l'immunité épithéliale chez l'enfant ses implications cliniques

Université de Liège
Faculté de médecine
Professeur Oreste Battisti

Exemples d'applications potentielles

- Maladies inflammatoires du tube digestif
- Maladies inflammatoires de l'appareil respiratoire
- Déficits immunitaires
- Cancers
- …
- → Par quel mécanisme ?
- → Comment ?

1. Focus sur l'immunité

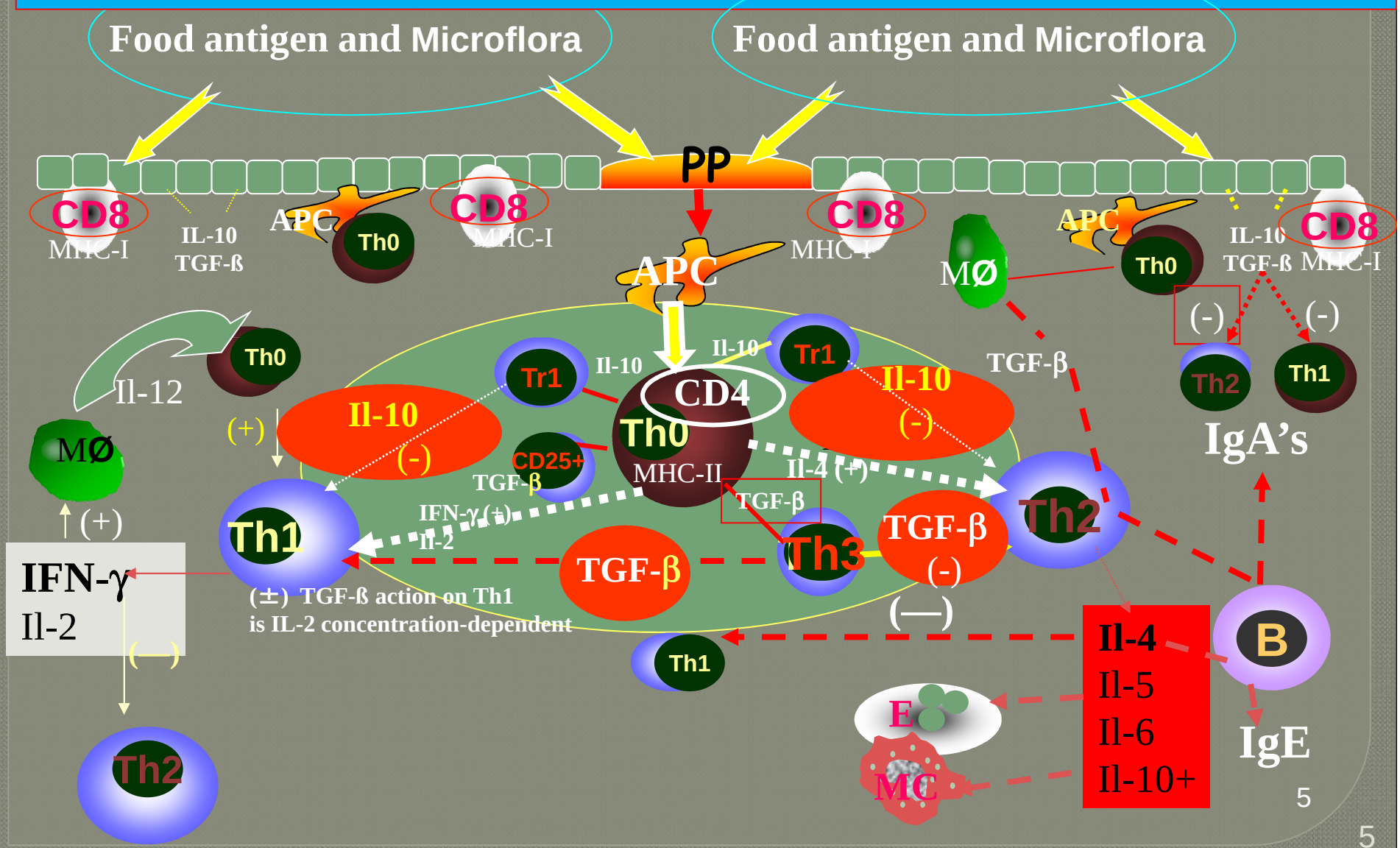
- Générale
- Épithéliale
- De l'appareil digestif
- De l'appareil respiratoire
- De l'appareil cutané

S’il faut Suivre un conducteur
Dans l’immunité épithéliale, la route
Semble fort complexe et compliquée.

Et si on « plonge » dans l’avenue, cela ressemble à ceci…

Food Antigen – Epithelial Cell – Microflora Interface :

Th1/Th2 optimal equilibrium and a high level of T-reg Cells activation process



5

Les postes frontières et le bureau d'immigration

- Le poste douane
- Les douaniers
- L'acceptation d'un visa de séjour
- Les bénéfices
- Le refus d'un visa de séjour
- Le retrait du visa de séjour
- Les sanctions

Et ici aussi, il y a douane et douane….


DOUANES FRANÇAISES


Douane Zoll Dogana
L'Auberson
Passage de la frontière autorisé :
avec documents de voyage reconnus et valables
marchandises :
RIEN À DÉCLARER


DOUANE

L'immunité:
innée et adaptative

- **« innée » (10 %):**

= Barrières anatomiques, cellules dendritiques, phagocytose, complément, interféron, CRP.

- **« adaptative »** .

En réponse lors d'une confrontation à des antigènes ou des provocateurs:

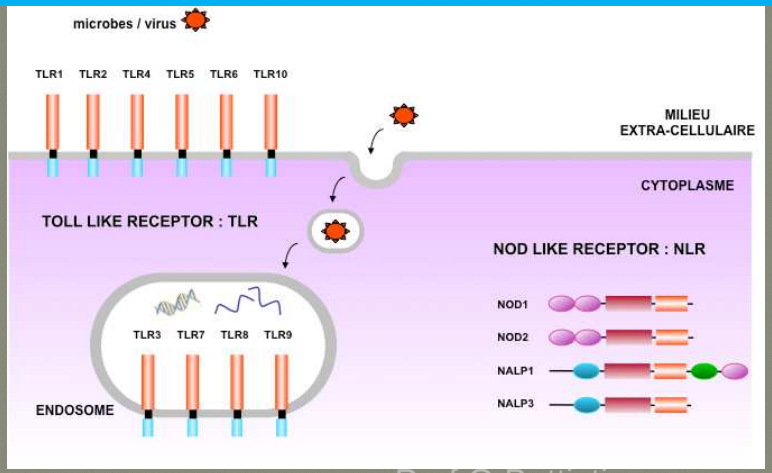
- Humorale (70 %)
- Cellulaire (20 %)

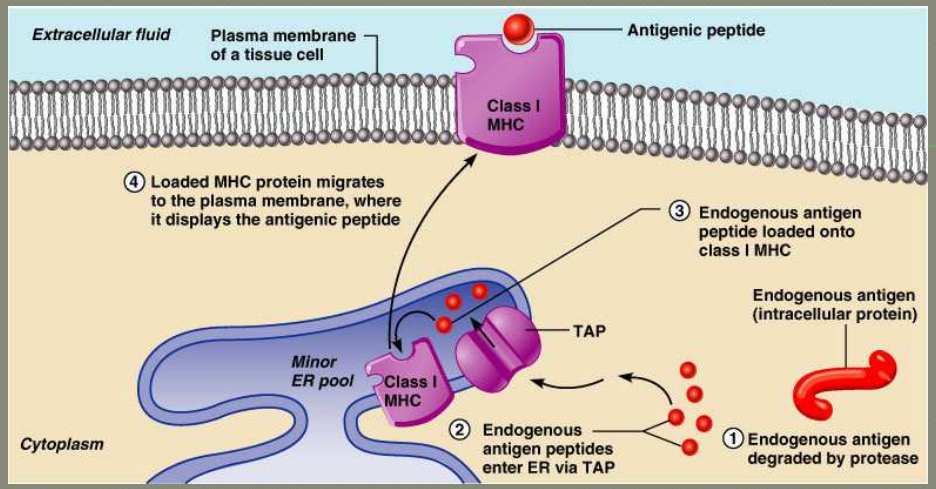
Les “barrières” épithéliales

- L’épithélium de la cavité utérine, le bouchon du col cervical, le trophoblaste
- Le liquide amniotique
- Peau: vernix caseosa, pH:3 à 5, sebum
- Muqueuse gastrique
- La Salive et les larmes,
- Epithelium et Mucus épithélial (voies urinaires, respiratoires et digestives), jonctions serrées, peptides antimicrobiens (défensines a et b, cathélicidines,…)

Les “Toll-like Receptors (TLRs)”

Toll = péage »





Il y a les "Class I and II MHC (self)" Protéines

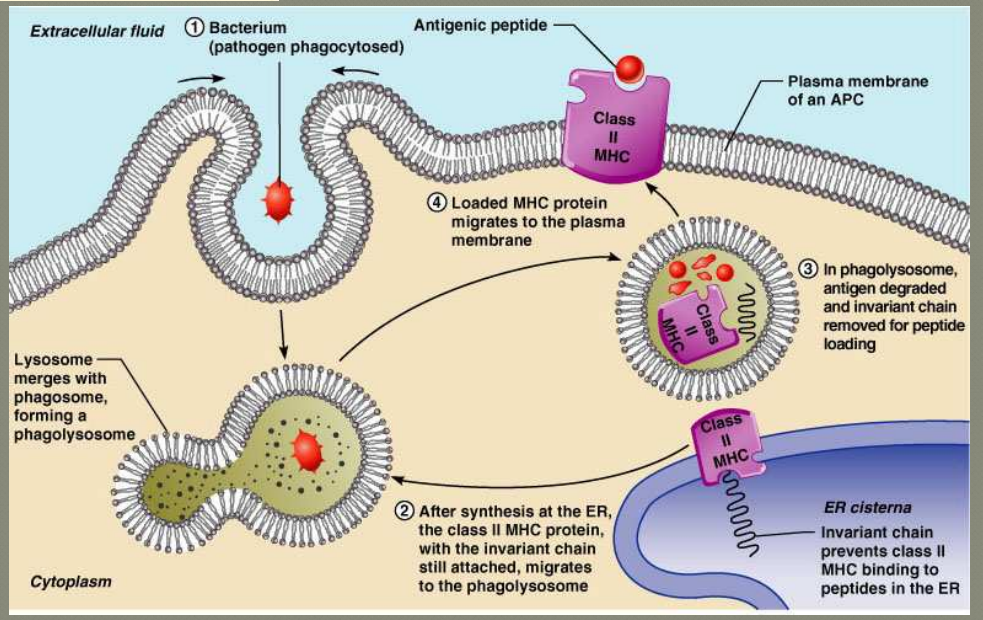


Figure 21.15a

Antigen Recognition and MHC Restriction

- Immunocompetent T cells are activated when the V regions of their surface receptors bind to a recognized antigen
- T cells must simultaneously recognize:
  - Nonself (the antigen)
  - Self (a MHC protein of a body cell)

Toll-like receptors in innate immunity

Functional characterization of Toll-like receptors (TLRs) has established that innate immunity is a skillful system that detects invasion of microbial pathogens. Recognition of microbial components by TLRs initiates signal transduction pathways, which triggers expression of genes. These gene products control innate immune responses and further instruct development of antigen-specific acquired immunity. TLR signaling pathways are finely regulated by TIR domain-containing adaptors, such as MyD88, TIRAP/Mal, TRIF and TRAM. Differential utilization of these TIR domain-containing adaptors provides specificity of individual TLR-mediated signaling pathways. Several mechanisms have been elucidated that negatively control TLR signaling pathways, and thereby prevent overactivation of innate immunity leading to fatal immune disorders. The involvement of TLR-mediated pathways in autoimmune and inflammatory diseases has been proposed.

TLR-mediated activation of innate immunity controls not only host defense against pathogens but also immune disorders.

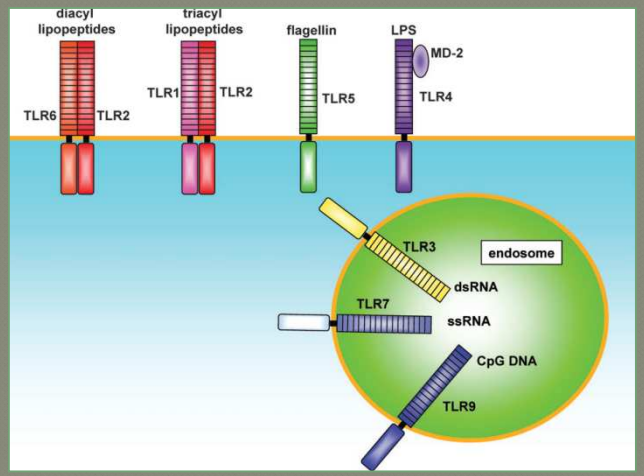
Innate immune: dendritic cells and M cells or macrophages

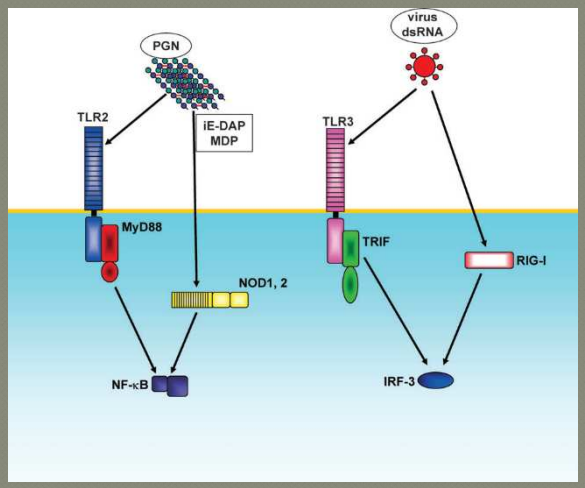
Phagocytosis of pathogens,

Presentation of pathogen-derived peptide antigens to naïve Tcells. TLRs recognize pathogen-derived components and induce expression of genes, such as co-stimulatory molecules and inflammatory cytokines. Phagocytosis-mediated antigen presentation, together with TLR-mediated expression of co-stimulatory molecules and inflammatory cytokines,

instruct development of antigen-specific adaptive immunity, especially Th1

the TLR family members recognize specific patterns of microbial components: peptoglycan, lipoprotein and RNA





Helper T Cells ($T_{H:\ TH17,\ldots}$)

- Regulatory cells that play a central role in the adaptive immune response
- Once primed by APC presentation of antigen, they:
  - Chemically or directly stimulate proliferation of other T cells
  - Stimulate B cells that have already become bound to antigen
- Without T_H, there is no immune response

Helper T Cells (T_H)

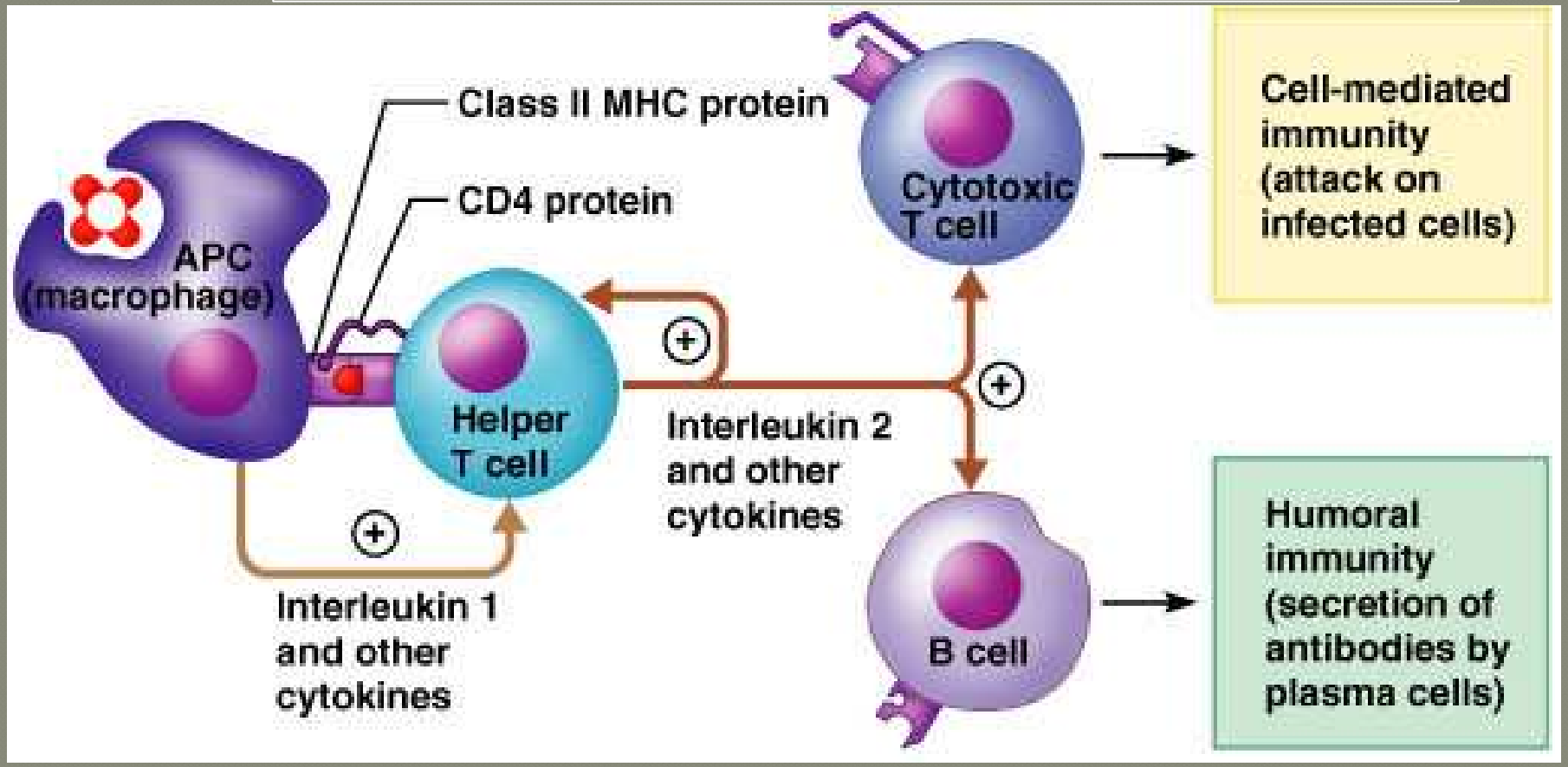


Figure 21.17a

Helper T Cells

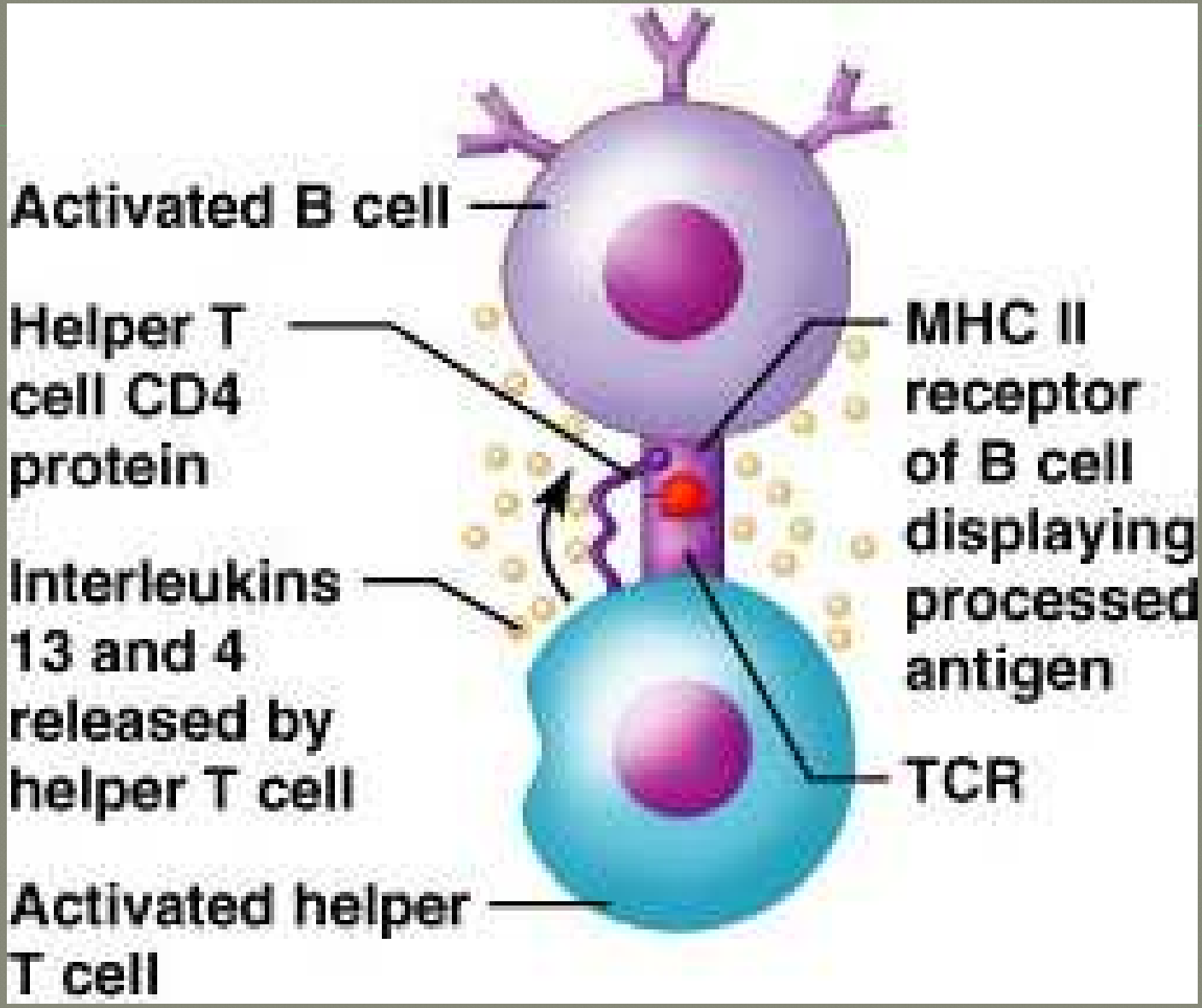


Figure 21.17b

Immunocompetent B or T cells

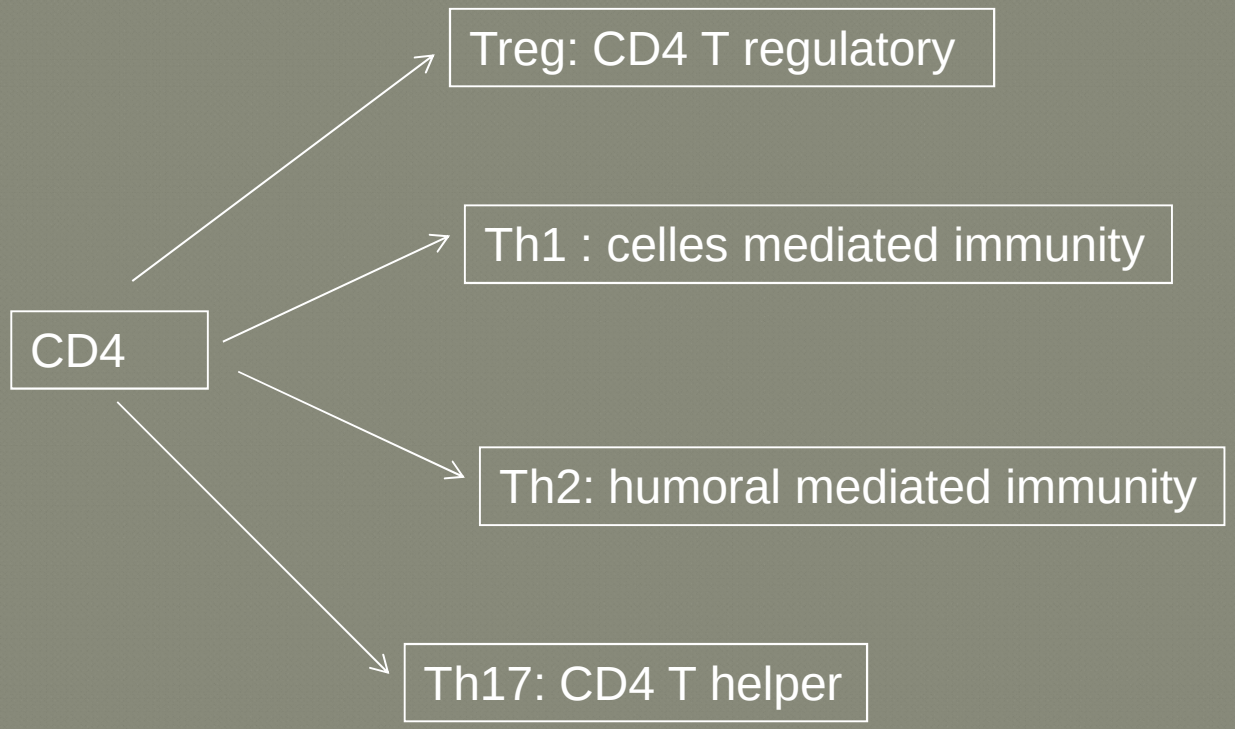
- Display a unique type of receptor that responds to a distinct antigen
- Become immunocompetent before they encounter antigens they may later attack
- Are exported to secondary lymphoid tissue where encounters with antigens occur
- Mature into fully functional antigen-activated cells upon binding with their recognized antigen
- It is genes, not antigens, that determine which foreign substances our immune system will recognize and resist

- Les épitopes reconnus par les lymphocytes T sont distincts de ceux reconnus par les lymphocytes B

TABLE 3-3 COMPARISON OF ANTIGEN RECOGNITION BY T CELLS AND B CELLS

Characteristic	B cells	T cells
Interaction with antigen	Involves binary complex of membrane Ig and Ag	Involves ternary complex of T-cell receptor, Ag, and MHC molecule
Binding of soluble antigen	Yes	No
Involvement of MHC molecules	None required	Required to display processed antigen
Chemical nature of antigens	Protein, polysaccharide, lipid	Mostly proteins, but some lipids and glycolipids presented on MHC-like molecules
Epitope properties	Accessible, hydrophilic, mobile peptides containing sequential or nonsequential amino acids	Internal linear peptides produced by processing of antigen and bound to MHC molecules

