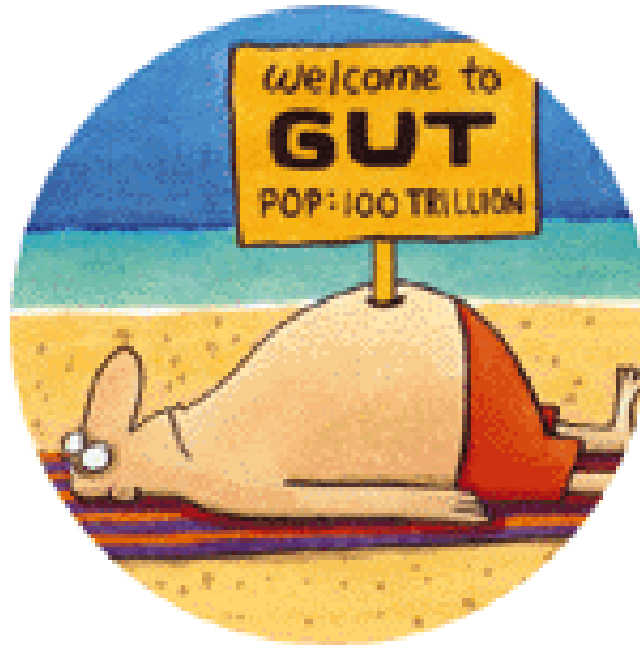
- We are a "super-organsim"
- The human gastrointestinal tract hosts a vast microbiome
  - >1000 different species; **>majority non-culturable**
- Establishes during the first week and stabilizes within the first 3-5 years of life
- Cooperative relationships have established through co-evolution to maintain homeostasis

The GIM a diverse community

Combined genome 150 fold size of
human genome

Stomach
10^1–10^3 cfu/ml

Duodenum
10^1–10^3 cfu/ml

Colon
10^{11}–10^{12} cfu/ml

Jejunum/ileum
10^4–10^7 cfu/ml

Bacteria (~1000 species)

- Firmicutes (*Ruminococcus, Clostridium, Eubacteria*)
- Bacteroidetes *(Bacteroides)*
- Actinobacteria (*Bifidobacteria*)
- Proteobacteria (*Enterobacteraceae*)
- Fusobacteria

Viruses

Fungi

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The gut flora as a forgotten organ, p688-693, Figure 1. Copyright (2006)
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Immune tissue development requires normal gut colonization
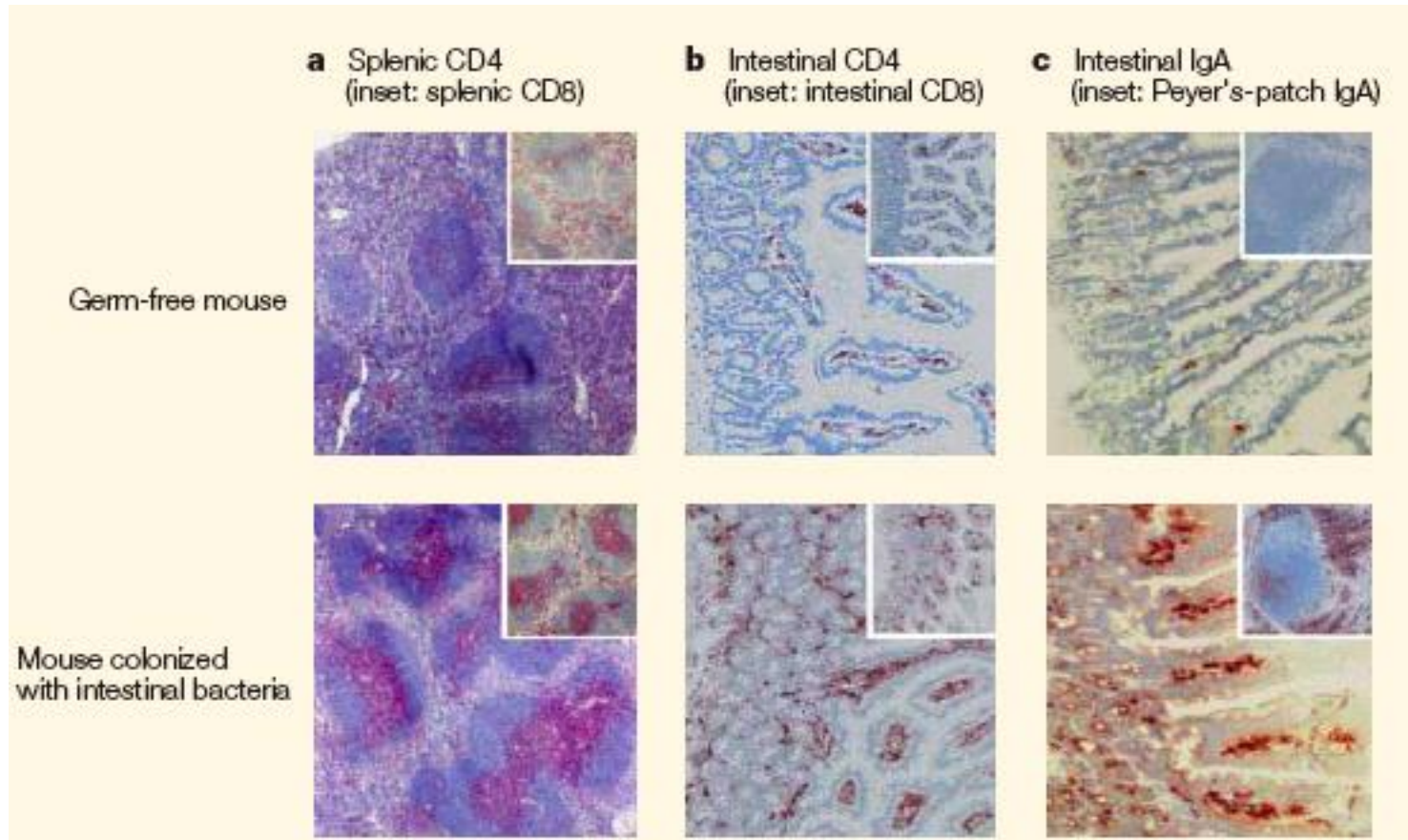
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Oral tolerance

