

**MasterClass**  
**Module 0 « Immunologie de l'allergie »**  
**15 décembre 2022**

**Le système immunitaire en action**  
**Induction et régulation**  
**de l'inflammation cutanée**

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# Team “Epidermal Immunity & Allergy”

## Research activities



### Main Features

- High prevalence
  - 10% of children (AD)
  - 1<sup>st</sup> occupational disease (ACD)
- Benign to very severe
- Localized acute or chronic lesions
- **Delayed-type allergy / Specific T cells**
- Breakdown of tolerance

### Allergens

- Chemicals/haptens & proteins
- Endowed with antigenic & adjuvant properties
- Skin or systemic route

### Objectives

- Decipher the pathophysiology
- Develop new diagnostic/predictive assays
- Develop new therapeutic strategies to restaure skin tolerance

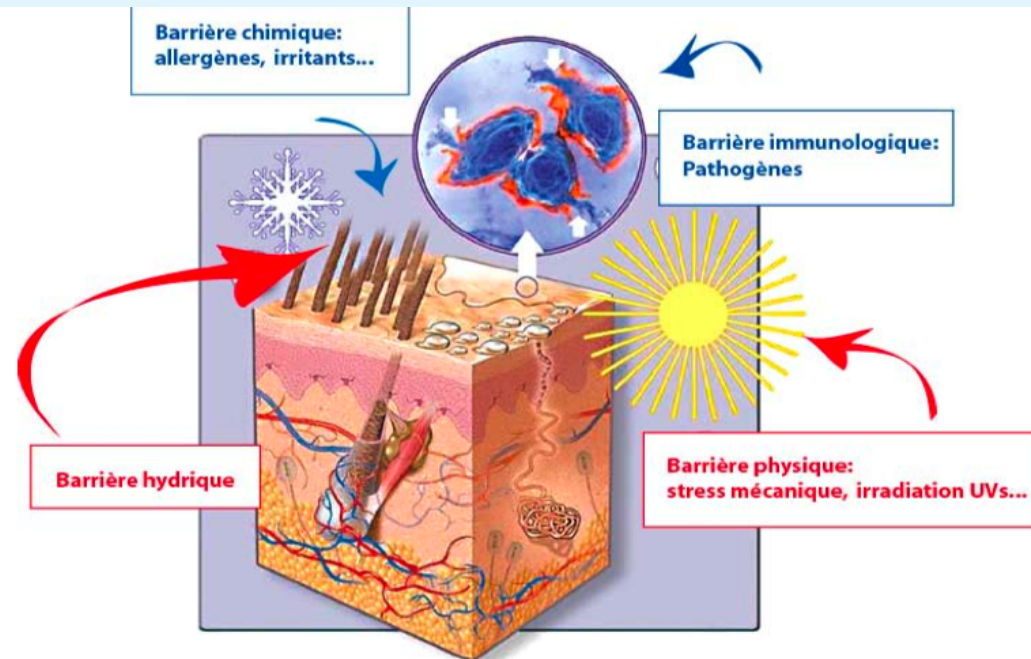
# PLAN

- Bases immunologiques de la réponse à l'interface cutanée
- Induction & régulation de l'inflammation cutanée : *exemple de l'eczéma de contact*

# PLAN

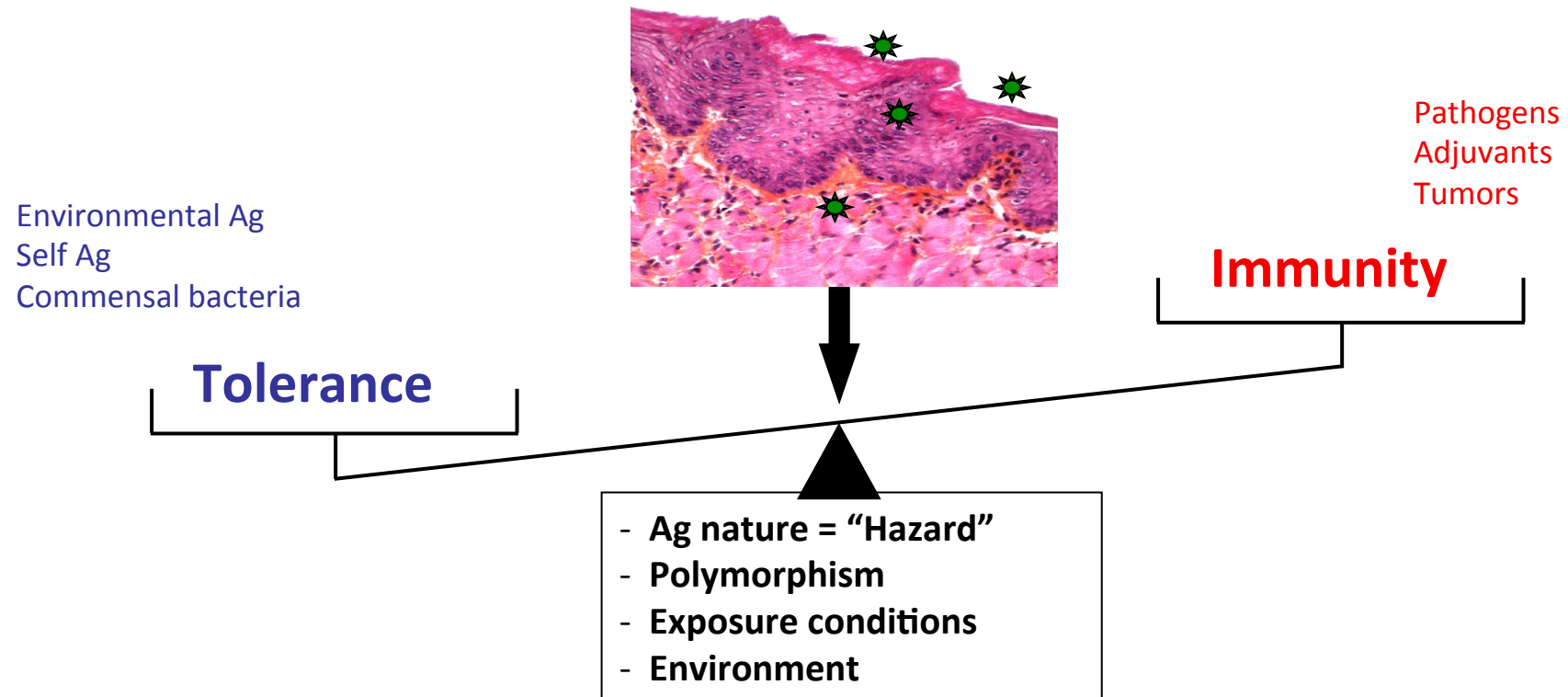
- Bases immunologiques de la réponse à l'interface cutanée
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# The skin: the multitasking organ



- Skin area=1.8 m<sup>2</sup>
- Being constantly exposed to potential hazards -> maintain homeostasis
- Examples of the non-immune functions of the skin:
  - Physical and biochemical barrier
  - Sensory-receptive area
  - Ensures hydratation
  - Allows synthesis of vitamins, hormones

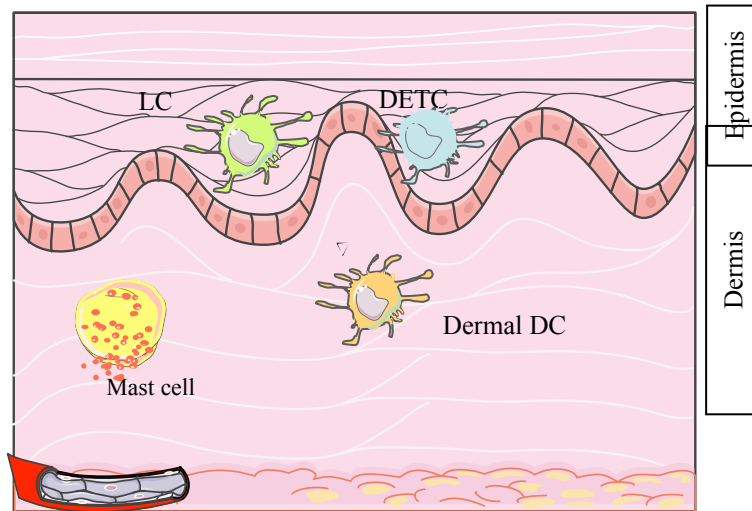
# The skin: an immuno-protective organ



- Serves as an immuno-protective organ that actively defends deeper body tissues against infectious agents. Privileged site for vaccination
- Maintains self-tolerance, preventing allergens and inhibiting autoimmunity<sup>6</sup>

# Induction of systemic immunity upon skin exposure/immunization

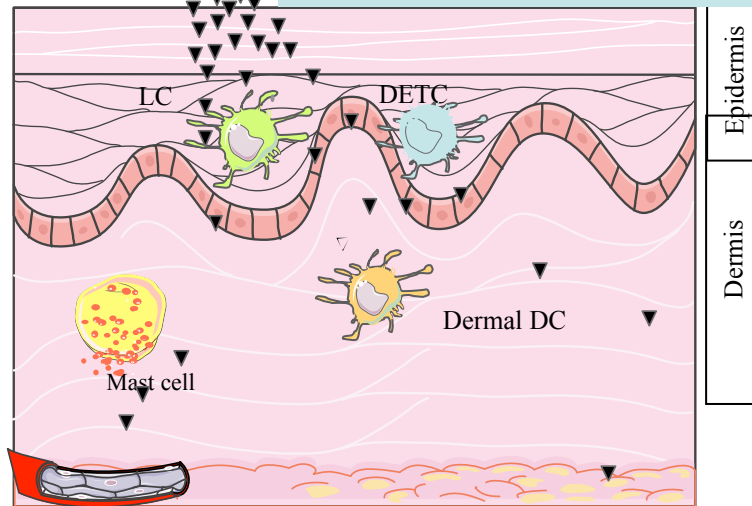
Skin exposure, immunization



# Induction of systemic immunity upon skin exposure/immunization

Skin exposure, immunization

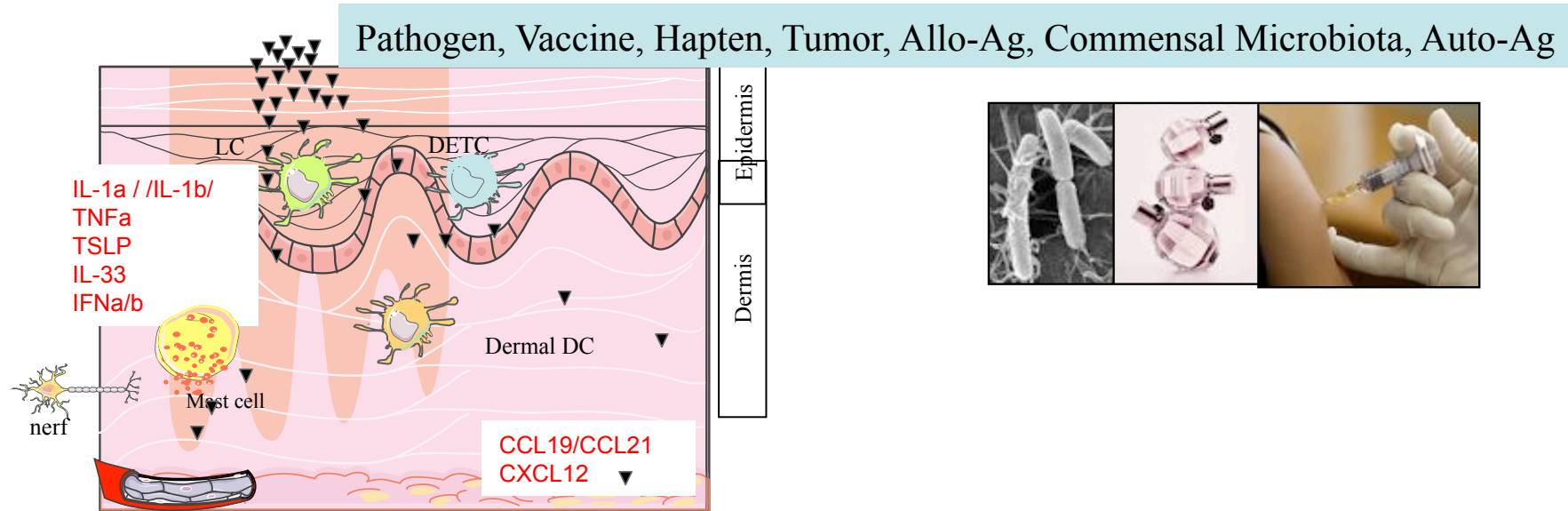
Pathogen, Vaccine, Hapten, Tumor, Allo-Ag, Commensal Microbiota, Auto-Ag





# Induction of systemic immunity upon skin exposure/immunization

Skin exposure, immunization



Innate immunity -> 1st line of defence  
Release of inflammatory mediators

# Inflammation

## General scheme

4 major inflammatory components

### Inducers

**Microbes,  
Allergens  
AlloAg**

### Tissue damage

Cell-derived  
Plasma-derived  
ECM-derived

### Sensors

#### P(athogen)AMPs

TLR, NLR...

#### D(amage)AMPs

TLR, NLR, RAGE...

#### Nociceptors

### Mediators

#### Cellular

Neutrophils, Eosinophils,  
Monocytes/Macrophages,  
T & B cells...

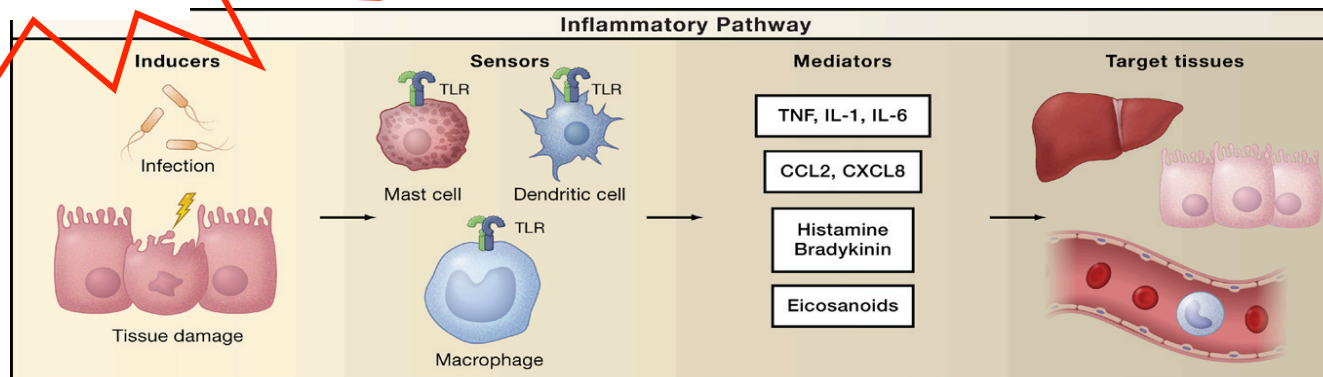
#### Molecular

Cytokines and chemokines,  
Vasoactive amines or peptides  
Complement fragments  
lipide mediators  
proteolytic enzymes

### Target tissues

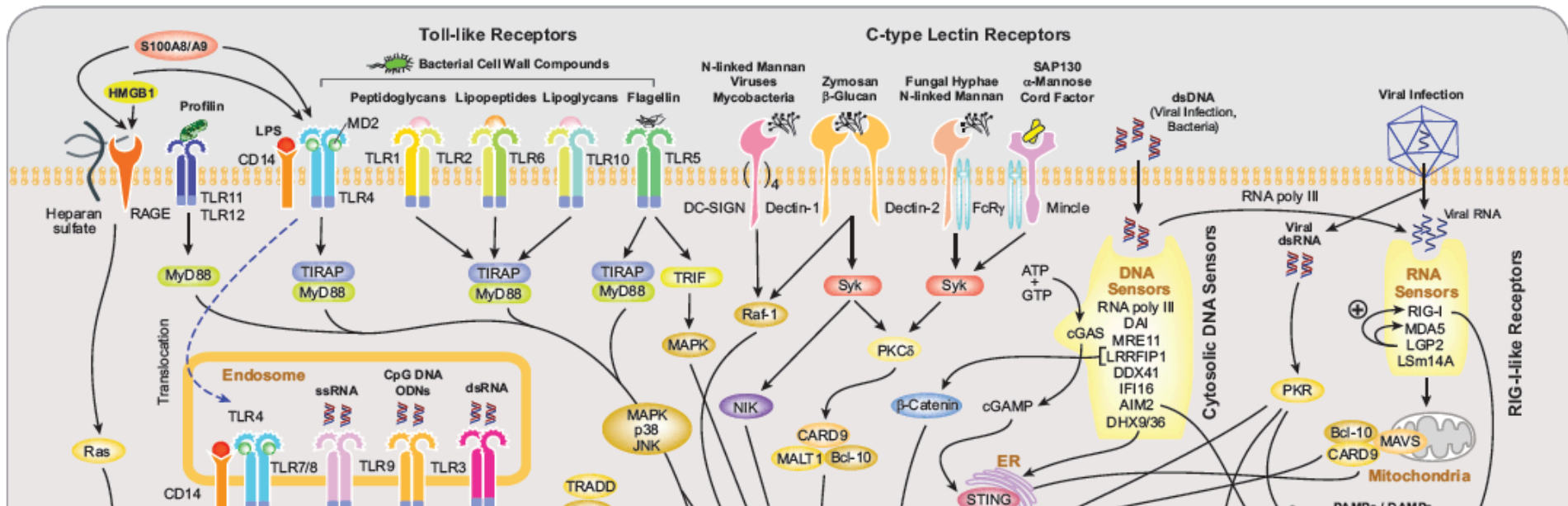
**Redness/Oedema  
Heat/Pain  
Loss of function**

**DANGER  
Hypothesis**



# Pathogen recognition receptors (PRRs)

- Microbial Pattern Recognition Receptors: TLR, RLR, NLR, CLR signaling (examples)



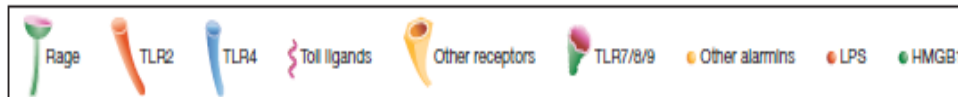
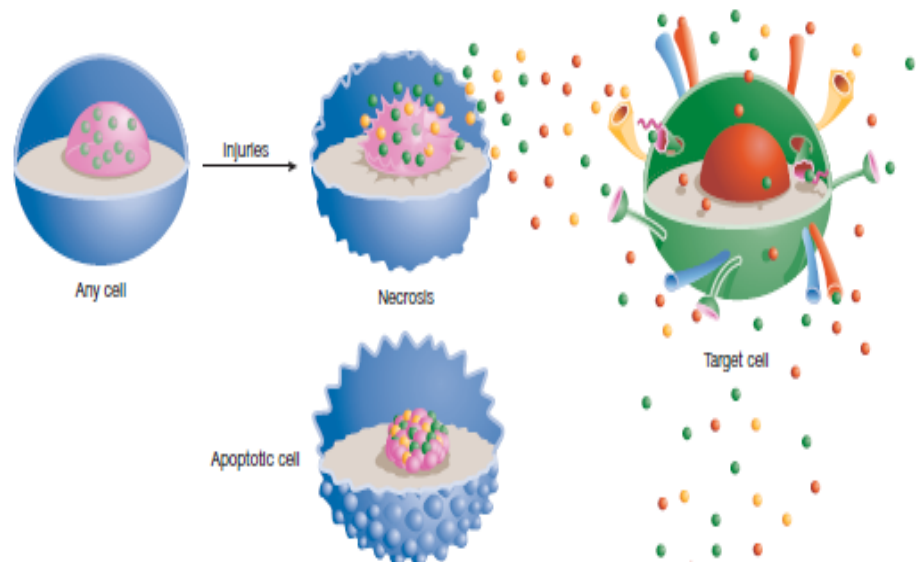
- Recognition of specific structures (polysach, nucleic acids, nucleotides lipoproteins, glycolipids)
- Cell compartment localisation, tissue-specific expression
- Cell intrinsic → infected cells, cell extrinsic → not infected cells; but most of PAMPs are detected by both
- Recognition of functional features (enzymatic activities, pore-forming toxins)

NOD-like Receptors

# Inflammation

## PAMPs – DAMPs and their sensors

### Intracellular DAMPs



DAMP	Adjuvant activity
HMGB1	<i>In vivo</i> : adjuvant activity of purified molecule; adjuvant activity shown by selective depletion  <i>In vitro</i> : DC activation
Uric acid (MSU)	<i>In vivo</i> : adjuvant activity shown by injection of purified molecule and selective depletion  <i>In vitro</i> : DC activation
Chromatin, nucleosomes and DNA	<i>In vivo</i> : DC maturation induced by purified molecule <i>In vitro</i> : DC activation induced by chromatin-IgG complexes
HSPs	<i>In vivo</i> : tumour immunogenicity enhanced by overexpressed molecule or addition of purified molecule (HSP70); DC migration to lymph nodes induced by purified molecule (gp96) <i>In vitro</i> : DC maturation (gp96 and HSP70)
Adenosine and ATP	<i>In vivo</i> : exacerbation or abrogation of bronchial asthma by purified molecule or specific inhibition, respectively <i>In vitro</i> : DC maturation
Galectins	<i>In vivo</i> : ND  <i>In vitro</i> : DC maturation
Thioredoxin	ND
S100 proteins	ND
Cathelicidins	<i>In vitro</i> : DC maturation; DC activation induced by LL37-self-DNA complex
Defensins	<i>In vivo</i> : adjuvant activity by co-administration of purified molecule <i>In vitro</i> : DC maturation
N-formylated peptides	<i>In vivo</i> : ND  <i>In vitro</i> : DC chemotaxis