MALT = mucosal associated lymphoid tissue
GALT = gut



In newborn

G + bacteria → skin → liver by circulation

G + bacteria → intestinal epithelium

cytokines

cytokine	Relative expression in NN	Effect of cAMP on production	General function	comment
TNF	↓	↓	Pro-inflammatory; activates neutrophils Th1 response	Spontaneous abortion or preterm labour
IFN alpha	↓	↓	Antiviral; vaccine response	Important for MHC class I expression
INFgamma	↓	↓	Activation of macrophage, Th1 response, IL12 induction	Impaired in neonates
IL 12	↓	↓	Activates celle mediated immunity, Th1 response	Impaired in neonates
IL 1 beta	↓	↓	Endothelial adhesion, fever, acute phase response	Impaired in neonates
IL6	↑	↑	Acute phase response, inhibits tissue neutrophilia, inhibits Treg and promotes Th17	
IL8	↑	<->	Neutrophil chemoattractant	Hypoxia enhances
IL10	↑	↑	Antiinflammatory, inhibits production of TNF, IL1 and IFN gamma	
IL23	↑	↑	Promotes Th17 celles	IL 17 enhances epithelial expression of antimicrobial peptides

Antimicrobial Proteins

- Enhance the innate defenses by:
  - Attacking microorganisms directly
  - Hindering microorganisms' ability to reproduce
- The most important antimicrobial proteins are:
  - Interferon
  - Complement proteins

Adaptive Immune Defenses

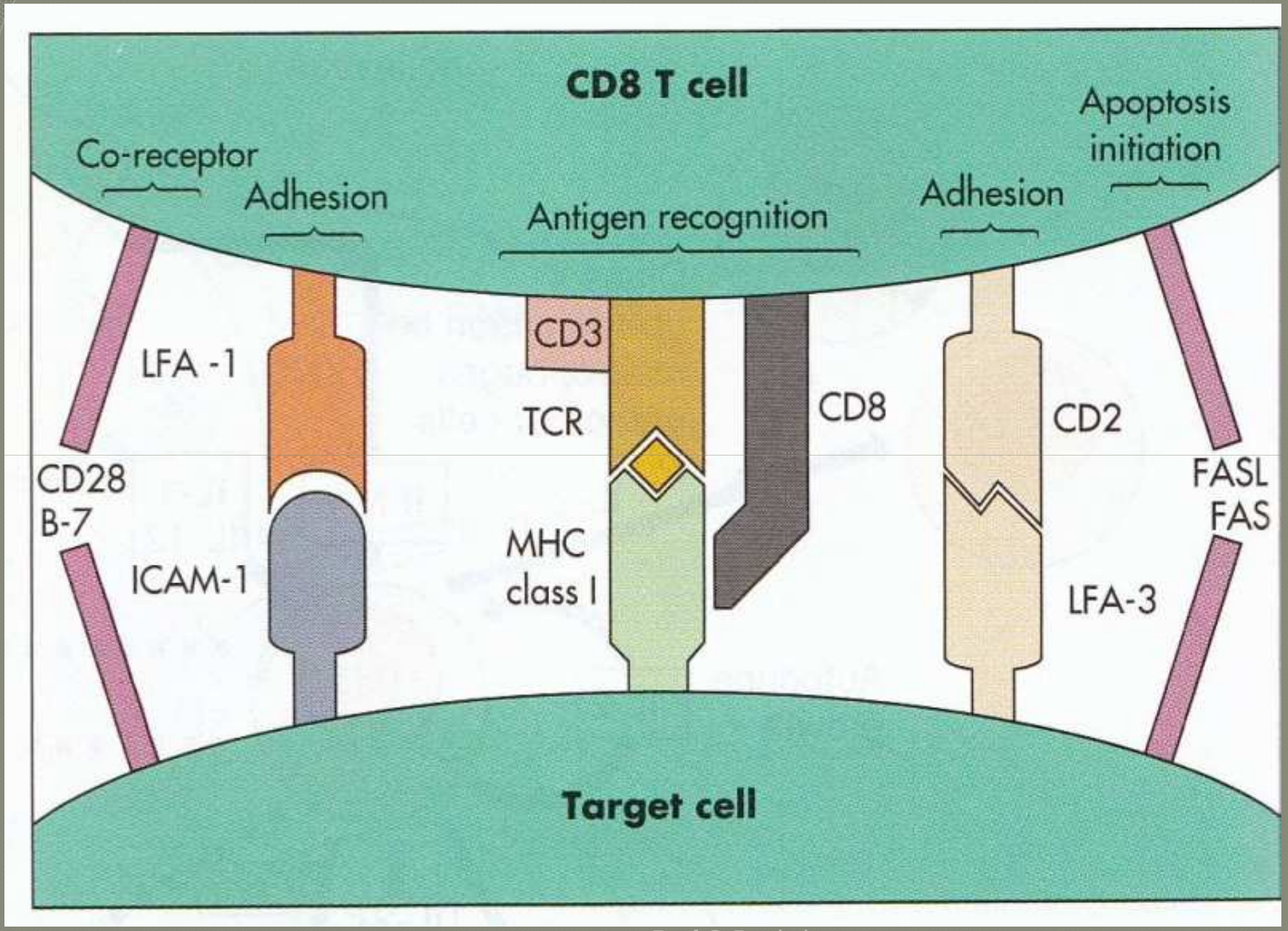
- The adaptive immune system is antigen-specific, systemic, and has memory
- It has two separate but overlapping arms
  - Humoral, or antibody-mediated (B Cell) immunity
  - Cellular, or cell-mediated (T Cell)

ADAPTIVE IMMUNE RESPONSE
Helper T cell Function

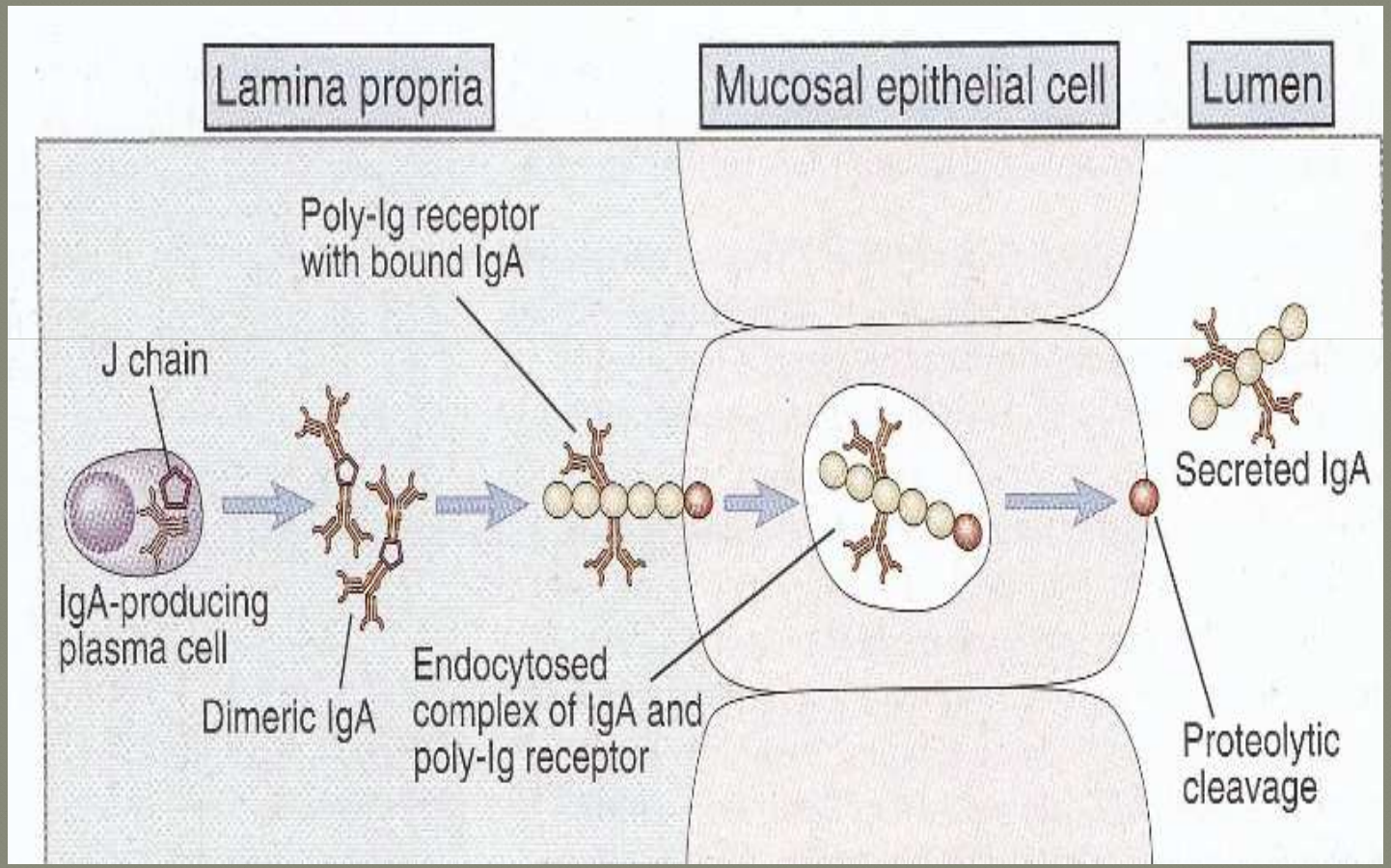
- TH1 and TH2 CD4 (helper) T cells
  - TH0 cells mature into TH1 or TH 2 depending on
    - Nature and concentration of antigen
    - How antigen presented
    - Type of APC
    - Cytokines
  - TH1 = IgM, IgG, activated Macs
  - TH2 = humoral response; IgA, IgE

ADAPTIVE IMMUNE RESPONSE
Cytotoxic T cells

- Activation of CD8 T cells = cytotoxic T cells (CTLs)
  - Precursors in nodes bind TCR + CD8 to MHC-1 of APC + costim
    - TCR recognizes foreign protein in self MHC molecule
  - Specific clone expands by ~100,000
  - Activated CTLs bind with target cell
  - Granulysin, granzymes and perforin released from granules = apoptosis
  - Also interaction of FasL on CTL with Fas on target = apoptosis
- Apoptosis
  - Cell DNA and internal membranes fragment
  - Shrink to "apoptotic bodies" which are easily phagocytosed
  - "Clean" cell death as apposed to necrosis


CD8 T cell
Co-receptor
Adhesion
Antigen recognition
Adhesion
Apoptosis initiation
LFA -1
CD3
TCR
CD8
CD2
CD28
B-7
ICAM-1
MHC class I
LFA-3
FASL
FAS
Target cell

Transport of IgA through Epithelial Cells



Environmental control of Th17 differentiation

- Th17 cells participate in the control of extracellular bacteria and fungi, but dysregulated Th17 cell activity can result in immunopathology
- microbes, dietary components and environmental toxins influence the Th17 response.
- aryl hydrocarbon receptor (AHR): Toxin receptors in the differentiation of Th17 cells: AHR as a nexus between environmental toxins, dietary compounds and infections and Th17 cells.
- The commensal flora triggers IL-25 secretion by intestinal epithelial cells, which then interferes with the differentiation of Th17 by DC (ATP secreted by commensal bacteria, bacterial TLR5 ligand flagellin
- Vit A: The RA produced by intestinal DC and macrophages has been shown to control the balance between Th17 and Treg at multiple points
- Vit D: similarly to what has been described for vitamin A in the gut, vitamin D drives the generation of functional Treg in the skin. Indeed, Vitamin D metabolites have been described to favor the expansion of Treg [20, 21] and boost their activity [22], while it interferes with the Th17 cell response

Regulatory T Cells Selectively Express Toll-like Receptors and Are Activated by Lipopolysaccharide

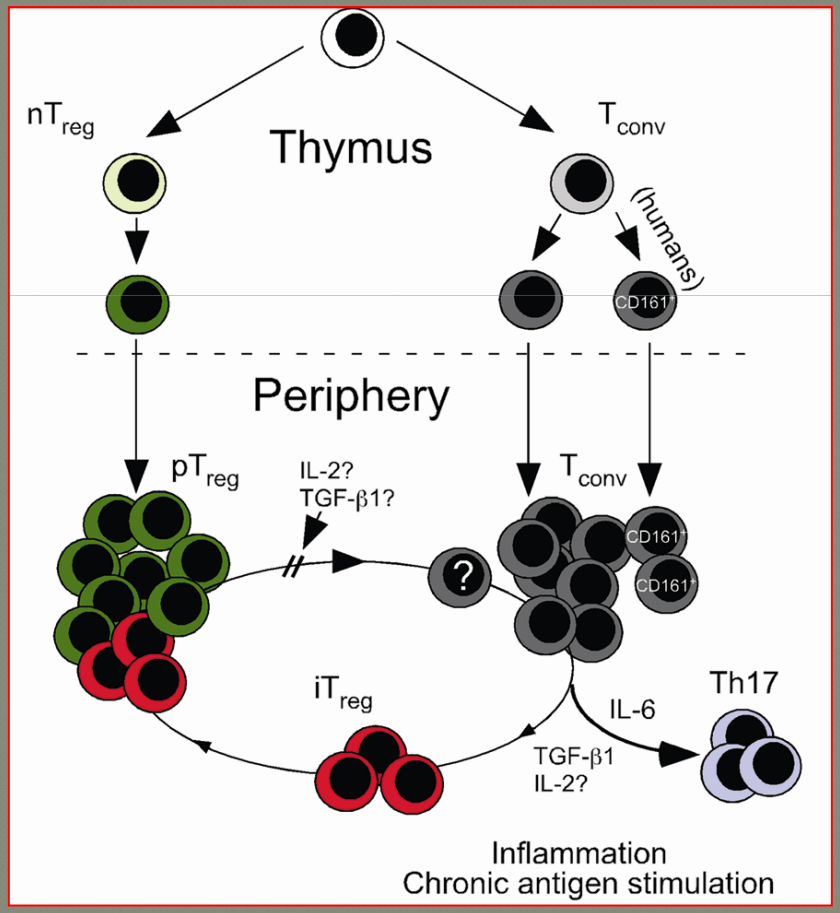
- Regulatory CD4 T cells (Treg) control inflammatory reactions to commensal bacteria and opportunist pathogens
- Treg respond directly to proinflammatory bacterial products, a mechanism that likely contributes to the control of inflammatory responses
- Oral administration of an IL-10–secreting Lactococcus lactis strain prevents food-induced IgE sensitization
- **Persistent Beneficial Effects of Breast Milk Ingested in the Neonatal Intensive Care Unit on Outcomes of**

Extremely Low Birth Weight Infants at 30 Months of Age

HUMORAL IMMUNE RESPONSE

- Antibodies inactivate microorganisms by
  - Agglutination
  - Neutralization
    - Antibody to toxins
    - Antibody to microbial surface molecules that bind to host cells
  - Opsonization
    - Natural killer cells have receptors for IgG
    - Eosinophils have receptors for IgG, IgA, and IgE
  - Complement fixation
    - Neutrophils, macrophages have receptors for C3b
    - Gram negative organisms susceptible to MAC

Induced Treg (iTreg) cells and Th17 cells are derived from a common naïve conventional T (Tconv) precursor population in mice.
In humans, Th17 cells may come from a unique CD161+ subset derived in the thymus.
Both iTreg and Th17 cells require TGFβ1 for their development, although the Th17 pathway is favored in the presence of IL-6.
The peripheral Treg
(pTreg) cell pool is comprised of iTreg cells and "natural" Treg (nTreg) cells derived in the thymus.
Both IL-2 and TGFβ-1 may stabilize Foxp3 expression, although some Treg cells can ultimately
lose it. The fate of cells that exit the pTreg compartment is unknown, although recent data
suggests they could survive and develop into another type of Th cell.



-**IL-17–producing Treg cells may play critical roles in antimicrobial defense, while controlling autoimmunity and inflammation.**
-composition of intestinal microbiota regulates the Th17:Treg balance in the LP and may thus influence intestinal immunity, tolerance, and susceptibility to inflammatory bowel diseases.
Th17 Cells Are Preferentially Present in the Small Intestine
Th17 Cells Are Specifically Enriched in the Small Intestinal Lamina Propria.
Specific Antibiotic Treatment Selectively Prevents Intestinal Th17 Cell Differentiation

Réponse normale

Lors d'une infection par un pathogène, la réaction inflammatoire associée conduit à la différenciation des cellules dendritiques matures.
Ces dernières activent alors les cellules T CD4 naïves et induisent leur différenciation en cellules effectrices (Th1, Th2 et TH17).

Summary of the Primary Immune Response

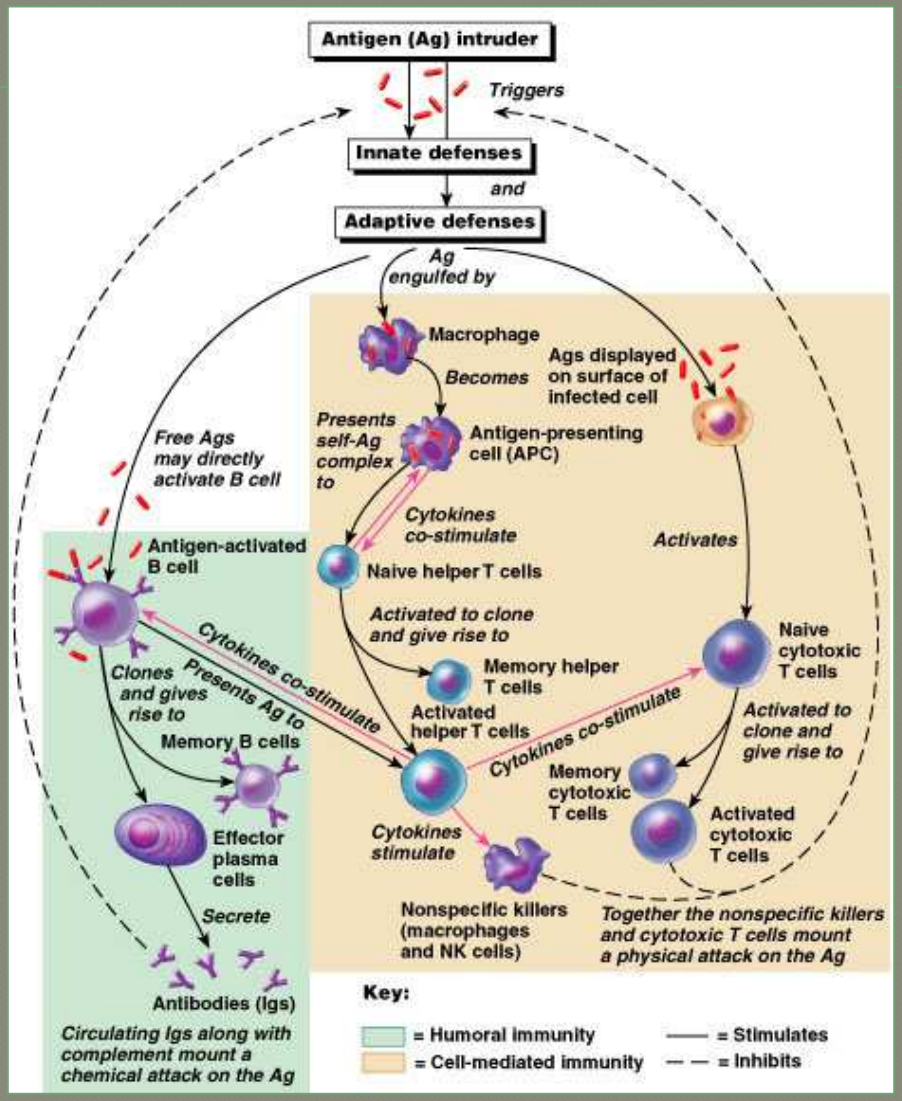
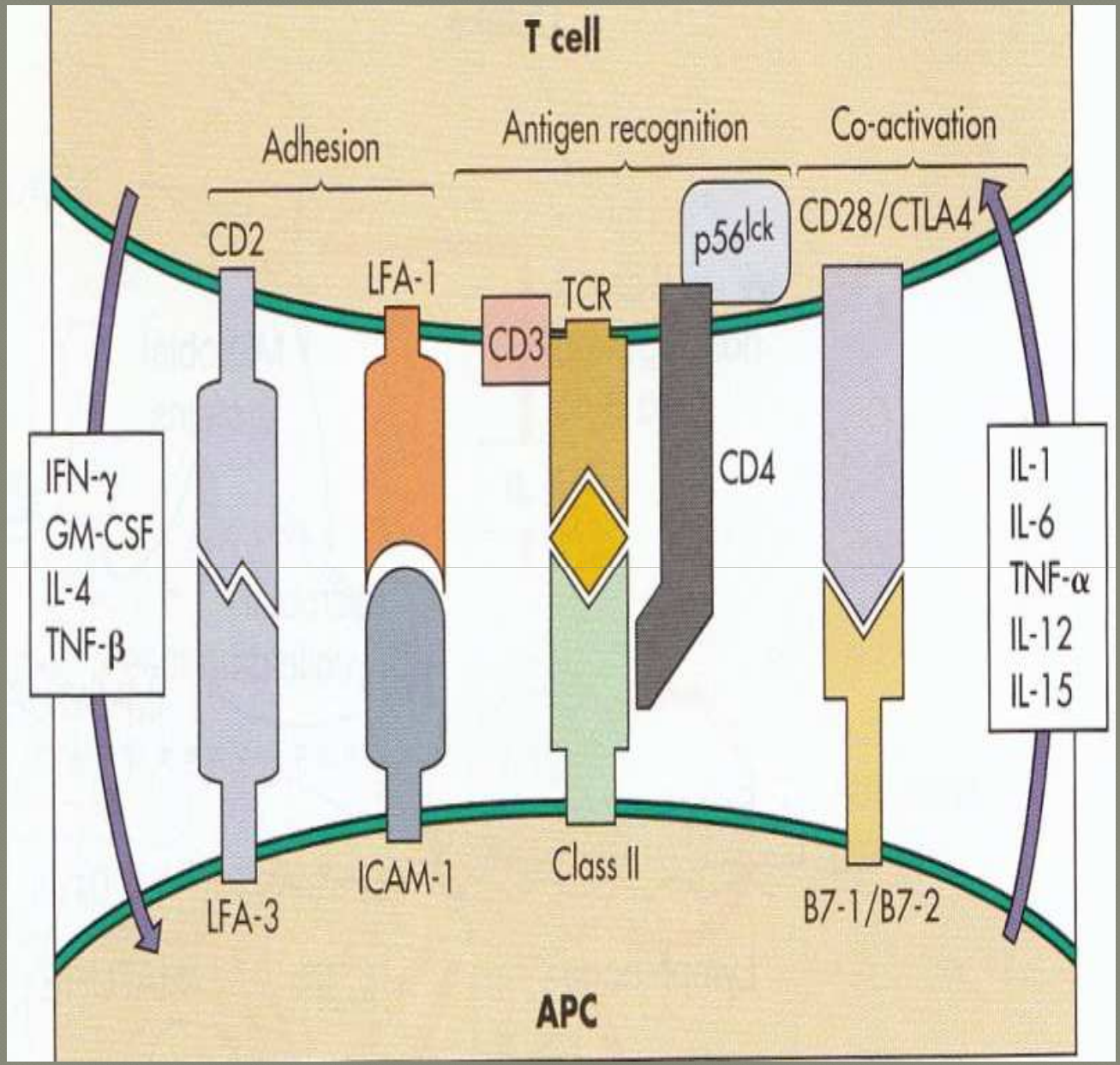


Figure 21.19


T cell
Adhesion
Antigen recognition
Co-activation
CD2
LFA-1
CD3
TCR
p56lck
CD28/CTLA4
CD4
IFN-γ
GM-CSF
IL-4
TNF-β
IL-1
IL-6
TNF-α
IL-12
IL-15
LFA-3
ICAM-1
Class II
B7-1/B7-2
APC

Cytotoxic T Cell (T_c)

- T_C cells, or killer T cells, are the only T cells that can directly attack and kill other cells
- They circulate throughout the body in search of body cells that display the antigen to which they have been sensitized
- Their targets include:
  - Virus-infected cells
  - Cells with intracellular bacteria or parasites
  - Cancer cells
  - Foreign cells from blood transfusions or transplants

Cytotoxic T Cells

- Bind to self-antiself complexes on all body cells
- Infected or abnormal cells can be destroyed as long as appropriate antigen and co-stimulatory stimuli (e.g., IL-2) are present
- Natural killer cells activate their killing machinery when they bind to MICA receptor
- MICA receptor – MHC-related cell surface protein in cancer cells, virus-infected cells, and cells of transplanted organs

Mechanisms of T_c Action

- In some cases, T_C cells:
  - Bind to the target cell and release perforin into its membrane
    - In the presence of Ca^{2+} perforin causes cell lysis by creating transmembrane pores
- Other T_C cells induce cell death by:
  - Secreting lymphotoxin, which fragments the target cell's DNA
  - Secreting gamma interferon, which stimulates phagocytosis by macrophages