- Induction of mucosal and systemic non-responsiveness to dietary antigens
- Active immune response that depends upon
  - tolerogenic dendritic cells (make IL-10),
  - $CD4^+CD25^+T$ regulatory cells (make TGFβ & IL-10)
- Mechanisms of tolerance:
  - Ignorance (no/low costimulatory signals)
  - Inhibition/Anergy (inhibitory signals)

Uptake of food antigens occurs via

- Transcytosis through M cells,
- Paracellular diffusion or transcytocis across intestinal villi ECs,
- Uptake of exosomes (MHC Class II loading compartments fused with endosomes) by lamina propria DC,
- Luminal capture by CXCR31^{high} DC/macrophages.

GIT mucosal defenses balance immune responsiveness to its flora

- Commensal non-pathogenic & pathogenic gut microbial species are recognized by pattern recognition receptors (e.g. TLR, NOD)
- Several mechanisms are involved in attenuating inflammatory responses against the commensal flora:
  - Different levels and cellular distribution of TLRs on epithelial cells (EC)
    - NOD2 modulation of TLR signaling (*thus mutations of NOD2 may contribute to IBD*)
  - Containment of luminal flora and defense against microbial translocation
    - Mucin production
    - Elaboration of paneth cell defensins
    - IgA secretion
  - Commensal bacteria mediated inhibition of NF-kβ signaling (*likely through soluble mediators released by bacteria*)
  - Inhibition of innate and adaptive immune responses by
    - TSLP (thymic stromal lymphopoietin) production by enterocytes
    - Regulatory T cells
    - Tolerogenic dendritic cells

Dysbiosis

- Generally describes the intestinal flora when it has been altered from a healthy equilibrium or baseline
- Combined with other environmental and genetic influences likely contributes to the development of intestinal and systemic diseases:
  - IBD
  - Diabetes
  - Metabolic syndrome and obesity
  - Food allergy