# Inflammation

## PAMPs – DAMPs and their sensors

### Extracellular DAMPs

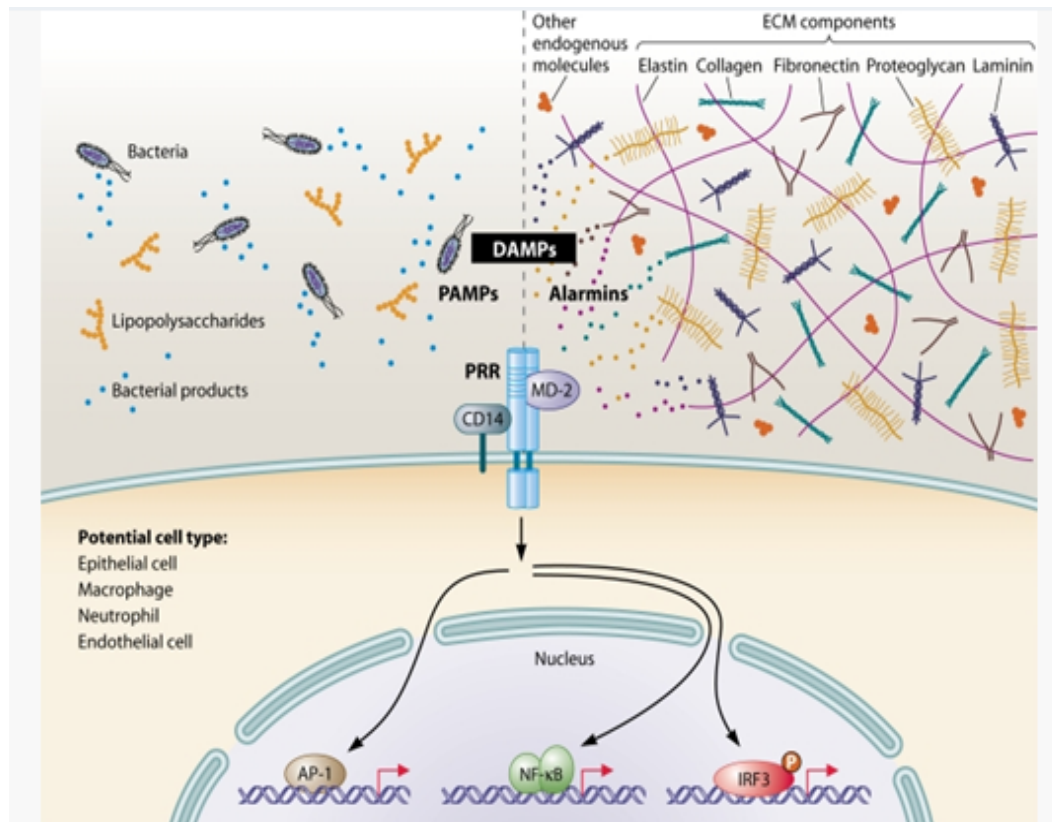
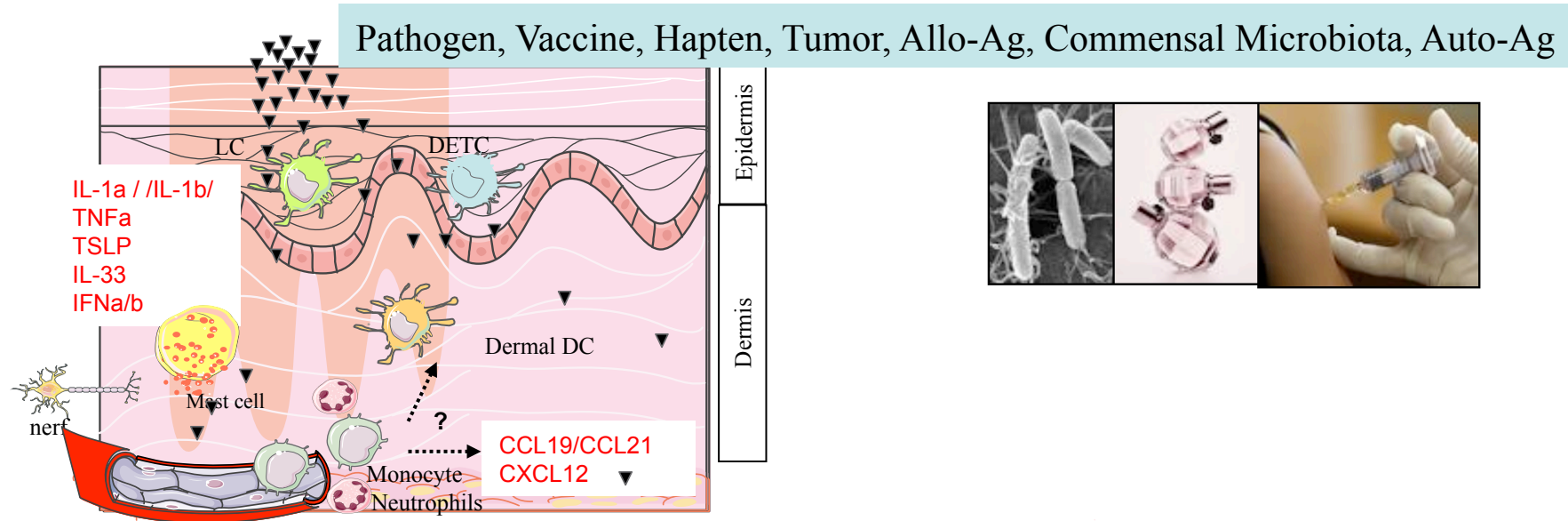


Table 2 | Adjuvant and pro-inflammatory activity of extracellular DAMPs

DAMP	Adjuvant activity
Hyaluronic acid	<i>In vivo</i> : inhibition of Langerhans-cell maturation by blocking peptide; adjuvant activity by administration of purified molecule  <i>In vitro</i> : DC maturation
Heparan sulphate	<i>In vitro</i> : DC maturation
Fibrinogen	<i>In vitro</i> : DC maturation
Collagen-derived peptides	<i>In vivo</i> : ND  <i>In vitro</i> : DC maturation
Fibronectin	<i>In vitro</i> : DC maturation
Elastin-derived peptides	<i>In vivo</i> : ND  <i>In vitro</i> : ND
Laminin	<i>In vivo</i> : ND  <i>In vitro</i> : ND

# Induction of systemic immunity upon skin exposure/immunization

Skin exposure, immunization



Innate immunity -> 1st line of defence

Release of inflammatory mediators

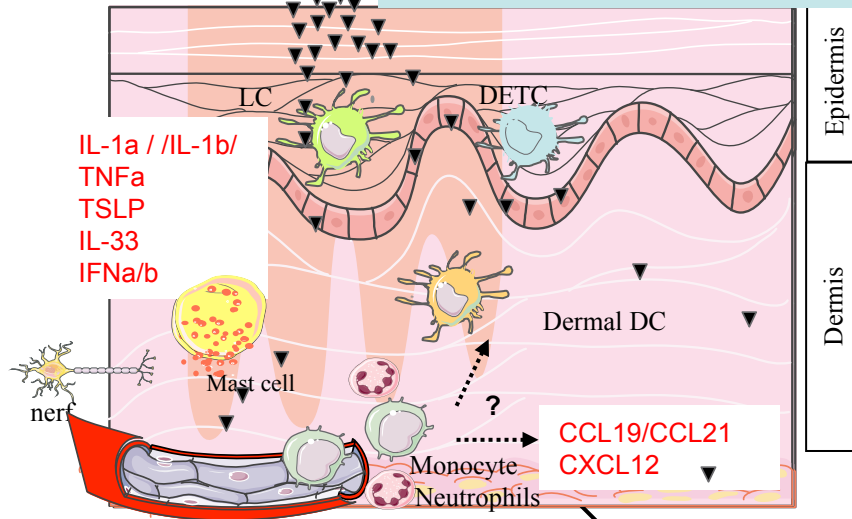
Coordinated cross-talk between epithelial and immune cells

Infiltration of blood leucocytes

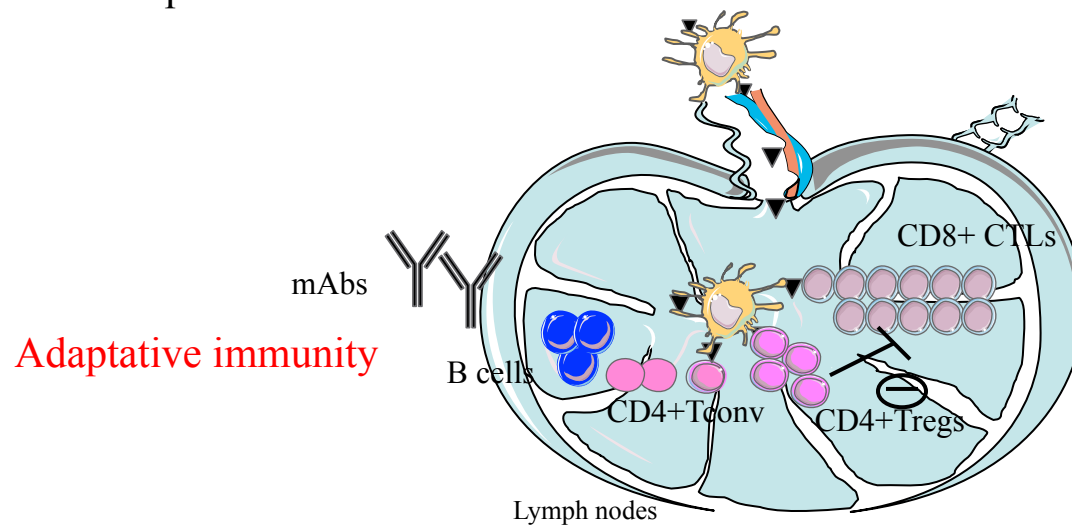
# Induction of systemic immunity upon skin exposure/immunization

Skin exposure, immunization

Pathogen, Vaccine, Hapten, Tumor, Allo-Ag, Commensal Microbiota, Auto-Ag



Innate response



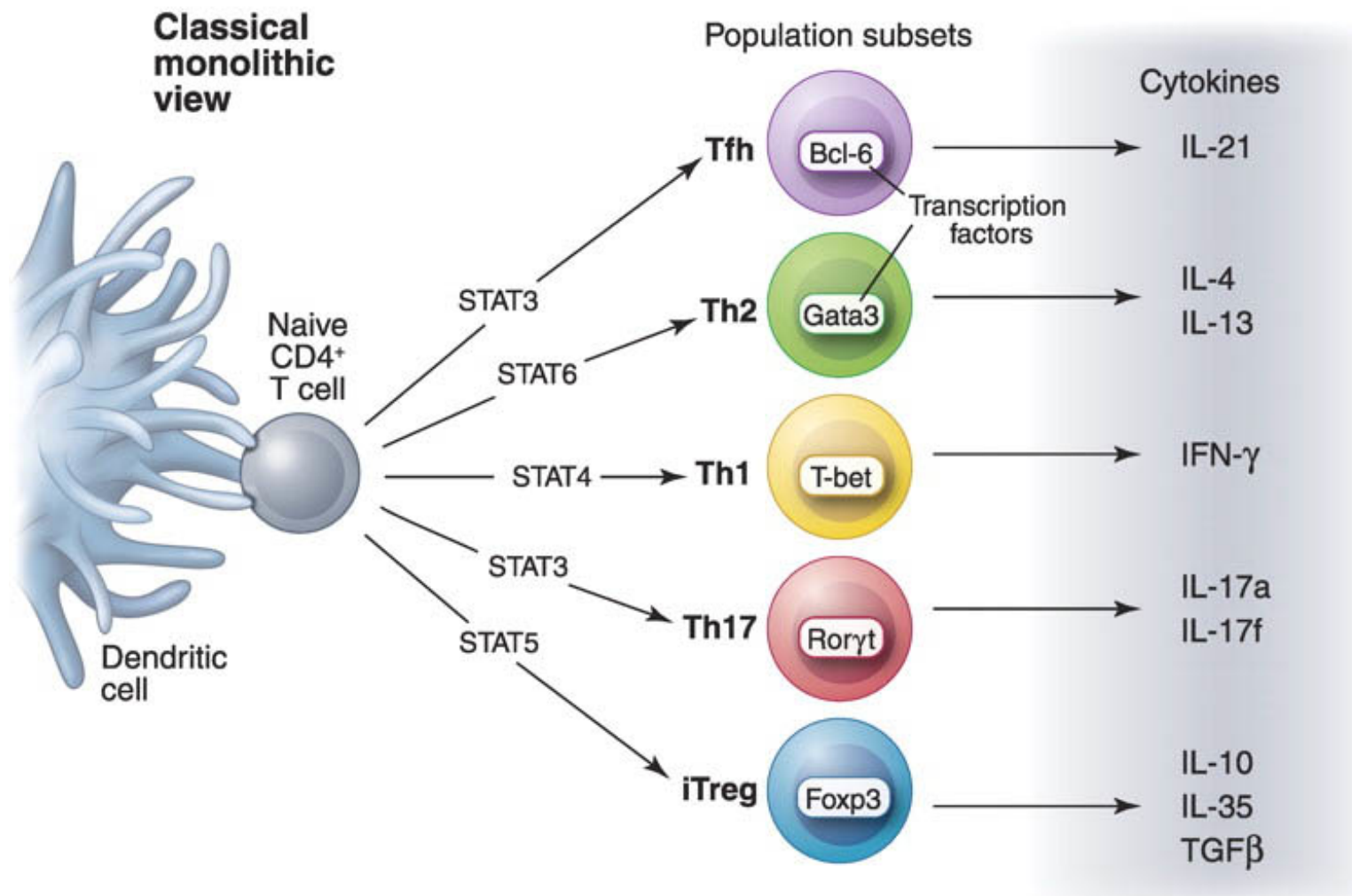
Adaptative immunity

Antibody production

Effector CD4+ & CD8+ T cells

Memory T cells, B cells & plasma cells

# Distinct T cells

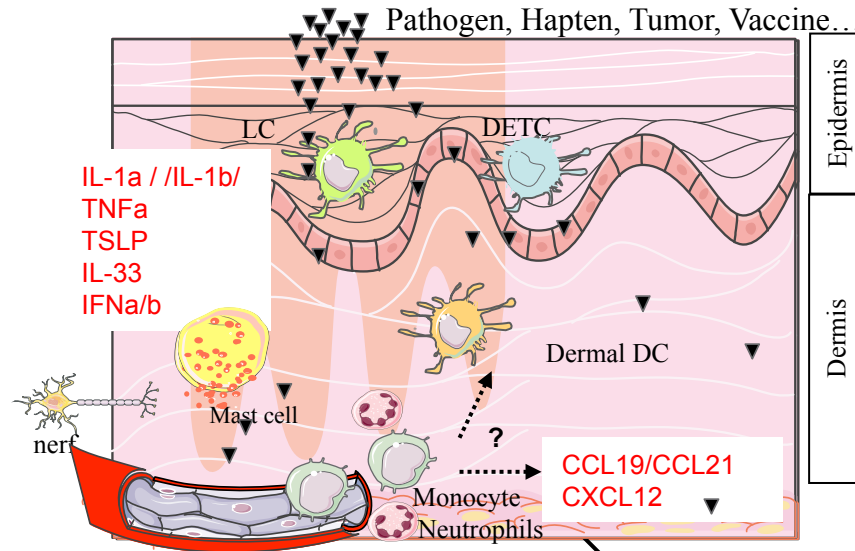




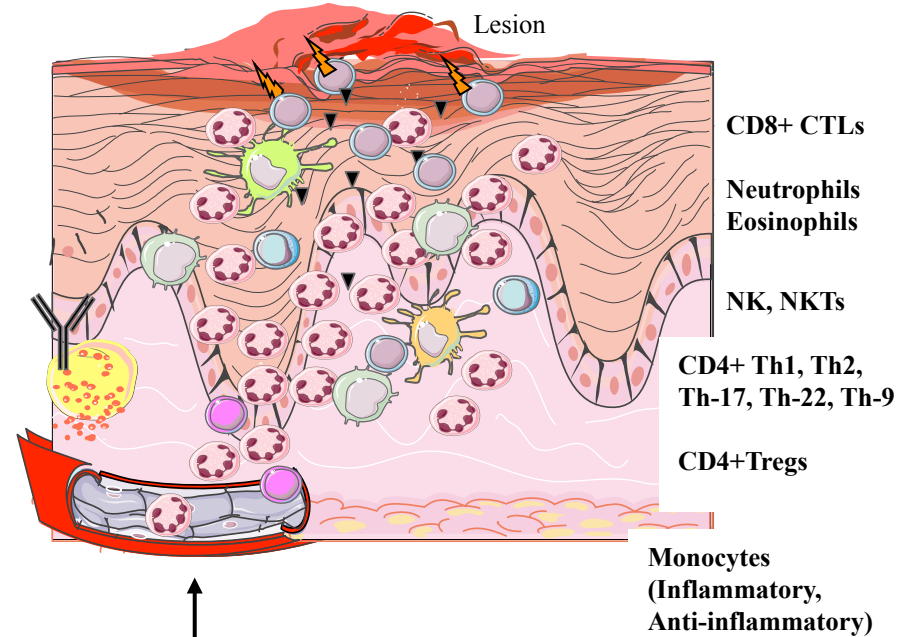
# Induction of systemic immunity upon skin exposure/immunization

Skin exposure, immunization

Persistence / Re-exposure → delayed-response (days)  
 Skin inflammation, elimination of infected cells  
 Tissue response/repair

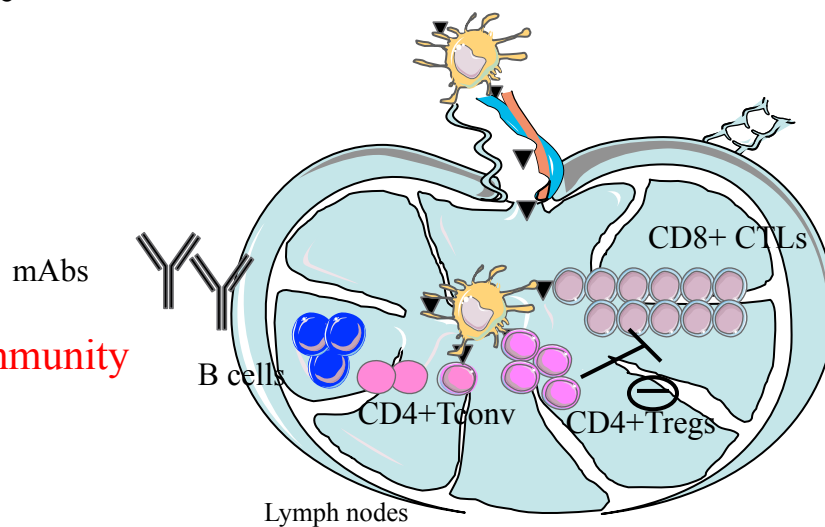


Innate response



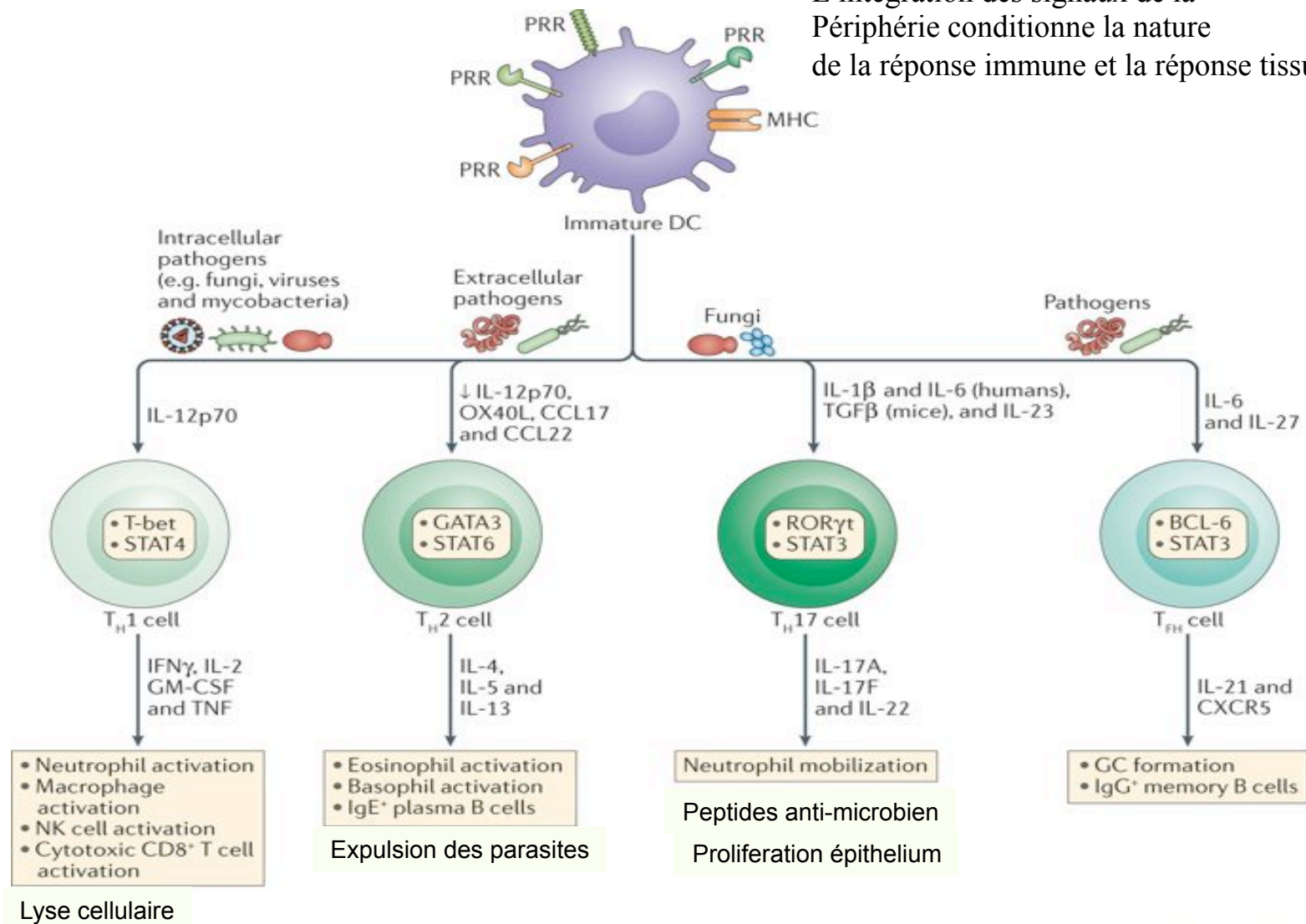
**Effector & memory response → 2<sup>nd</sup> line of defence**

**Adaptative immunity**



# Different mode of recognition by the innate immunity → different layers of sensing by the immune system → different effector response

L'intégration des signaux de la Périphérie conditionne la nature de la réponse immune et la réponse tissulaire



# PLAN

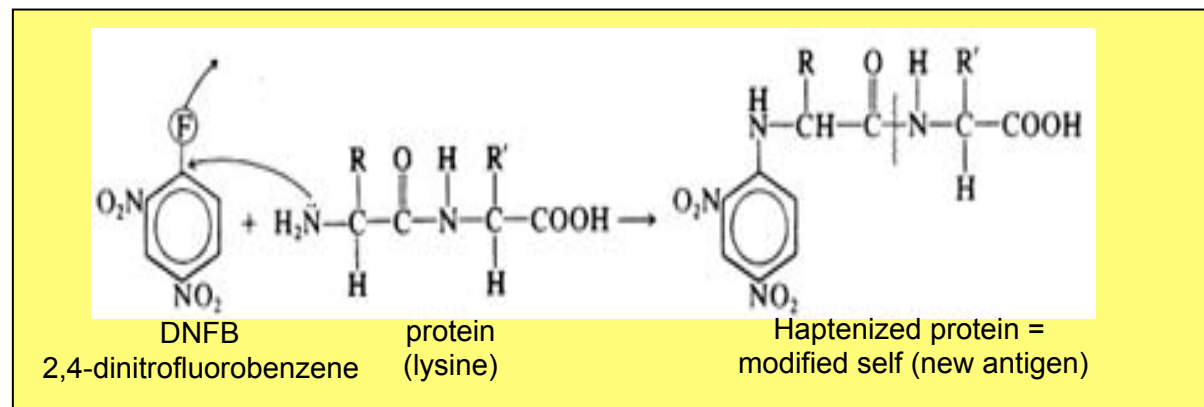
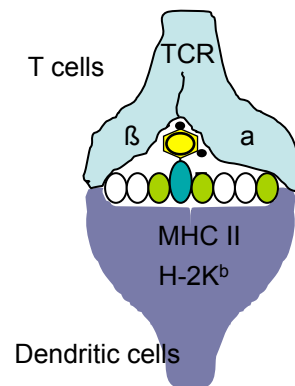
- Bases immunologiques de la réponse à l'interface cutanée
- Induction & régulation de l'inflammation cutanée : exemple de l'eczéma de contact

# Allergic Contact Dermatitis (ACD): Generalities



## Features

- High prevalence, 1st occupational disease
- Repeated exposure to environmental allergens (cosmetics, jewels, drugs...)
- Breakdown of skin tolerance
- Delayed-type allergy:  
→ infiltration and activation of allergen-specific T cells