Mechanisms of T_c Action

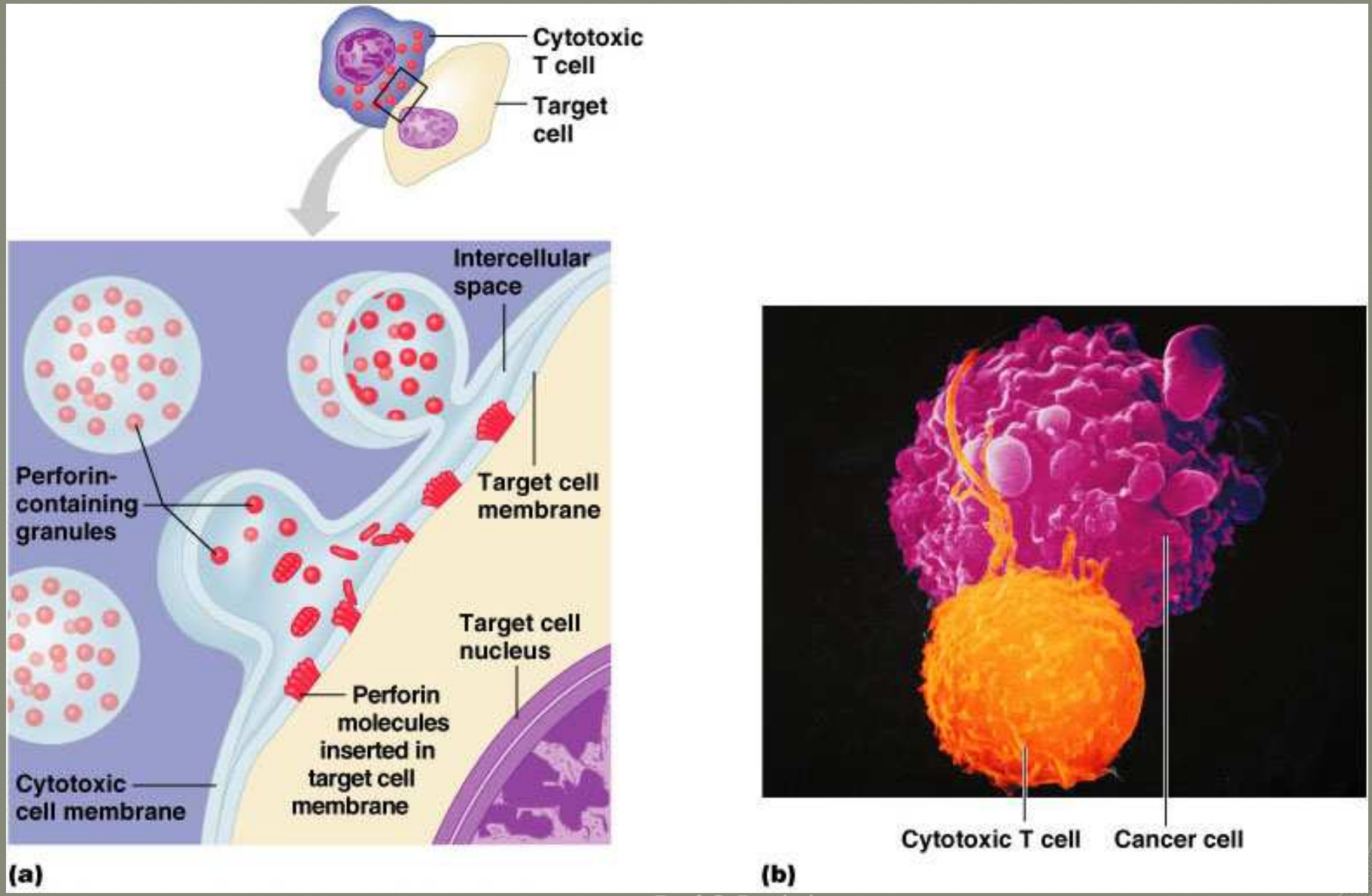


Figure 21.18a, b

Cytokines

- Mediators involved in cellular immunity, including hormonelike glycoproteins released by activated T cells and macrophages
- Some are co-stimulators of T cells and T cell proliferation
- Interleukin 1 (IL-1) released by macrophages co-stimulates bound T cells to:
  - Release interleukin 2 (IL-2)
  - Synthesize more IL-2 receptors

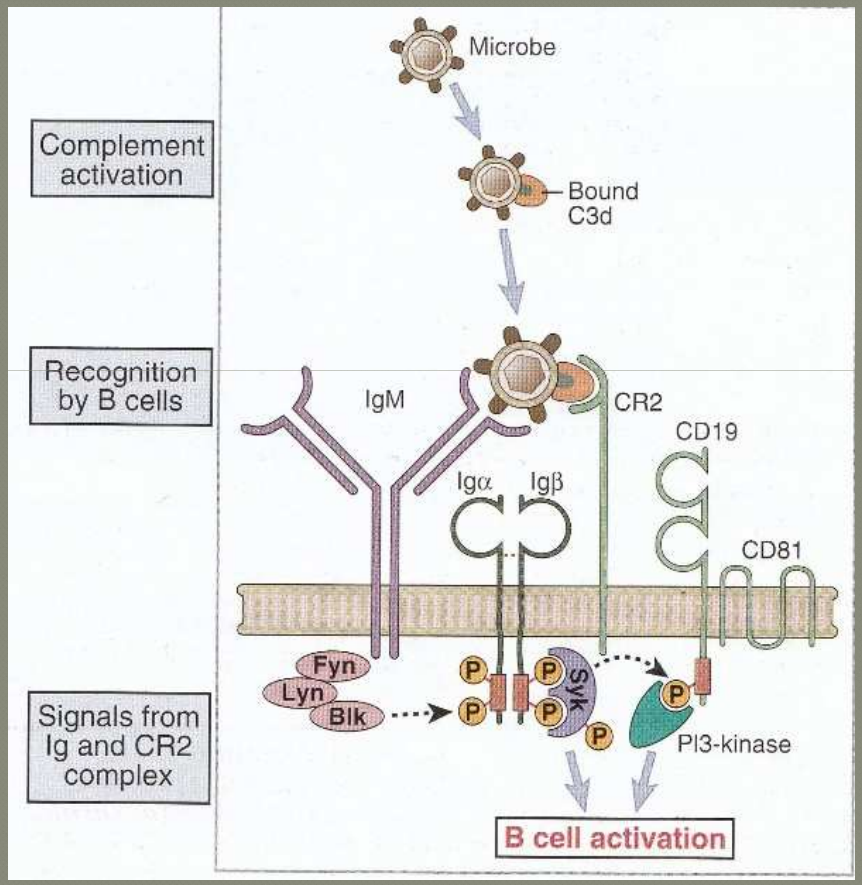
Cytokines

- IL-2 is a key growth factor, which sets up a positive feedback cycle that encourages activated T cells to divide
  - It is used therapeutically to enhance the body's defenses against cancer
- Other cytokines amplify and regulate immune and nonspecific responses

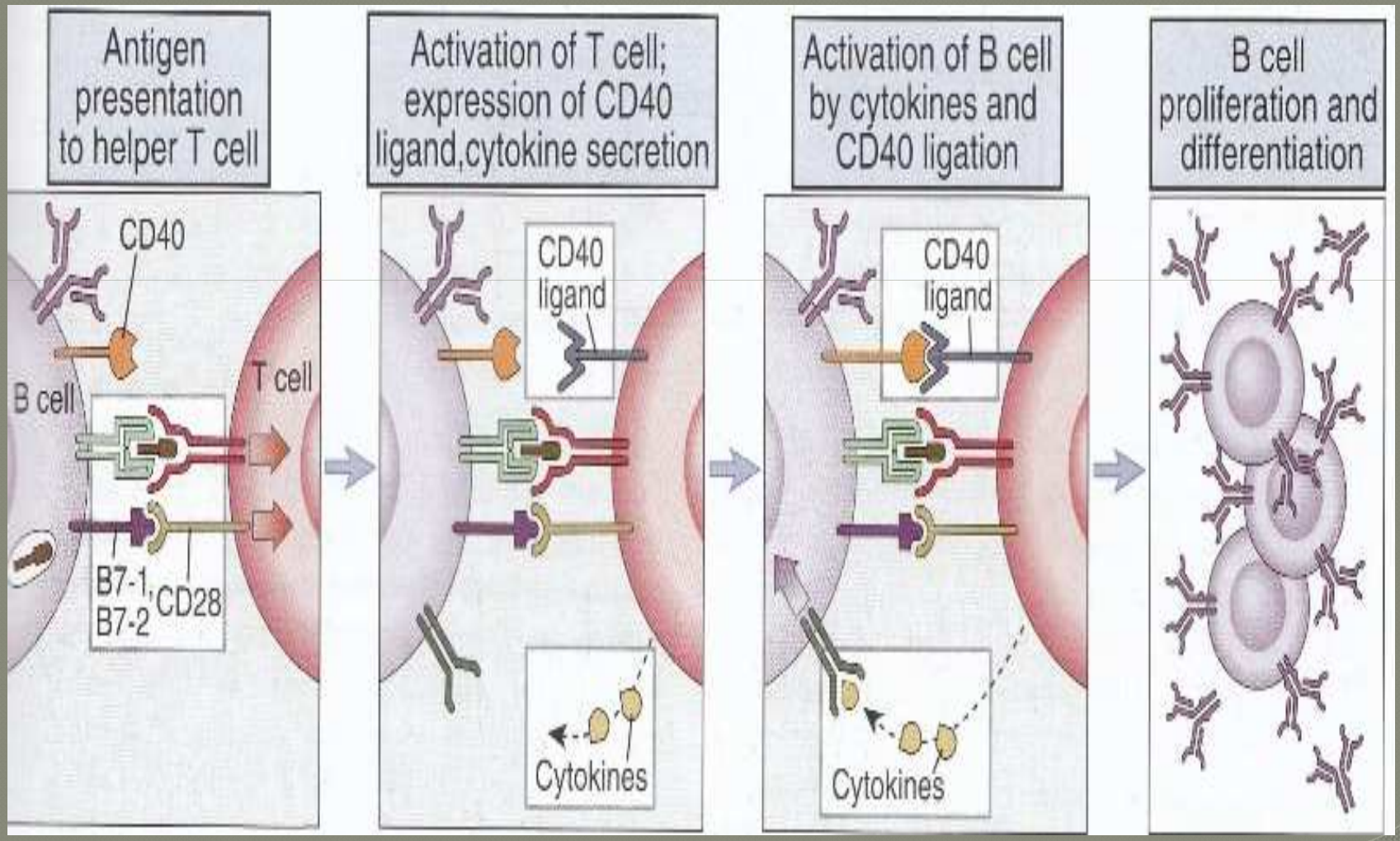
Other T Cells

- Suppressor T cells (T_S) – regulatory cells that release cytokines, which suppress the activity of both T cells and B cells
- Gamma delta T cells (T_{gd}) – 10% of all T cells found in the intestines that are triggered by binding to MICA receptors

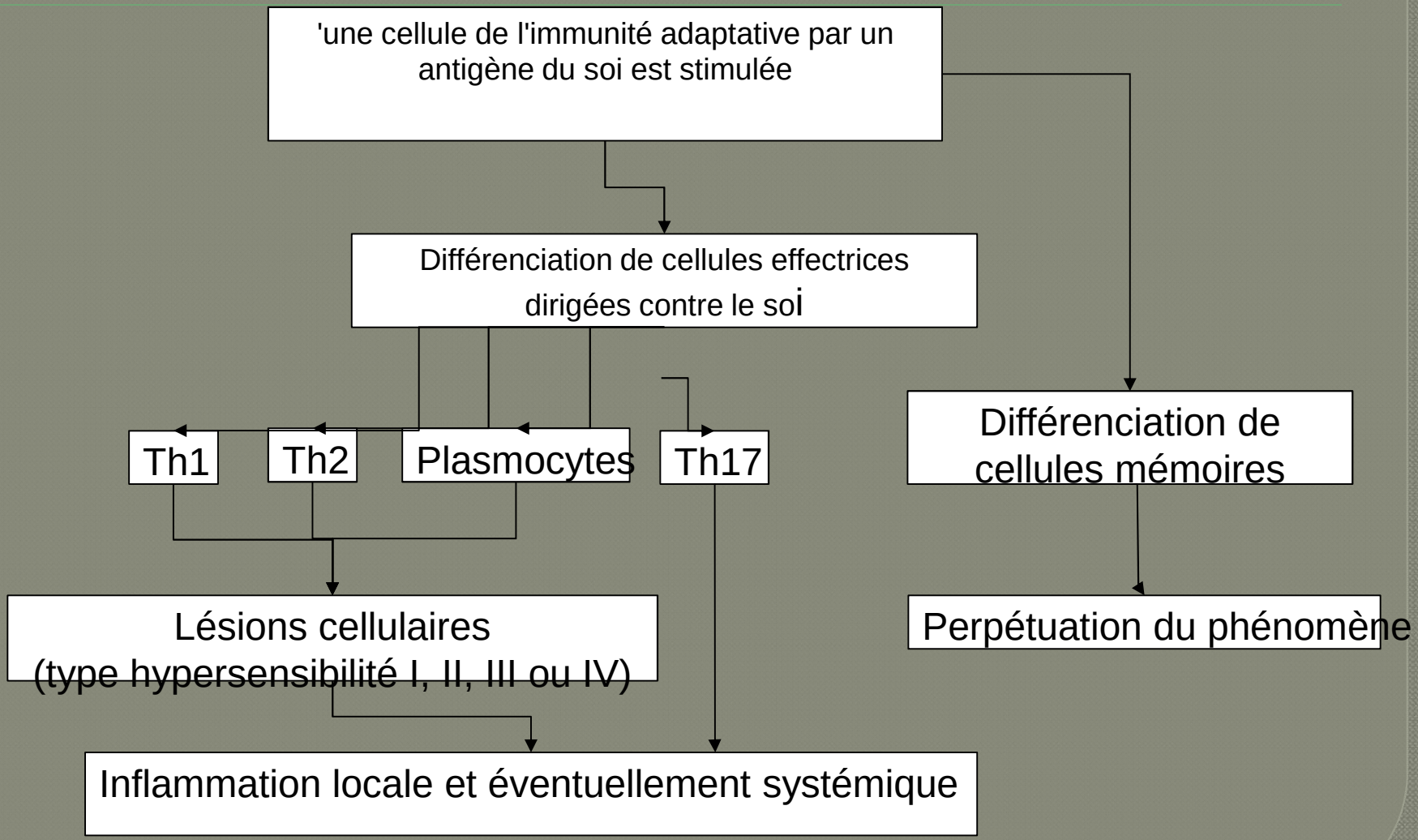
C' in B Cell Activation



T Cell - Mediated B Cell Activation

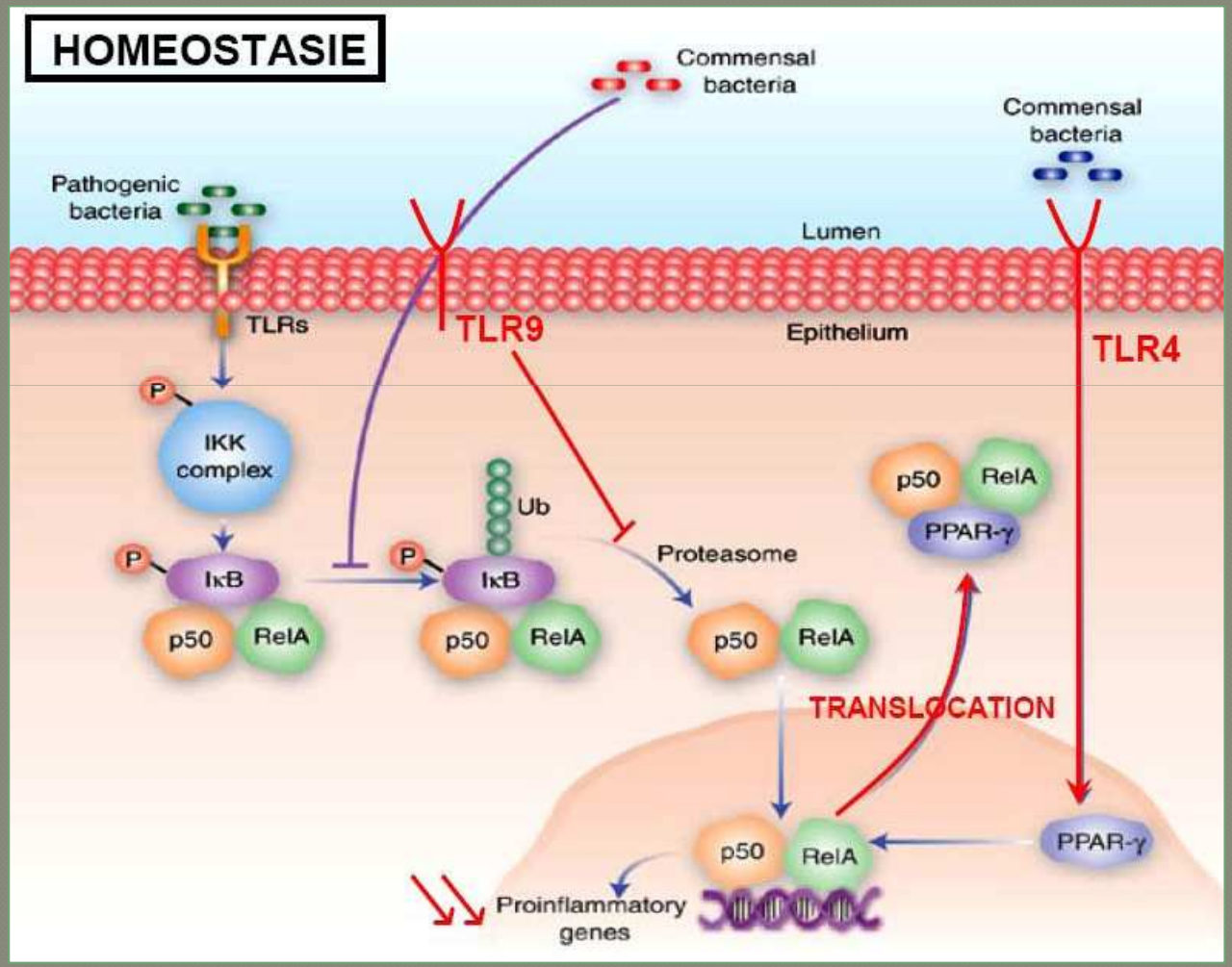


Réponse anormale = l'autoimmunité

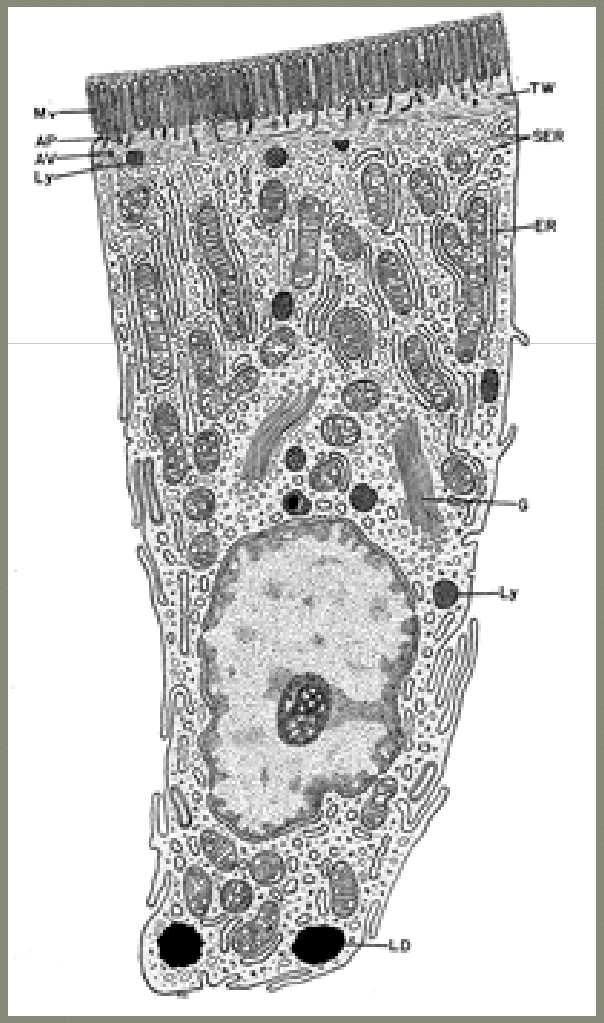


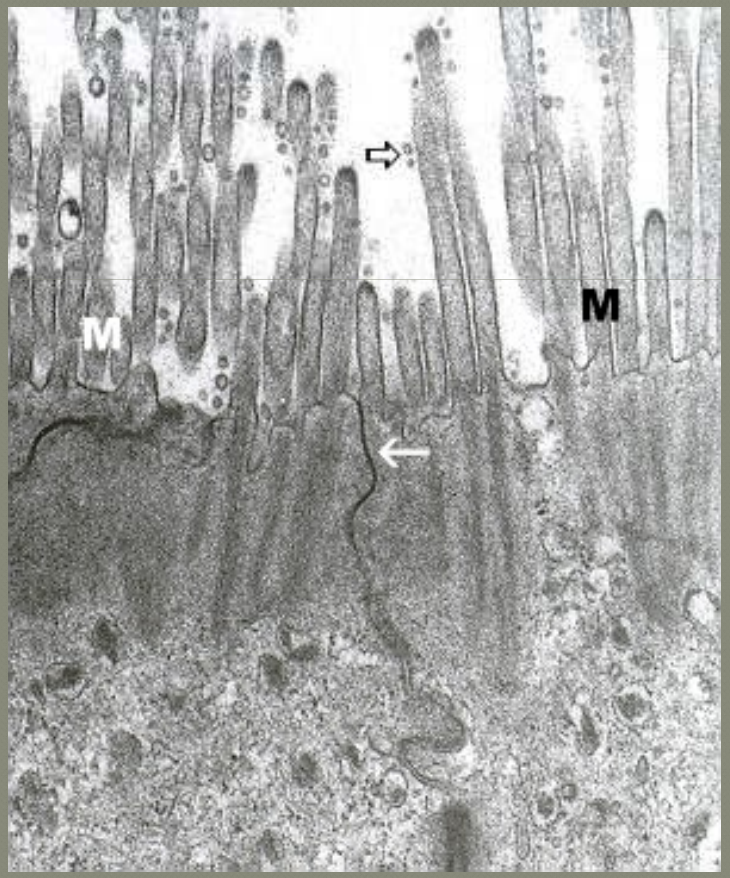
- L’autoimmunité peut s’installer:
  - Soit parce que le seuil est anormalement bas
    - Prédisposition génétique (« hyperexcitabilité » intrinsèque, manque de lymphocytes T régulateurs, etc.)
    - Faiblesse spécifique de la barrière pour un autoantigène donné
  - Soit parce que le signal du soi est trop intense
    - Rupture de barrière (y compris acides nucléiques du noyau)
    - Contexte inflammatoire (cytokines inflammatoires telles que IFN-α, génération de second signaux via CD28+++)
    - Mimétisme moléculaire

Épithelium intestinal

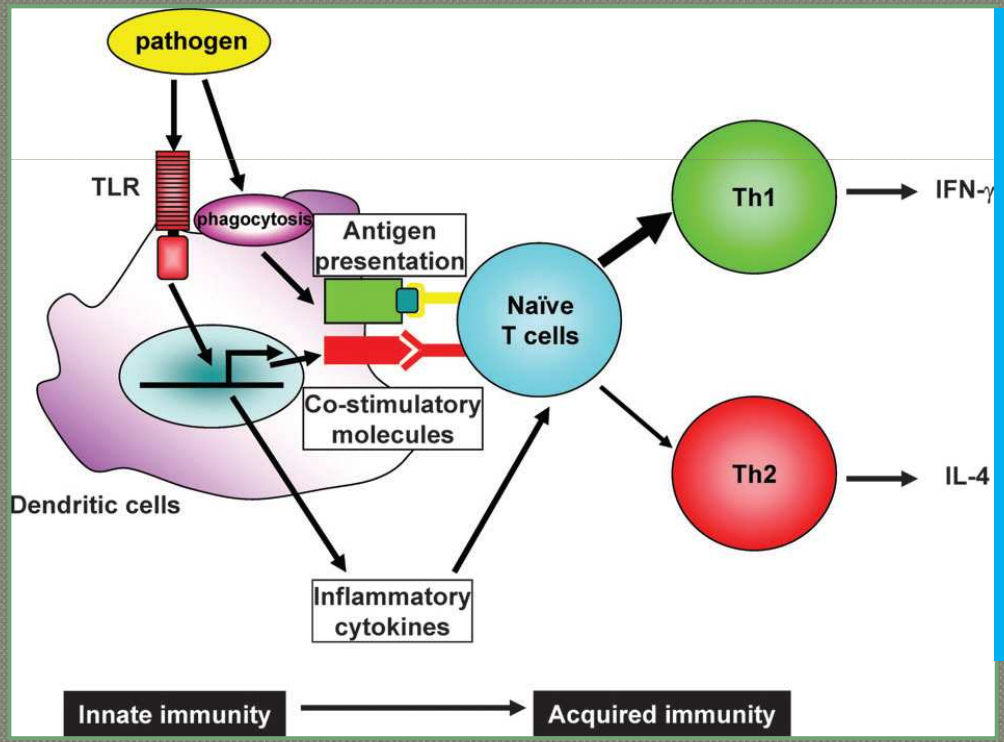


L'épithélium intestinal se régénère tous les 3 à 5 jours





“The Gut-Immune Interface” ou l’immunité et les muqeuses.

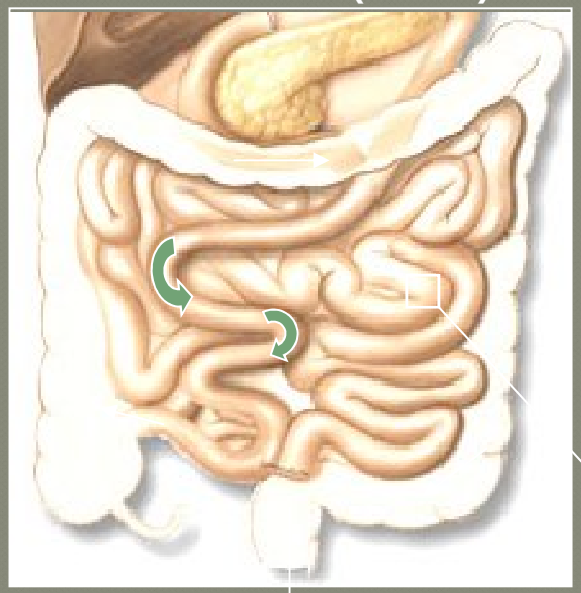


- La signalisation par les protéines Réceptrices du soi sont situées
À la surface de l’épithélium : TLR
et dans le cytosol: NOD 1 et NOD2
-Lors d'une infection par un pathogène,
-Ou lors de la perte de la tolérance,

la réaction inflammatoire associée conduit à la différenciation des cellules dendritiques matures.
Ces dernières activent alors les cellules T CD4 naïves et induisent leur différenciation en
cellules effectrices (Th1, Th2 et TH17).