Dysbiosis and IBD
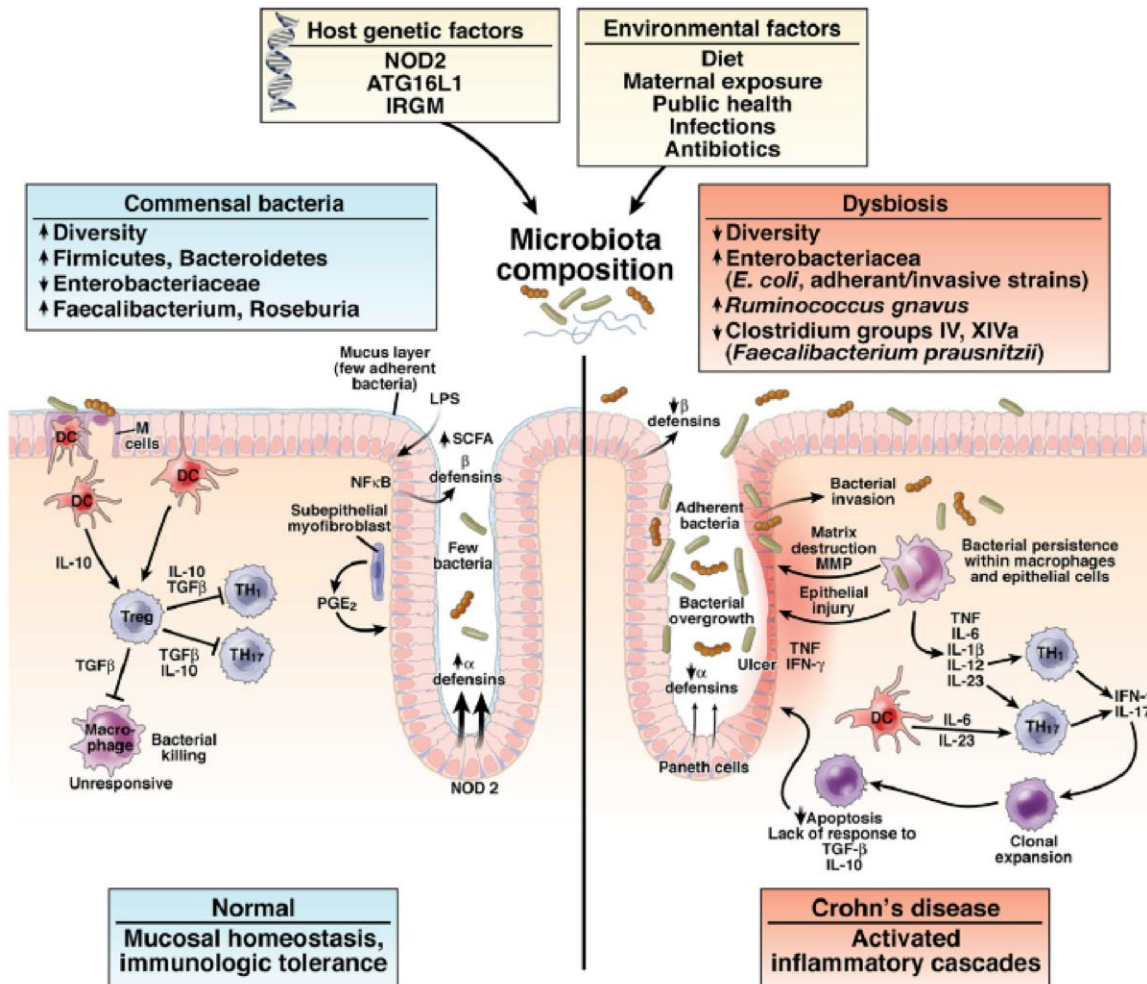
Summary Points

- The goal of GIT immunity is to recognize and maintain a healthy microbiome while also recognizing and expelling microbes that cause disease.

- GIT immune function derives from the immune cells and their interactions within the gastrointestinal associated lymphoid tissue, or **GALT**.

- The GALT includes **innate** immune function, provided by cells that quickly respond to threats and re-establish equilibrium.

- The GALT also includes **adaptive** immune function, provided by cells that respond to threats and generate a mucosal immune response that is specific and robust.

- **Cross talk** between innate and adaptive systems is crucial for avoiding unnecessary or ineffective inflammatory responses to ingested antigens and the re

Summary Points- 2

- **Immune induction** occurs in peyers patches and mesenteric lymph notes. Specialized cells in the mucosa overlying the patches are integral participants in the adaptive immune response.

- Immune **effector function** occurs in the lamina propria, and includes specific lymphocytes and plasma cells that product IgA.

- **IgA** serves to neutralize toxins and invasive organisms while also maintaining keeping commensal organisms in check.

Summary Points-3

- GIT microbiome has evolved to benefit us, and we have evolved to benefit its constituents.

- Derangements in the GIT microbiome are associated with inflammatory, allergic, and metabolic diseases.

- Oral tolerance of ingested protein is established by interaction between particular antigen presenting cells and modulatory T cells. Derangements can result in allergy.

Case #1

- 12 year old male presents with a one year history of
  - Stunted growth
  - Diarrheal stools
  - Occasional oral apthous sores
- What is the likely diagnosis?
- What deficiency may be noted on histologic examination of ileal biopsies?
- Which identified PRR or cytokine gene mutations could contribute to his diagnosis?

Case #2

- 3 year old toddler develops lip swelling, shortness of breath, and blotchy skin rash a few hours after eating a strawberry

- What is the likely diagnosis? And what immune process has broken down?

- What maternal factors would protect against development of this diagnosis?

- Which mucosal immune compartment and cell population contributes to regulated immunity against the strawberry?

Case #1

- 12 year old male presents with a one year history of
    - Stunted growth
    - Diarrheal stools
    - Occasional oral apthous sores
- What is the likely diagnosis? – *Crohn's Disease*
- What deficiency may be noted on histologic examination of ileal biopsies? *Paneth Cells*
- Which identified PRR or cytokine gene mutations could contribute to his diagnosis? *NOD2, IL-10, IL23/Th17 axis. Defects in the expression of these gene products contribute to driving chronic mucosal inflammation by impairing host control of the intestinal flora (NOD2) or by resulting in over-exuberant inflammatory responses against relatively innocuous gut bacteria (IL23/Th17 axis) or by allowing normal inflammatory responses to proceed unchecked due to ineffective negative regulation (IL10).*

Case #2

- 3 year old toddler develops lip swelling, shortness of breath, and blotchy skin rash a few hours after eating a strawberry
- What is the likely diagnosis? *Food Allergy*
- And what immune process has broken down? *Oral tolerance*
- What maternal factors would protect against development of this diagnosis? *Breast feeding and transfer of maternal secretory IgA*
- Which mucosal immune compartment and cell population contributes to regulated immunity against the strawberry? *Mesenteric lymph nodes, Foxp3 T regulatory cells, and IgA B cells*

Please send any questions or comments to:

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  or

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