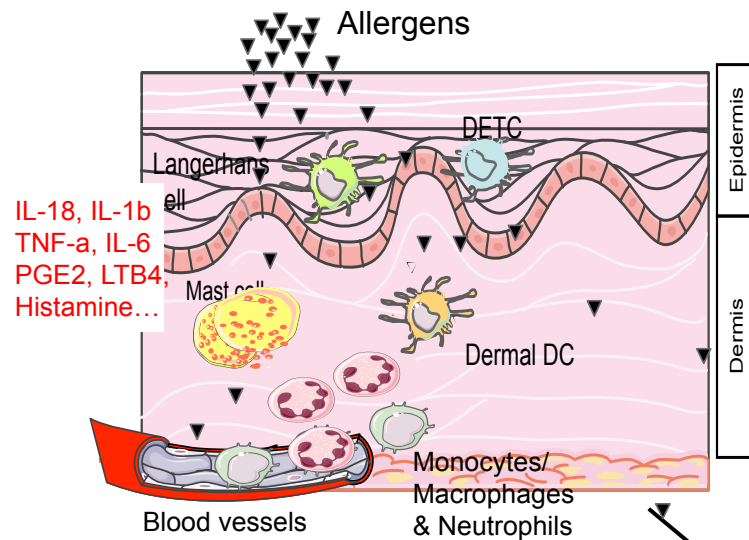


Presentation of haptene peptides

# Pathophysiology of skin allergy

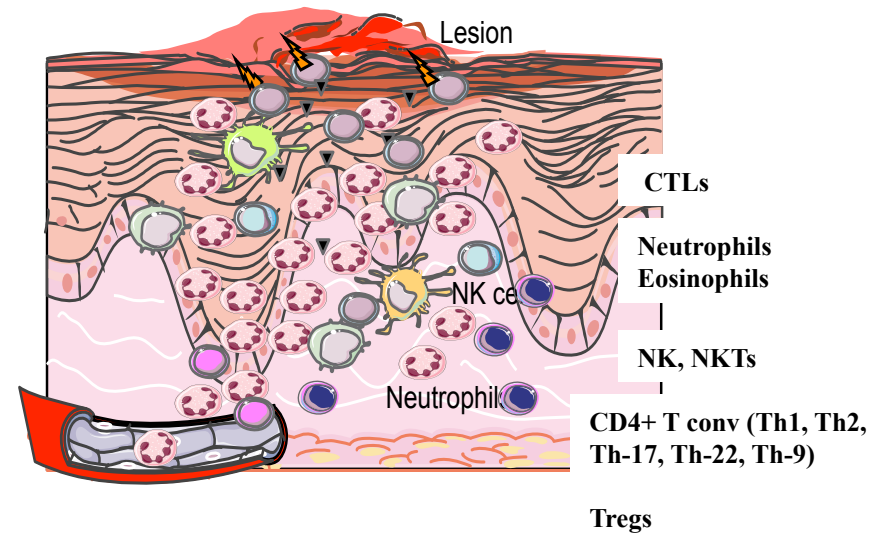
## 1- Sensitization phase

Innate immunity/ T cell priming

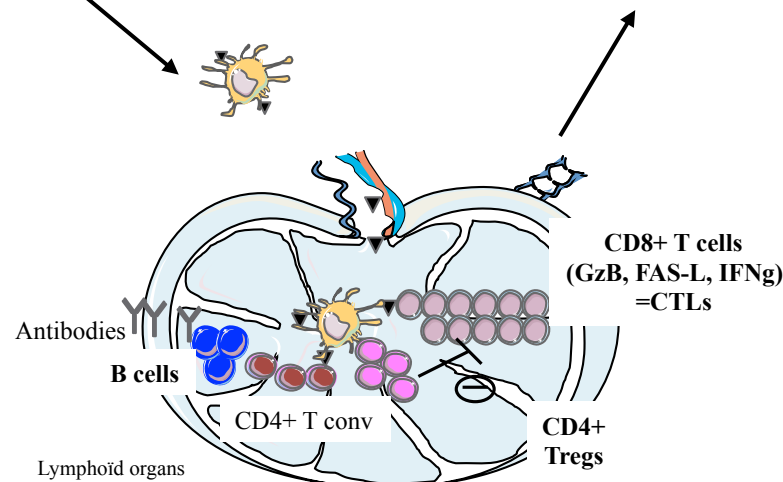


## 2- Elicitation phase

Effector response/ Polymorphic recruitment



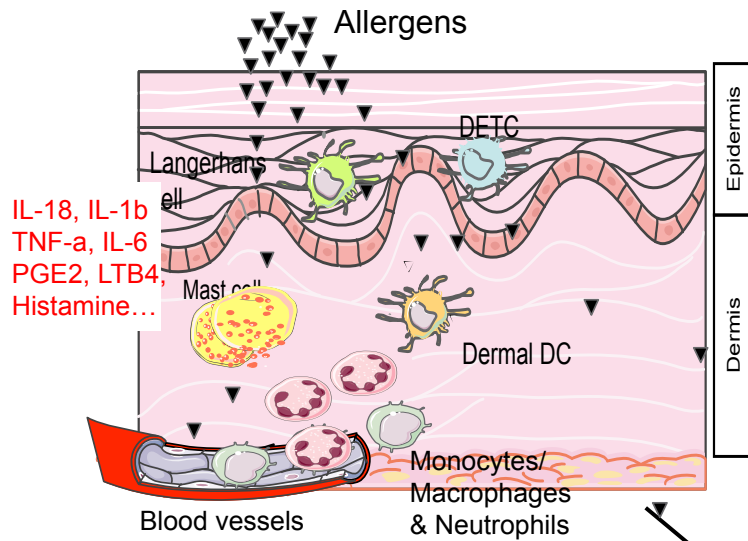
Monocytes  
(Inflammatory,  
Anti-inflammatory)



BOUR et al. *Eur J Immunol*, 1995  
 KRASTEVA et al. *J Immunol*, 1998  
 KEHREN et al. *J Exp Med*, 1999  
 AKIBA et al. *J Immunol*, 2002  
 SAINT-MEZARD et al. *J Immunol*, 2003  
 AKIBA et al. *J Invest Dermatol*, 2004  
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 GOUBIER et al. *J Invest Dermatol*, 2013  
 CORTIAL et al. *Nanomedicine*, 2015  
 GAMRADT *J Allergy Clin Immunol* 2019

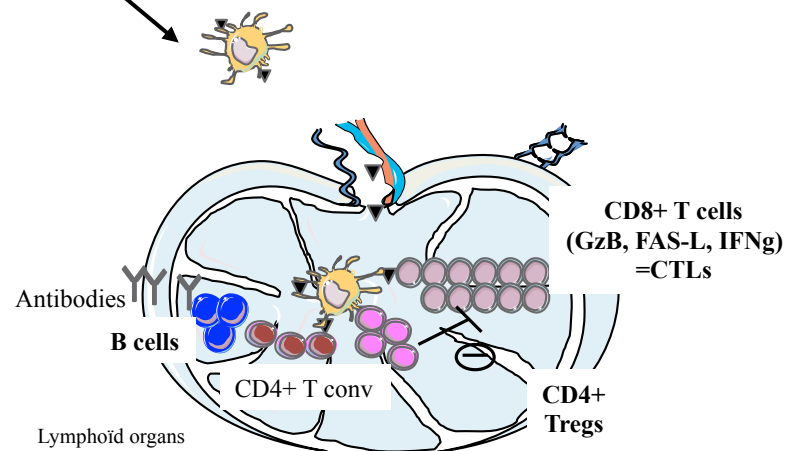
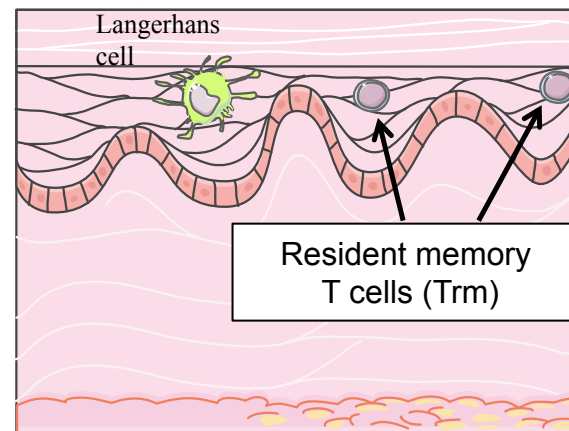
## 1- Sensitization phase

Innate immunity/ T cell priming



## 3- Resolution of skin inflammation

Healed lesion/ Persistence of skin T<sub>RM</sub>

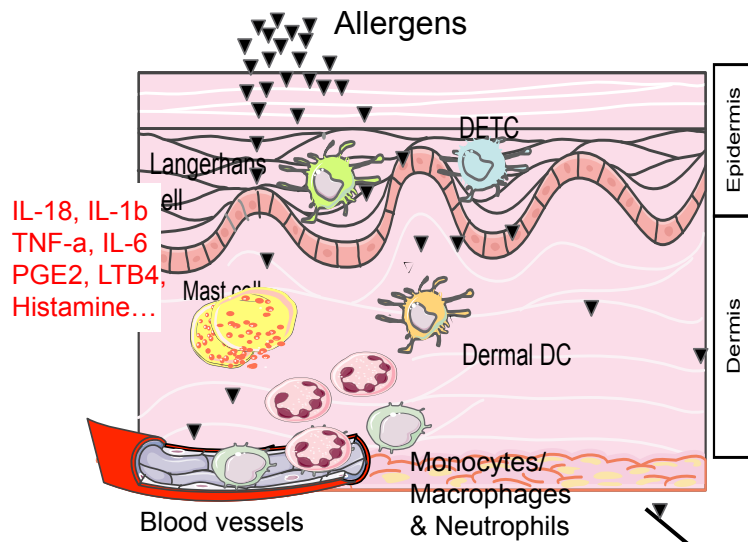


BOUR et al. *Eur J Immunol*, 1995  
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 GAMRADT *J Allergy Clin Immunol* 2019

# Pathophysiology of skin allergy

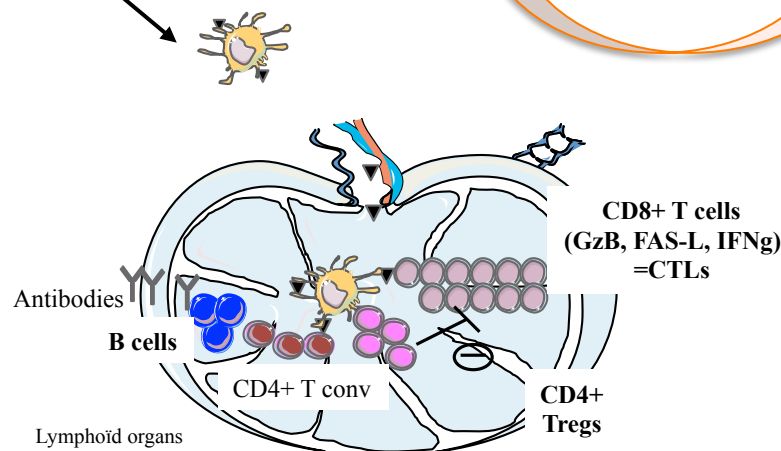
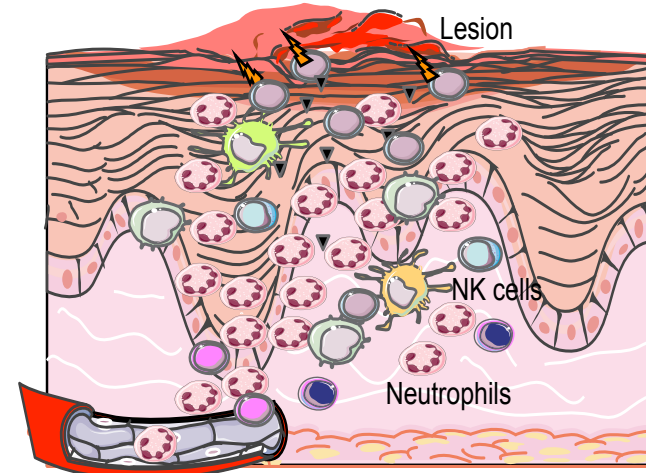
## 1- Sensitization phase

Innate immunity/ T cell priming



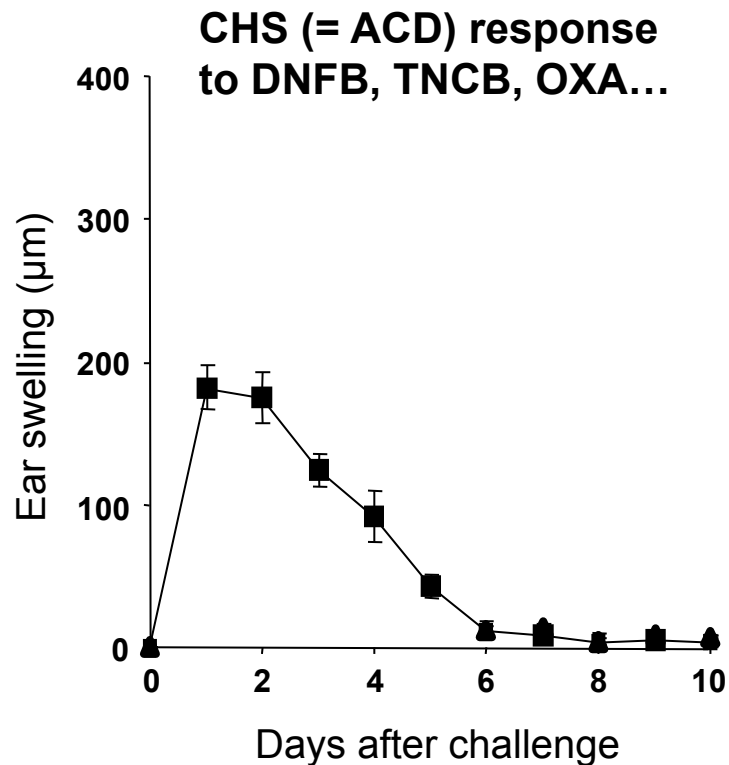
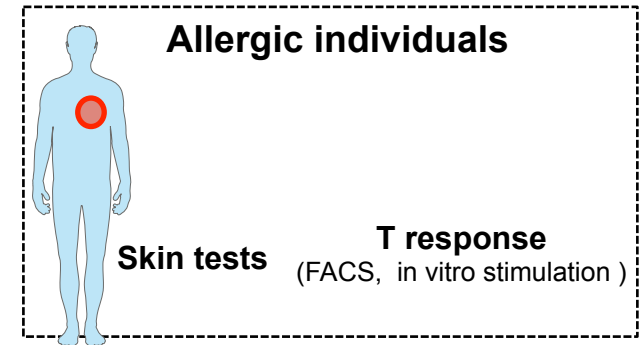
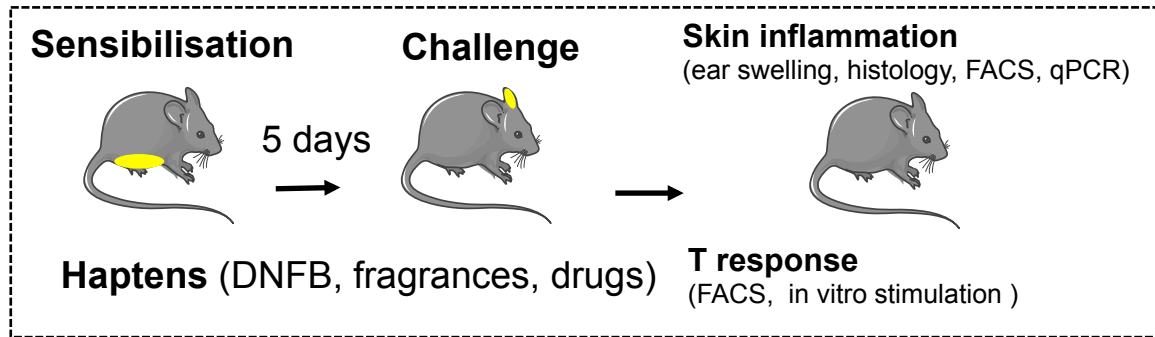
## 4- Recurrence / Severity / Chronicity

New exposure / Flares



BOUR et al. *Eur J Immunol*, 1995  
 KRASTEVA et al. *J Immunol*, 1998  
 KEHREN et al. *J Exp Med*, 1999  
 AKIBA et al. *J Immunol*, 2002  
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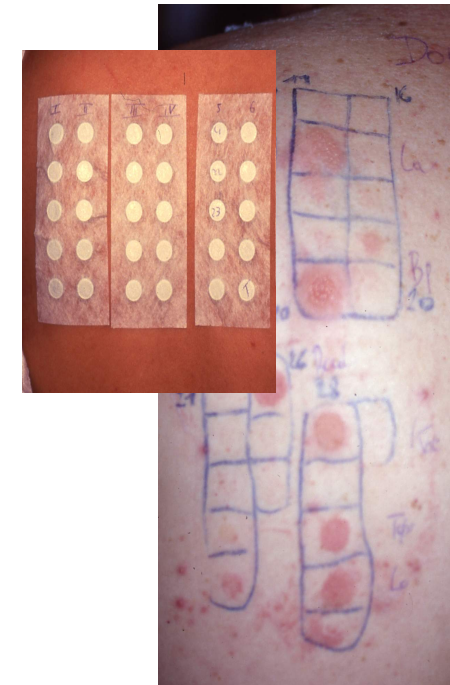
# Experimental models of ACD in mouse, in human



**ACD lesions**



**Positive patch-tests to reference allergens**





# Permeation of haptens into the skin

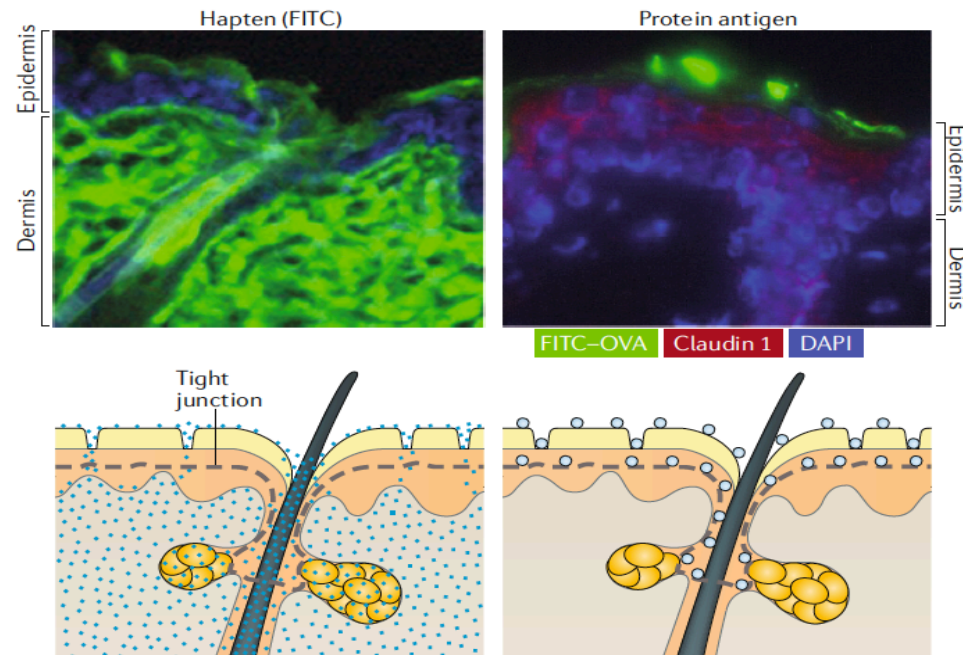
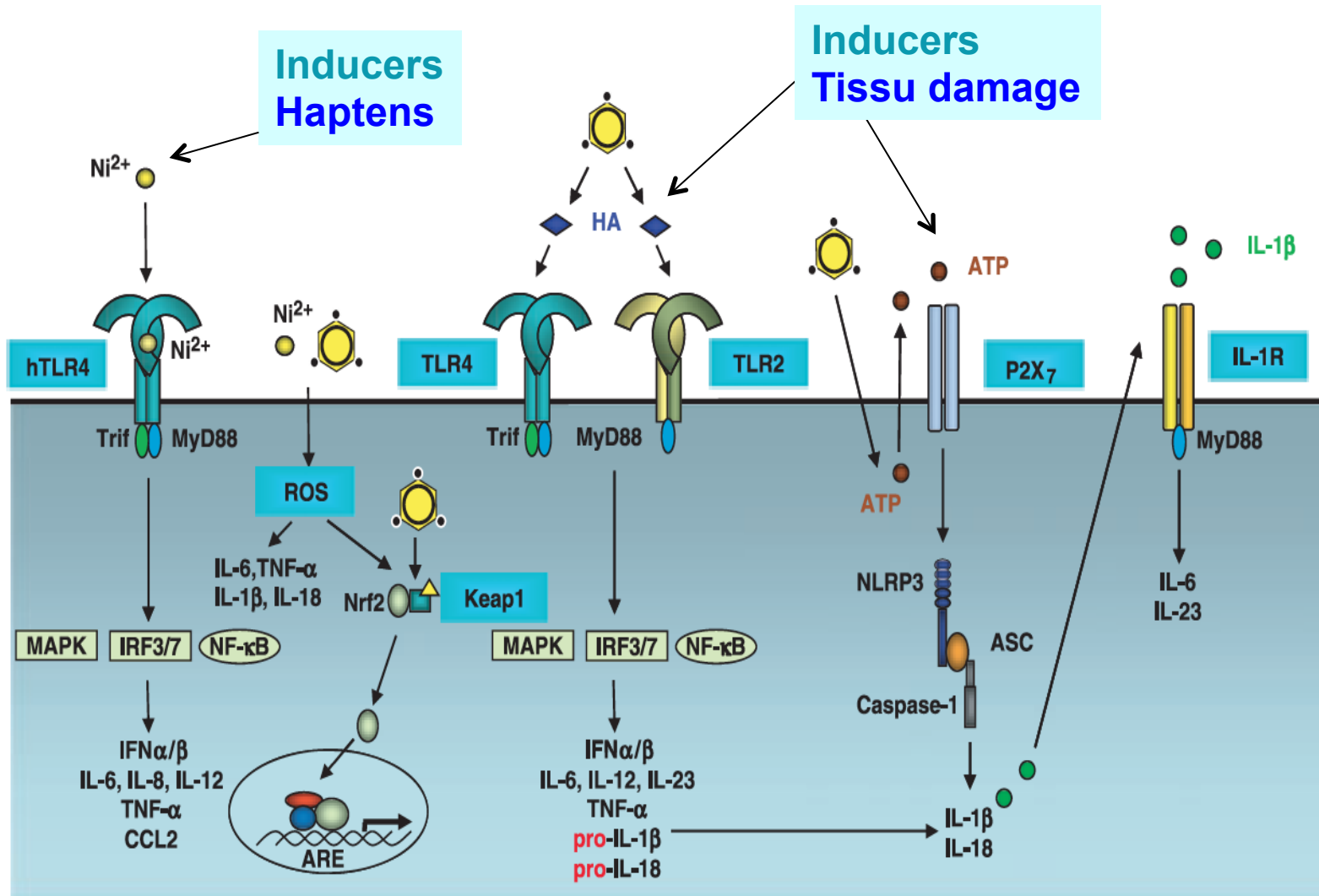


Fig. 4 | **Penetration of haptens and proteins into the skin.** A hapten (fluorescein isothiocyanate (FITC); molecular mass = 389; left) or FITC-conjugated ovalbumin (FITC-

- Les haptènes sont pour la plus part des substances hydrophobes
- Pénétration dépend de l'hydrophobicité (LogP), mais aussi de la présence de groupes chargés, la taille (poids moléculaire < 1000 Daltons), la forme moléculaire et du véhicule.
- Les peaux altérées (blessures physiques, chimiques ou anomalie génétique) favorisent l'apparition d'un eczéma de contact

# How haptens activate innate immunity?



# Les diverses étapes de la sensibilisation : activation de l'immunité innée

- Rôle crucial de la structure du TLR4 humain sur le développement de la réponse d'EAC

nature  
immunology

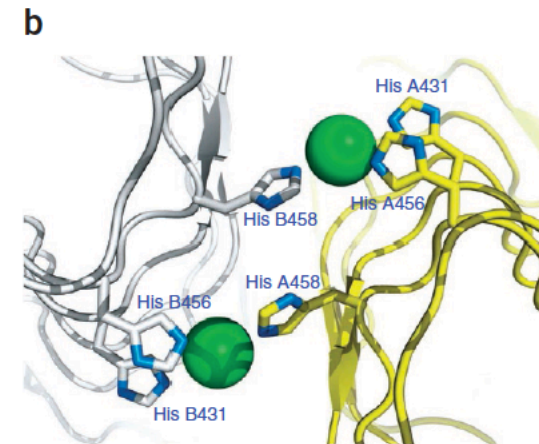
## Crucial role for human Toll-like receptor 4 in the development of contact allergy to nickel

Marc Schmidt<sup>1,2</sup>, Badrinarayanan Raghavan<sup>1,2</sup>, Verena Müller<sup>1,2</sup>, Thomas Vogl<sup>3</sup>, György Fejer<sup>4</sup>, Sandrine Tchapchet<sup>4</sup>, Simone Keck<sup>4</sup>, Christoph Kalis<sup>4</sup>, Peter J Nielsen<sup>4</sup>, Chris Galanos<sup>4</sup>, Johannes Roth<sup>3</sup>, Arne Skerra<sup>5</sup>, Stefan F Martin<sup>6</sup>, Marina A Freudenberg<sup>4</sup> & Matthias Goebeler<sup>1,2</sup>