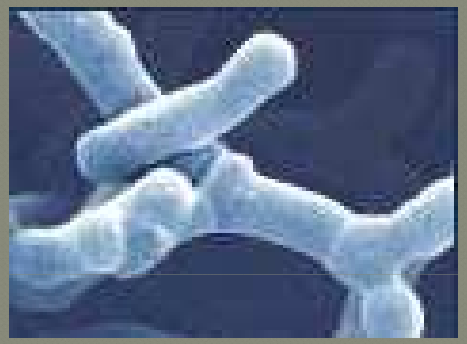
La barrière intestinale implique:

1- La motricité (transit)



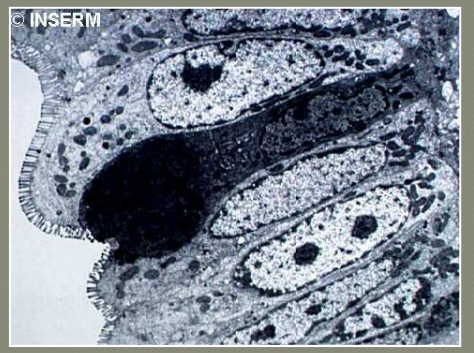
Evite l'adhésion des bactéries pathogènes aux cellules épithéliales

2- La flore intestinale



S'oppose à la colonisation par les bactéries étrangères

3- Le mucus (cellules caliciforme



Protection de l'épithélium

4- L'épithélium (entérocytes)

Transport sélectif trans- et paracellulaire

5- Les cellules immunitaires

Défense rapide, non spécifique

Effecteurs immunitaires de la muqueuse intestinale

- **Lamina propria:**

Lymphocytes T CD4+ / T CD8+
Plasmocytes IgA+
DC
Macrophages
Innate Lymphoid cells

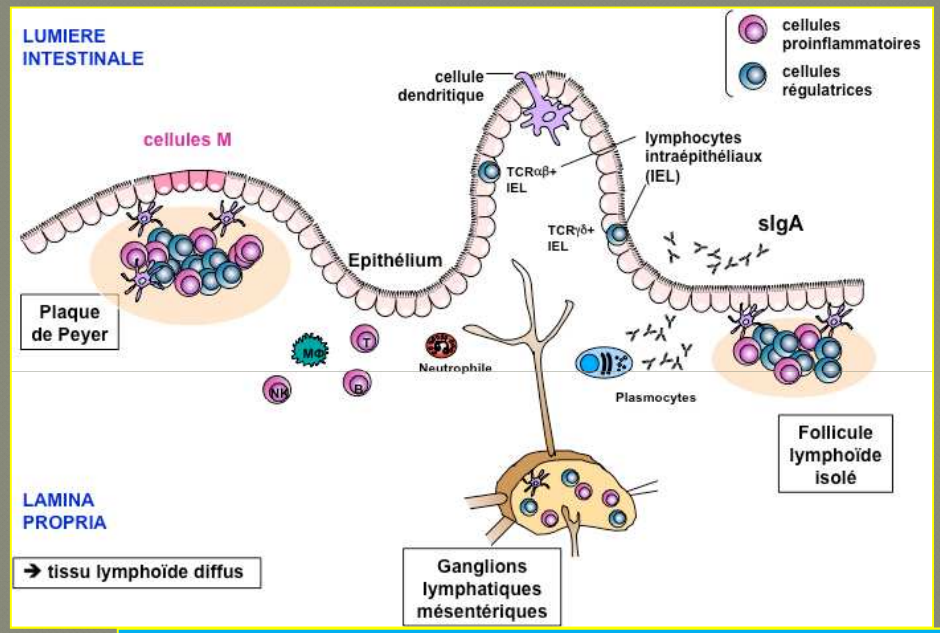
- **Plaques de Peyer:**

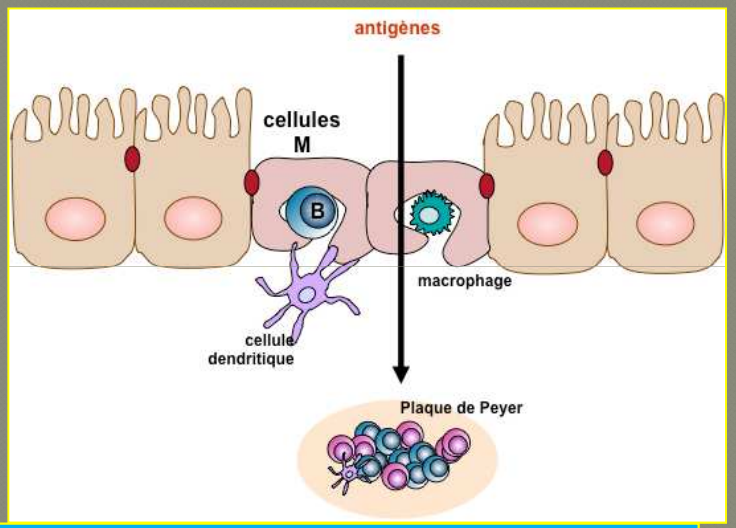
DC
Lymphocytes T CD4+ / T CD8+
Lymphocytes B

- **Epithelium:**

Enterocytes
Goblet cells
Paneth cells
Intra-Epithelial Lymphocytes

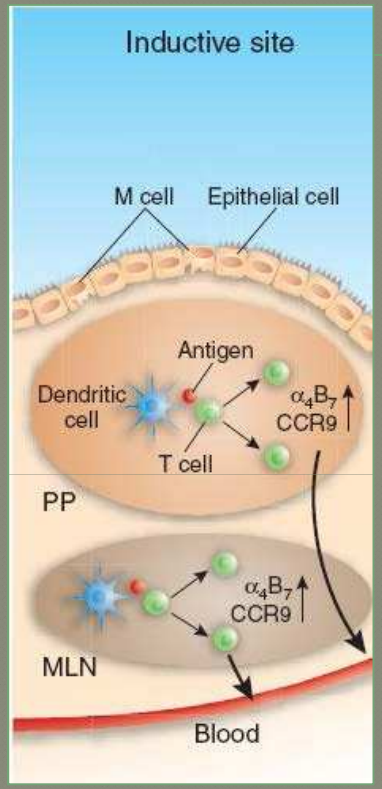
La mise en route de la réponse immunitaire épithéliale

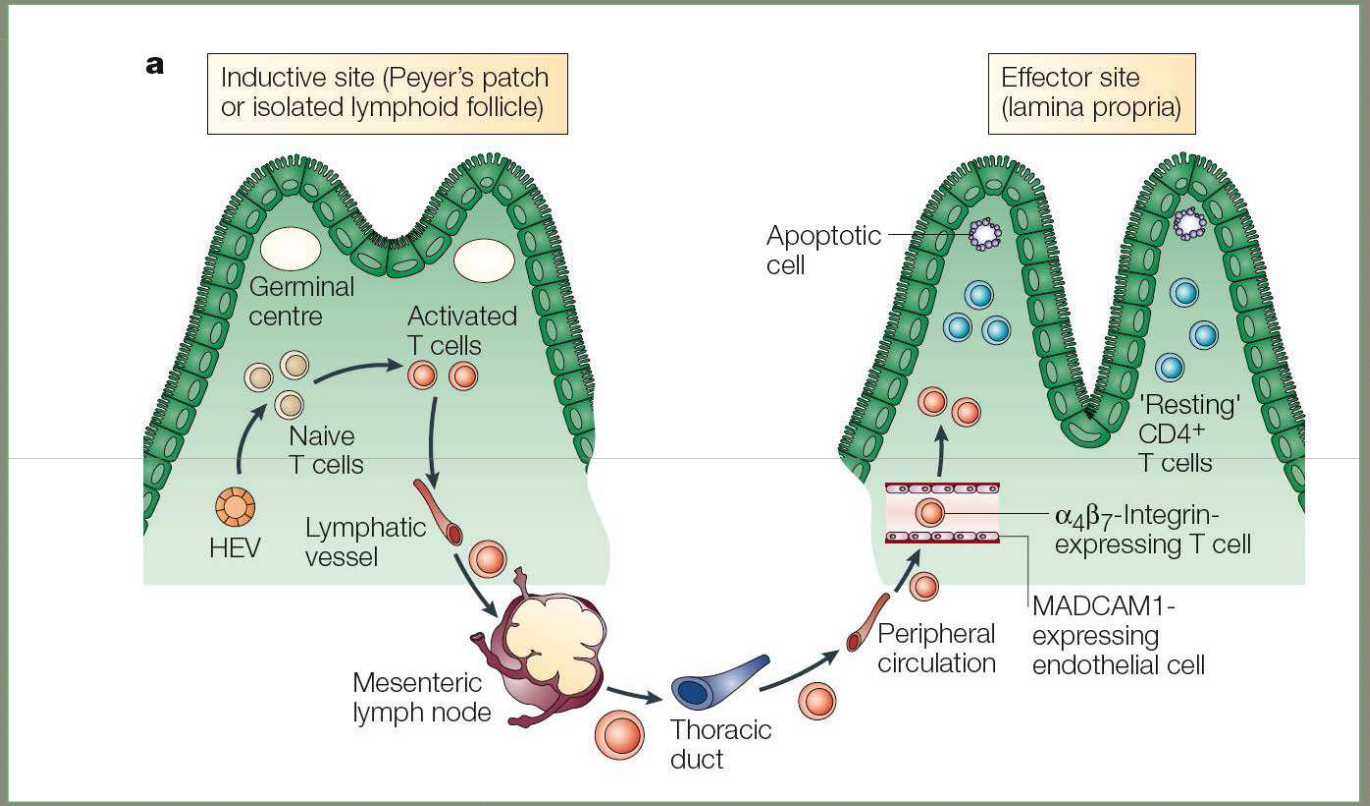




Il existe deux modes de capture des antigènes présents dans la lumière :
- endocytose des antigènes par les cellules M (figure 3) de la muqueuse intestinale puis capture par les cellules dendritiques sous-jacentes.
- au niveau des villosités, des cellules dendritiques émettent des pseudopodes entre les cellules épithéliales pour sonder en permanence les microorganismes qui sont dans la lumière

Marqueurs de homing intestinal

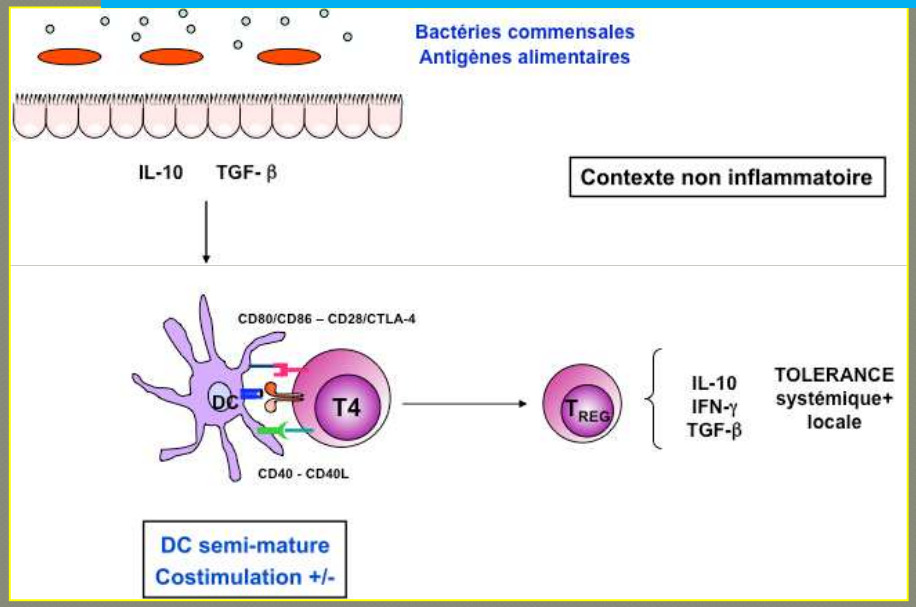


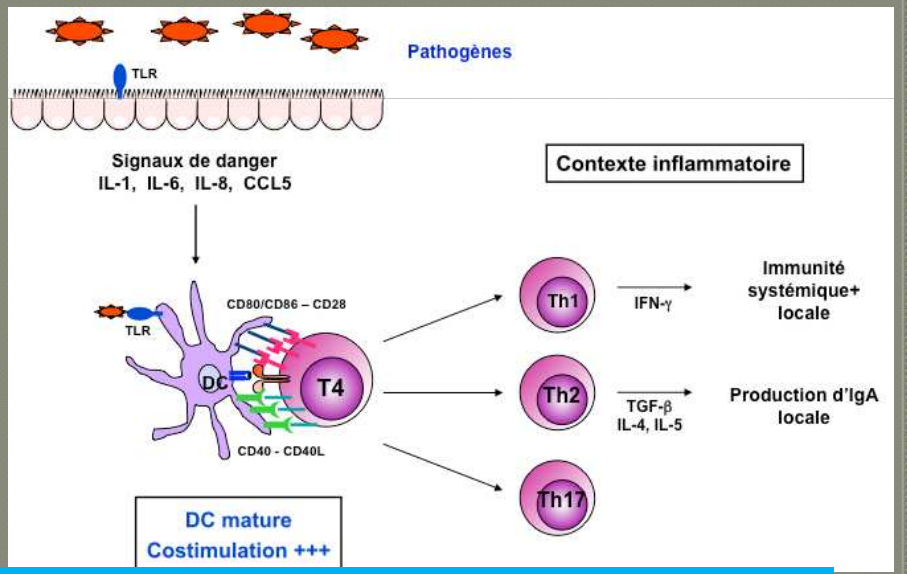


Les lymphocytes CCR9+ α4β7+ périphériques sont destinés à migrer vers la muqueuse de l'intestin grêle
→ Rôle de CCR9 et α4β7

Tolérance ou intolérance

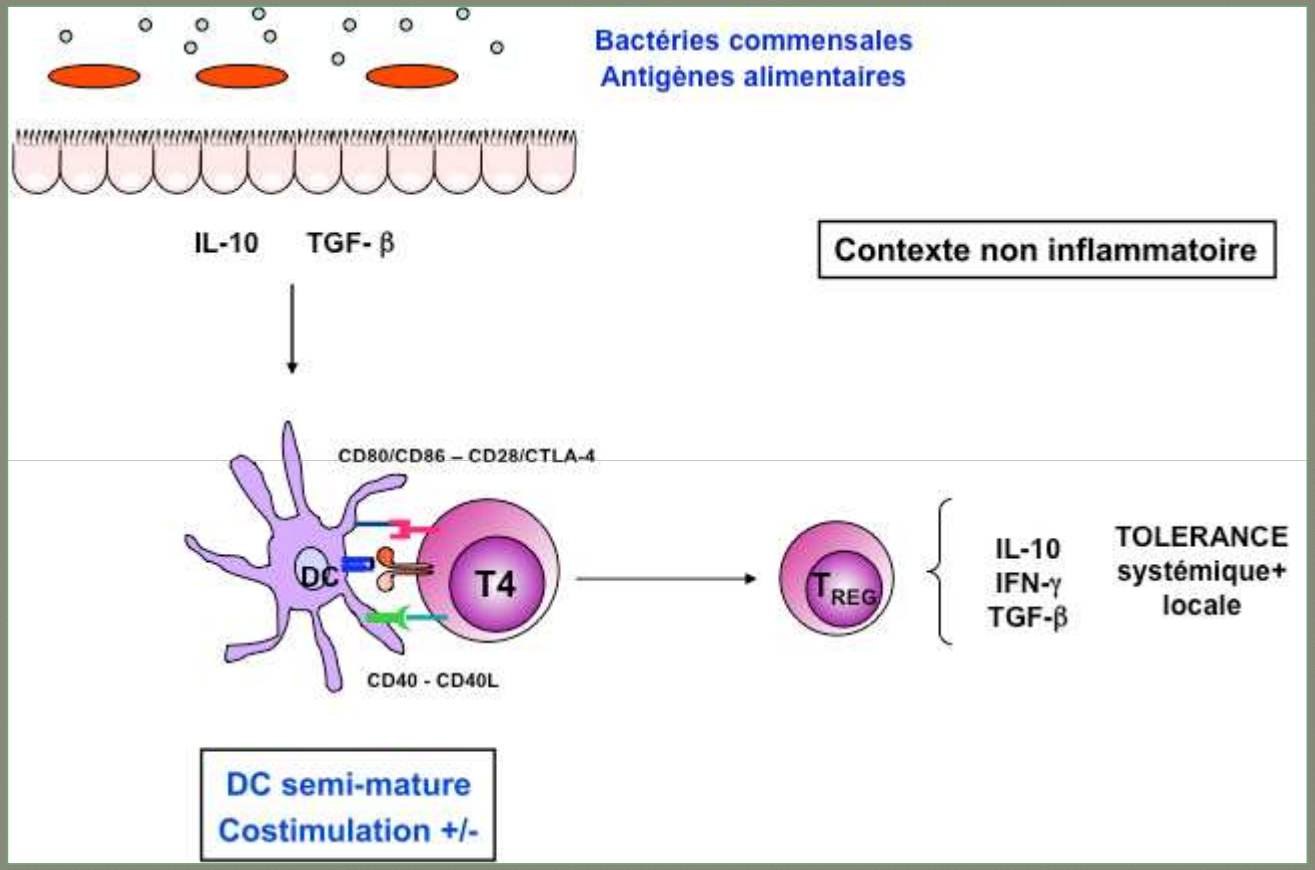
Dans un contexte non inflammatoire, la reconnaissance de bactéries commensales ou de protéines alimentaires conduit à la différenciation des cellules T CD4 naïves en cellules T régulatrices (Treg) capables d'inhiber la réponse immunitaire des cellules T effectrices





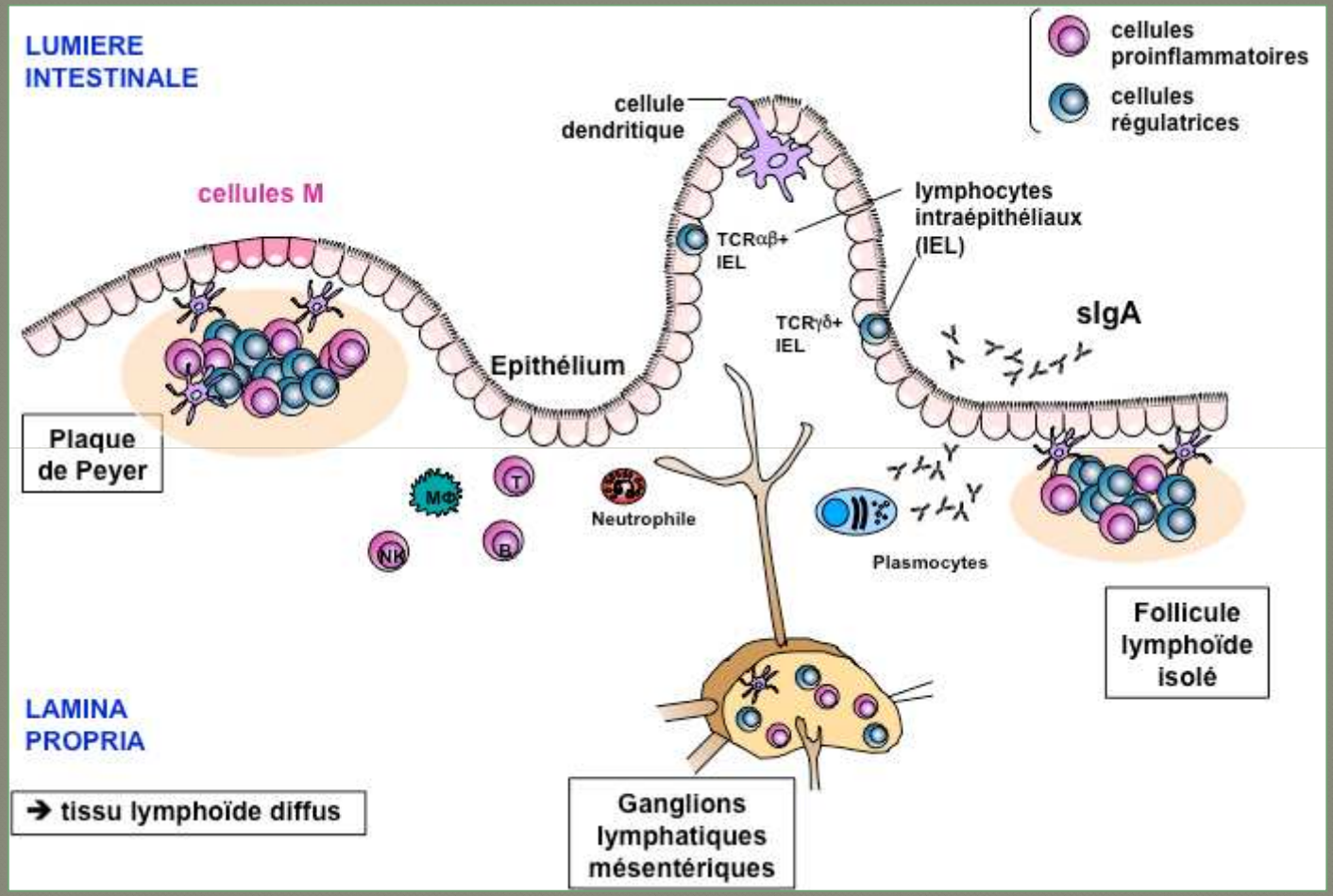
Lors d'une infection par un pathogène, la réaction inflammatoire associée conduit à la différenciation des cellules dendritiques matures. Ces dernières activent alors les cellules T CD4 naïves et induisent leur différenciation en cellules effectrices (Th1, Th2 et TH17).

Tolérance vis-à-vis des bactéries commensales ou d'antigènes alimentaires

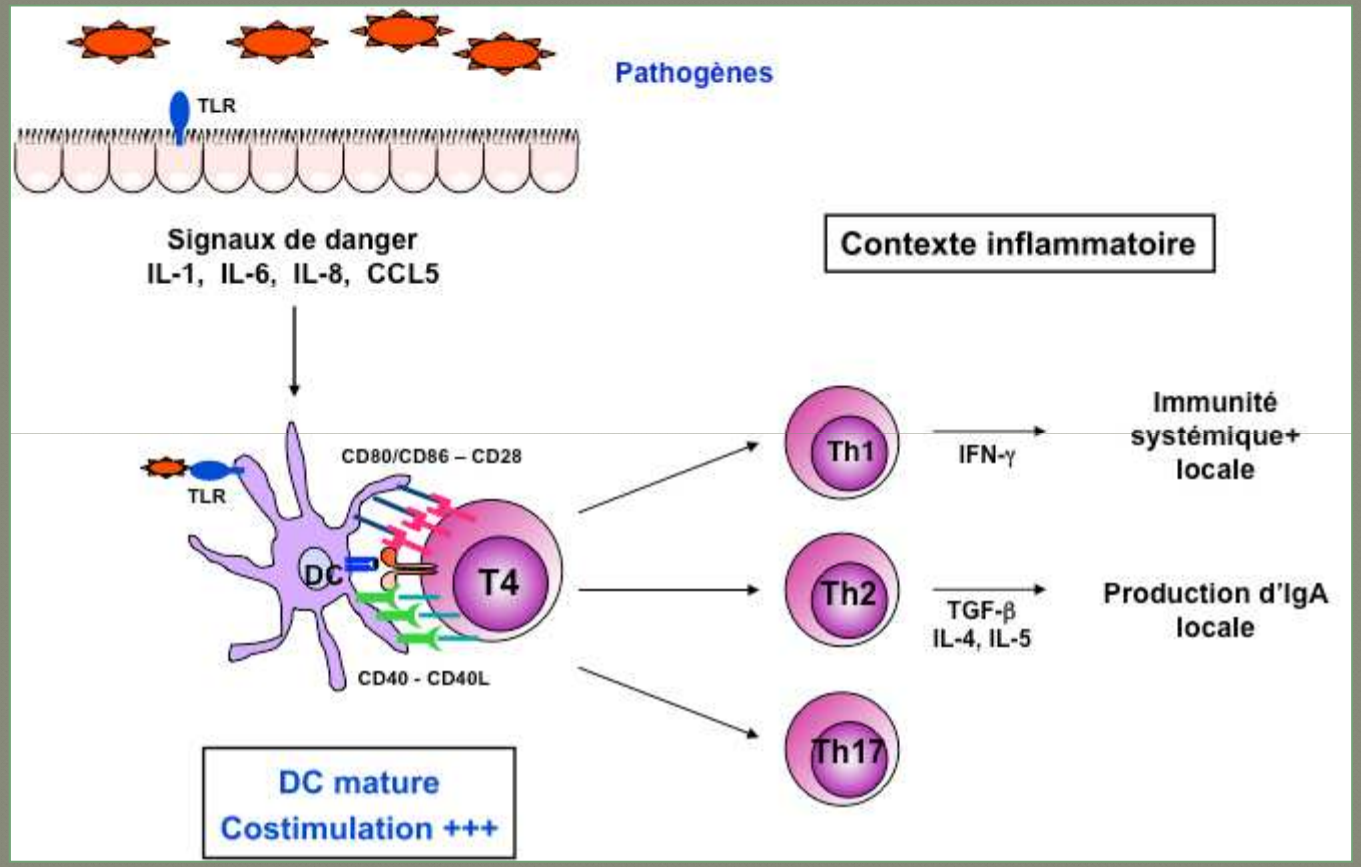


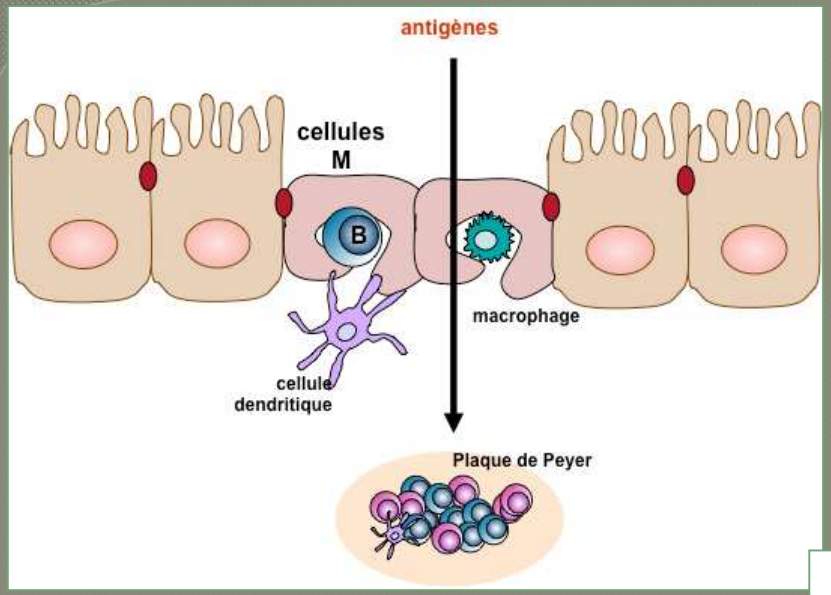
Dans un contexte non inflammatoire, la reconnaissance de bactéries commensales ou de protéines alimentaires conduit à la différenciation des cellules T CD4 naïves en cellules T régulatrices (Treg) capables d'inhiber la réponse immunitaire des cellules T effectrices..