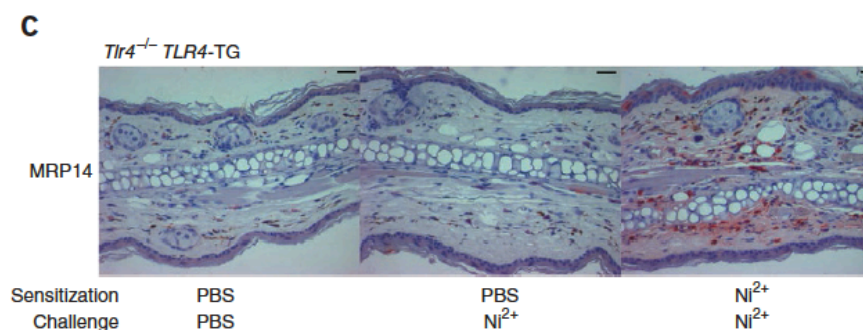
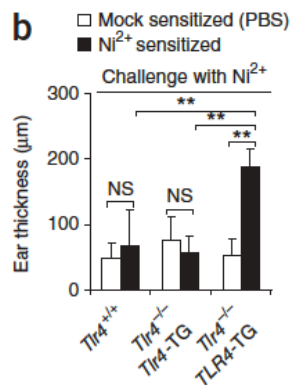
Conserved histidines on human TLR4 as potential binding sites for nickel

**a**

hTLR4	LRR14	DLPSLEFLDLSRNLGSLFKGCCSQSDF	396
mTLR4	LRR14	ALPFLSYLDLSRNALSFSGCCSYSDL	394
hTLR4	LRR15	GTTSLKYLDLSFNGVITMSSNFL	419
mTLR4	LRR15	GNLSLRHLDLSFNGAIIIMSANFM	417
hTLR4	LRR16	GLEQLE <sup>431</sup> LDFO <sup>431</sup> SNLRKQMFSEFVFL	444
mTLR4	LRR16	GLEELO <sup>431</sup> LDFO <sup>431</sup> STLKRVTFFSAPL	442
hTLR4	LRR17	SLRNLIVLDL <sup>455</sup> ST <sup>455</sup> IRVAENGIFN	468
mTLR4	LRR17	SLEKLLVLDL <sup>455</sup> ST <sup>455</sup> IRKIDFDGIFL	466
hTLR4	LRR18	GLSSLEVLKMGNSFQENFLPDIPT	493
mTLR4	LRR18	GLTSLNLTLMAGNSFKDNTLSNVFA	491
hTLR4	LRR19	ELRNLTFLDLSQCQLEQLSPTAFN	517
mTLR4	LRR19	NTNLTFLDLSKQLEQISNGVFD	515
hTLR4	LRR20	SLSSIQVLNMS <sup>539</sup> HNFFSLDTFPYK	541
mTLR4	LRR20	TLHRLQLLNMS <sup>539</sup> HNLLFLDSSHYN	539
hTLR4	LRR21	CLNSLQVLDYSLN <sup>563</sup> MTSKQELQ <sup>563</sup>	566
mTLR4	LRR21	QLYSLTLDCSFL <sup>563</sup> MTSKGI-LQ <sup>563</sup>	563
hTLR4	LRR22	FPSSLAFNLNTQNDFA	582
mTLR4	LRR22	FPKSLAFFNLNTNSVA	579
hTLR4	LRRCT	CTCE <sup>607</sup> HSFLQWIKDQRQLLVEVERM	607
mTLR4	LRRCT	CICE <sup>607</sup> HSFLQWVKEQKQFLVNVEM	604

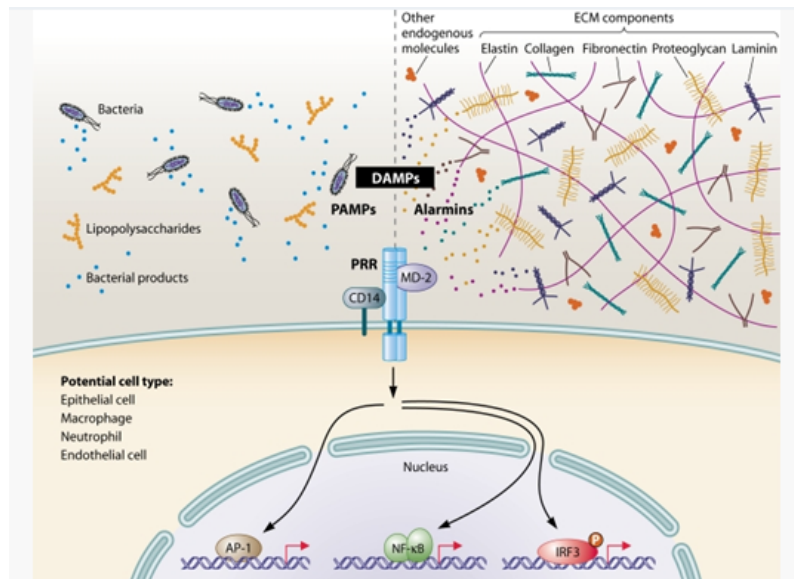


Transgenic expression of human TLR4 in mice confers reactivity toward nickel

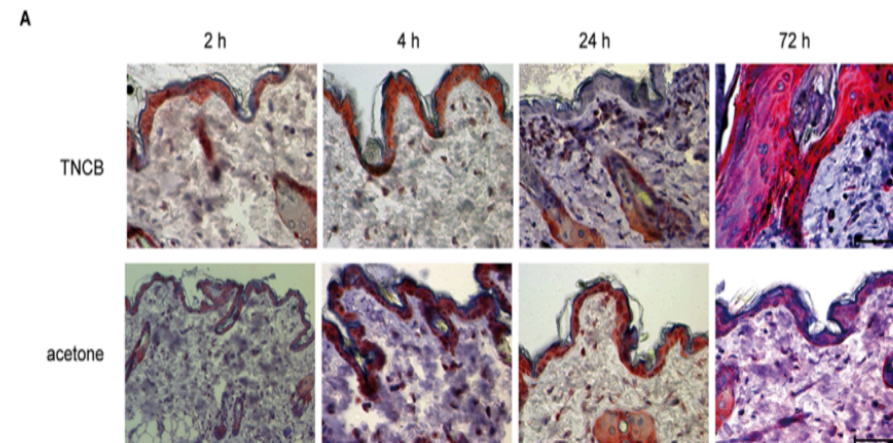


# Les diverses étapes de la sensibilisation : activation de l'immunité innée

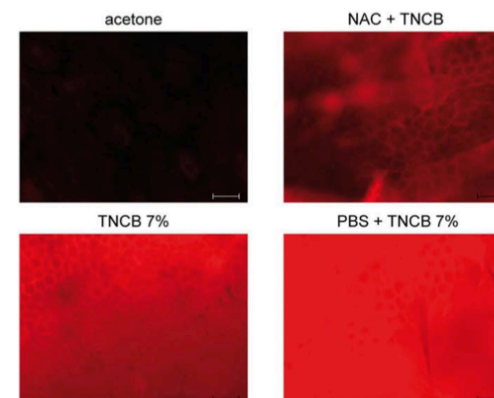
- Impact des médiateurs reconnus par les TLRs sur le développement de la réponse d'EAC



Dégradation Acide Hyaluronique ht PW, 24h après application

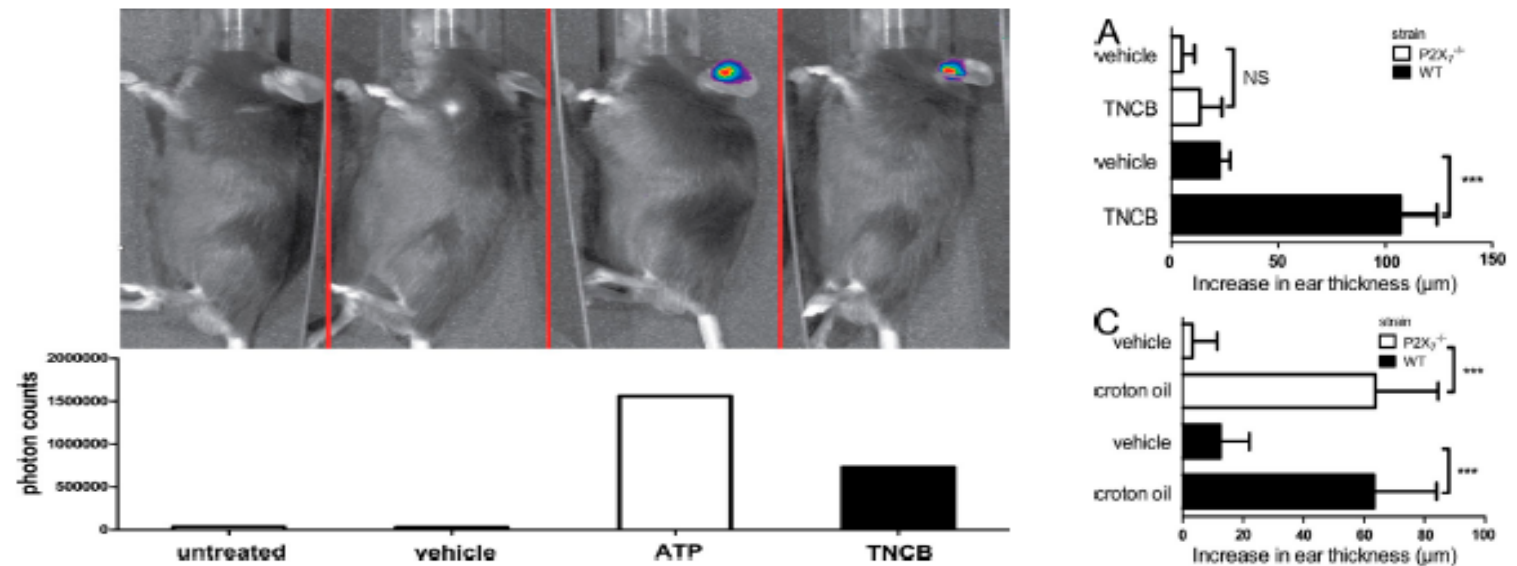


Production ROS, peau challengée



# Les diverses étapes de la sensibilisation : activation de l'immunité innée

- Impact des médiateurs reconnus par les NLRs sur le développement de la réponse d'EAC



Relargage ATP, peau challengée

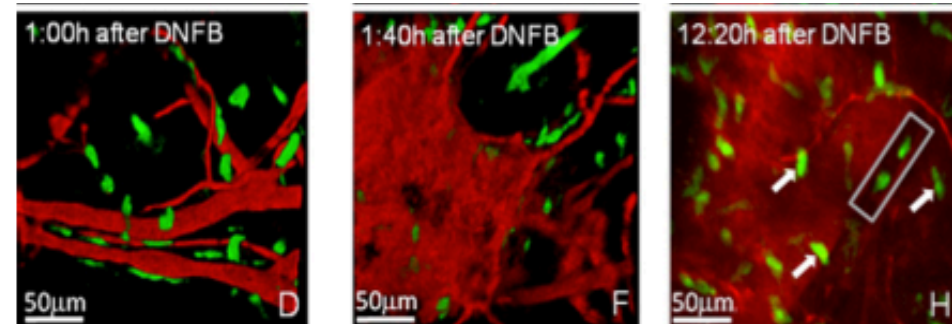
# Contribution of innate cells? Mast cells

Ear skin mast cells and blood vessels respond to hapten

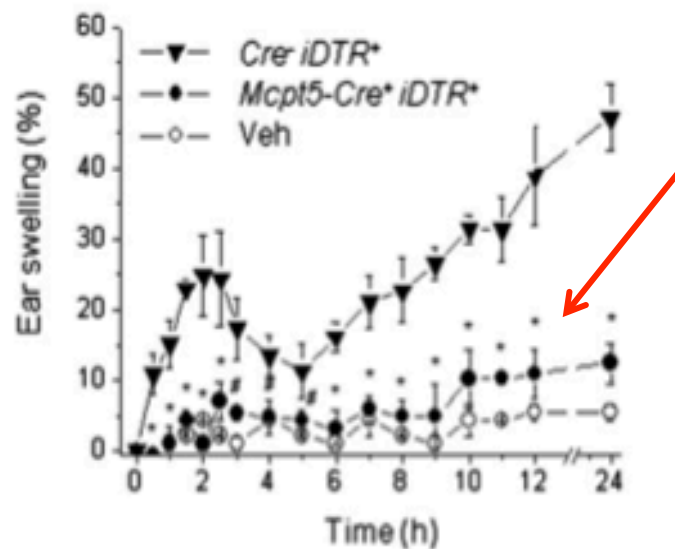
Immunity  
Article

## Mast Cells Are Key Promoters of Contact Allergy that Mediate the Adjuvant Effects of Haptens

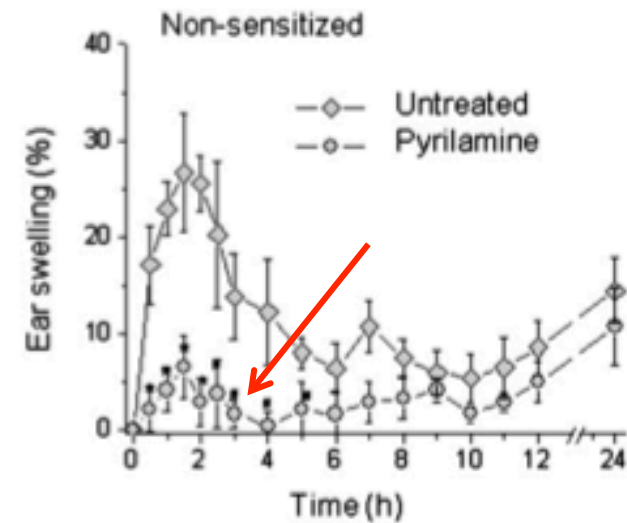
Anne Dudeck,<sup>1,8</sup> Jan Dudeck,<sup>1,8</sup> Julia Schoiten,<sup>2,8</sup> Anke Petzold,<sup>1</sup> Sangeetha Surianarayanan,<sup>1</sup> Anja Köhler,<sup>3</sup> Katrin Peschke,<sup>1</sup> David Vöhringer,<sup>4</sup> Claudia Waskow,<sup>5</sup> Thomas Krieg,<sup>2</sup> Werner Müller,<sup>6</sup> Ari Waisman,<sup>7</sup> Karin Hartman Matthias Gunzer,<sup>3,6,\*</sup> and Axel Roers<sup>1,8,\*</sup>



Dramatic decrease of ACD response in animals conditionally depleted in mast cells



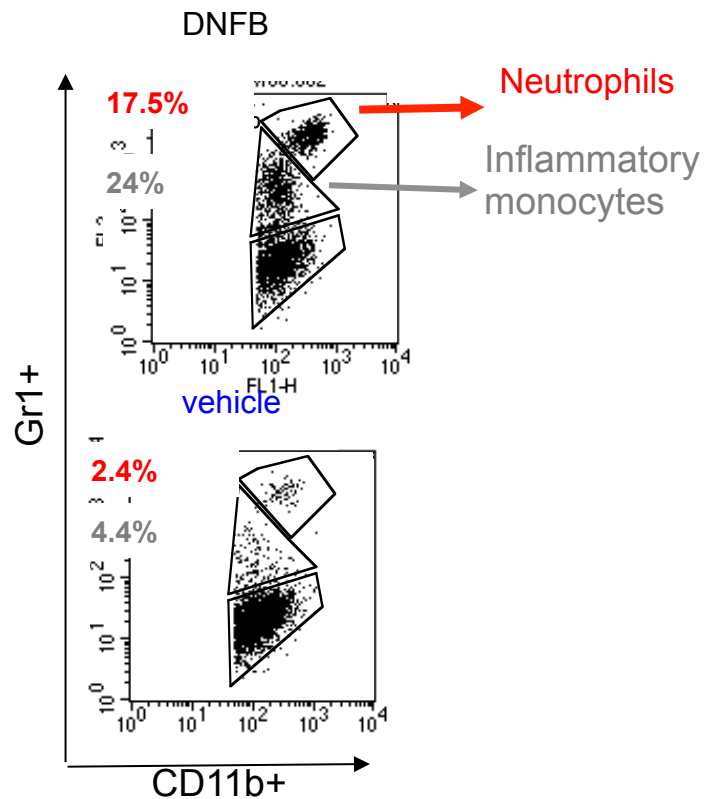
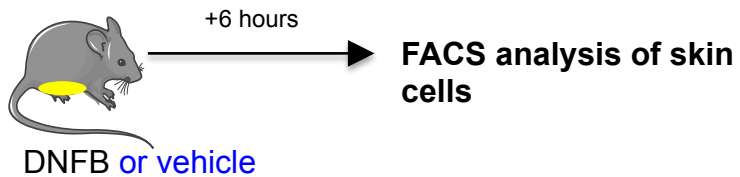
Skin inflammation is histamine-dependent



# Contribution of innate cells? Neutrophils

Large infiltration of neutrophils in the hours following hapten application

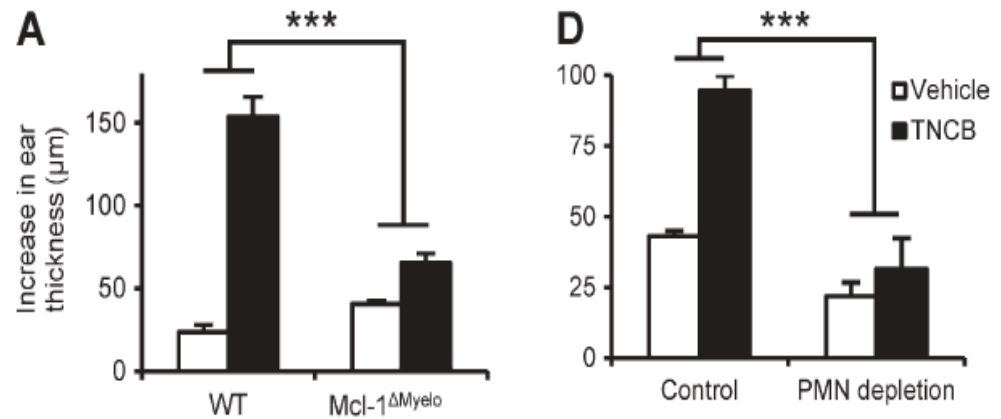
Lack of neutrophils (depletion, transgenic animals) prevents T cell priming and development of skin inflammation



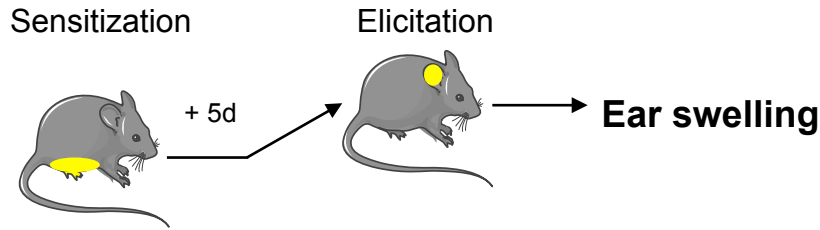
JEM Brief Definitive Report

Neutrophils are required for both the sensitization and elicitation phase of contact hypersensitivity

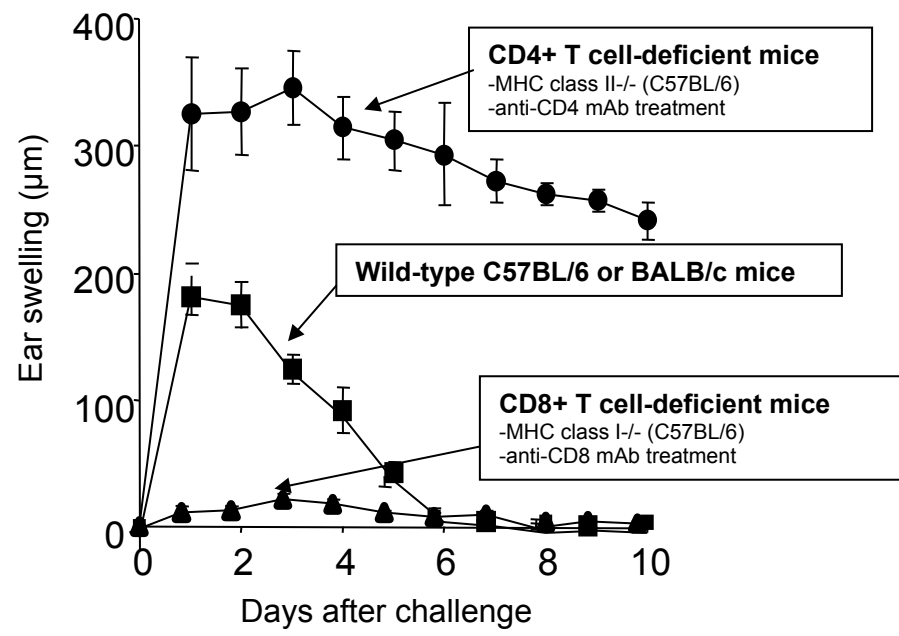
Felix C. Weber,<sup>1,2,3\*</sup> Tamás Németh,<sup>3,4\*</sup> Janka Z. Csepregi,<sup>3,4\*</sup> Anne Dudeck,<sup>5</sup> Axel Roers,<sup>5</sup> Béla Oszvári,<sup>6</sup> Eva Oswald,<sup>1</sup> László G. Puskás,<sup>6</sup> Thilo Jakob,<sup>1</sup> Attila Mócsai,<sup>3,4\*\*</sup> and Stefan F. Martin<sup>1\*\*</sup>



# Main effectors? CD8+ CTLs



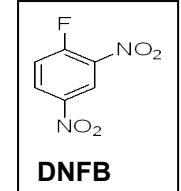
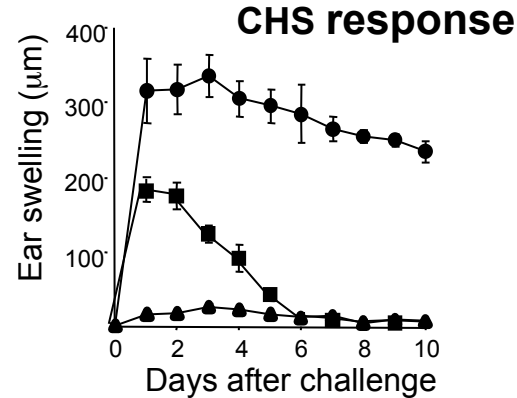
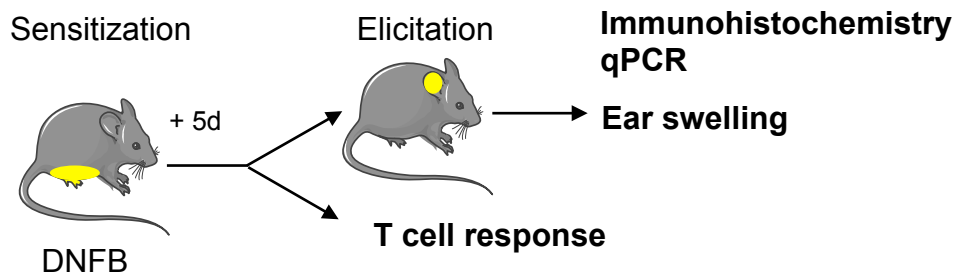
**Strong haptens: DNFB, TNCB, OXAZOLONE...**



- CD8+ T cells are effector cells
- CD4+ T cells comprise regulatory T cells



# Main effectors? CD8+ CTLs

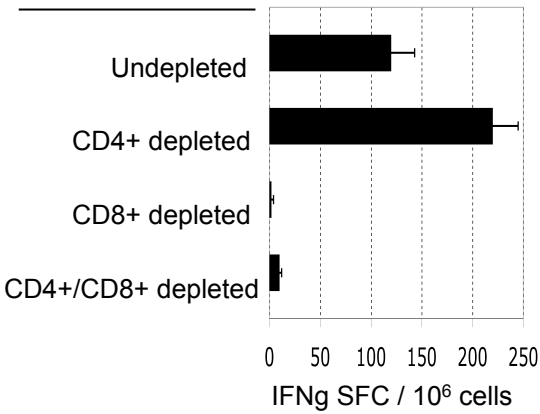


## Priming of IFN $\gamma$ -producing CD8+ T cells

**T cell response**  
(draining lymph nodes)

*Elispot assay*

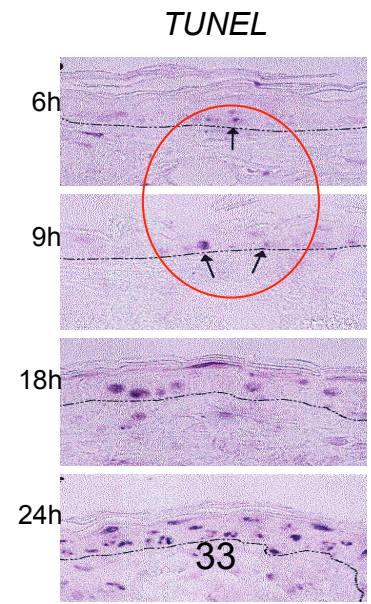
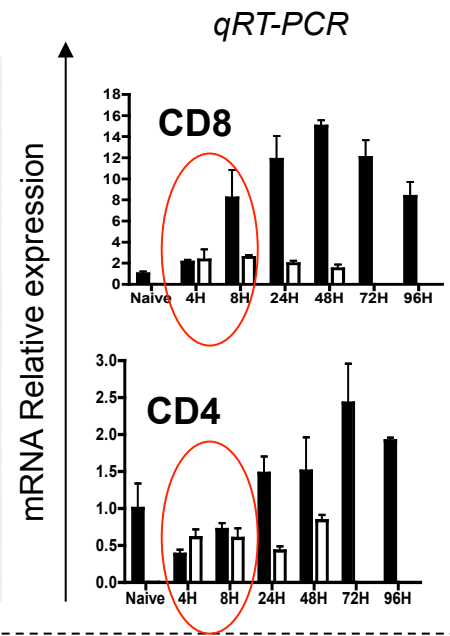
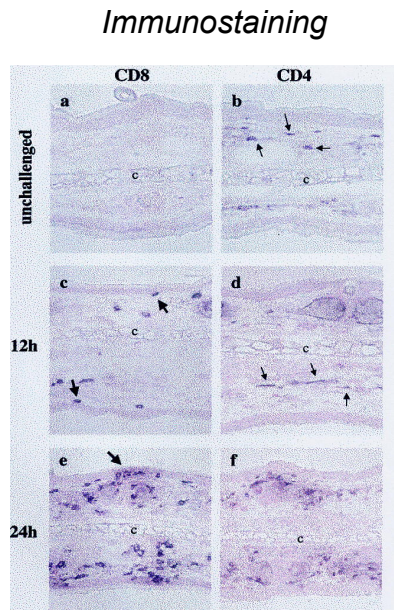
**mAb Treatment**



## Early recruitment of CD8+ T cells initiates eczema

**T cells recruitment**  
(challenged ears)

**Keratinocytes:**  
target of CTLs



# Main effectors? CD8+ CTLs

## Recurrence, chronicity

### Inhibitory checkpoint receptors control CD8<sup>+</sup> resident memory T cells to prevent skin allergy

J ALLERGY CLIN IMMUNOL  
2019

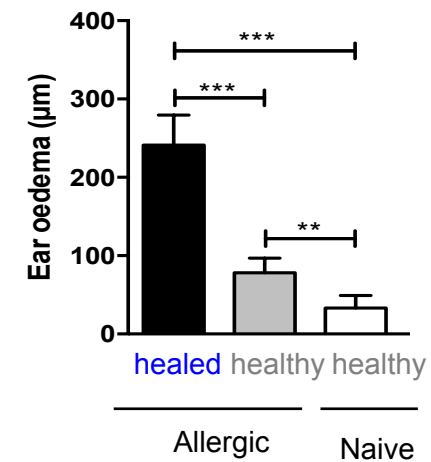
Pia Gamradt, PhD,<sup>a,b,c,d,e\*</sup> Léo Laoubi, MSc,<sup>a,b,c,d,e\*</sup> Audrey Nosbaum, MD, PhD,<sup>a,b,c,d,e</sup> Virginie Mutez, MSc,<sup>a,b,c,d,e</sup> Vanina Lenief, MSc,<sup>a,b,c,d,e</sup> Sophie Grande, MD,<sup>f</sup> Daniel Redoulès, PhD,<sup>g</sup> Anne-Marie Schmitt, MD, PhD,<sup>h</sup> Jean-François Nicolas, MD, PhD,<sup>a,b,c,d,e,f</sup> and Marc Vocanson, PhD<sup>a,b,c,d,e</sup> Lyon, Pierre-Benite, and Toulouse, France

Healthy skin

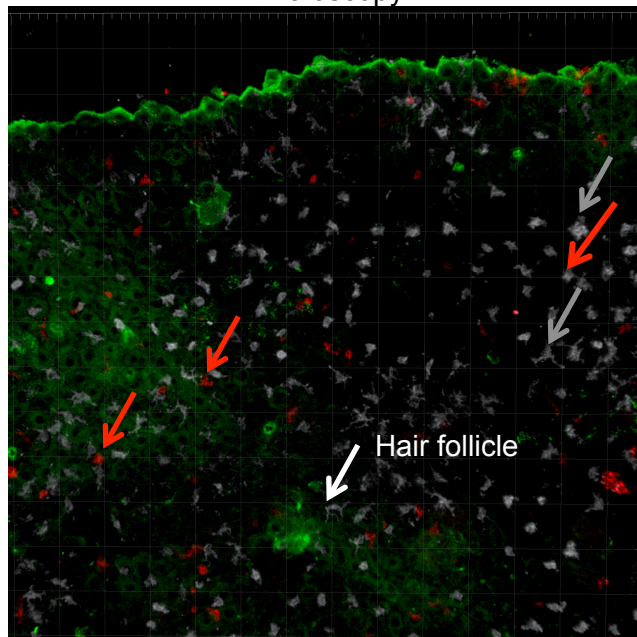


allergic animal

Flare-up reaction



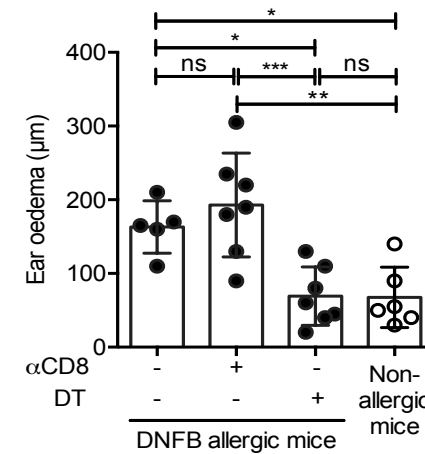
Epidermal sheet, Confocal microscopy



Skin edge

CD8+ T cells  
Ag = DNP moities  
DETC

Acute depletion of CD8+Trm abrogates flares



Injection of diptheria toxin or anti-CD8+ mAbs  
IDTR transgenic animals

# Main effectors? NK cells

- NK cells are far less important than CD8+ CTLs for eczema

European Journal of Immunology

**Natural killer cells and T cells induce different types of skin reactions during recall responses to haptens**

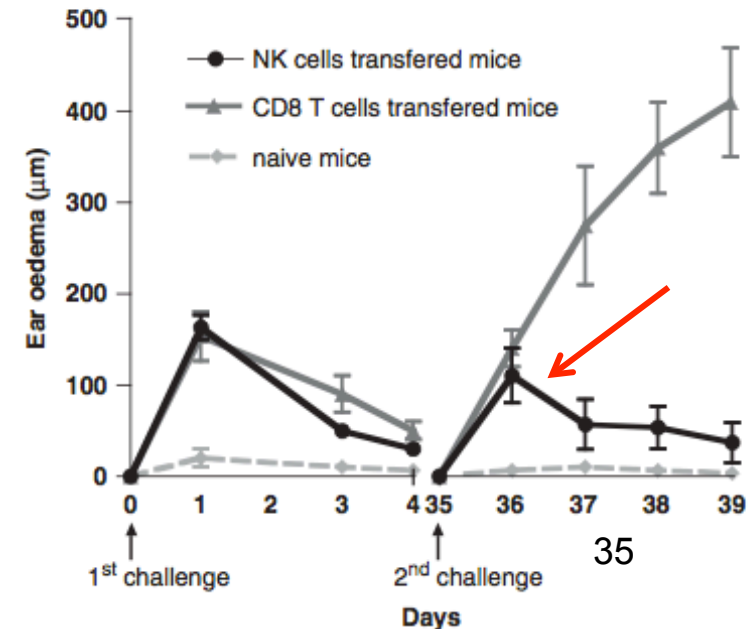
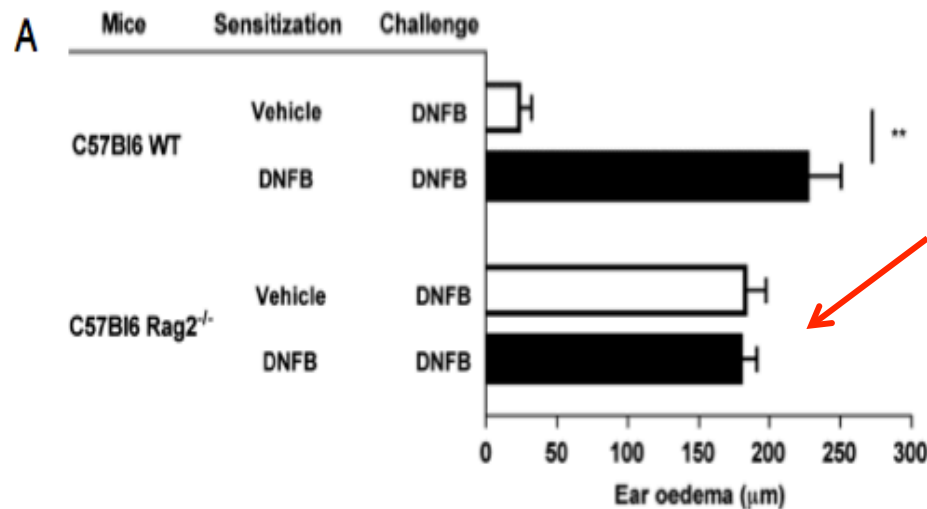
Paul Rouzairé<sup>1,2,3</sup>, Carmelo Luci<sup>4</sup>, Elisabeth Blasco<sup>1,2,3</sup>, Jacques Bienvenu<sup>1,2,3</sup>, Thierry Walzer<sup>1,2</sup>, Jean-François Nicolas<sup>1,2,5</sup> and Ana Hennino<sup>1,2</sup>

Paulst S. Nat Immunol 2011

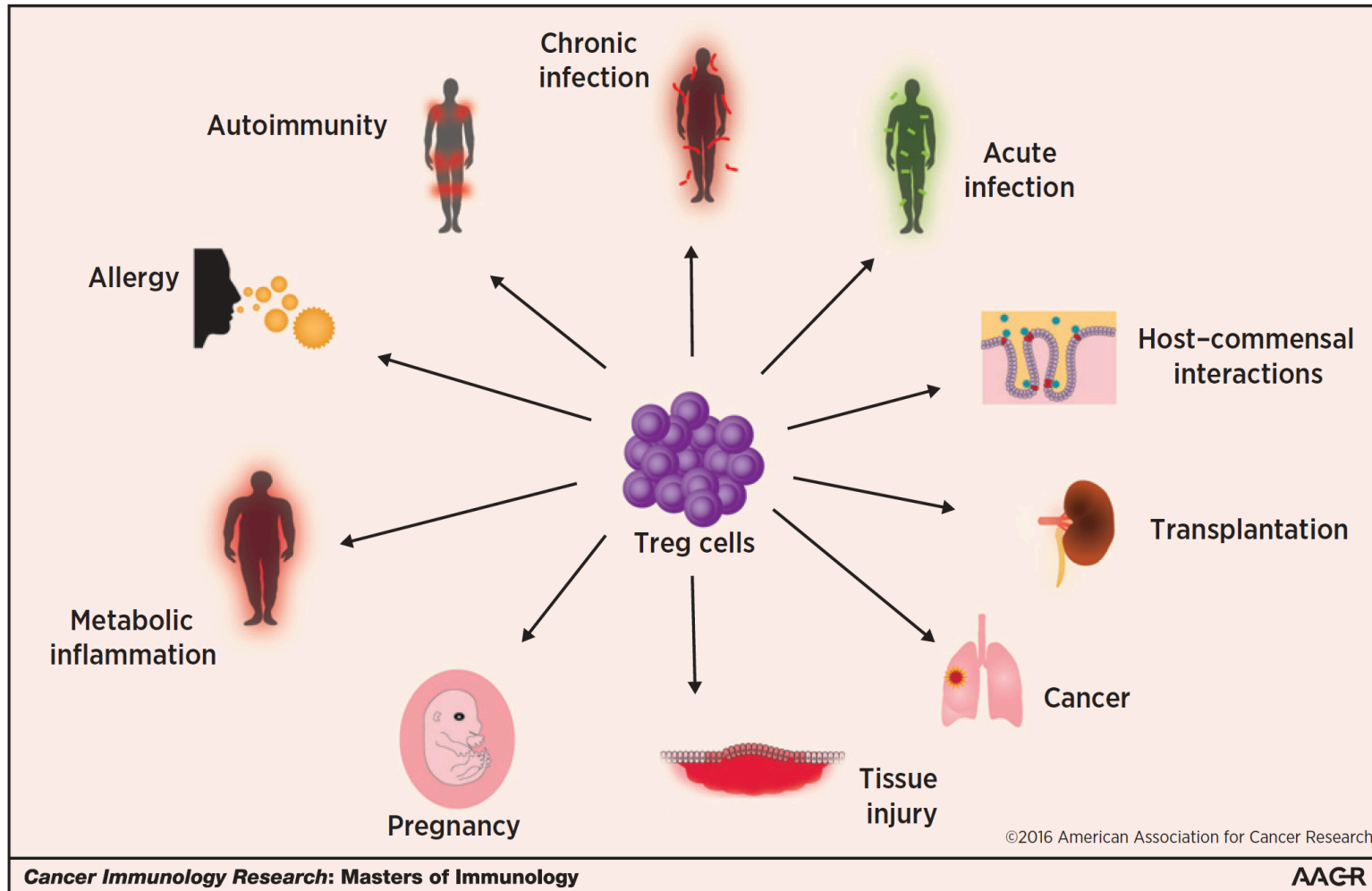
NK cells confer CHS and recall responses, when extracted from liver and transferred into recipient animals

-> NK cell « memory »

No CHS response in T cell-deficient strains

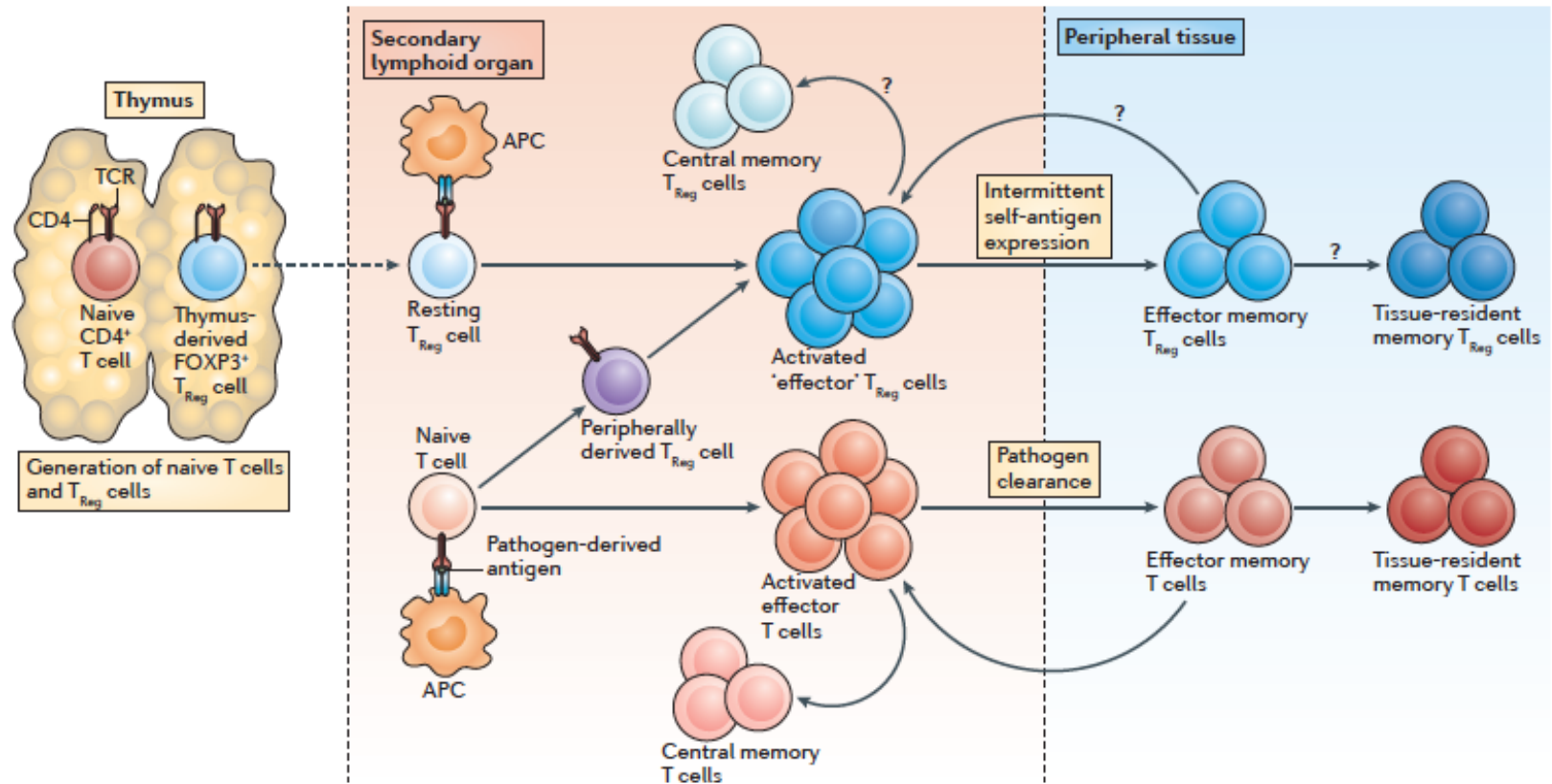


# Les lymphocytes T régulateurs FoxP3+



# Les lymphocytes T régulateurs FoxP3+

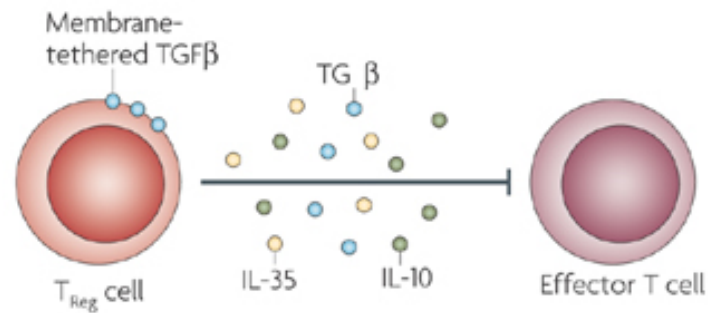
## Ontogeny: Life Cycle of Regulatory and Conventional T cells



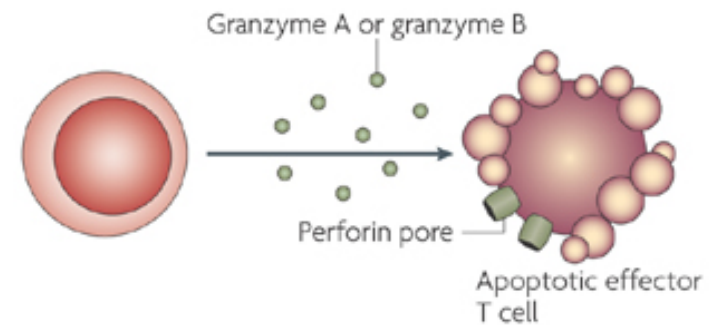
# Les lymphocytes T régulateurs FoxP3+

## Suppressive mechanisms used by Tregs

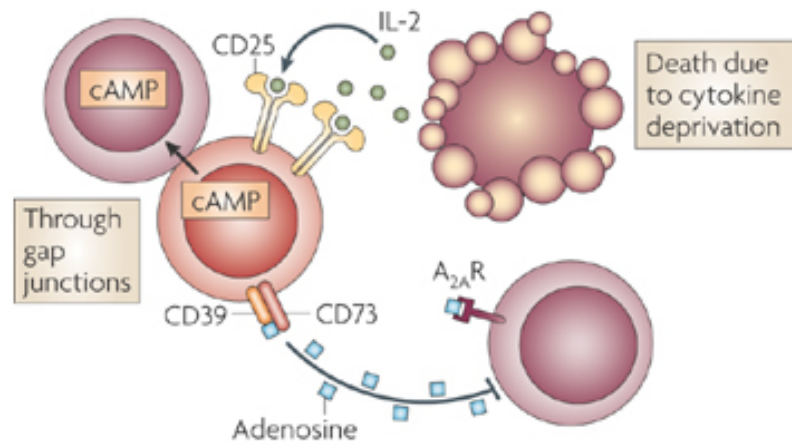
**a Inhibitory cytokines**



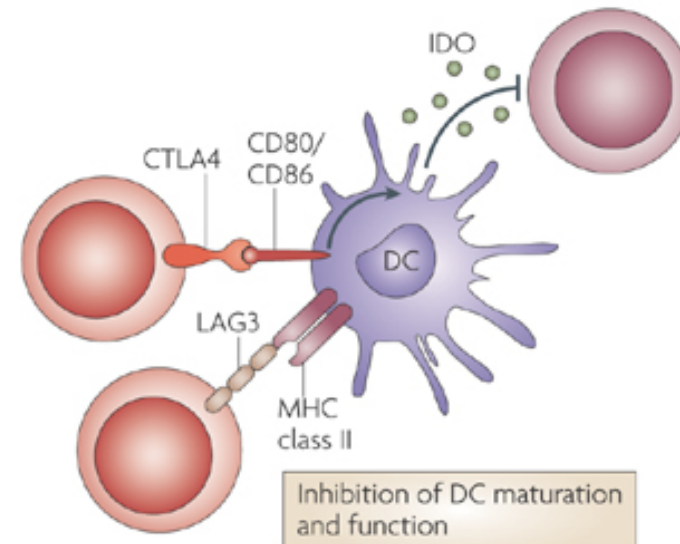
**b Cytolysis**



**c Metabolic disruption**



**d Targeting dendritic cells**



# Main regulatory cells? FoxP3+Tregs

- Multifunctional FoxP3+ICOS+ regulatory T cells control CTL-induced skin inflammation

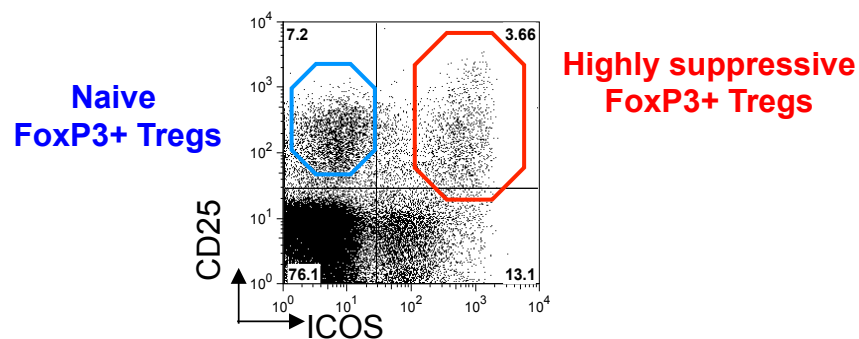
**Inducible costimulator (ICOS) is a marker for highly suppressive antigen-specific T cells sharing features of T<sub>H</sub>17/T<sub>H</sub>1 and regulatory T cells**

J ALLERGY CLIN IMMUNOL  
VOLUME 126, NUMBER 2

Marc Vocanson, PhD,<sup>a,b,c</sup> Aurore Rozieres, PhD,<sup>a,b,c</sup> Anca Hennino, PhD,<sup>b,c</sup> Gaelle Poyet, MSc,<sup>a,b,c</sup>  
Vincent Gaillard, BSc,<sup>a,b,c</sup> Sarah Renaudineau, MSc,<sup>b,c</sup> Amine Achachi, PhD,<sup>b,c</sup> Josette Benetiere, BSc,<sup>a,b,c</sup>  
Dominique Kaiserlian, PhD,<sup>b,c</sup> Bertrand Dubois, PhD,<sup>b,c</sup> and Jean-François Nicolas, MD, PhD<sup>a,b,c,d</sup> Lyon, France