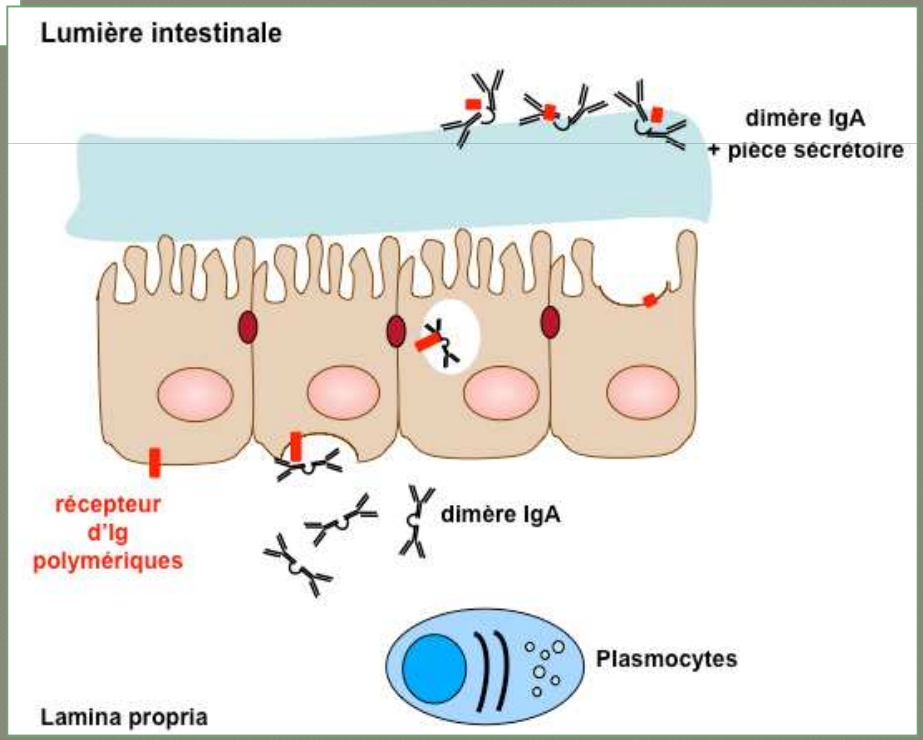
Mise en route d'une réponse immunitaire



Réponse immunitaire dirigée contre un micro-organisme pathogène




antigènes
cellules
M
B
macrophage
cellule
dendritique
Plaque de Peyer


Lumière intestinale
dimère IgA
+ pièce sécrétoire
récepteur
d'Ig
polymériques
dimère IgA
Plasmocytes
Lamina propria

Types of Acquired Immunity

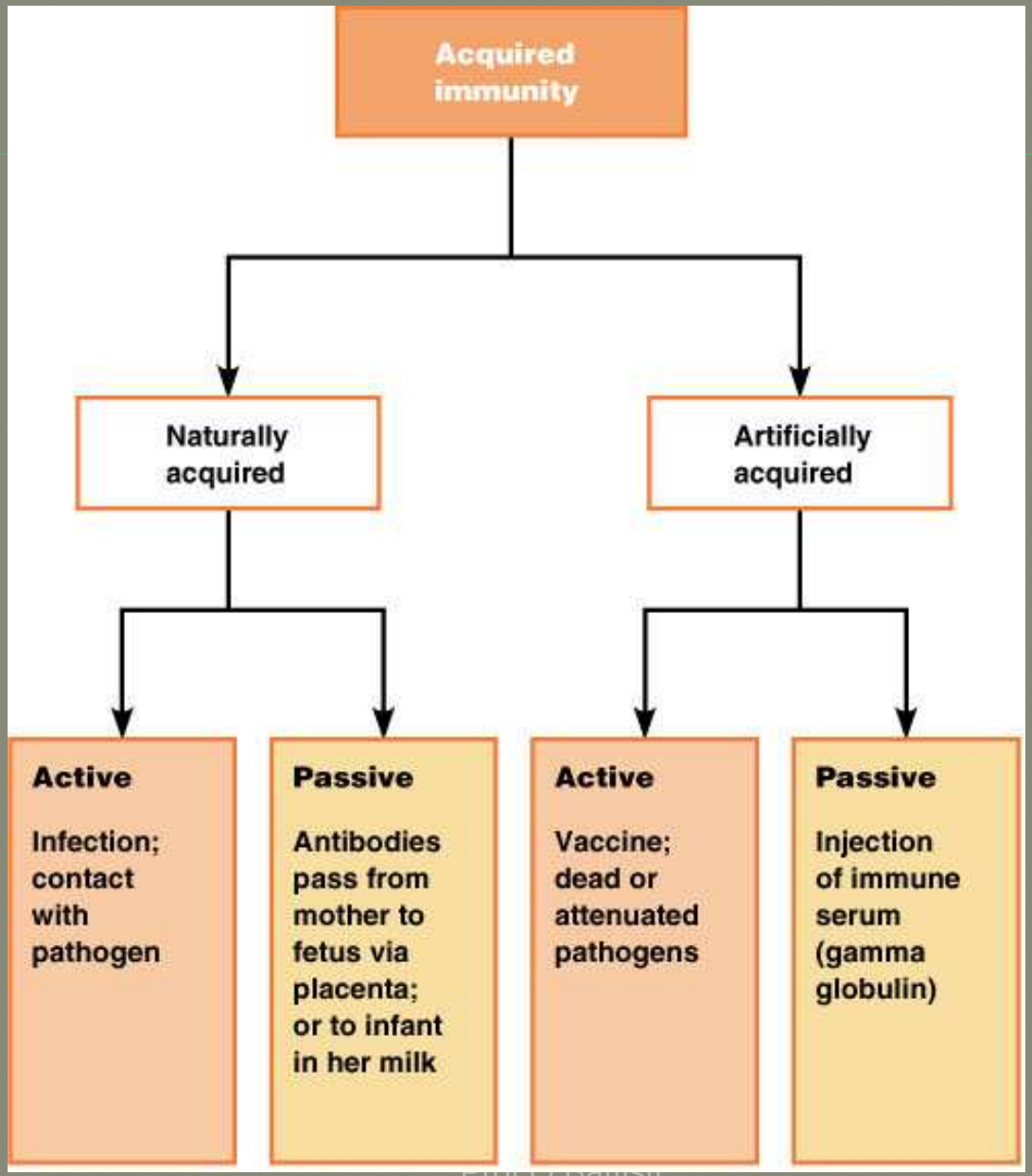
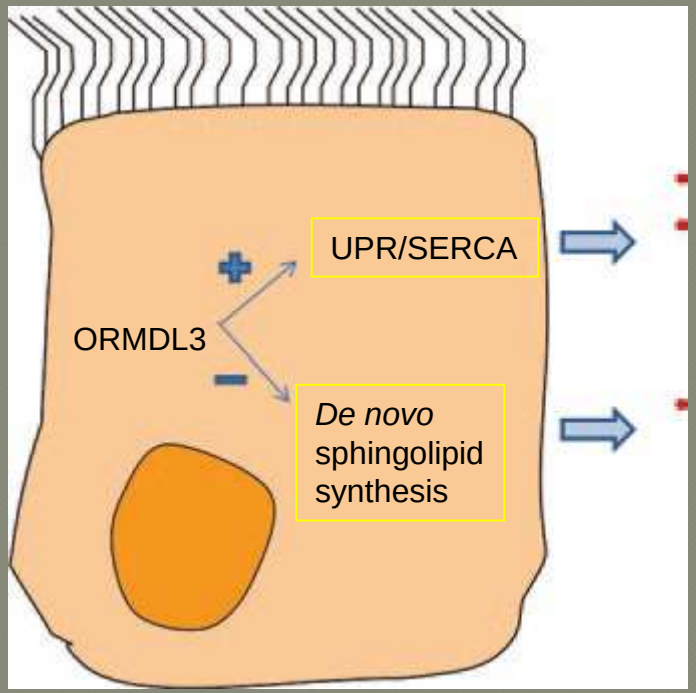


Figure 21.11

L'épithélium respiratoire

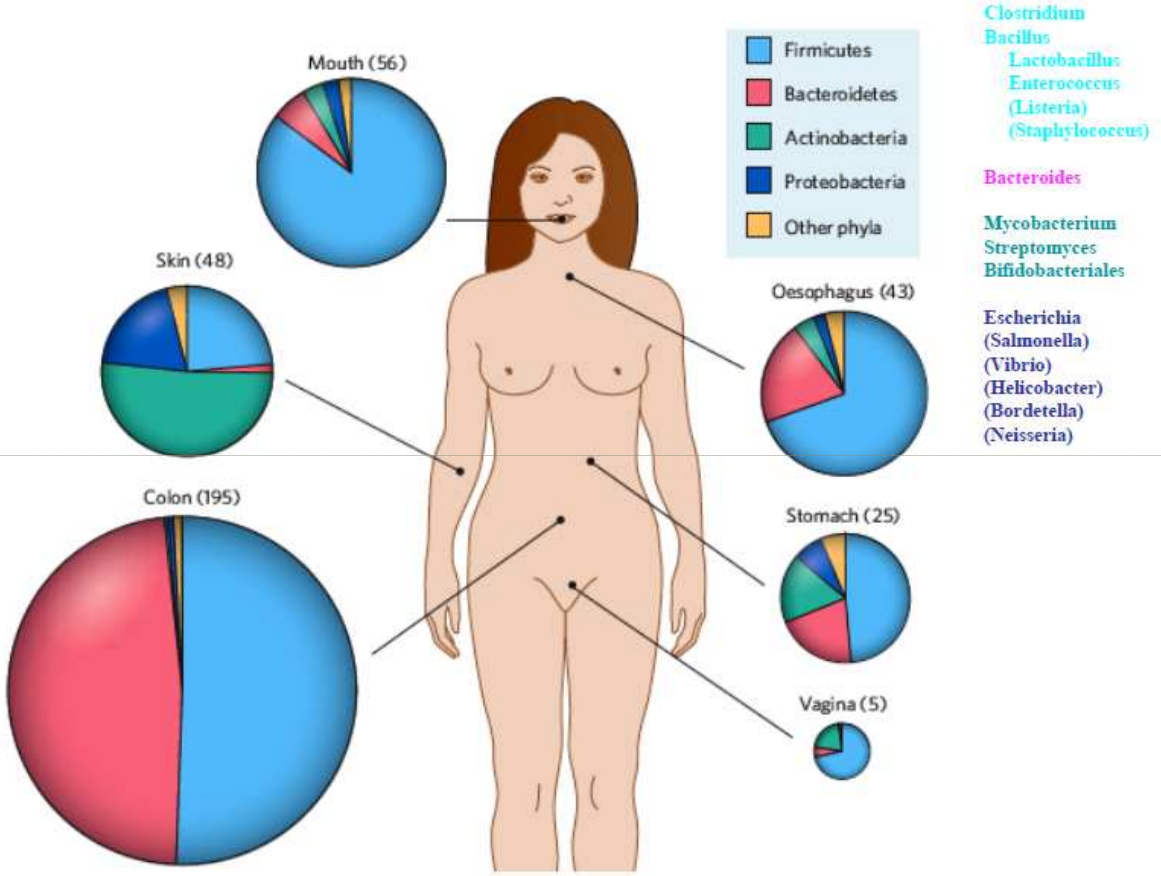
- Adenoids and tonsils increase chitinase activity
- 17q21 locus modulates ORMDL3 activity
- Les infections virales RSV et rhinovirus
- Vitamine A et D et différentiation Th17



Inflammation
Key features of asthma
Cell proliferation/
airway remodeling

Bronchial
hyperreactivity

Bacterial commensals of the human body



Clostridium
Bacillus
Lactobacillus
Enterococcus
(Listeria)
(Staphylococcus)

Bacteroides

Mycobacterium
Streptomyces
Bifidobacteriales

Escherichia
(Salmonella)
(Vibrio)
(Helicobacter)
(Bordetella)
(Neisseria)

L'épithélium intestinal

- L'épithélium intestinal Possède des TLR qui reconnaissent les bactéries commensales

Au niveau apical, TLR moins réactifs
Au niveau basal, TLR plus réactifs
→ HOMEOSTASIE

- **Le TLR a un rôle dans la maturation et l'éducation du système immunitaire pour la modulation de la réponse effectrice→ tolérance des bactéries commensales + réaction aux bactéries pathogènes**
- Colonisation intestinale: Les bactéries commensales participent à la mise en place d'une muqueuse mature
- Les bactéries commensales conduisent à l'expansion des LT CD4+ ainsi que celle des LT régulateurs (CD4+ et CD8+) dans les ganglions mésentériques.

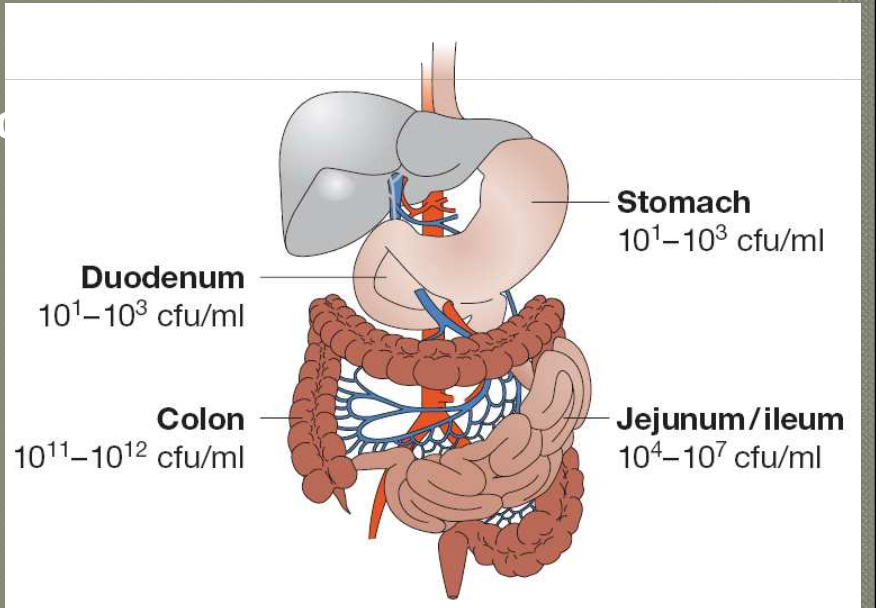
->Education des cellules T:
->induction de facteurs proinflammatoires (cytokines).
->Production de IL10 et stimulation de cellules T reg -> immunosuppression et tolérance
En absence de bactéries commensales : mauvaise/absence de mise en place de structures immunitaires spécifiques à l'intestin
La bactérie commensale éduque le système immunitaire en vue d'induire une tolérance

- Quels sont les effets du microbiote sur l'immunité?
- Quand absence de TLR : colonisation de la rate par les bactéries les bactéries sont passées dans la circulation par franchissement de la barrière épithéliale non du à une rupture de la barrière épithéliale
- L'absence de TLR (Myd88 -/-) augmente la production d'anticorps en présence de bactéries commensales → L'immunité adaptative est activée dans un cadre où elle ne l'est normalement pas
- Les bactéries de la famille des Clostridium(SFB) induiraient la réponse immunitaire intestinale;
- La colonisation par SFB induit la production de IL17, IL10 et IFNγpar des cellules CD4.**SFB est un composant de la microflore indispensable à la maturation des cellules T de l'intestin;**
- SFB induit l'expression de IL17, IL10, du ligand de CD40 et IFNgdans les plaques de Peyer**SFB a un effet stimulateur sur l'immunité au niveau des plaques de Peyer**

Gut microbiota

- more than 1000 species
- collective weight of about 1kg in human intestine
- colonization begins immediately after birth
- symbiotic bacteria provide benefits to the host:
  - nutrient supply
  - pathogen defense
  - immune system development/ func

Anaerobic genera	Aerobic genera
Bifidobacterium	*Escherichia*
Clostridium	*Enterococcus*
Bacteroides	*Streptococcus*
Eubacterium	*Klebsiella*



O´Hara and Shanahan, EMBO reports, 2006

Gut-associated lymphoid tissue (GALT)

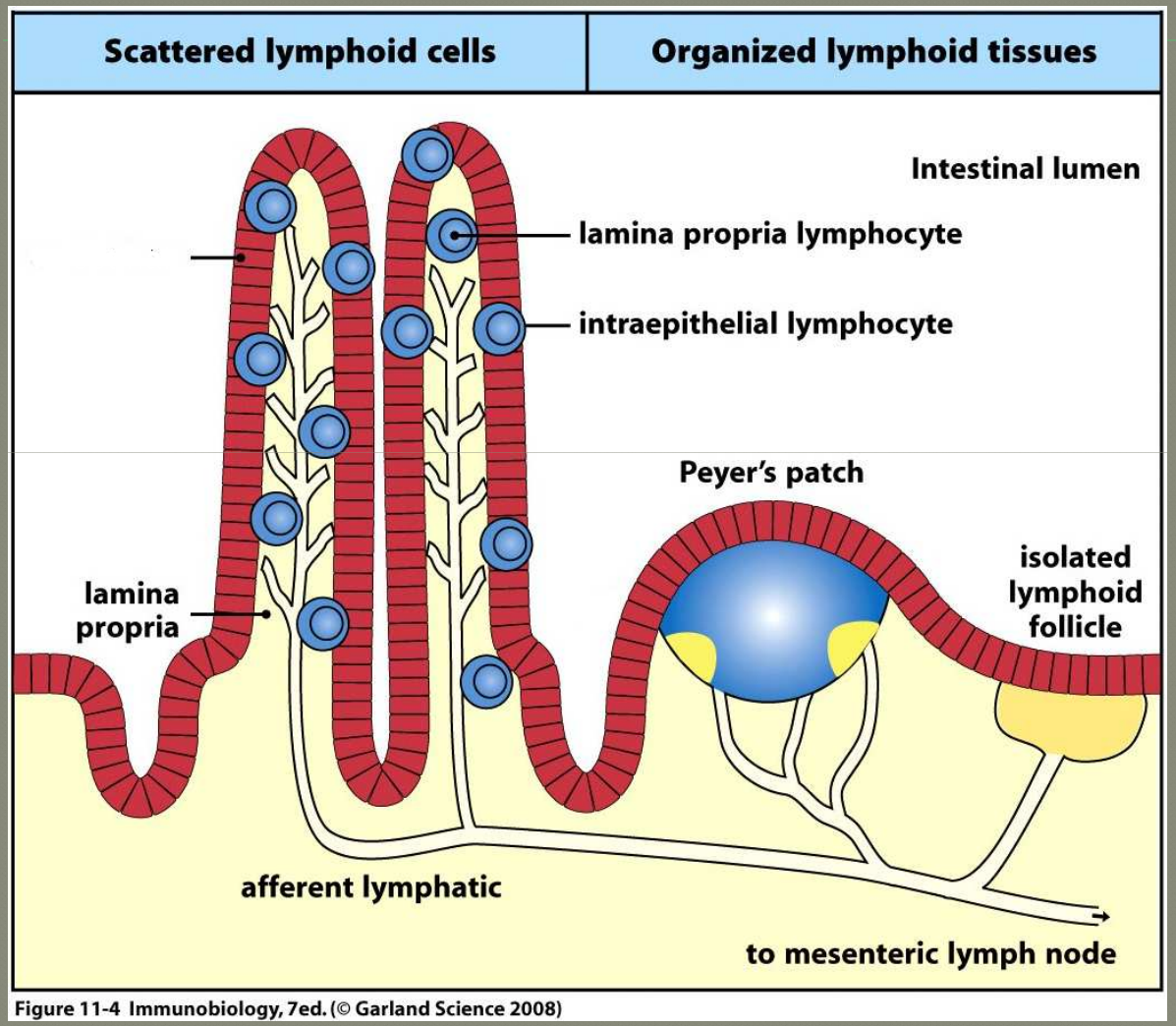


Figure 11-4 Immunobiology, 7ed. (© Garland Science 2008)

GALT

- **Peyer`s patches**
- **appendix**
- **isolated lymphoid follicles**

= T-cell areas

= B-cell follicles

Defects in immune response in germ-free mice

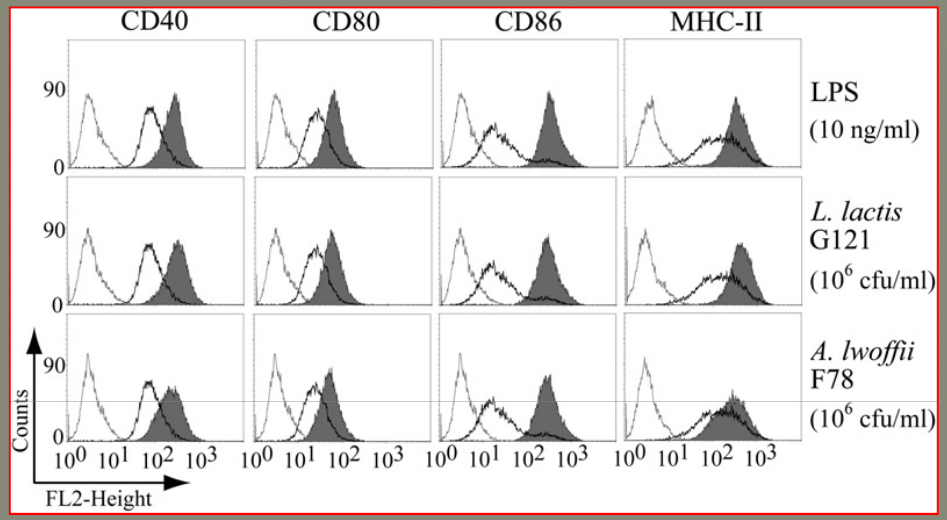
- **decreased immune resistance to infection with *Shigella flexneri***
- **decreased bacterial clearence upon *Listeria monocytogenes* infection**
- ***Salmonella enterica* serovar Typhimurium exploits deficiency in colonization resistance to establish infection**

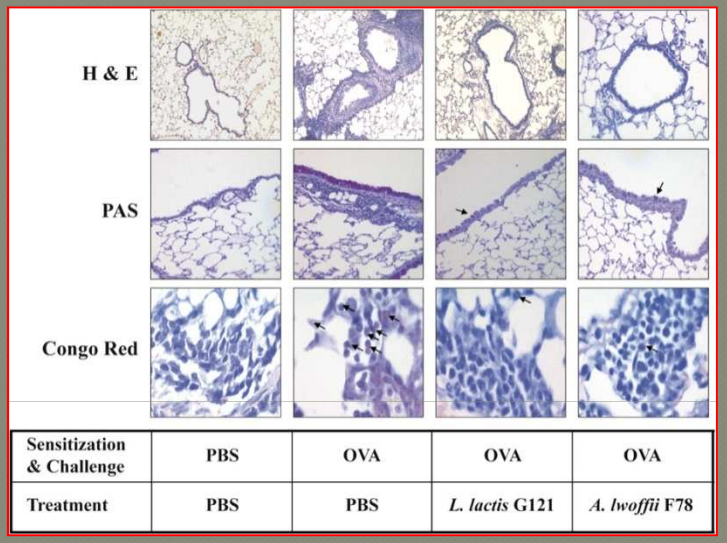
Developmental defects in germ-free mice

- IEC show decreased rates of cell-turnover and reduced expression of MHC class II molecules, TLR9 and antimicrobial proteins
- fewer lymphocytes in lamina propria and epithelium
- fewer and smaller Peyer`s patches, isolated lymphoid follicles and mesenteric lymph nodes
- reduced levels of secretory IgA

Macpherson and Harris,
Nat rev Immunol., 2004

Acinetobacter lwoffii and Lactococcus lactis strains isolated from farm cowsheds possess strong allergy-protective properties (J ALLERGY CLIN IMMUNOL VOLUME 119, NUMBER 6)

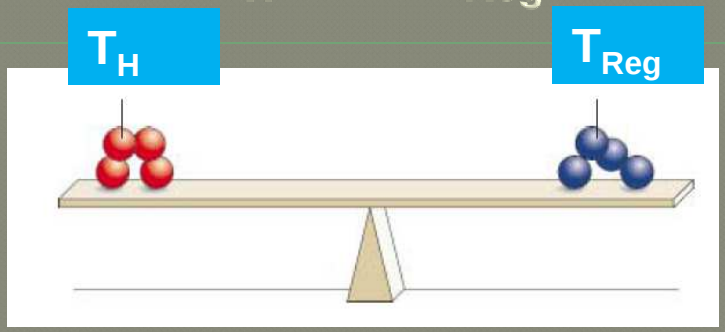




Both bacteria induce upregulation of costimulatory molecules in human moDCs. Analysis of CD40, CD80, CD86, and MHC class II expression in human moDCs after treatment (shaded histograms) with indicated stimuli for 24 hours is shown. Gray open histograms indicate the isotype control, and black open histograms indicate untreated moDCs. Data are representative for 3 independent experiments with moDCs from different healthy donors

Intranasal treatment with L lactis G121 and A lwoffii F78 inhibits development of airway inflammation and goblet cell hyperplasia. Mice were treated as described in Fig 4. Shown are representative microphotographs of airways from mice after hematoxylin and eosin staining, periodic acid–Schiff staining of goblet cells (arrows point to individual goblet cells), and Congo Red staining of eosinophils (arrows).