Activation of CD4+CD25+FoxP3+ICOS+ Tregs in the draining lymph nodes of hapten-sensitized mice

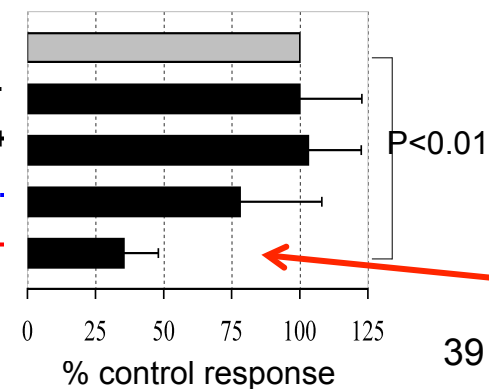
Transfer of FoxP3+ICOS+ Tregs prevents the priming of CD8+ CTLs and the development of skin inflammation in an antigen-dependant manner



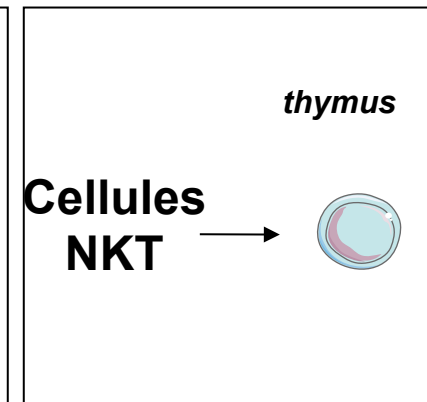
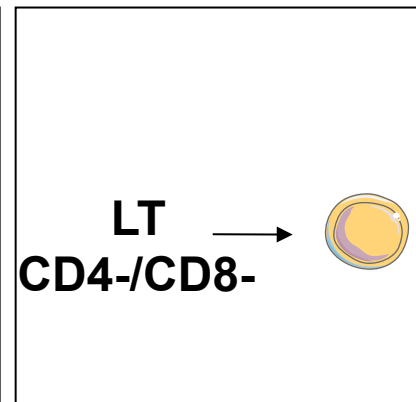
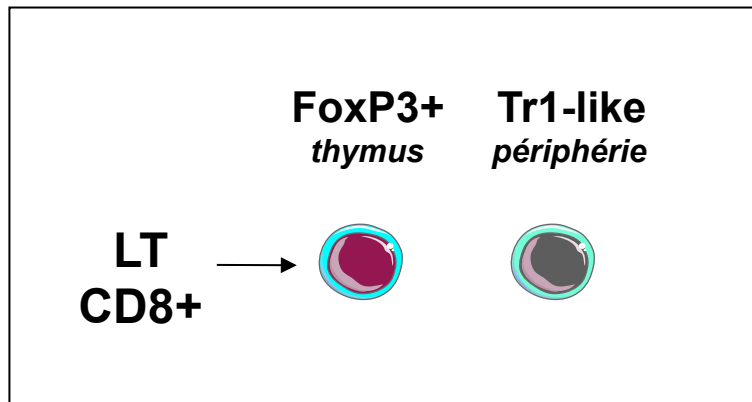
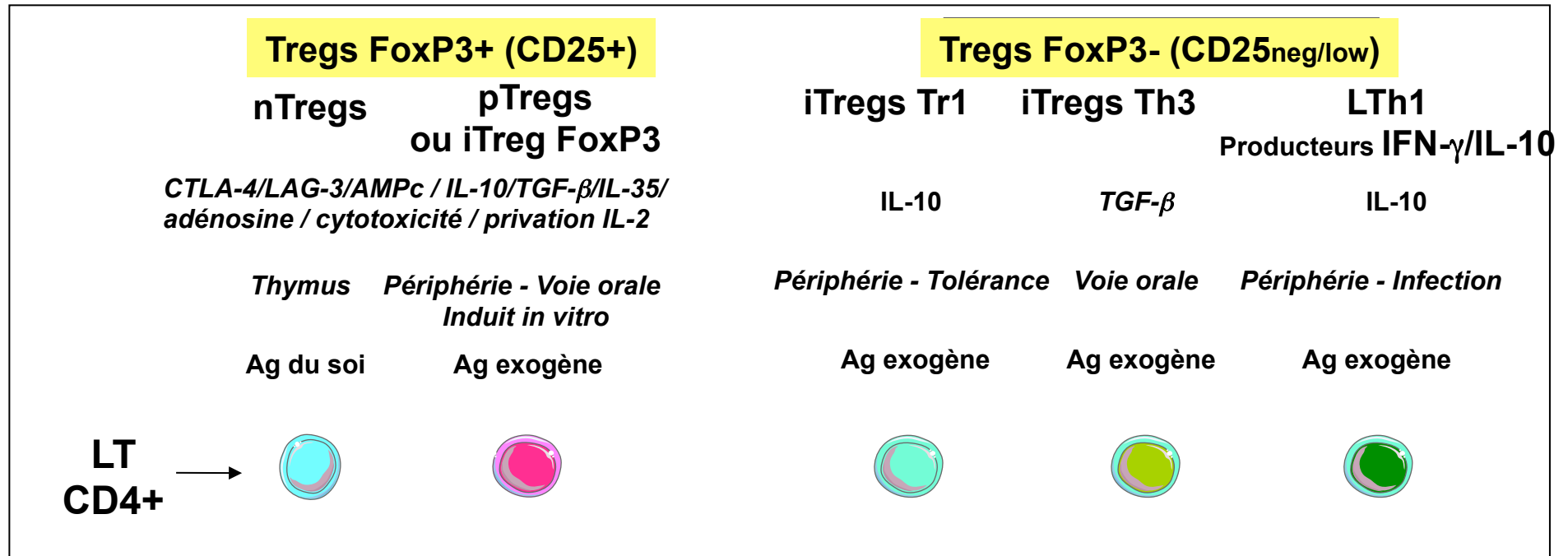
Transferred cells

CHS response 48h

PBS  
CD4+CD25-ICOS-  
CD4+CD25-ICOS+  
CD4+CD25+ICOS-  
CD4+CD25+ICOS+

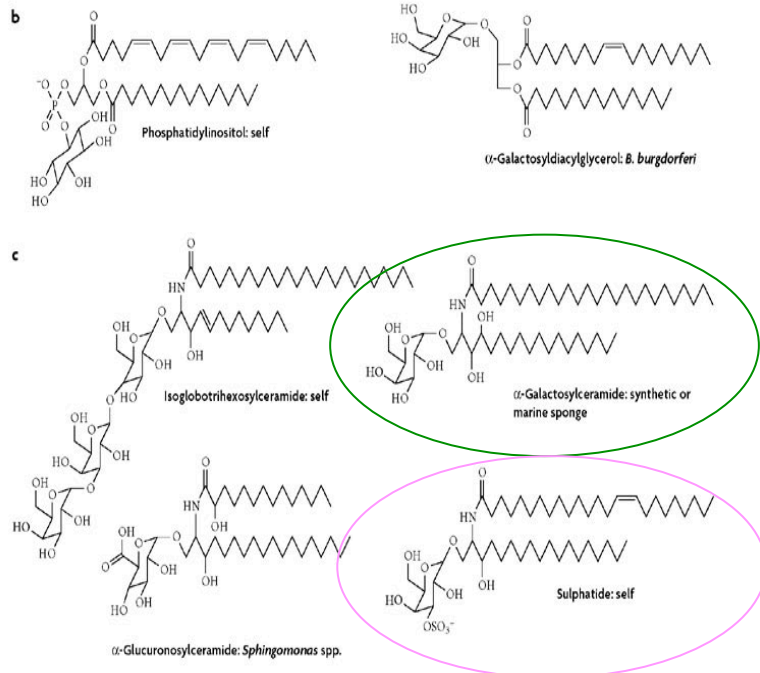
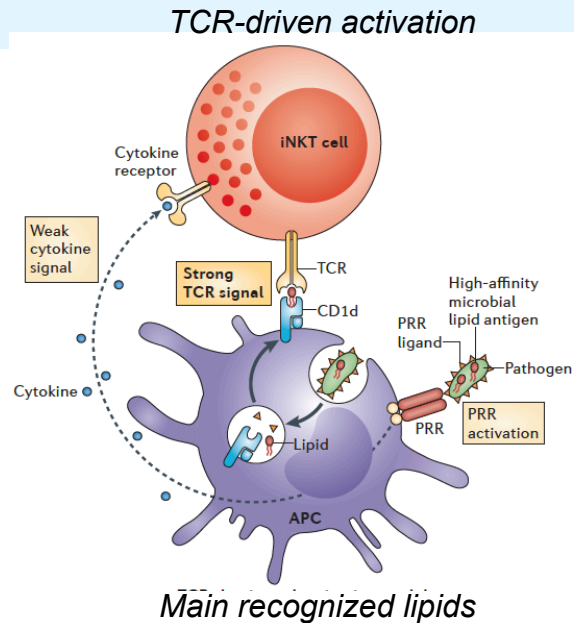


# De nombreux lymphocytes régulateurs





# Les lymphocytes non conventionnels : les cellules NKTs



## Main features

- 2 groups of NKT cells:
  - ✓ invariant NKT cells (iNKT cells) = TCR  $V\alpha 24J\alpha 18$  and mainly  $V\beta 11$
  - ✓ non-invariant NKT cells (oligoclonal)
- iNKT predominant in mice, few in humans
- NKT cells promote immunity against cancers and microbes but suppress autoimmunity
- Functional versatility → different subsets (NKT1, NKT2, NK17, NKT<sub>FH</sub>...)
- iNKT cells respond to self and microbial lipids similar to the glycosphingolipid  $\alpha$ -GalCer
- Non-invariant NKT cells respond to lipids similar to sulphatide
- CD1d-restriction

# Other regulatory cells? iNKT cells

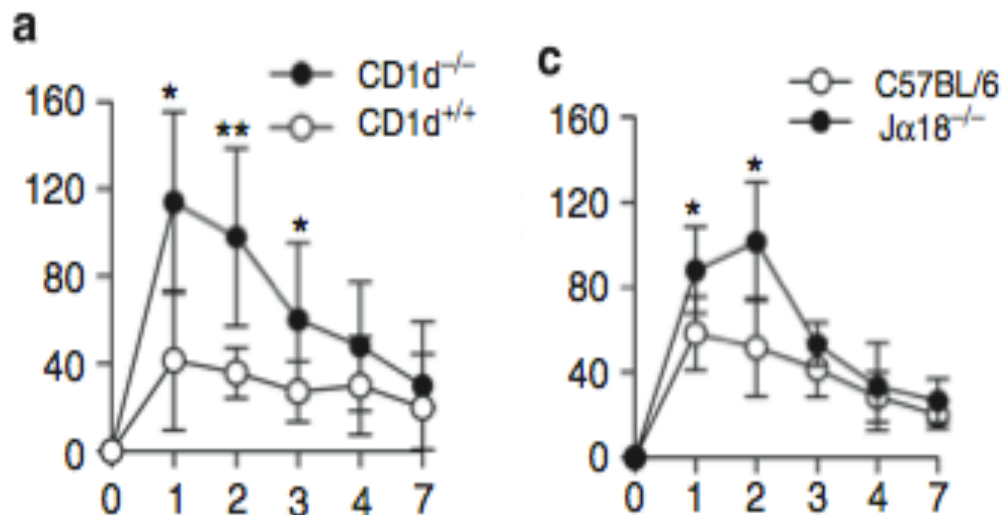
- iNKT cells are non-redundant downregulators of CTL-mediated CHS responses

## Invariant NKT Cells Suppress CD8<sup>+</sup> T-Cell-Mediated Allergic Contact Dermatitis Independently of Regulatory CD4<sup>+</sup> T Cells

Anne Goubier<sup>1,2,3,6</sup>, Marc Vocanson<sup>1,2,3,6</sup>, Claire Macari<sup>1,2,3</sup>, Gaëlle Poyet<sup>1,2,3</sup>, André Herbelin<sup>4,5</sup>, Jean-François Nicolas<sup>1,2,3</sup>, Bertrand Dubois<sup>1,2,3,6</sup> and Dominique Kaiserlian<sup>1,2,3,6</sup>

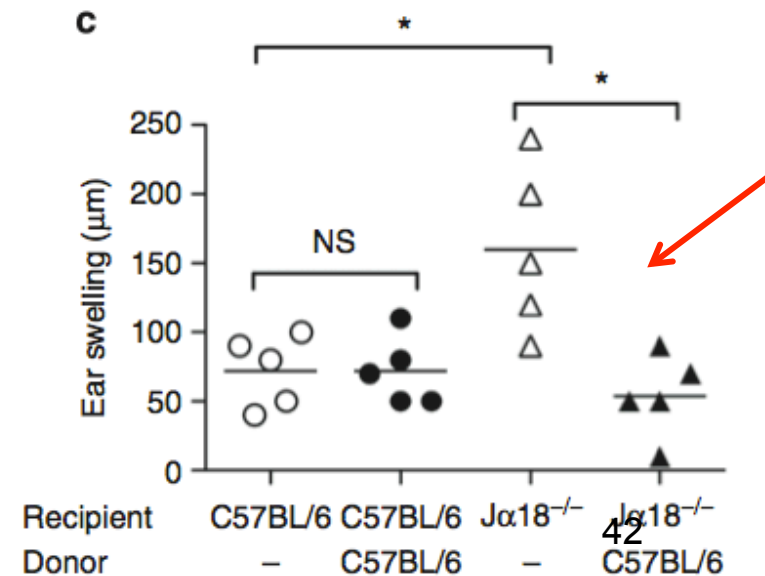
*Journal of Investigative Dermatology* (2013) 133, 980–987; doi:10.1038/jid.2012.404; published online 29 November 2012

Decreased CHS to DNFB response in NKT deficient mice (B6)



Other studies argue against the regulatory functions of iNKT cells and suggest stimulatory functions

Adoptive transfer of iNKT in Ja18<sup>-/-</sup> mice normalises CHS response



# Other regulatory cells? B cell subsets

- Other regulatory cells (peritoneal B-1a cells) participate to the resolution of skin inflammation

**THE JOURNAL OF IMMUNOLOGY**

This information is current as of June 24, 2014.

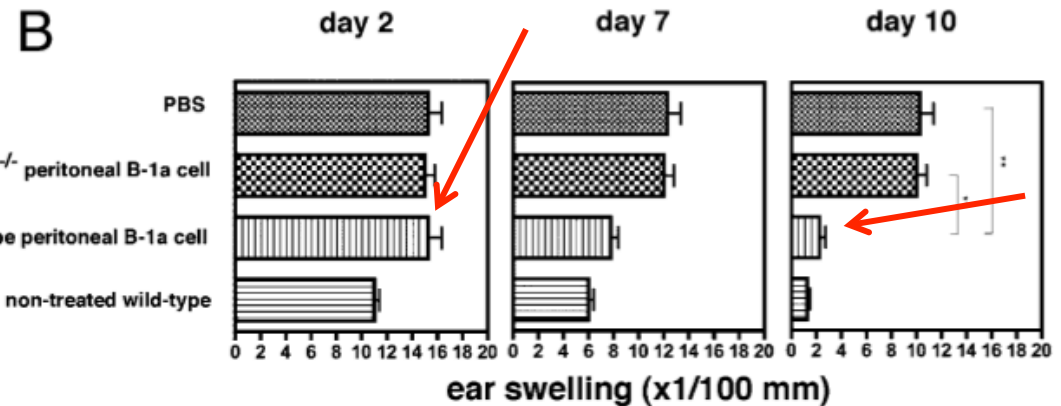
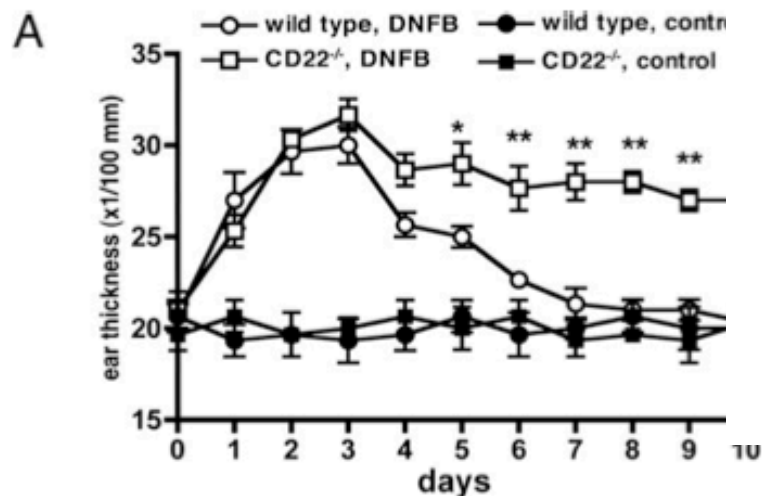
**CD22 Expression Mediates the Regulatory Functions of Peritoneal B-1a Cells during the Remission Phase of Contact Hypersensitivity Reactions**

Hiroko Nakashima, Yasuhito Hamaguchi, Rei Watanabe, Nobuko Ishiura, Yoshihiro Kuwano, Hitoshi Okochi, Yoshimasa Takahashi, Kunihiko Tamaki, Shinichi Sato, Thomas F. Tedder and Manabu Fujimoto

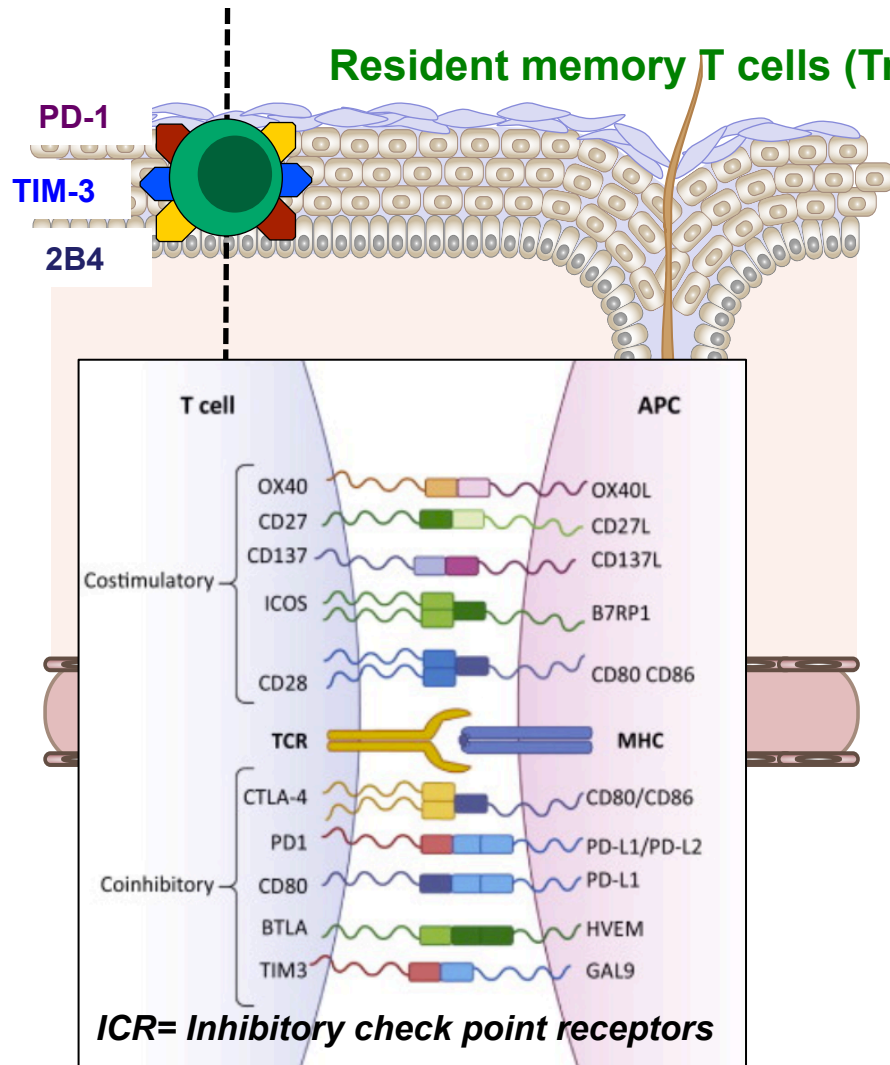
*J Immunol* 2010; 184:4637-4645; Prepublished online 24 March 2010;  
doi: 10.4049/jimmunol.0901719  
<http://www.jimmunol.org/content/184/9/4637>

Absence of CHS resolution in CD22<sup>-/-</sup> animals

Adoptive transfer of B1-a cell promotes the resolution of skin inflammation in CD22<sup>-/-</sup> animals



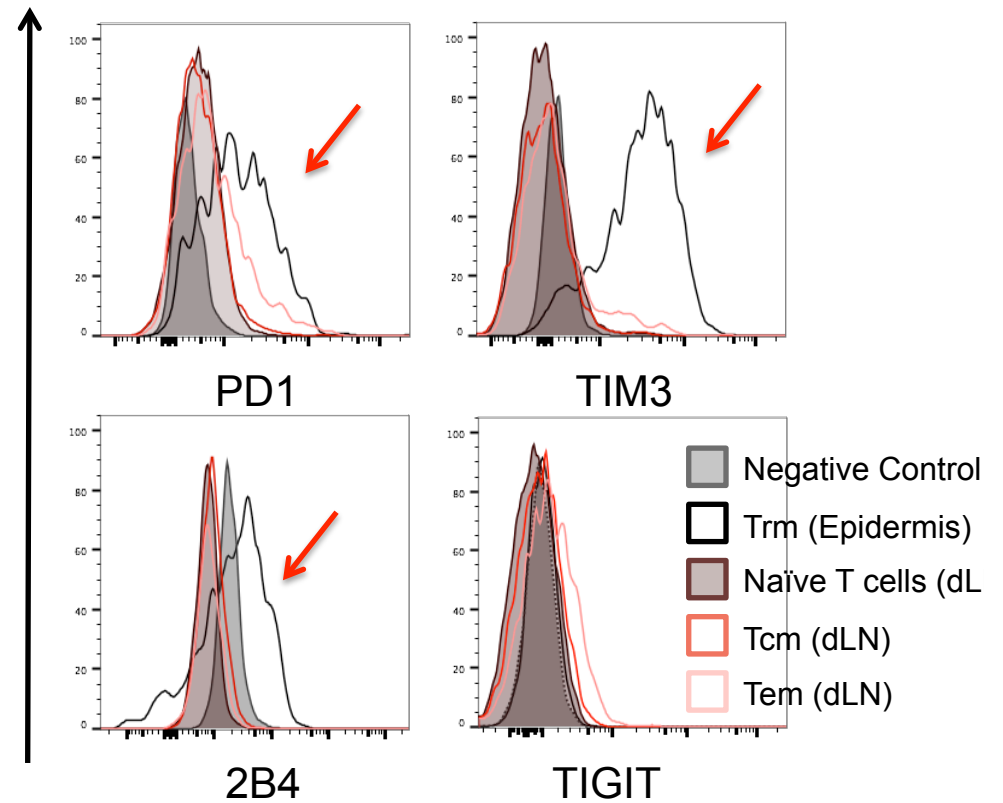
# Les mécanismes de régulation intrinsèques: Les récepteurs inhibiteurs



DNFB allergic mice  Healed

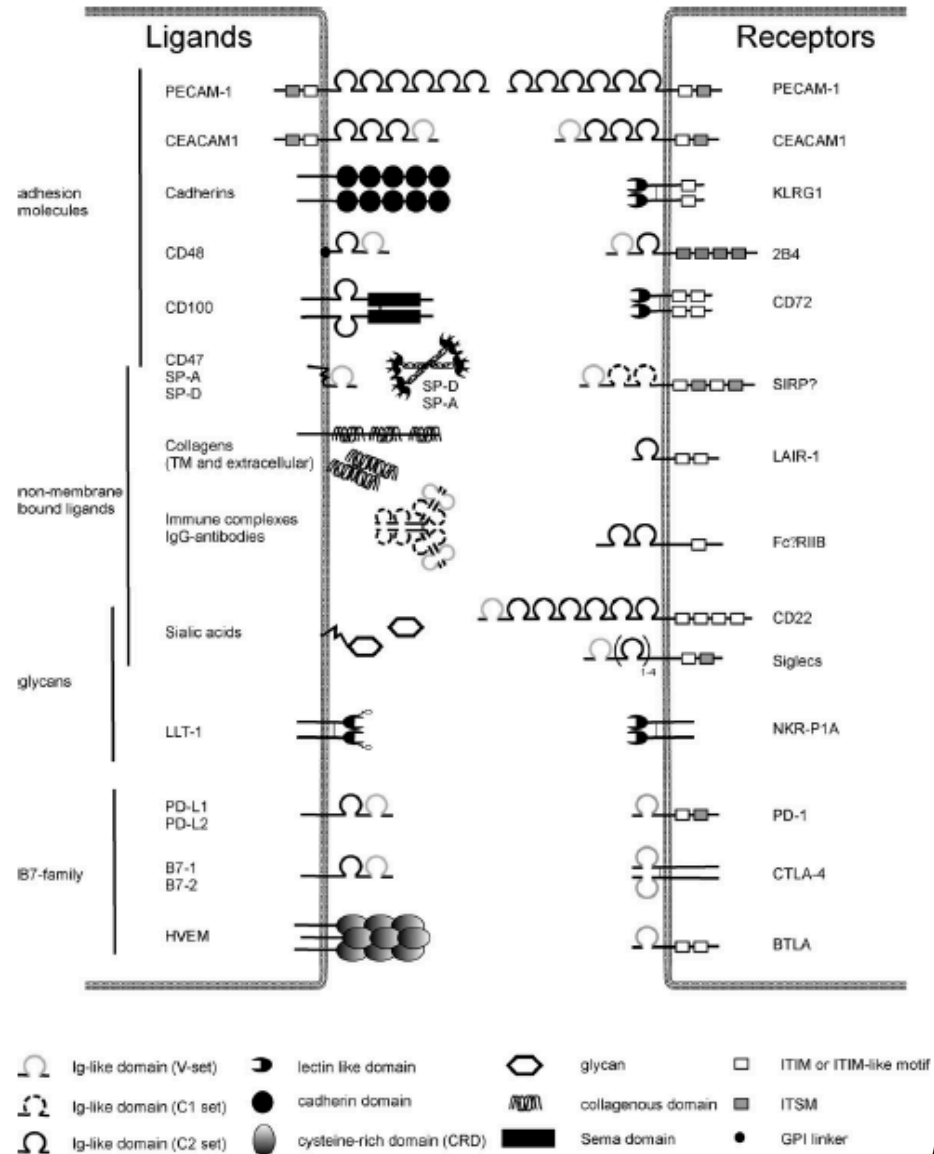
→ FACS

Expression of ICRs on skin CD8+ Trm



- Certain ICRs are expressed on skin CD8+ Trm but not on circulating memory T cells → they limited recurrences & exacerbations

# D'autres mécanismes de régulation par le tissu?

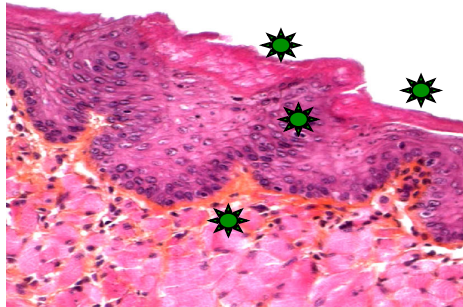


# Eczéma allergique de contact : les facteurs de risques

Ignorance?



Tolérance



Sensibilisation  
Eczéma

- > la nature de l'antigène = "le danger"
- > les conditions d'exposition (dose, fréquence, durée, route)
- > le polymorphisme génétique (barrière cutanée, enzymes de détoxification...), âge, sexe
- > l'environnement (maladie sous-jacente, stress, pollution...)

# Les facteurs génétiques

5. Studies on probably functionally relevant polymorphisms in contact allergic patients from our study, from more recent studies (columns I–IV, rows 1, 5, 6, and 9), and replication studies (column V)

	II	III	
	Polymorphisms	Results	
	Filaggrin null mutations (combined)	Results inconclusive	contact dermatitis vs. controls: risk
<b>Table 3.</b>	Difference in sensitization rates between children of sensitized and non-sensitized parents. 'The potent allergen DNCB is probably overpowering genetic influences'; Walker et al. (21)		contact dermatitis reaction to nickel
	Percentage of children sensitized		nickel
Status of parents	DNCB	NDMA	sensitizers'
Sensitized	65	51	based; increased.
Not sensitized	52	29	VAT2*4
	$p < 0.10$	$p < 0.01$	11 patients found as group allergic
	DNCB, 2,4-dinitrochlorobenzene; NDMA, p-nitroso-dimethylaniline.		acid – 9 reference
	Cytokines: <i>ILB</i> – 511, <i>ILB</i> +3953, <i>ILRA</i> , <i>IL6</i> – 174, <i>TNFA</i> – 238, <i>TNFA</i> – 308	<i>TNFA</i> – 308 (G → A): increased (in polysensitized individuals) <i>TNFA</i> – 308 G/G and <i>ILRA</i> polymorphism (77) increased in Turkish patients ( $n = 50$ )	gene pairs in intron (y) increased
	Cytokine: <i>IL-16</i>	<i>IL16</i> – 295 (T → C) increased (in polysensitized individuals)	
	Cytokine <i>IL-4</i>	No difference between chromate allergic and controls with regard to <i>IL4</i> – 590 polymorphism	

## Le pouvoir sensibilisant des haptènes varient en fonction de la nature des molécules

Chimique	Secteur	Pouvoir sensibilisant
Oxazolone	Chimie	Extrême
2,4-Dinitrofluorobenzène	Chimie	Extrême
2,4-Dinitrochlorobenzène	Chimie	Extrême
Glutaraldehyde	Conservateur, antiseptique	Fort
Formaldehyde	Cosmétique, Colorant	Fort
Cinnamaldehyde	Parfum, arôme	Modéré
Hexyl cinnamaldehyde	Cosmétique, Parfum	Modéré/Faible
Eugenol	Cosmétique, Parfum	Faible
Hydroxycitronellal	Cosmétique, Parfum	Faible
Linalool	Cosmétique	Faible
Citral	Parfum, arôme	Faible
Vanillin	Parfum, arôme	Faible
2,4-Dinitrocyano benzène	Chimie	Faible
Amoxicilline, cyanamide, ceftrimide	Médicament	Faible

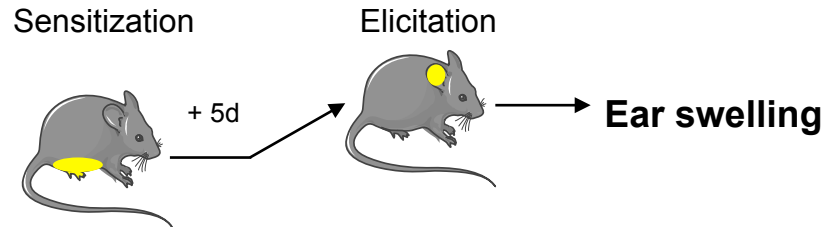


# Main effectors? CD8+ CTLs

*Journal of Investigative Dermatology* (2006) 126, 815-820. doi:10.1038/sj.jid.5700174; published online 2 February 2006

## CD8+ T Cells Are Effector Cells of Contact Dermatitis to Common Skin Allergens in Mice

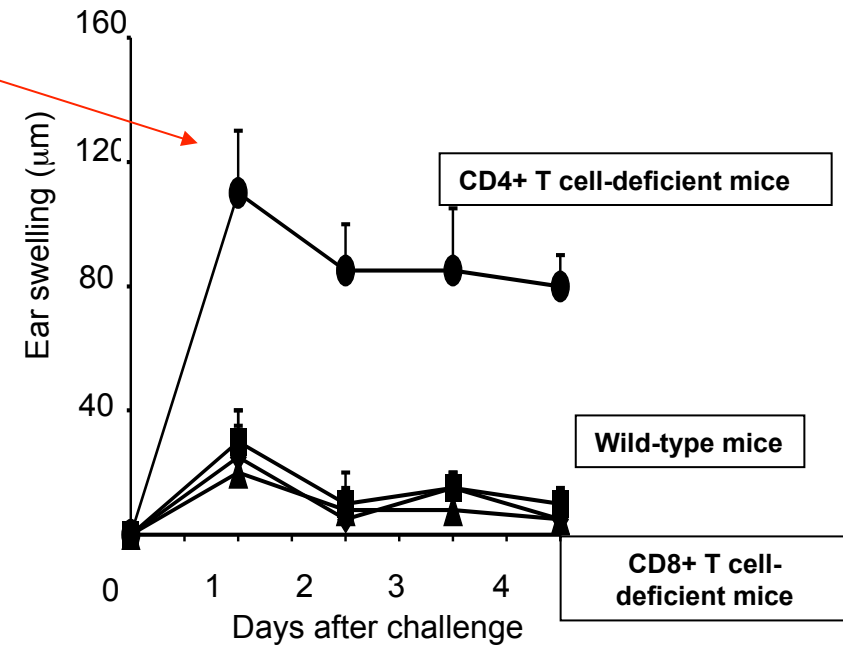
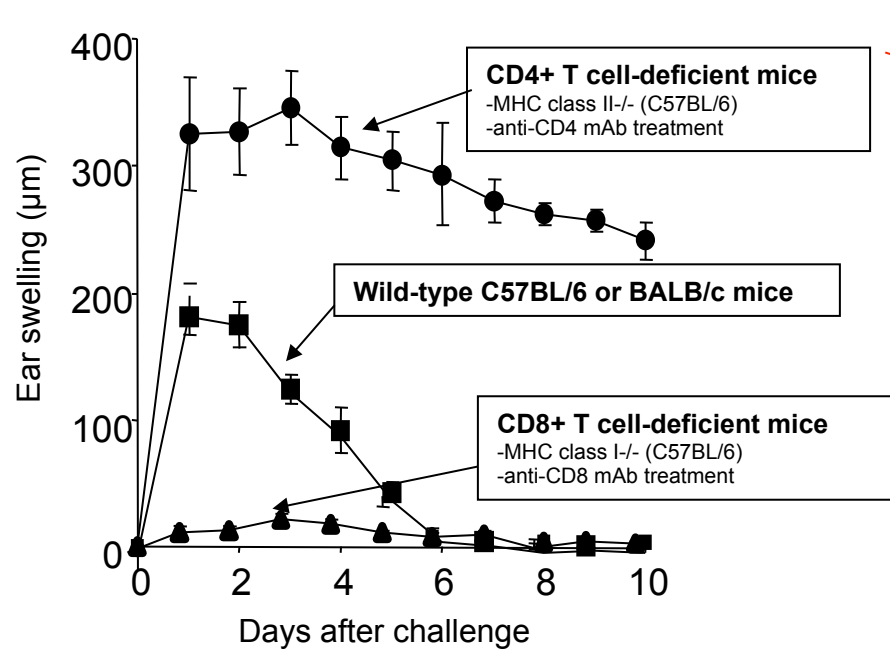
Marc Vocanson<sup>1</sup>, Anca Hennino<sup>1</sup>, Magalie Cluzel-Tailhardat<sup>1</sup>, Pierre Saint-Mezard<sup>1</sup>, Josette Benetiere<sup>1</sup>, Cyril Chavagnac<sup>1</sup>, Frederic Berard<sup>1,2</sup>, Dominique Kaiserlian<sup>3</sup> and Jean-François Nicolas<sup>1,2</sup>



**Strong haptens: DNFB, TNCB, OXAZOLONE...**

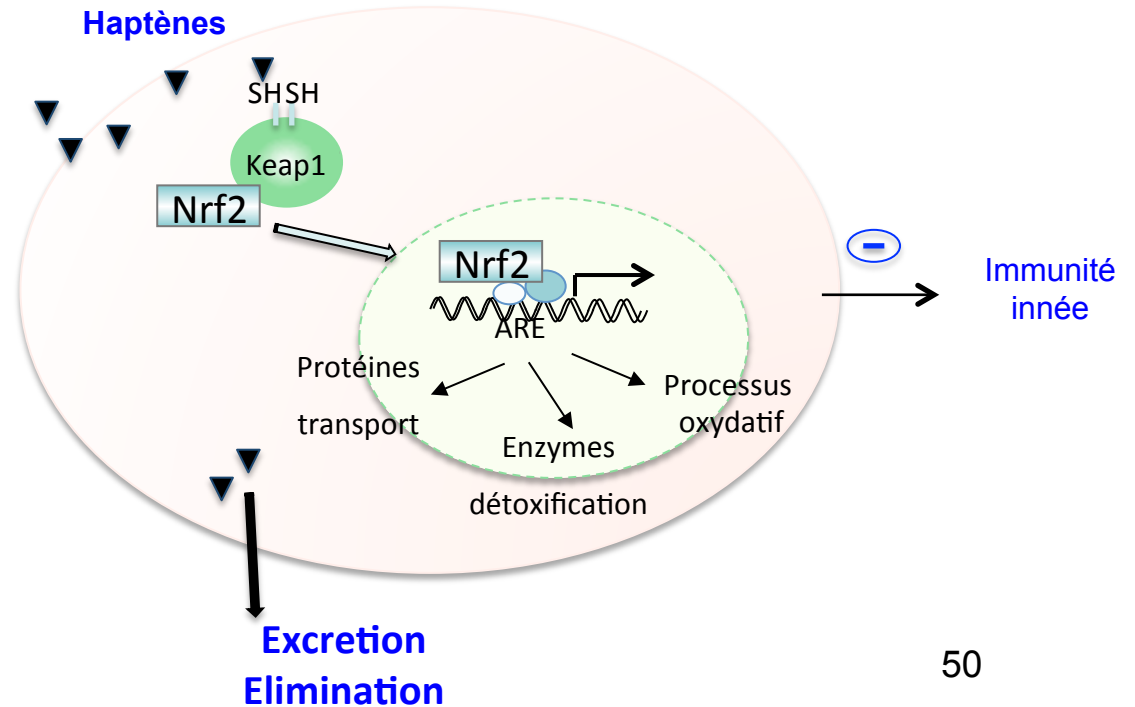
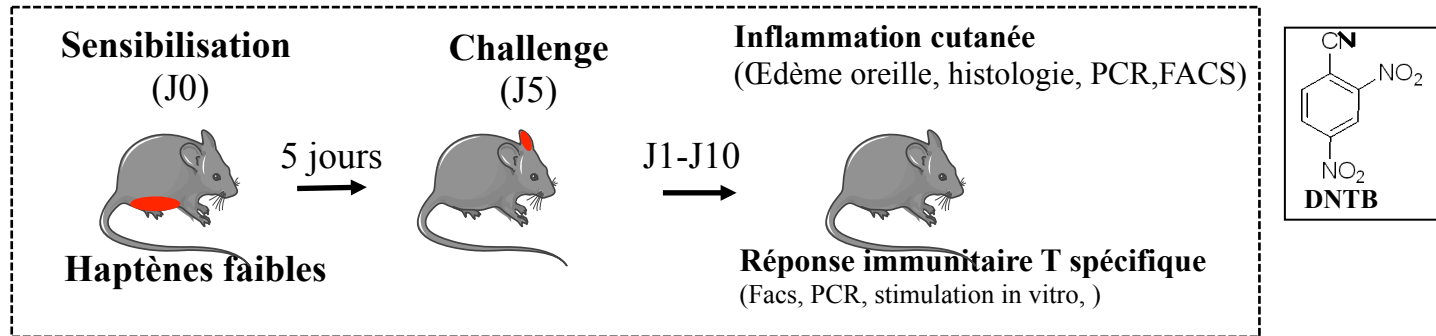
### Weak haptens

- Fragrances (Hexylcinnamaldehyde, Hydroxycitronellal, Eugenol, Dihydrocoumarin, Isoeugenol),
- Dye (paraphenylenediamine)
- Drugs (Amoxicillin, Rosephin, Phenytoin, Sulfasalazin)

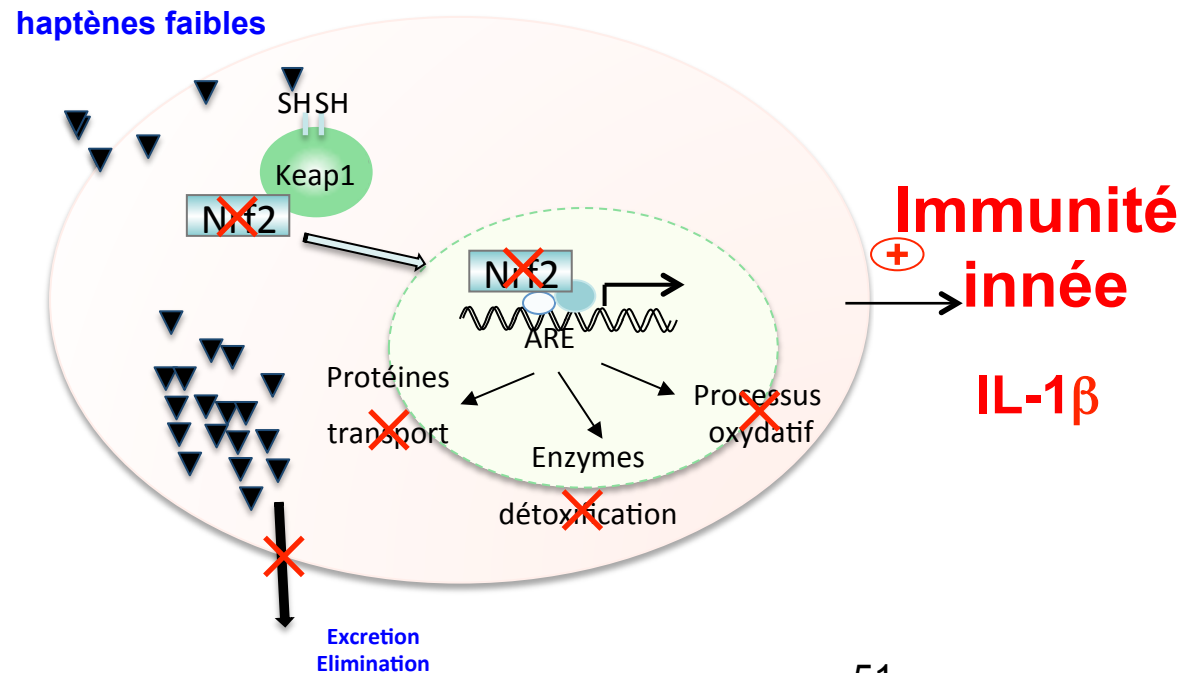
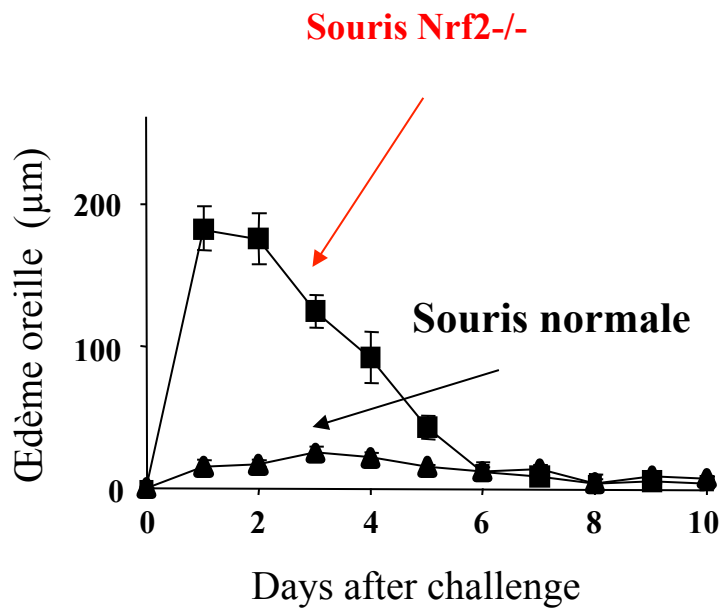
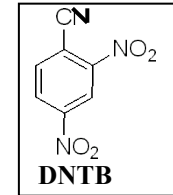
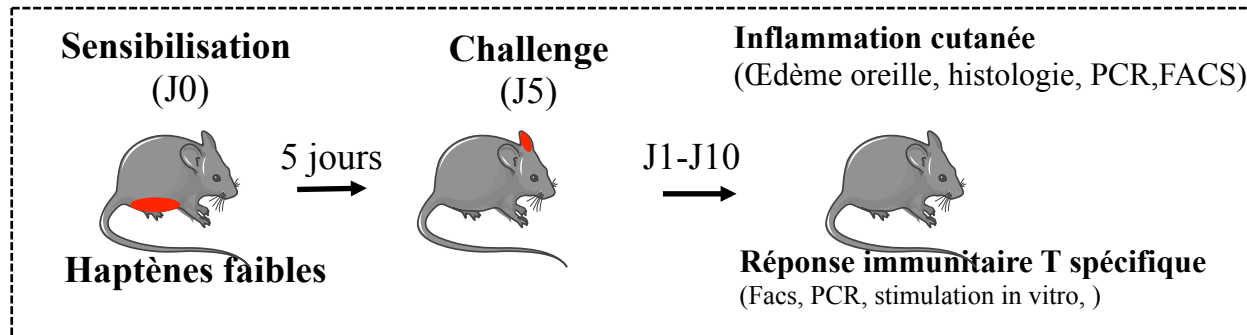


- CD8+ T cells are effector cells
- CD4+ T cells comprise regulatory T cells

# Un défaut de détoxification conduit à une rupture de tolérance vis-à-vis des haptènes faibles

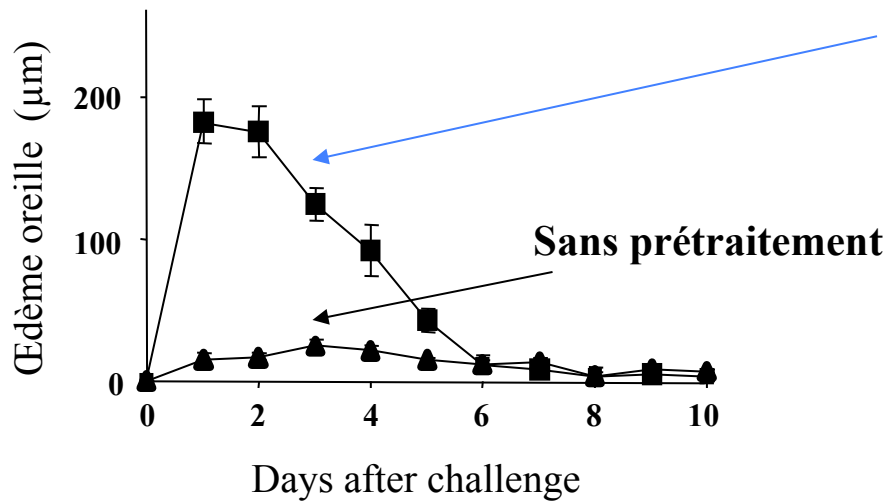
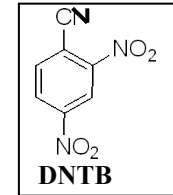
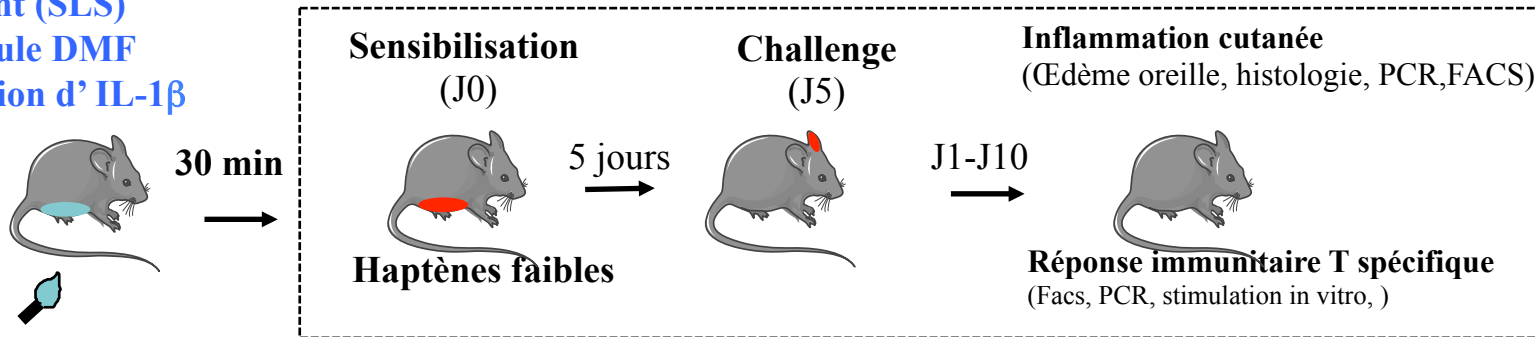