Involvement of the microbiota in regulating the balance between T_H and T_{Reg} cell subsets in the gut

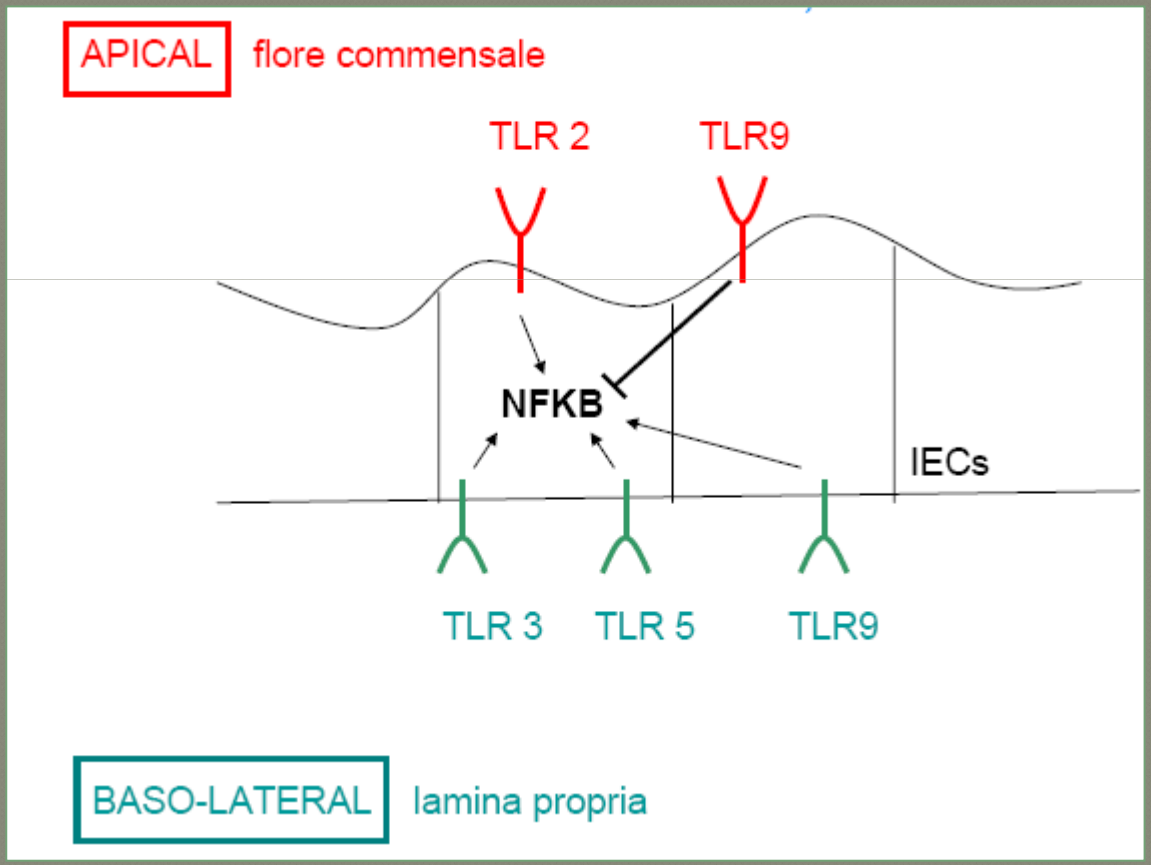


- germ-free animals show defective T_H17 cell development in the small intestine
- ATP generated by intestinal bacteria increases the production of IL-17 in the colon

- germ-free mice have reduced numbers of T_{Reg} cells in the mesenteric lymph nodes
- increased numbers of T_{reg} cells in the small intestine of germ-free mice

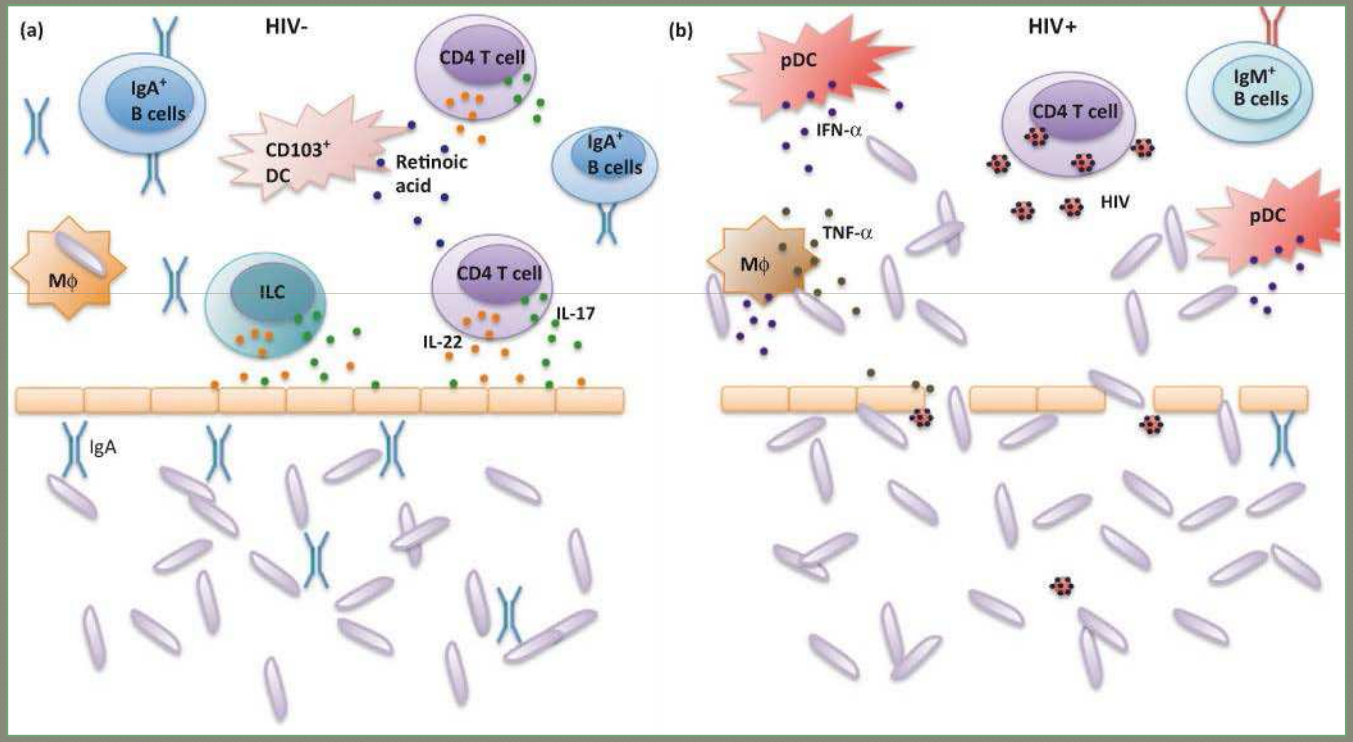
→ Intestinal bacteria direct the differentiation of both pro- and anti-inflammatory T cell populations and may therefore play a crucial role in IBD

Comment les bactéries commensales induisent l'immunité?



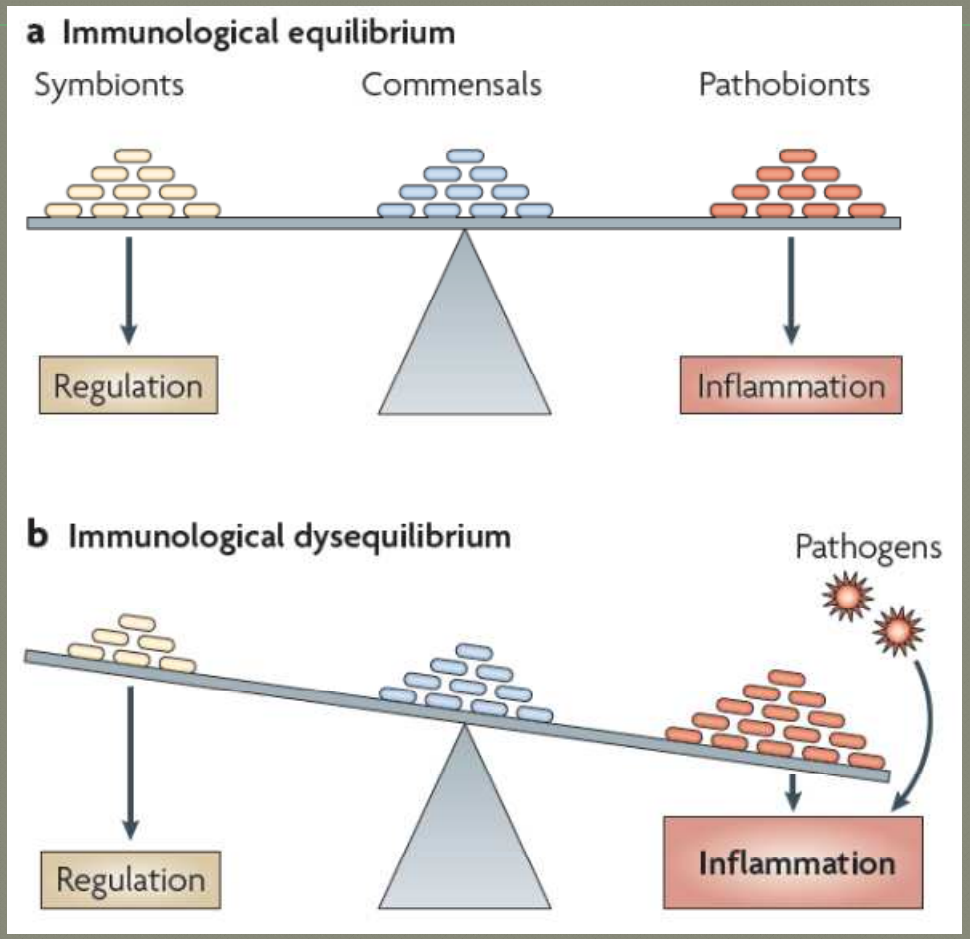
Translocation microbienne

NI inflammatoire



Le déséquilibre Th17 / Treg intervient dans la pathogénèse des maladies inflammatoires digestives

Immunological dysregulation associated with dysbiosis of the microbiota



Symbionts

→ health promoting functions

Commensals

→ provide no benefit or detriment

Pathobionts

→ have the potential to induce pathology

→ Inflammatory responses in IBD are directed towards pathobionts

Macrophage migration inhibitory factor (MIF)

Une cytokine de distribution ubiquitaire,

responsable de la réaction inflammatoire

Immunocytes:
- Lymphocytes T.
-Macrophages > cellules dendritiques - mastocytes - éosinophiles…

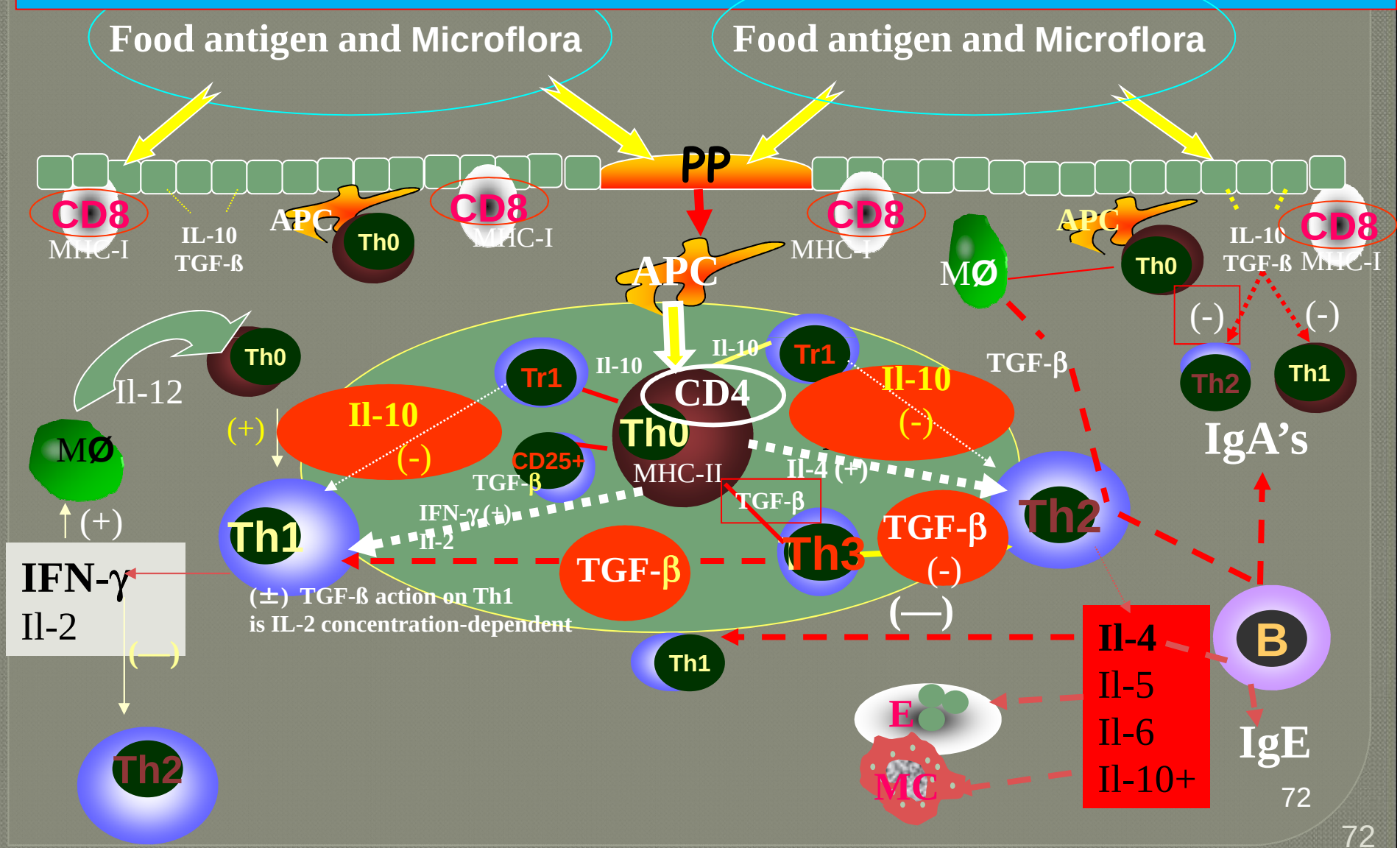
Tractus digestif:
- Cellules épithéliales et muqueuses gastrique, intestinale et colique.

Sphère uro-génitale:
- Cellules épithéliales de l'uretère, de la vessie, de l'utérus et du vagin,
- Glandes endométriales, ovaires, testicules et prostate.

Système nerveux:
- Axe cérébro-spinal (hypothalamus, astrocytes, moelle épinière lombo-sacrée…),
- Système nerveux autonome (afférences, efférences, cellules gliales ….).

Food Antigen – Epithelial Cell – Microflora Interface :

Th1/Th2 optimal equilibrium and a high level of T-reg Cells activation process



72

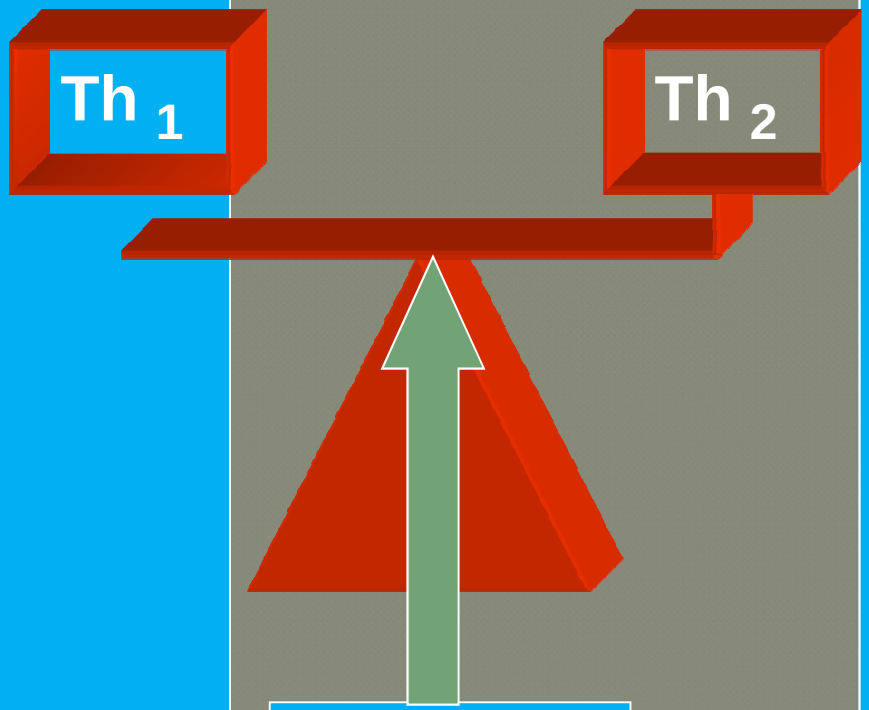
Facteurs permettant la tolérance immune intestinale

- Cellules épithéliales
- Cellules dendritiques
- Cellules T régulatrices
- Rôle probable de la flore intestinale, plus de bactérie dans le colon, que de cellules dans le corps..
- Lactobacillus GG dans le lait maternel.

La flore intestinale et l'immunité

Th $_1$ response

- **IFN - gamma**
- **TNF - alpha**
- **IL – 1**
- **IL - 2**
- **IL - 12**
- **IL – 18**
- **- Peptic ulcer**
- **- Cholestatic disease**
- **-Crohn 's disease**
- Ulcerative colitis



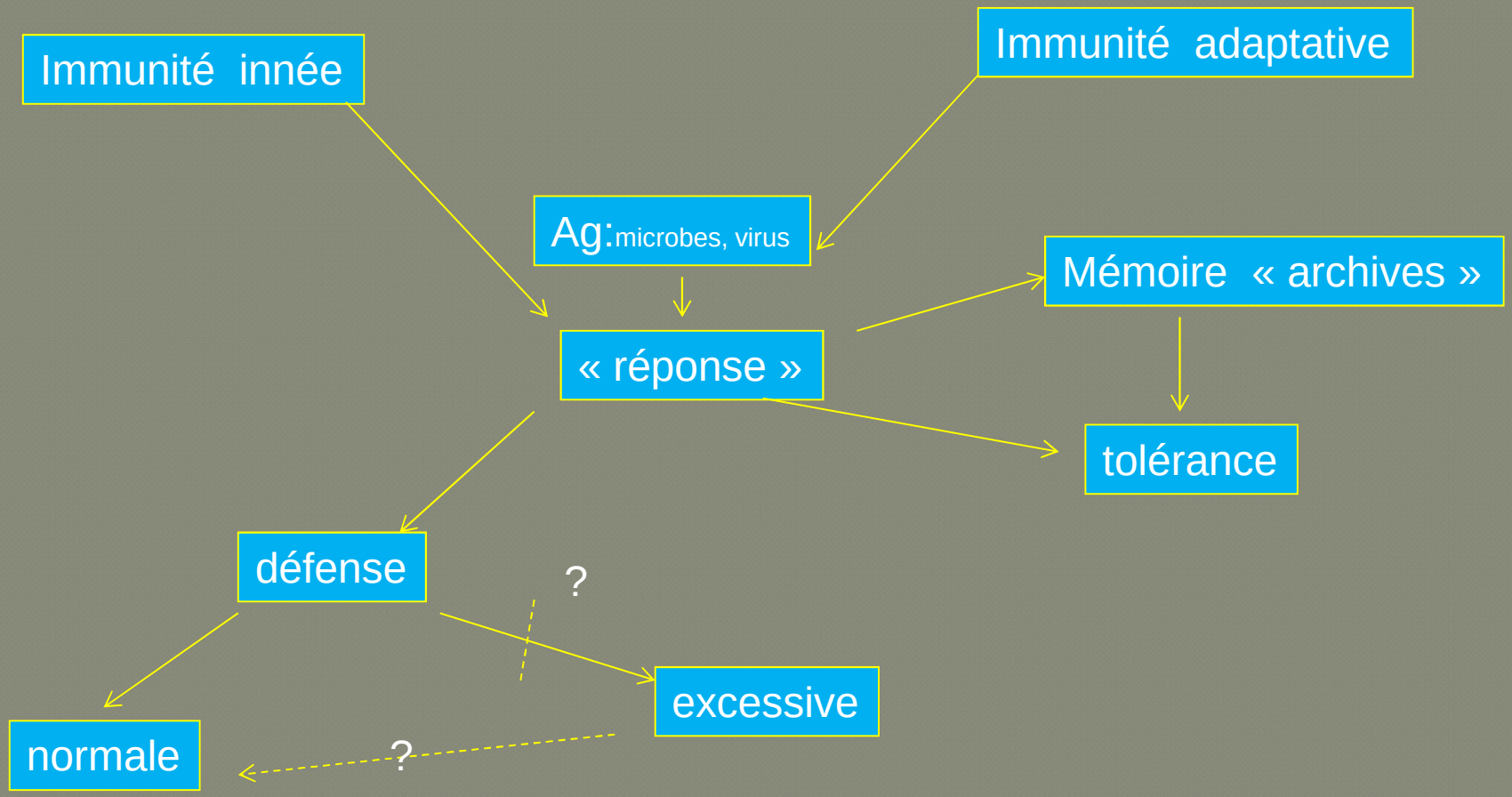
T Reg Cells :
- Th3 (TGF-ß)
- Tr-1 (Il-10)

Th $_2$ response

- **IL - 3**
- **IL - 4**
- **IL - 5**
- **IL - 9**
- **IL - 10**

- Food allergy

74


Immunité innée
Immunité adaptative
Ag: microbes, virus
Mémoire « archives »
« réponse »
tolérance
défense
?
excessive
normale
?

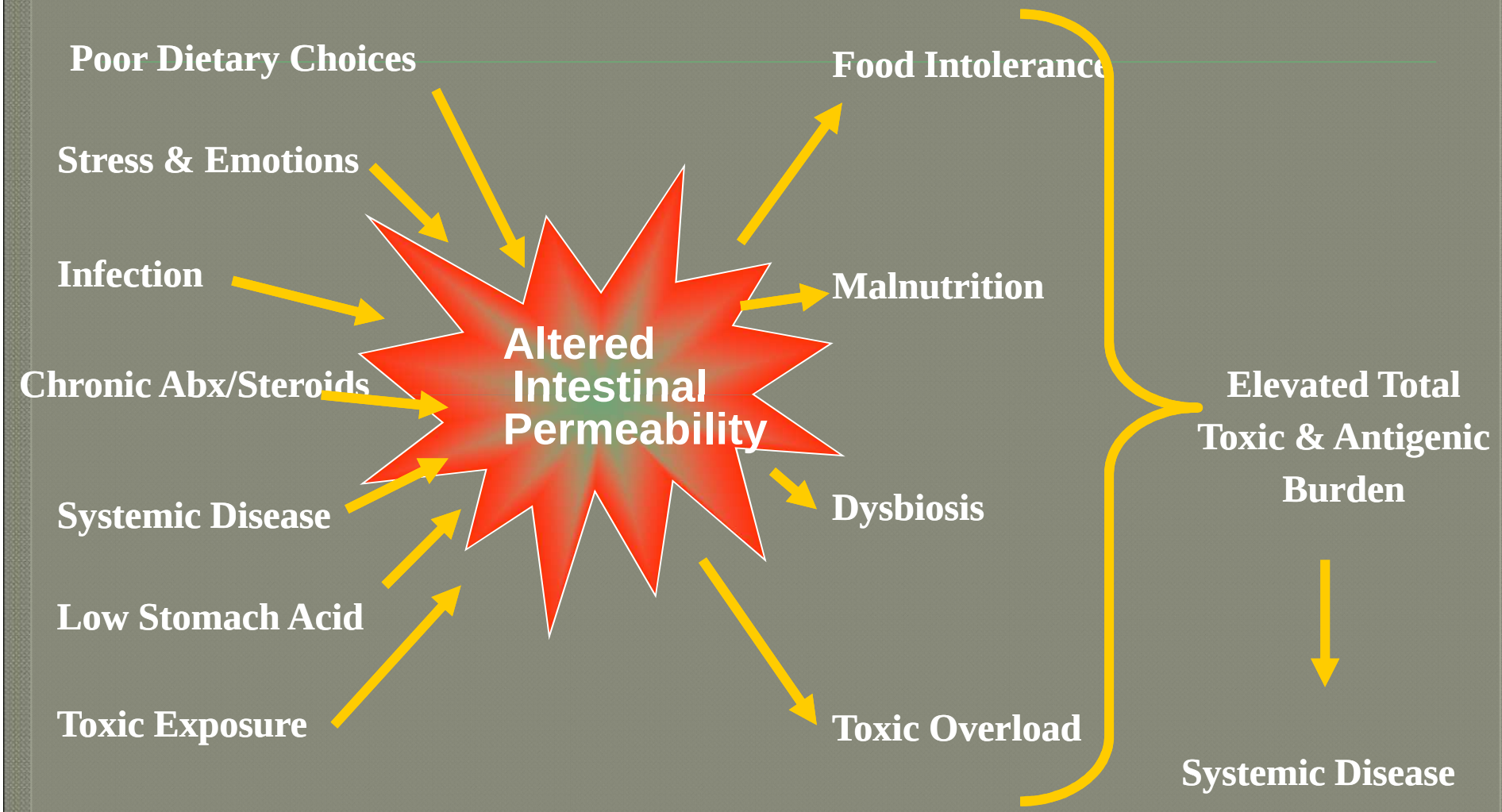
LA REPONSE IMMUNITAIRE

- innée

- adaptative

Réponses:
-inflammation → perméabilité augmentéearmeability
- arrivée et mise en oeuvre des phagocytes

↑ Intestinal Permeability - Pathophysiology



Chez le fetus et le nné il y a des différences notables avec l'adulte

- Expression réduite du gène 88 et du gène 35→ réduction de la réponse TLRc
- Réduction de la production de cytokines lors d'une provocation par liposaccharide
- Taux élevé d'adénosine →Taux élevé d'AMPc intracellulaire → réduction de la production de cytokines par les TLRc

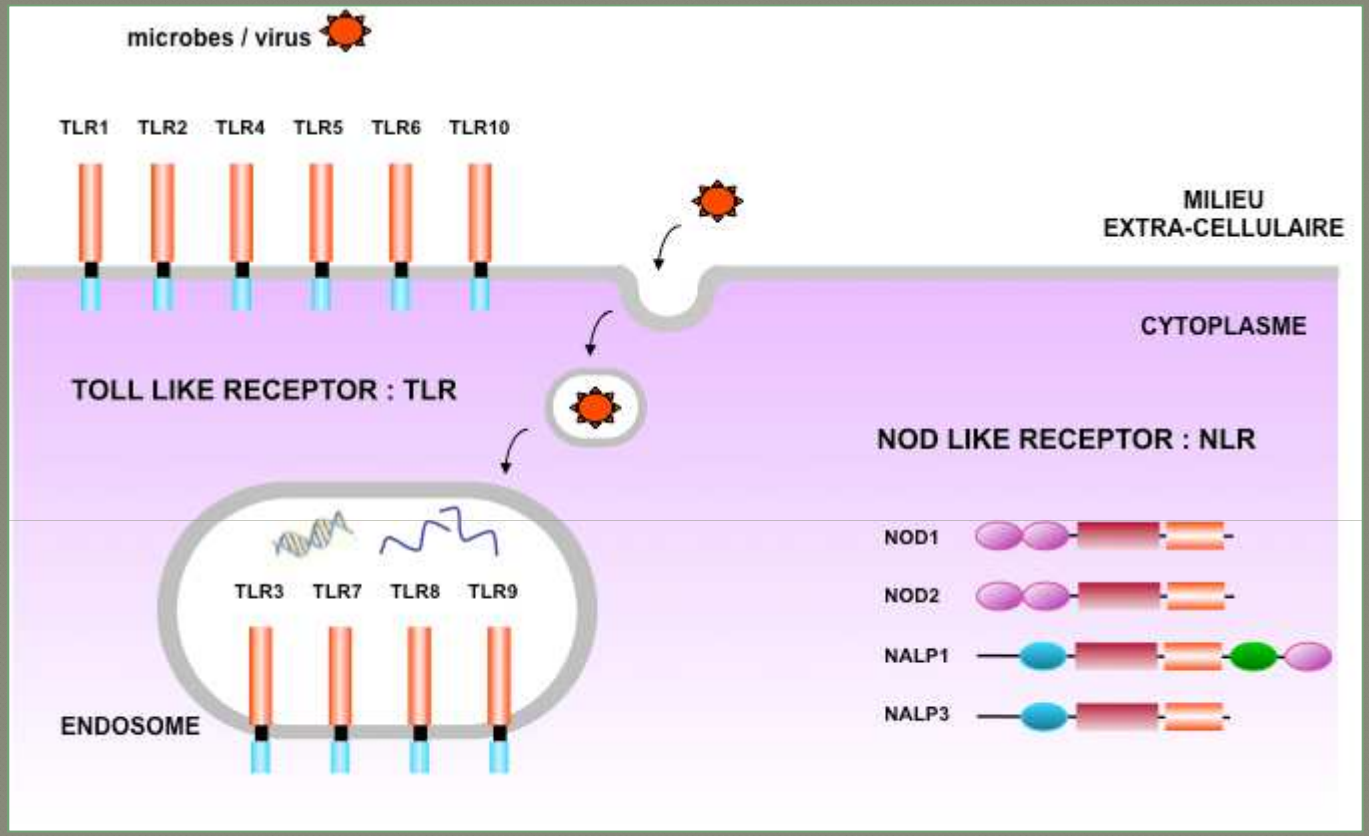
Evidence for the involvement of commensal bacteria in IBD

- IBD patients have increased antibody titres against indigenous bacteria
- inflammatory lesions are pronounced in areas with high numbers of bacteria
- genetic variants that are highly linked to IBD include mutations in genes that are involved in bacterial sensing (*NOD2*) and T cell immunity (*IL23R*)
- IBD patients show abnormal microbial composition (= dysbiosis)
- mice studies showed that dysbiosis alone may be important for the induction of IBD

Lactococcus lactis strain prevents food-induced IgE sensitization

- Nonpathogenic IL-10–producing microorganisms in the gut could have a potential to prevent systemicfood-induced anaphylaxis.(JAllergyClinImmunol 2007;119:952-9

Reconnaissance des microbes et immunité innée



Probiotics in primary prevention of atopic disease: A randomised placebo-controlled trial

Lancet, 2001. Vol. **357** (**9262**)

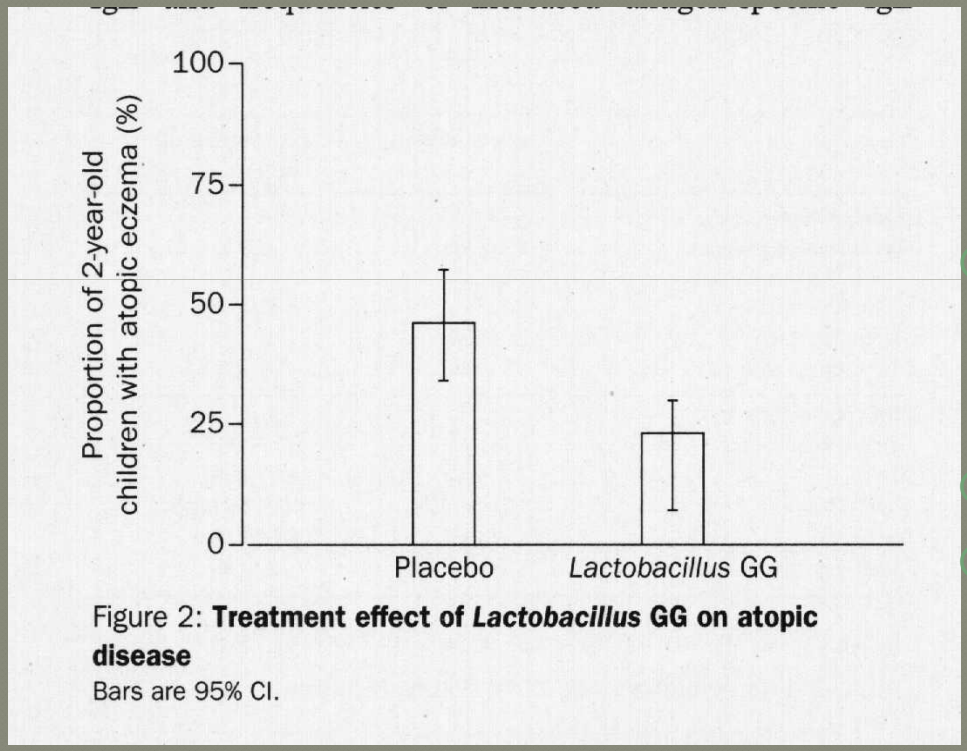


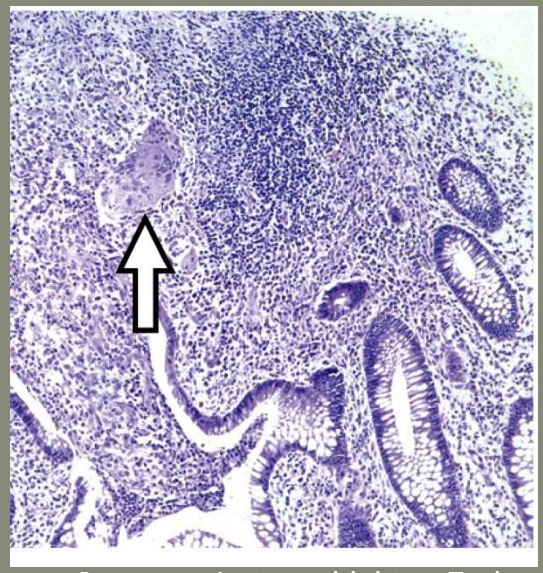
Figure 2: **Treatment effect of *Lactobacillus* GG on atopic disease**
Bars are 95% CI.

- Probiotics given prenataly and postnataly for a total of 6 months.
- Lactobicillus GG reduced eczema by 50%
- NNT = 4.5
- Follow-up study showed that effect persisted to 5 yrs of age.

Inflammatory bowel disease (IBD)

- chronic inflammation of the gastointestinal tract
- two main forms:
  1. Crohn`s disease
     - affects all layers of the bowel wall
     - granuloma formation in up to 60% of patients
  2. Ulcerative colitis
     - affects superficial mucosal layers
- no pathogen has been conclusively shown to be the causative agent
- incidence is the highest in developed countries

Crohn´s disease



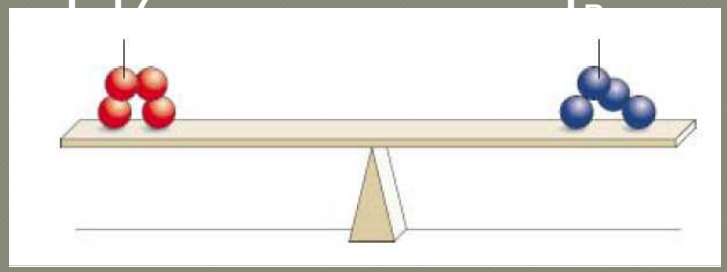
Janeway, Immunobiology, 7ed.

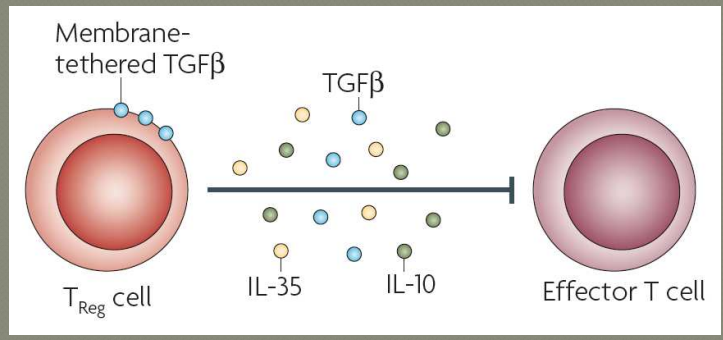
IBD is driven by T cells

mucosal homeostasis

→ cytokine production by regulatory (T_{Reg}) T cells supresses pro-inflammatory responses

T_H1, T_H2,

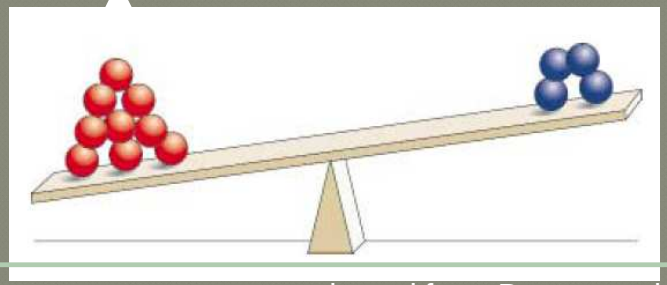




mucosal inflammation

→ increased production of pro-inflammatory cytokines by T helper (T_H) cells

TNF, IFNγ, IL-17



← T_{Reg} transfer can prevent the induction of experimental colitis

adapted from Bouma and Strober, Nat rev Immunol., 2003 and Vignali et al., Nat rev Immunol., 2008

Different bacterial species ameliorate the symptoms of IBD

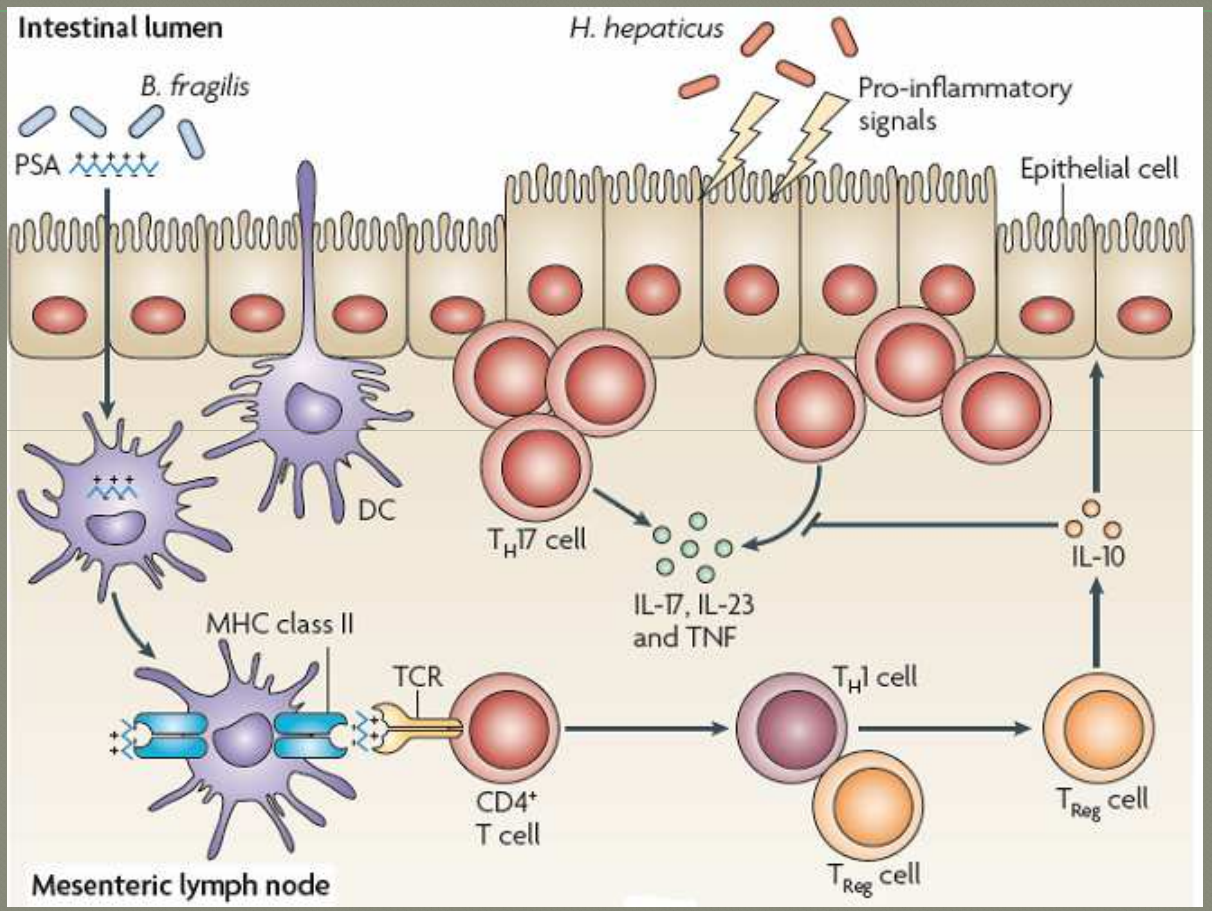
Probiotics

- dietary microorgansims that are beneficial to the health of the host
- act on serveral cell types (epithelial cells, DCs, T cells)
- ability to limit inflammation by induction of T_{Reg} cells

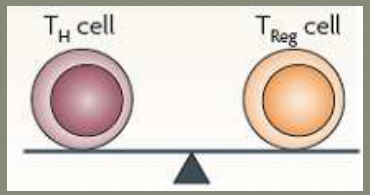
Bacterial strain	Model system	Disease type or model	Mechanism of disease suppression
VSL#3	Human and mouse	Pouchitis, ulcerative colitis and TNBS-induced colitis	Induction of IL-10- and TGFβ-expressing T cells
Bifidobacteria infantis	Mouse	*Salmonella enterica*-induced enteritis	Induction of T_{Reg} cells and inhibition of NF-κB activation
Bacteriodes fragilis	Mouse	T cell transfer and TNBS-induced colitis	Production of $CD4^+$ T cell-derived IL-10

VSL#3: mixture of *Lactobacillus* spp., *Bifidobacterium* spp., *Streptococcus salivarius*

Model for *Bacteroides fragilis*-mediated protection from disease induced by *Heliobacter hepaticus*



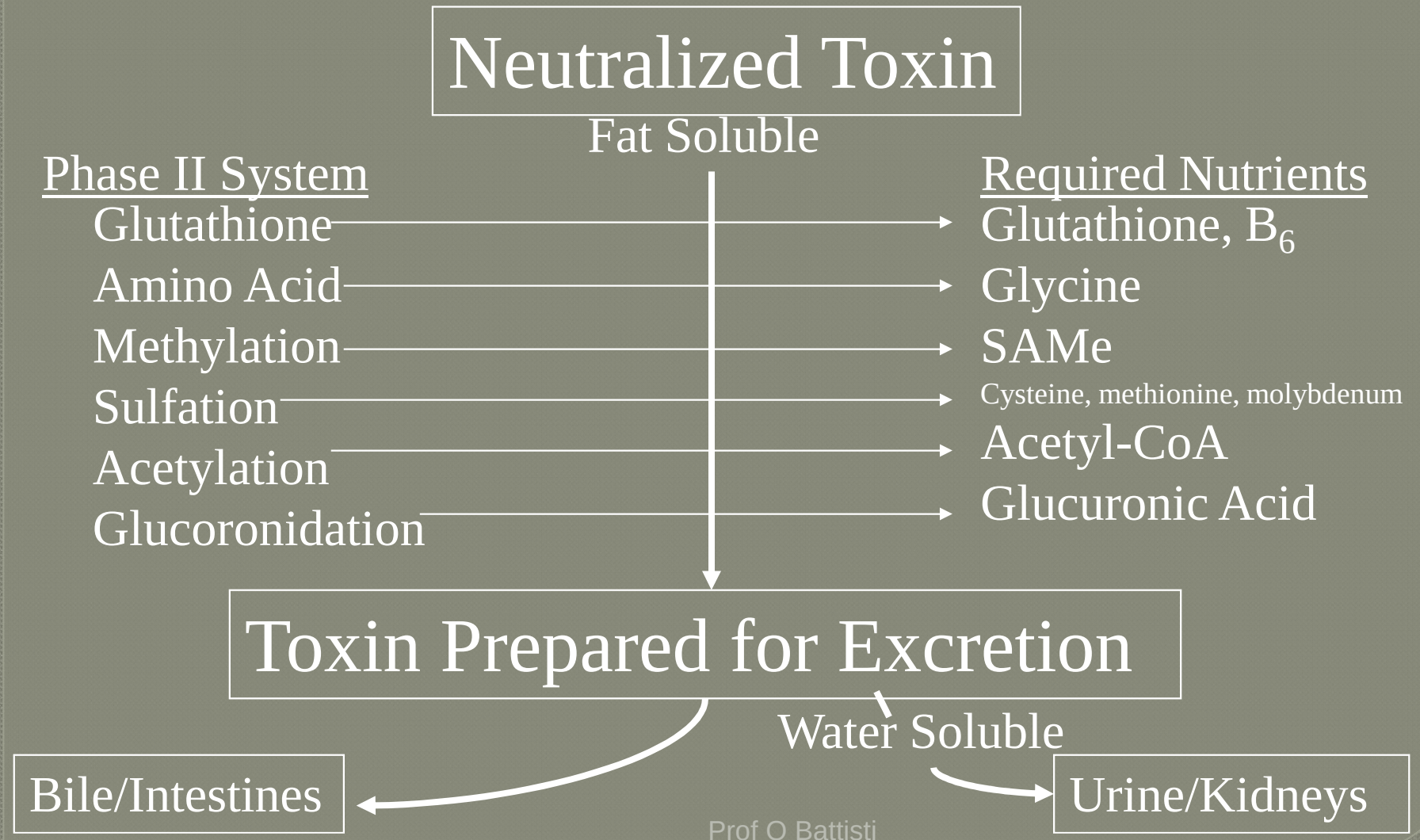
PSA: polysaccharid A (= symbiotic factor)



→ symbiotic factors actively maintain health

Phase 2 Detoxification

Conjugation



Increasing Glutathione

Safe with long-term use;

- Vitamin C 500 mg/d
- Vitamin E 800 IU/d (Mixed Tocopherols)
- B-Complex Vit. *(Thiamine & B6)*
- Milk Thistle

Needs Further Study;

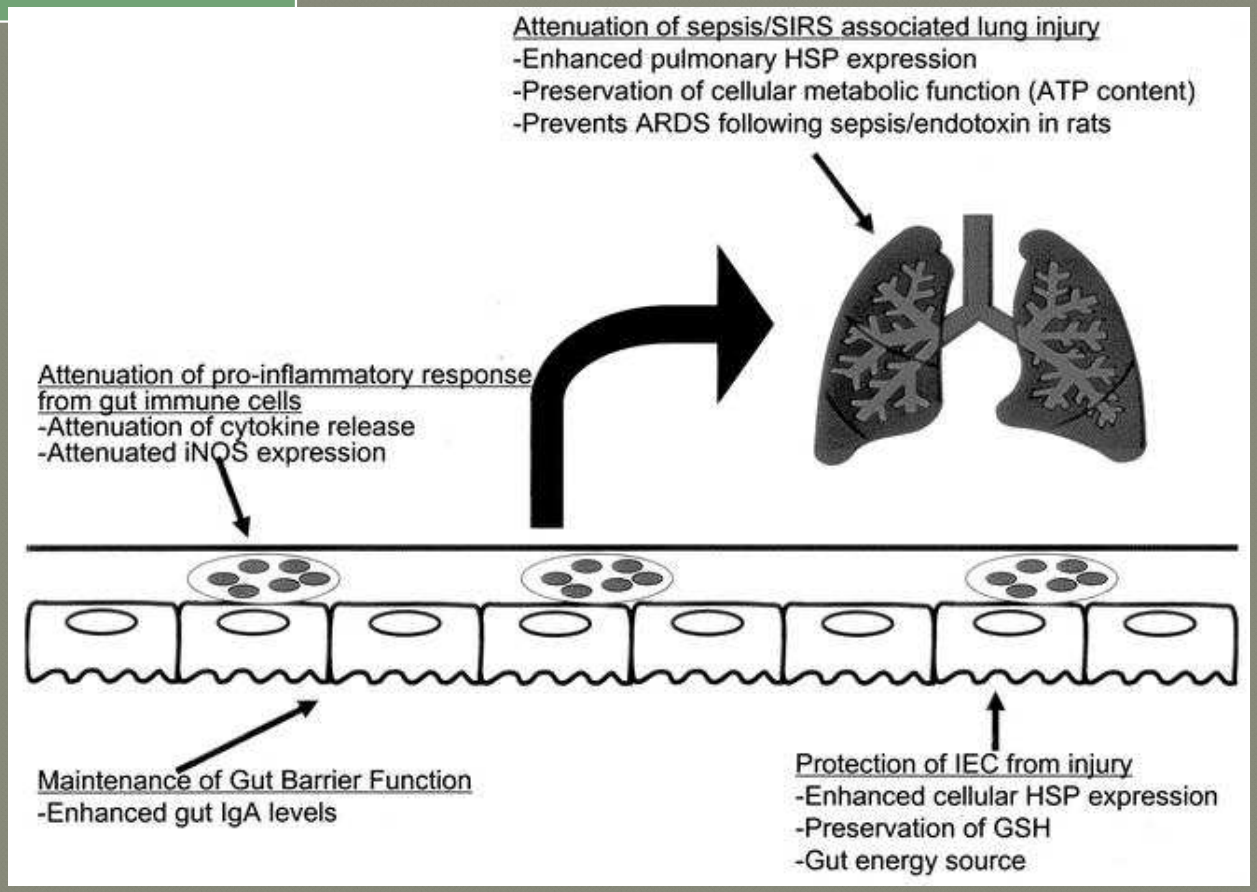
- NAC *(N-Acetylcysteine aka; Mucomyst)*
- Glutathione *(Good in food: fruits, vegetables & fish. Or a Mediterranean type diet)*

Glutamine MOA

- Reduces Inflammation by reducing gut associated cytokine release
  - Wischmeyer PE. Can glutamine turn off the motor that drives systemic inflammation? Crit Care Med. 2005; 33(5):1175-1178.
- Reduces risk of gut derived septicemia in critically ill
- Antioxidant, Increases Glutathione (GSH)
- Enhances Heat Shock Protein (HSP) that protects enterocytes from injury.
- Does not reduce permeability in those without inflammation who are nutritionally deprived
  - Hulsewe KW, et al. Clin Nutr. 2004; 23(5):1217-1225.

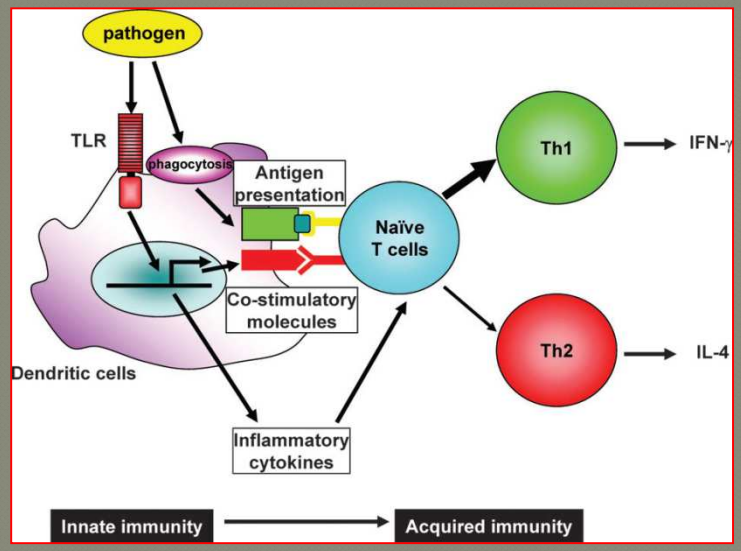
Glutamine's Benefit in the ICU

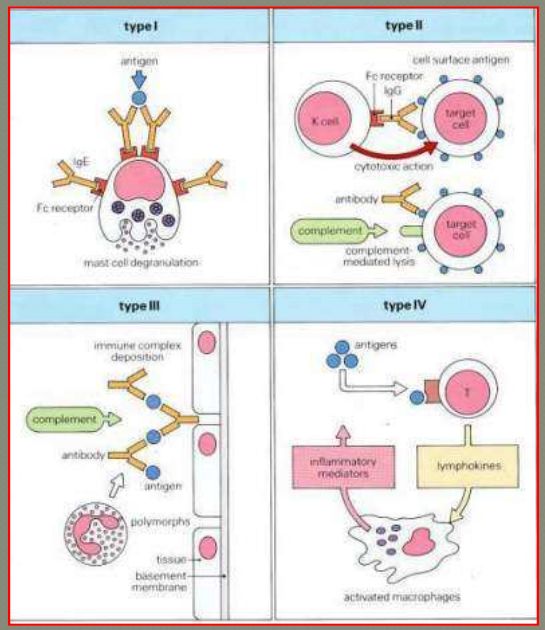
De-Souza DA, Crit Care Med. 2005; 33(5):1125-1135.