# Un défaut de détoxification conduit à une rupture de tolérance vis-à-vis des haptènes faibles



# L'irritation favorise la rupture de tolérance vis-à-vis des haptènes faibles

Prétraitement  
Irritant (SLS)  
Véhicule DMF  
Injection d'IL-1 $\beta$



L'application d'un irritant SDS / un véhicule différent (DMF) / l'injection d'une cytokine proinflammatoire comme IL-1 $\beta$  avant la sensibilisation induit une réponse d'eczéma vis-à-vis d'un allergène faible

L'inverse est vrai pour un haptène fort : bloquer la production d'IL-1 $\beta$  prévient la sensibilisation et favorise la tolérance

## L'irritation fait le lit de l'allergie



Maçon de 48 ans,  
eczéma de contact irritatif depuis des années,  
aggravation depuis 3 mois  
→ Eczéma allergique au chrome

# Département d'Immuno-Allergologie



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**Inserm**

Institut national  
de la santé et de la recherche médicale

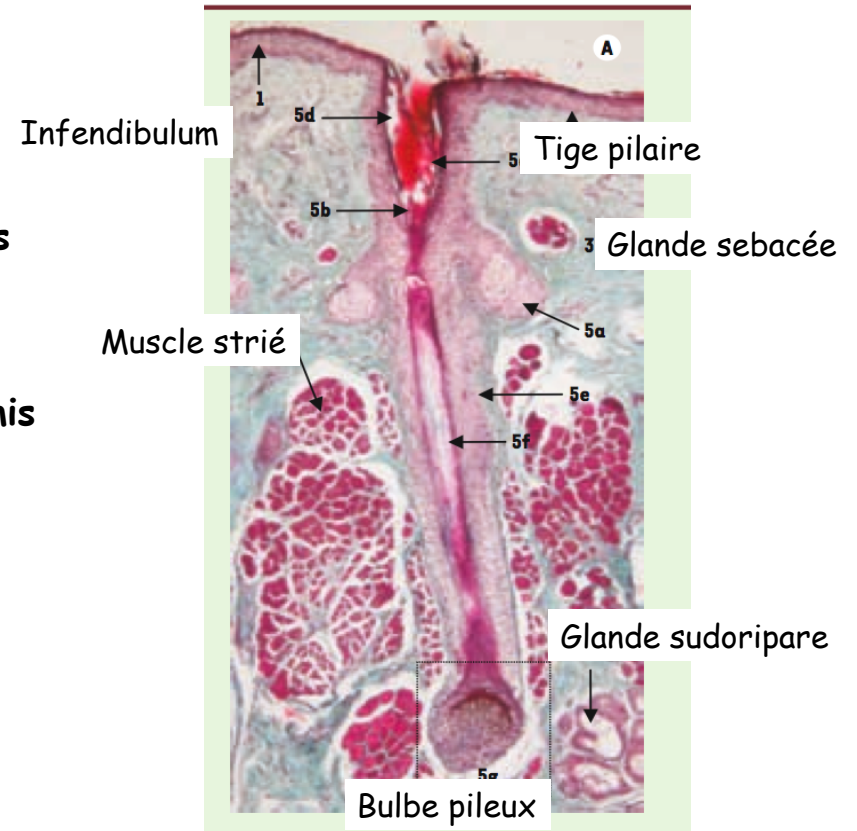
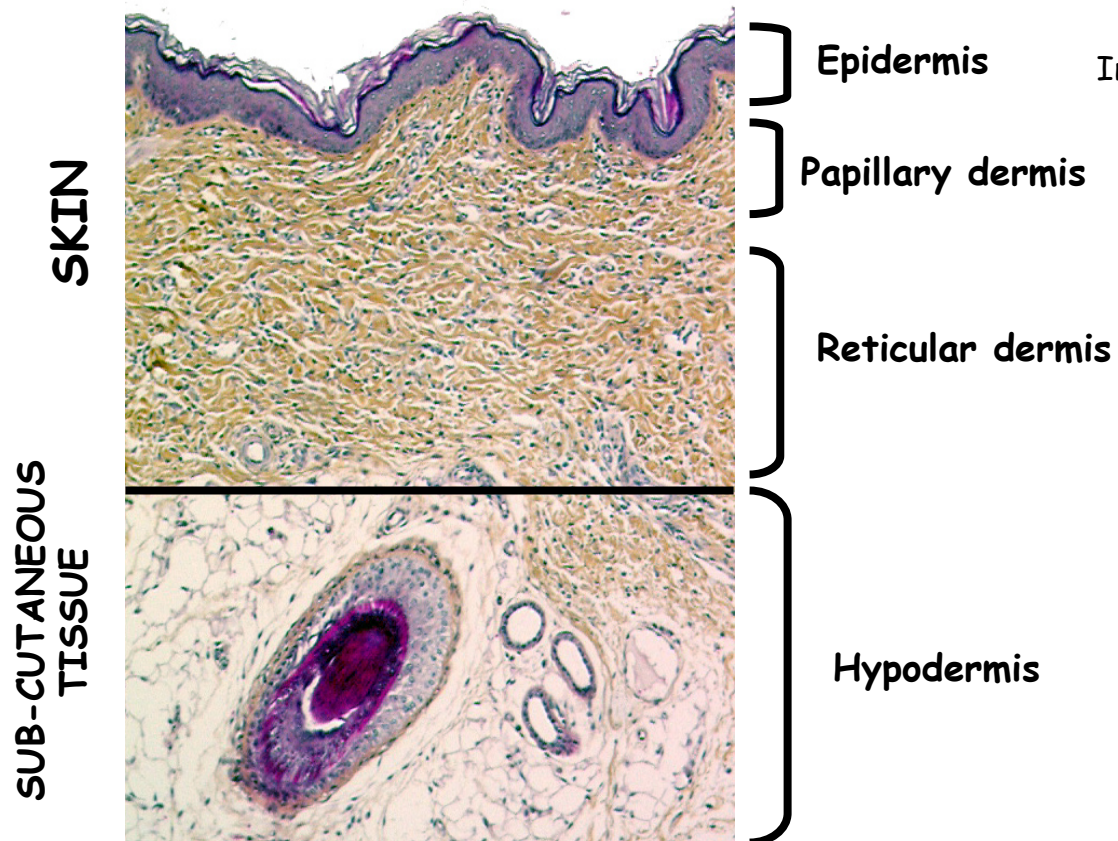


Centre  
International  
de Recherche  
en Infectiologie



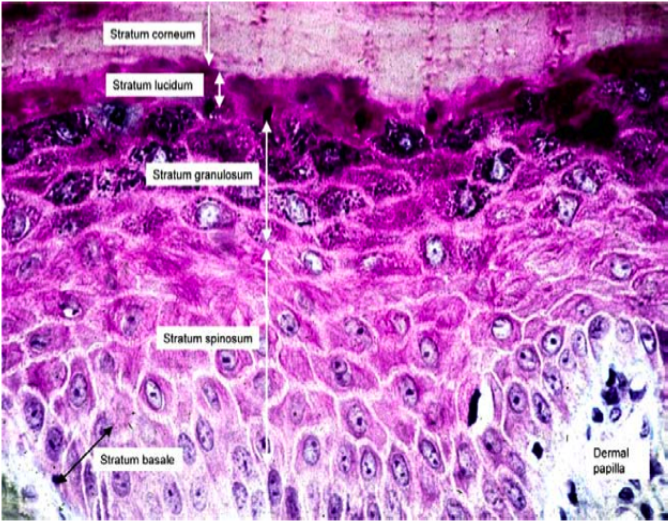
Hôpitaux de Lyon

# Anatomy of the skin

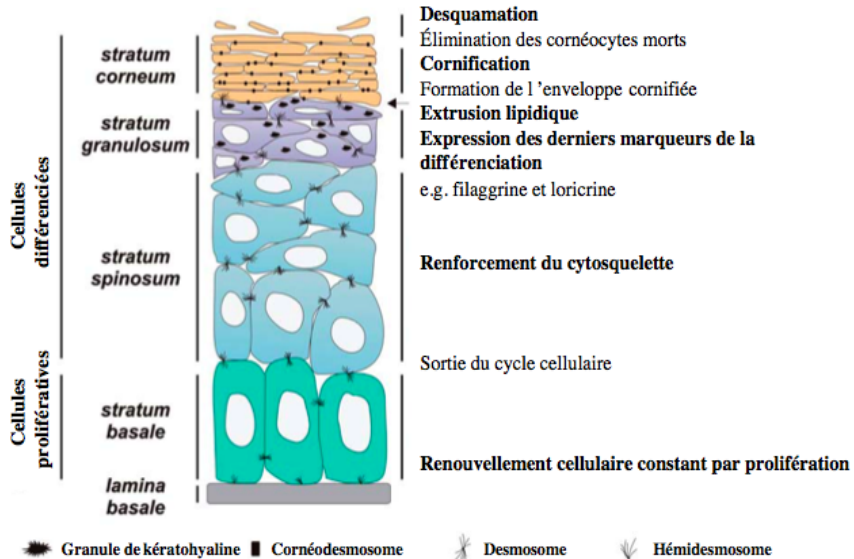


**Follicule pilo-sebacé**

# Anatomy of the epidermis



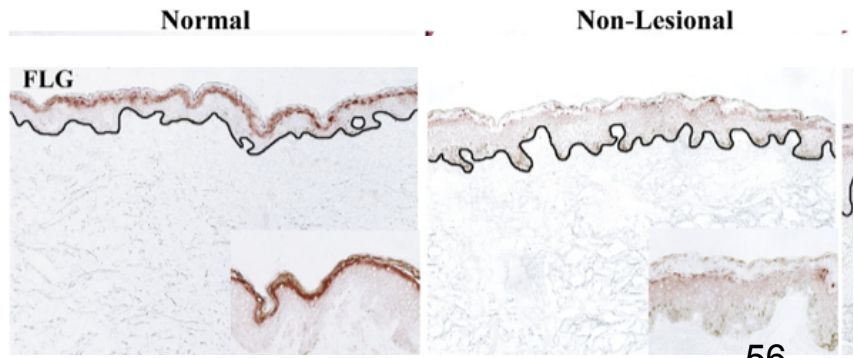
Epiderme - coloration Hematoxylline-Eosine



Différentiation épidermique - schéma



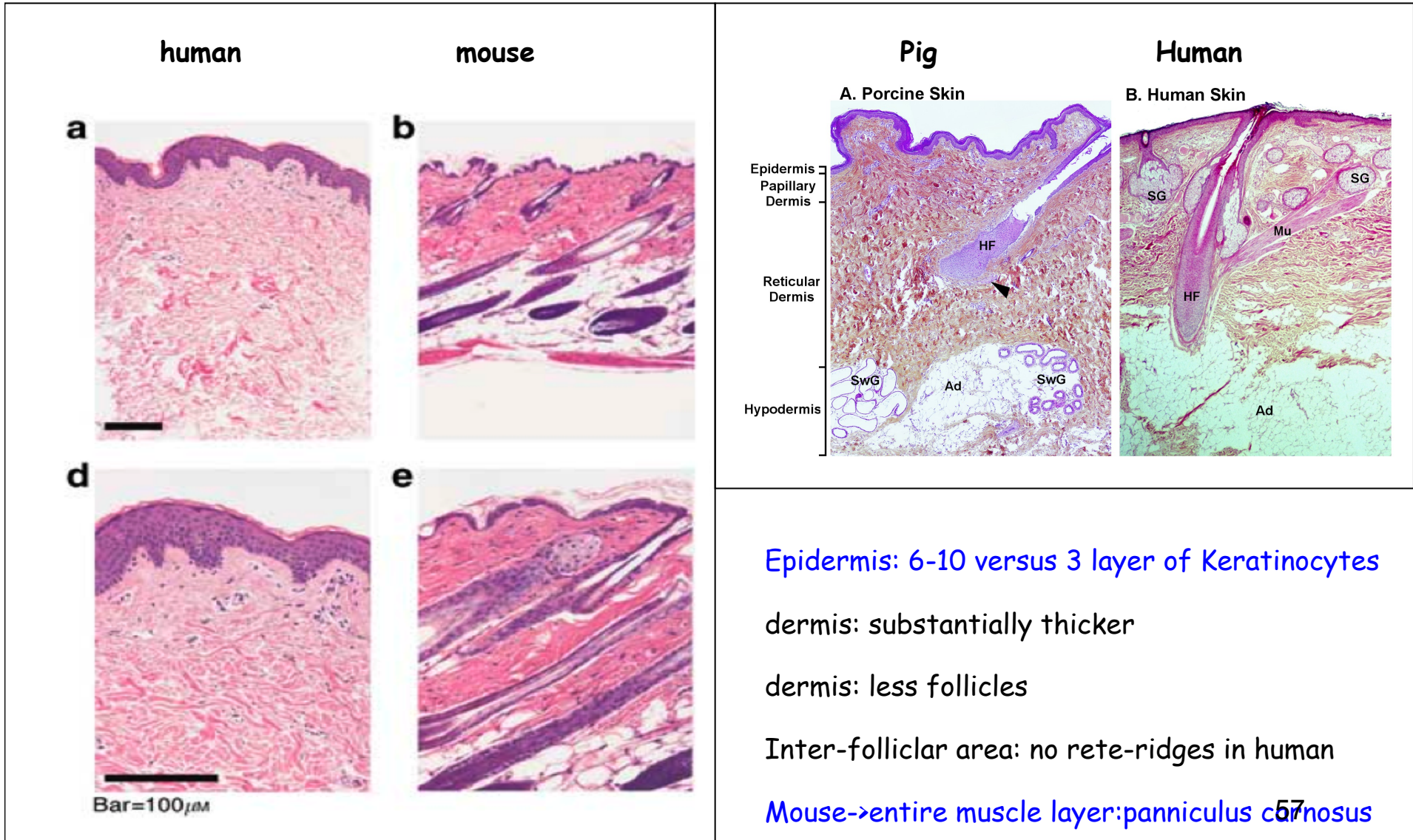
Atopic dermatitis



IHC staining of filaggrin, Suarez-Farinas et al. JACI 2010

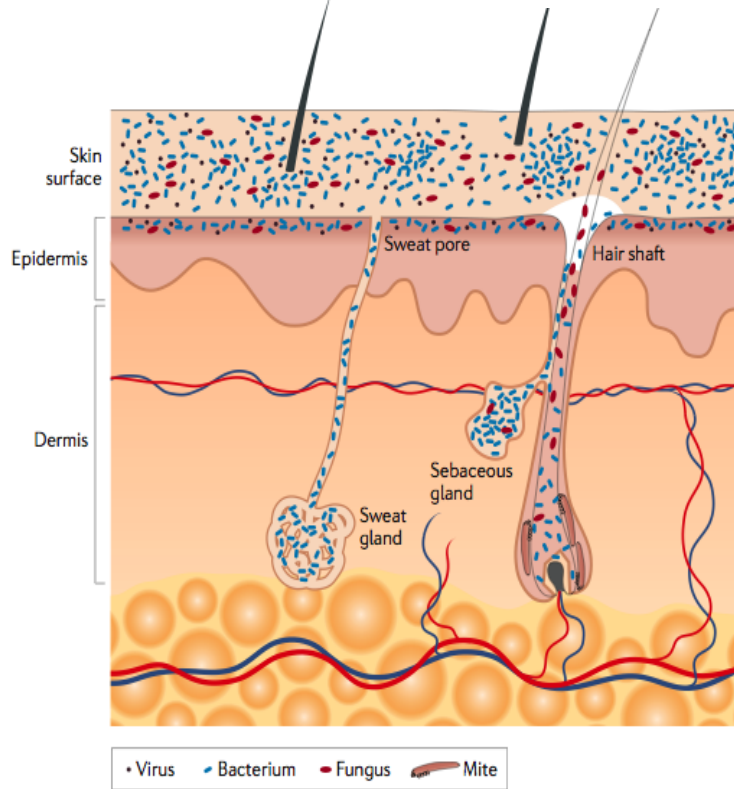


# Anatomy of the skin - Comparison human / mouse / Pig

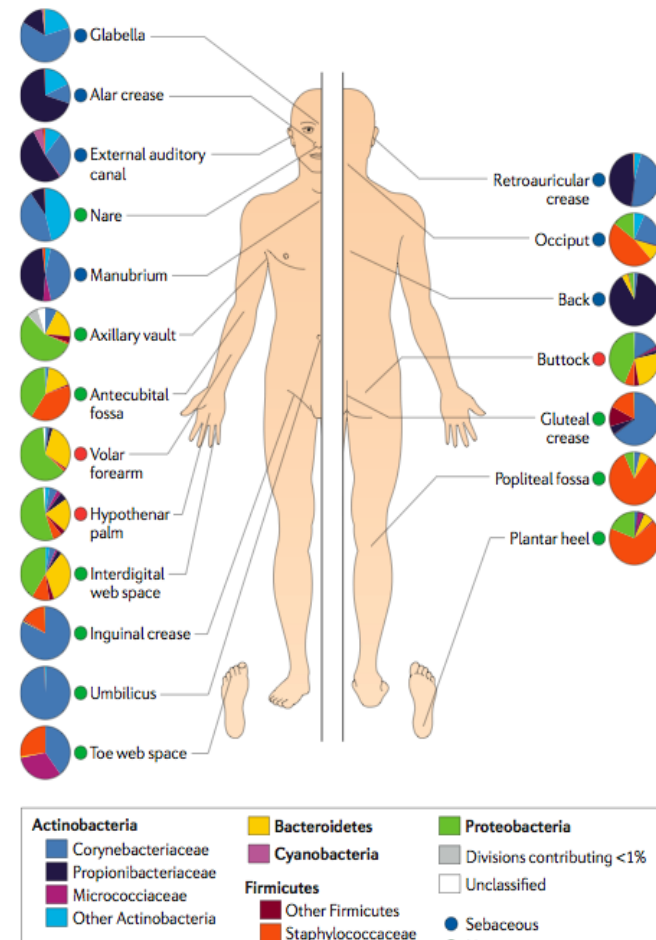


# The skin microbiome

Skin microbes and their regional localisations



Grice et al. Nat rev microb 2011



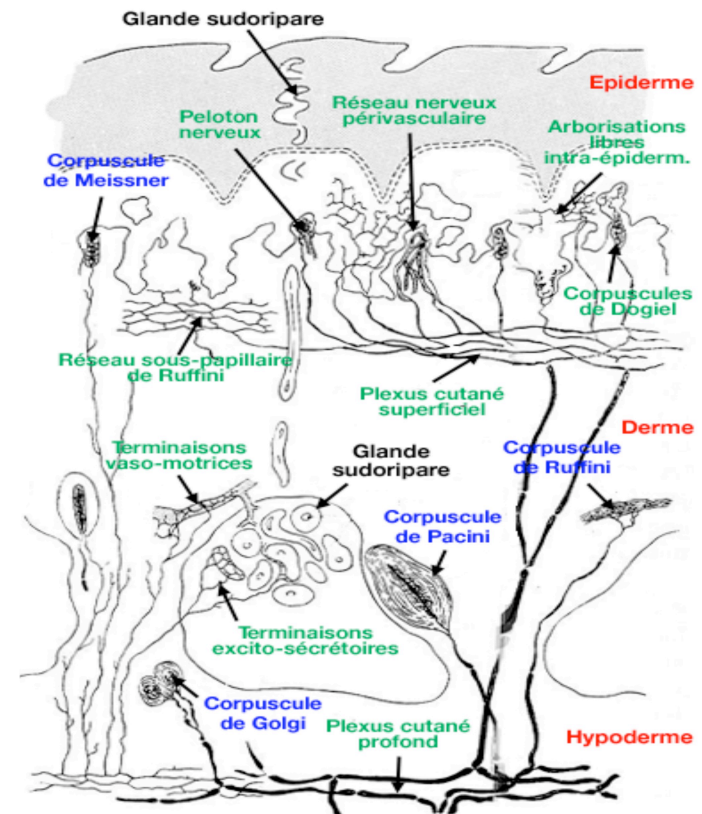
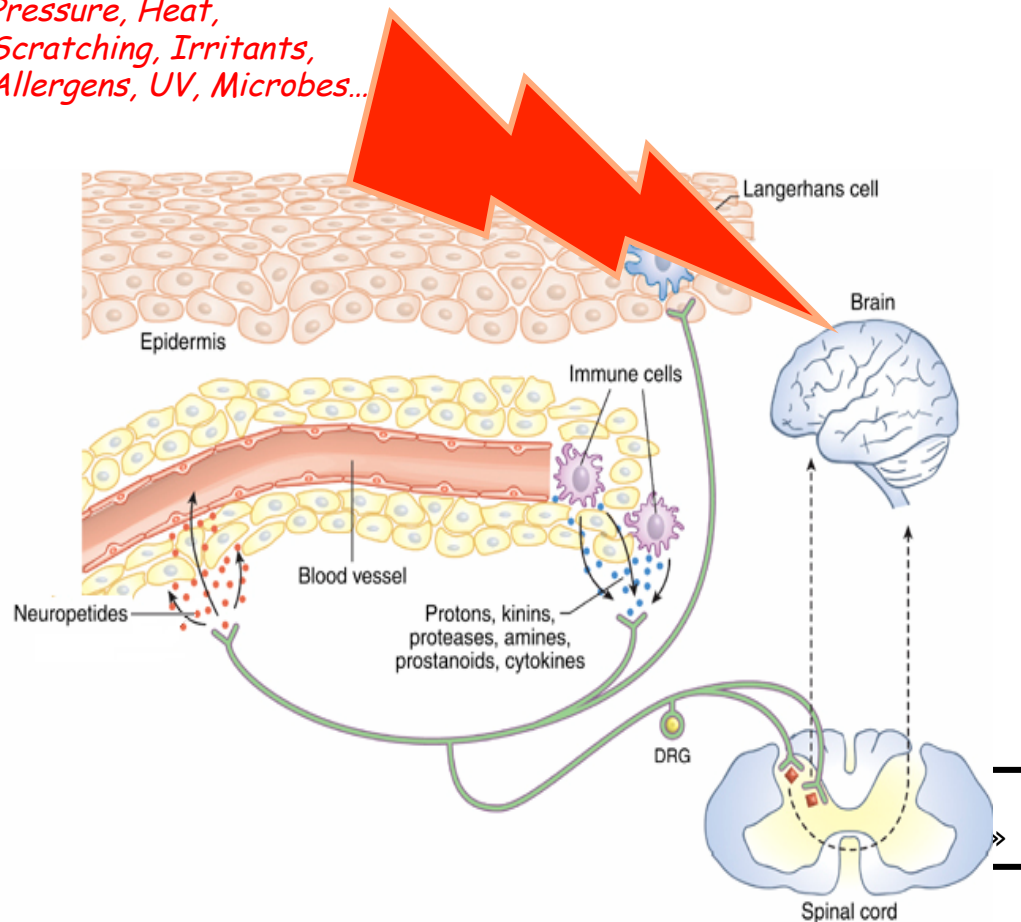
Up to  $10^{12}$  resident bacteria/m<sup>2</sup>

3 species particularly well-adapted to the acidic PH environment and host AMPs: *Staphylococcus*, *Propionibacterium*, *Corynebacterium*

# Neurogenic connection of the skin

## Pain, Pruritus, Sensorial... responses

*Pressure, Heat,  
Scratching, Irritants,  
Allergens, UV, Microbes...*



**Récepteurs simples**  
 -terminaisons nerveuses libres  
 -organes terminaux encapsulés

Mechano, thermo, chimioreceptors