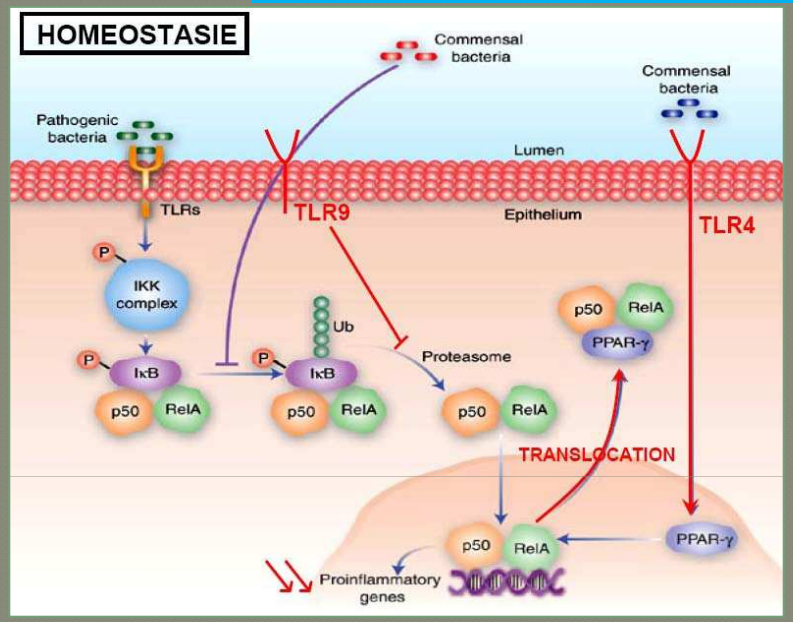
L'hyper réactivité immunitaire

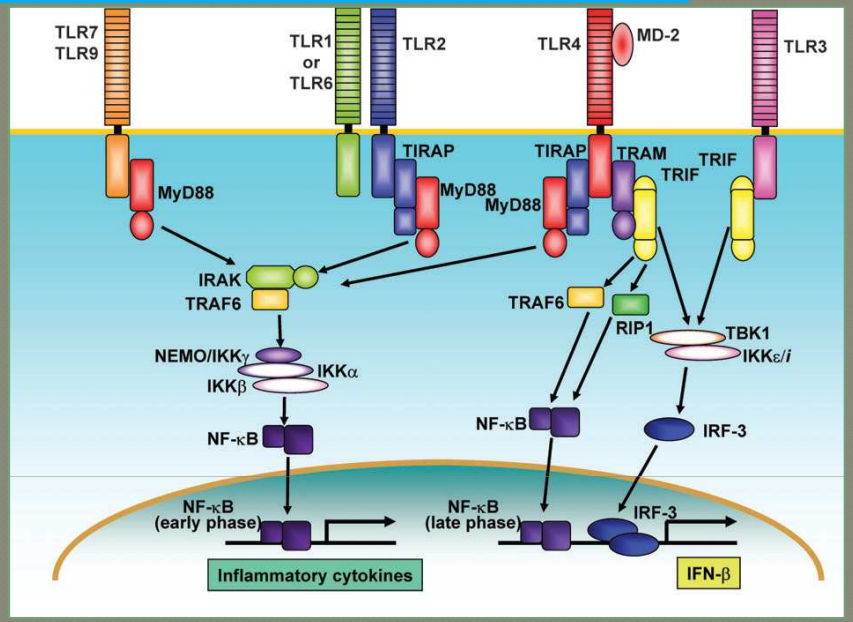
- Elle concerne la peau, l'épithélium respiratoire et l'épithélium bronchique
- Quelle est la place des
  - infections virales précoces ?
  - Antibiothérapies précoces ?
  - Certains facteurs périnataux ?
  - La flore commensale maternelle et infantile ?
  - L'obésité ?





L’absence de bactéries commensales entraîne une diminution de la synthèse des cytokines





Le TLR a un rôle dans la maturation et l’éducation du système immunitaire pour la modulation de la réponse effectrice ce qui permet
-la tolérance des bactéries commensales
-la réaction aux bactéries pathogènes
-Ce sont les composants du microbiote intestinal qui permettent cela, en stimulant les plaques de Peyer: stimulation des Treg, production de IL10, production d’IgA et empêchement de production d’IgG

Facteurs favorisants

Oui pour:

Usage du tabac pendant la grossesse et après…

early-life:

- **Prematurity and fetal growth retardation**
- **environmental tobacco smoke exposure**

And risk of wheeze or bronchial asthma *BMC Pediatrics 2012,* ***12:187 doi:10.1186/1471-2431-12-187***

Flore intestinale précocément perturbée (césarienne, antibiothérapie, nutrition)

Bronchiolite voire rhinite

Non pour

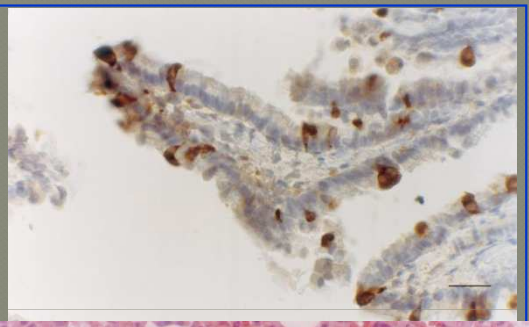
Paracetamol use in early life and asthma: prospective birth cohort study

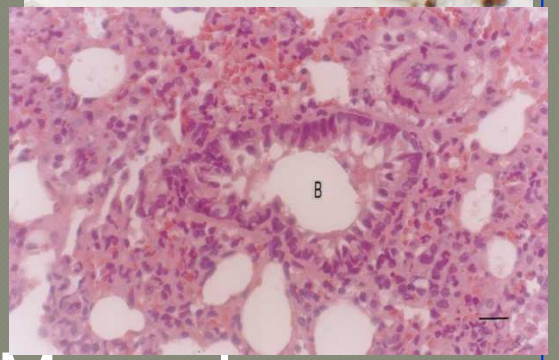
Adrian J Lowe, John B Carlin, Catherine M Bennett, Clifford S Hosking, Katrina J Allen, Colin F Robertson, Christine Axelrad, Michael J Abramson, David J Hill, Shyamali C Dharmage

BMJ 2010;341:c4616 (Published 15 September 2010)

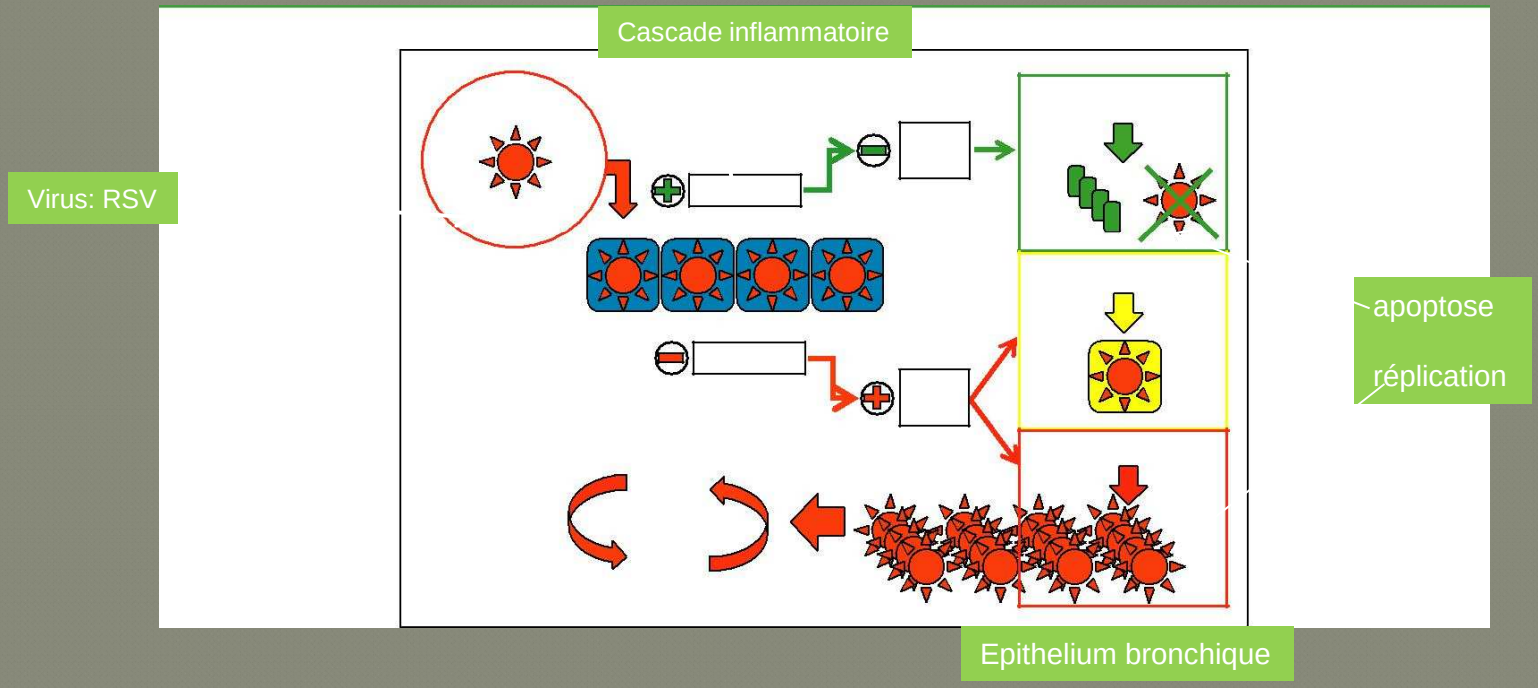
Quelle est la place de la Bronchiolite et de la rhinite

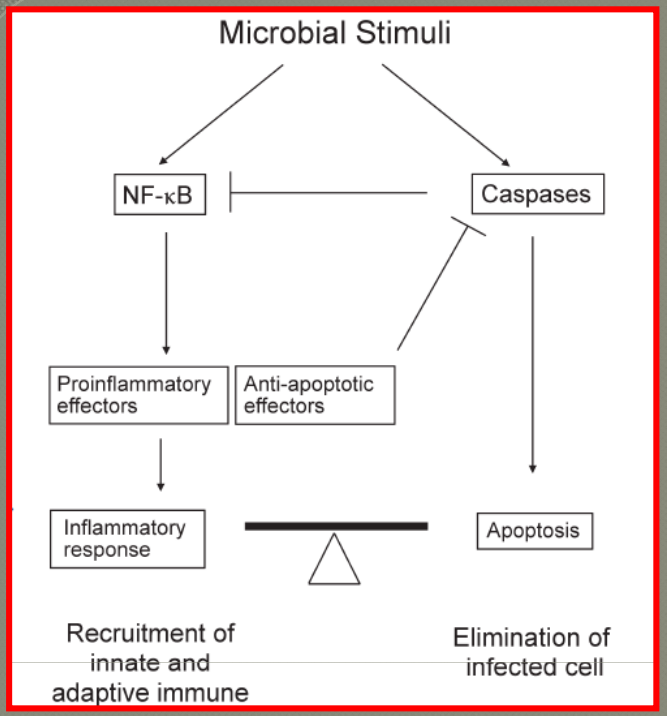
- Les virus
  - RSV
  - Parainfluenza
  - Metapneumovirus
  - Influenza
  - Rhinovirus
  - Coronavirus
  - bocavirus
- Parfois des germes atypiques Mycoplasma pneumoniae, chlamydia trachomatis





The consequent upregulation of the NGF-TrKA axis not only potentiates the local nociceptive innervation and neurogenic inflammation in distal airways, but also functions as a critical virulence mechanism implemented by RSV to coax host cells to resist apoptosis and persist latently in the lungs, and/or in a safe extrapulmonary niche within the bone marrow mesenchyme wherein it avoids detection by the immune system. Persistence of RSV virions and chronic upregulation of the NGF–TrKA axis may turn on lytic replication and inflammation in response to viral reinfection or reactivation, contributing to persistent airway hyperreactivity (AHR) and obstructive lung disease.





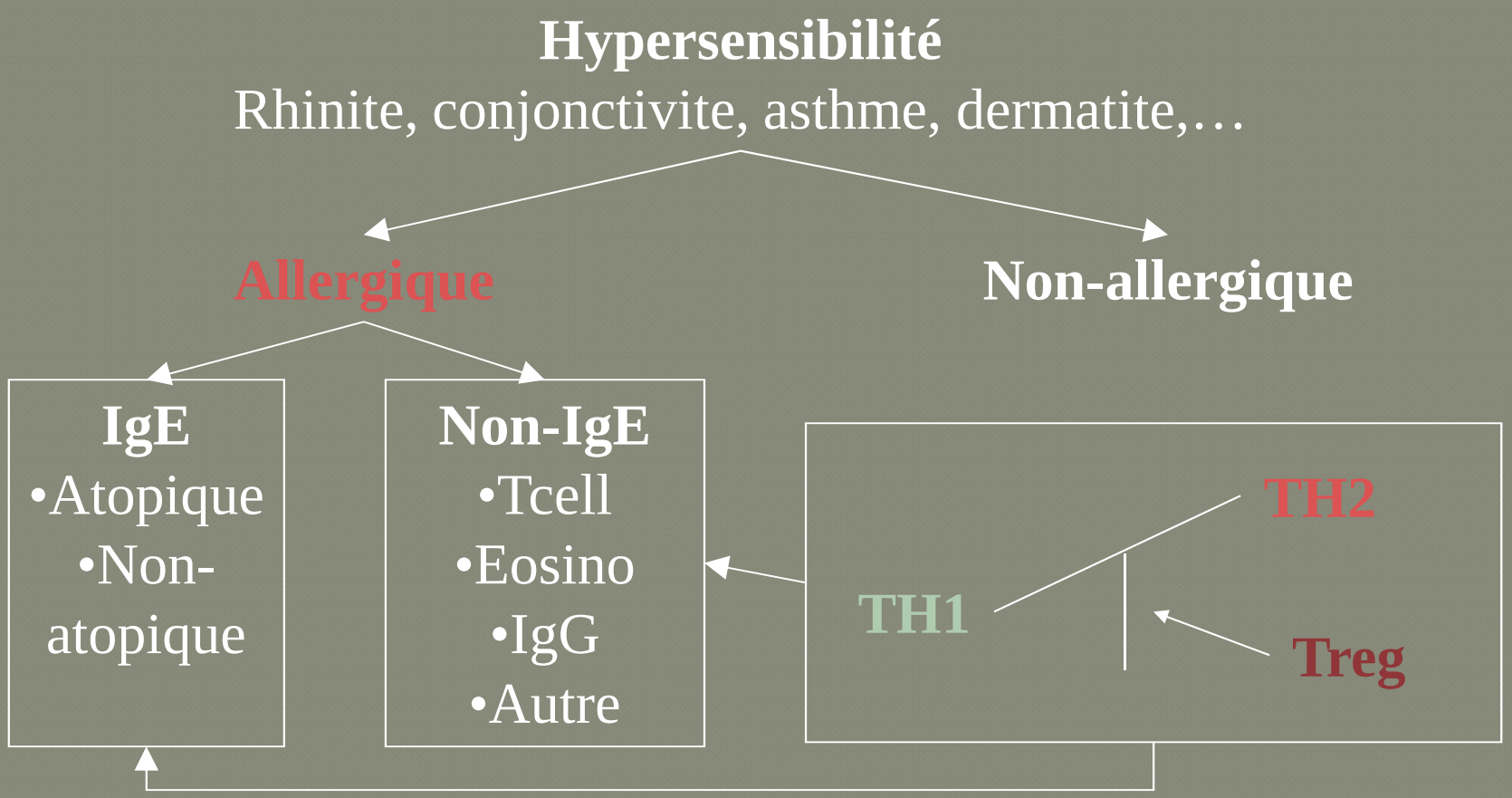
Les bactéries commensales conduisent à
l'expansion des LT CD4+ ainsi que celle des LT régulateurs (CD4+ et CD8+)
dans les ganglions mésentériques.

- Expression de gènes différents chez les bactéries commensales :
Moins de gènes de virulence et de pathogénicité (moins bonne survie si infection) -

- Digestion de l'amidon par les commensales
forme des petites unités de butyrate qui vont inhiber la production
de cytokines pro inflammatoires
induite par la présence de cette flore.
Cela augmente la production d'IL10 régulatrice.

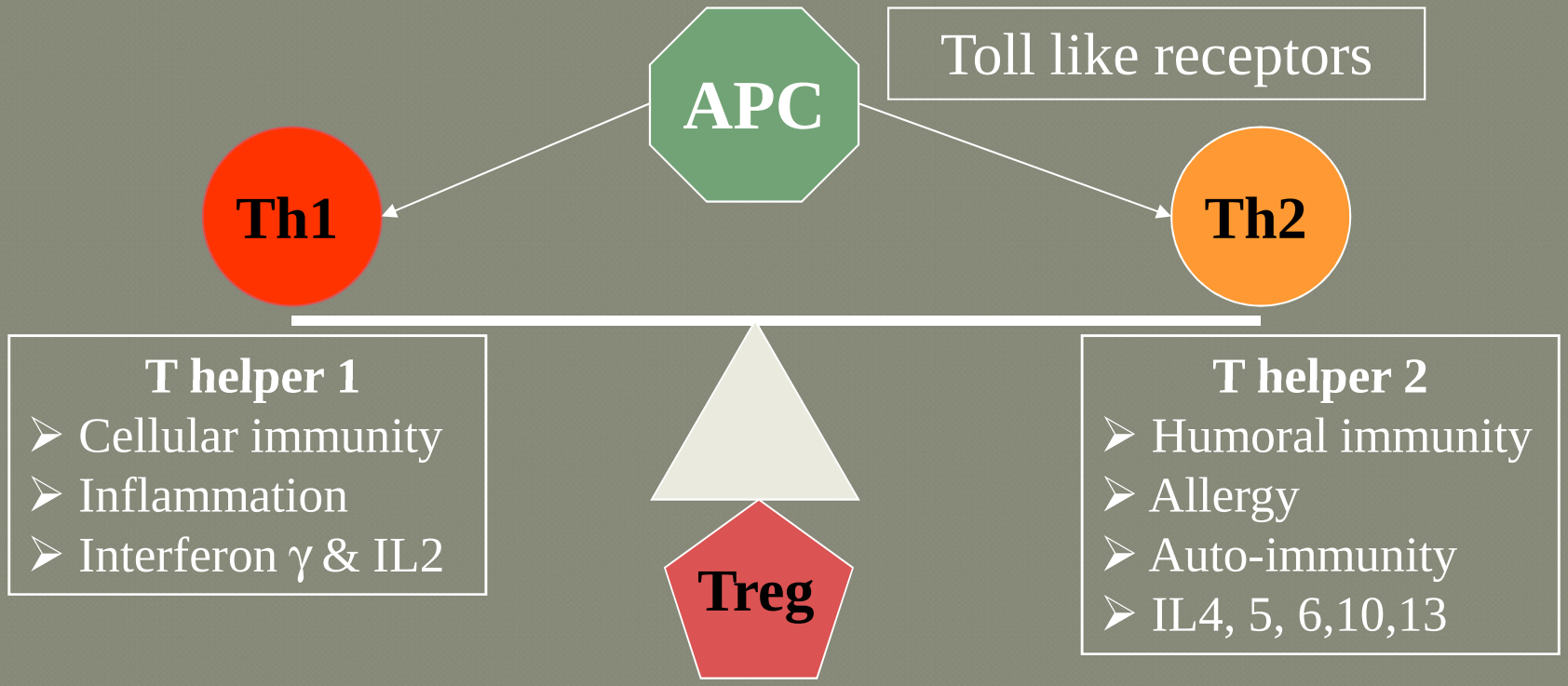
La flore commensale maternelle a une importance primordiale
Dans l'installation de l'immunité adaptative de l'enfant

Les Mécanismes de L'hypersensibilité



Gore et al.
Allergy 2004;59:151-61

La Balance Immunitaire



Facteurs influençant l'allergie

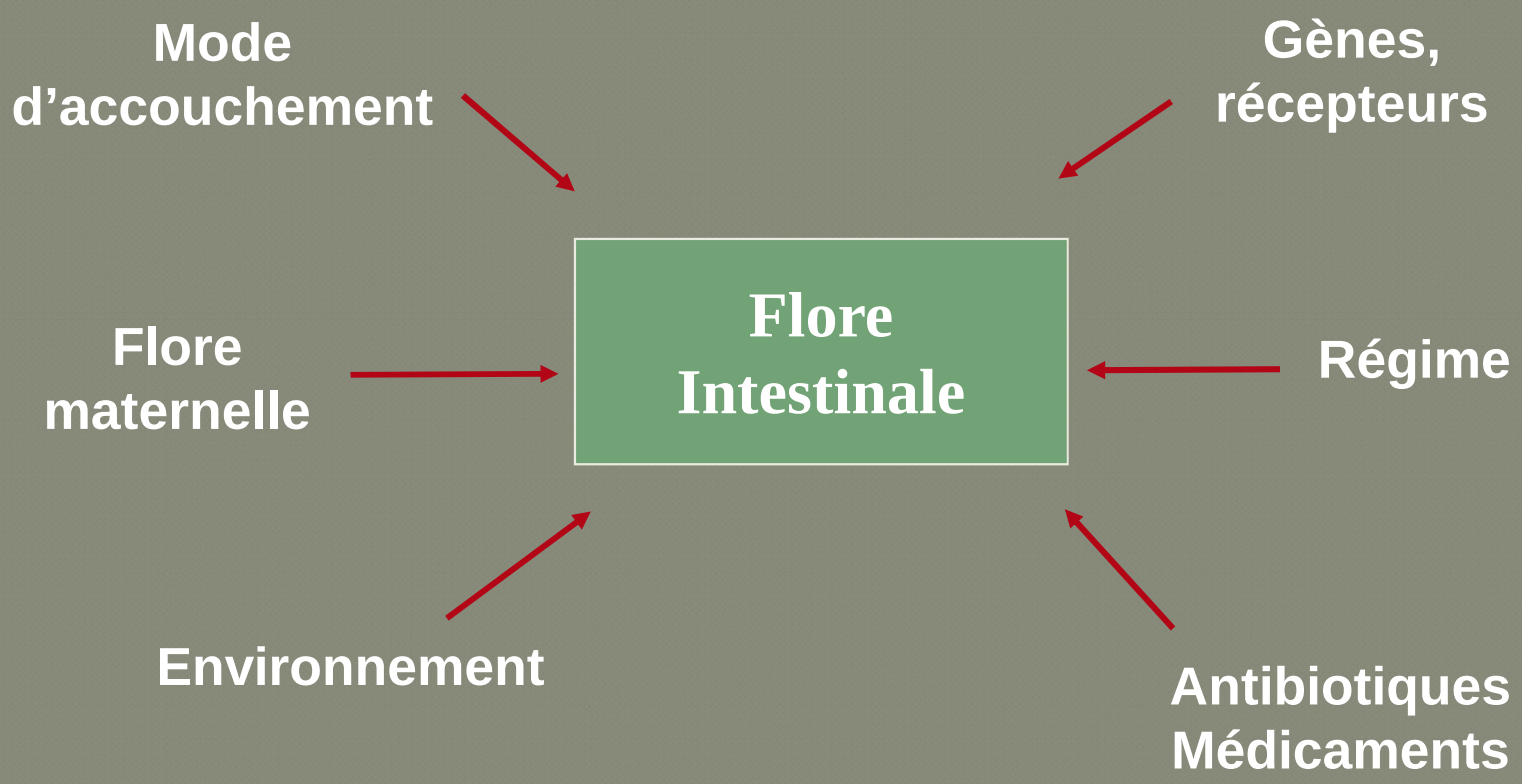
Facteurs	Influence	Evidence
Génétiques	Prédisposition	En ⇑
Exposition allergènes	Sensibilisation, pas maladie	+/-
Pollution	Surtout particules fines	En ⇑
Tabagisme passif	Augmente risque d'asthme	+++
Régime alimentaire	Plausabilité biologique	+/-
Obésité, ⇓ activité	Surtout chez les femmes	En ⇑
⇓ infections, ⇑ antibio	Rôle protecteur des infections	En ⇑
Vie à la ferme	Protection par endotoxines	En ⇑

Rôle De La Flore Intestinale

- 10^{14} microbes, > 1000 espèces différentes
- ++ Lactobacilles et bifidobactéries
- Protection contre les infections
- Rôle majeur dans l'immunité adaptative
  - Toll-like recepteurs, CD14
  - Th1 : IFN γ, IL 12, NF-κB
  - Th2 : IL 4, IL 5, IL 6
  - Treg : TGFβ, IL 10

J-P Langhendries
Arch Pediatr 2004; 11: 542-4

Facteurs Impliqués Dans La Colonisation Intestinale



Champs et modes d'application

- Conditions périnatales particulières
- Conditions nutritionnelles et infectieuses durant les 2 premières années de vie
- Probiotiques, prébiotiques, fibres, Zn, glutamine,…

Pour rendre les pratiques « sérieuses » Il faudra:
-Ajuster les combinaisons
-Ajuster le dosage pour chaque combinant
-Instaurer et rendre la prescription médicale