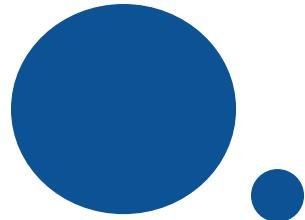




Inserm



Centre Hospitalier Universitaire Dijon

Physiopathologie de l'artérite à cellules géantes

Dr Maxime Samson et Bernard Bonnotte

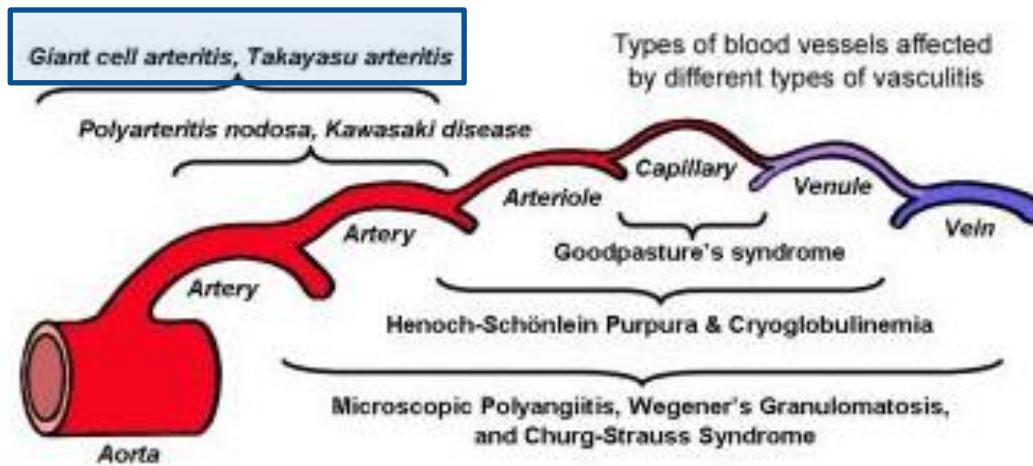
« Immunopathology, Immunoregulation » - INSERM U1098

Department of Internal medicine and clinical immunology

CHU Dijon

bernard.bonnotte@chu-dijon.fr

Artérite à cellules géantes

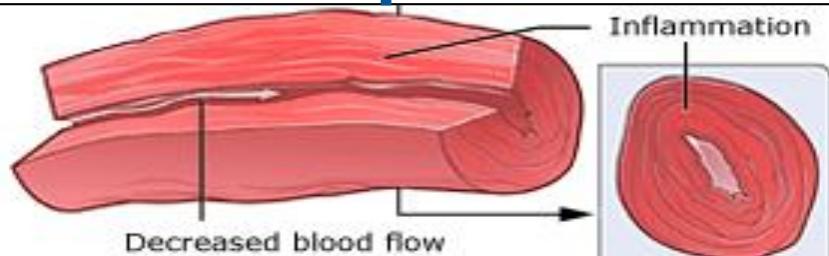


- Vascularite granulomateuse des gros vaisseaux
- Aorte et ses collatérales, branches extracrâniennes de la carotide externe, artères vertébrales
- Patients >50 ans

Manifestations cliniques

Signes systémiques

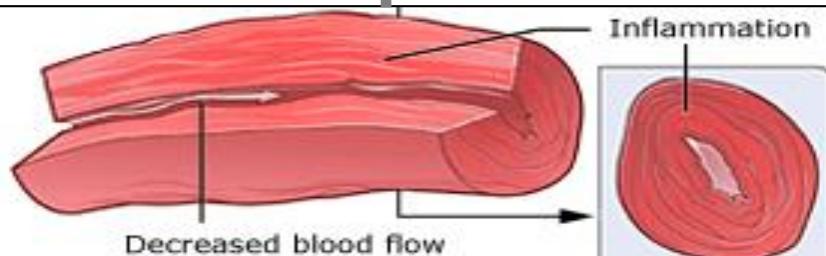
Amaigrissement, PPR, syndrome inflammatoire, fièvre, asthénie



Manifestations cliniques

Signes systémiques

Amaigrissement, PPR, syndrome inflammatoire, fièvre, asthénie

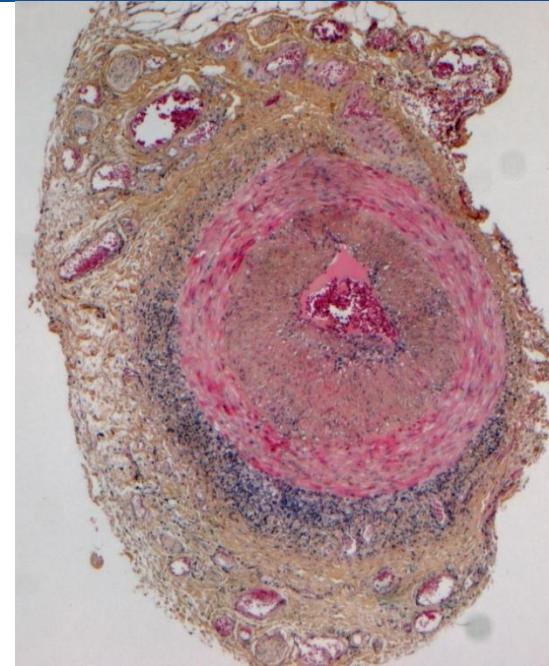


Signes ischémiques

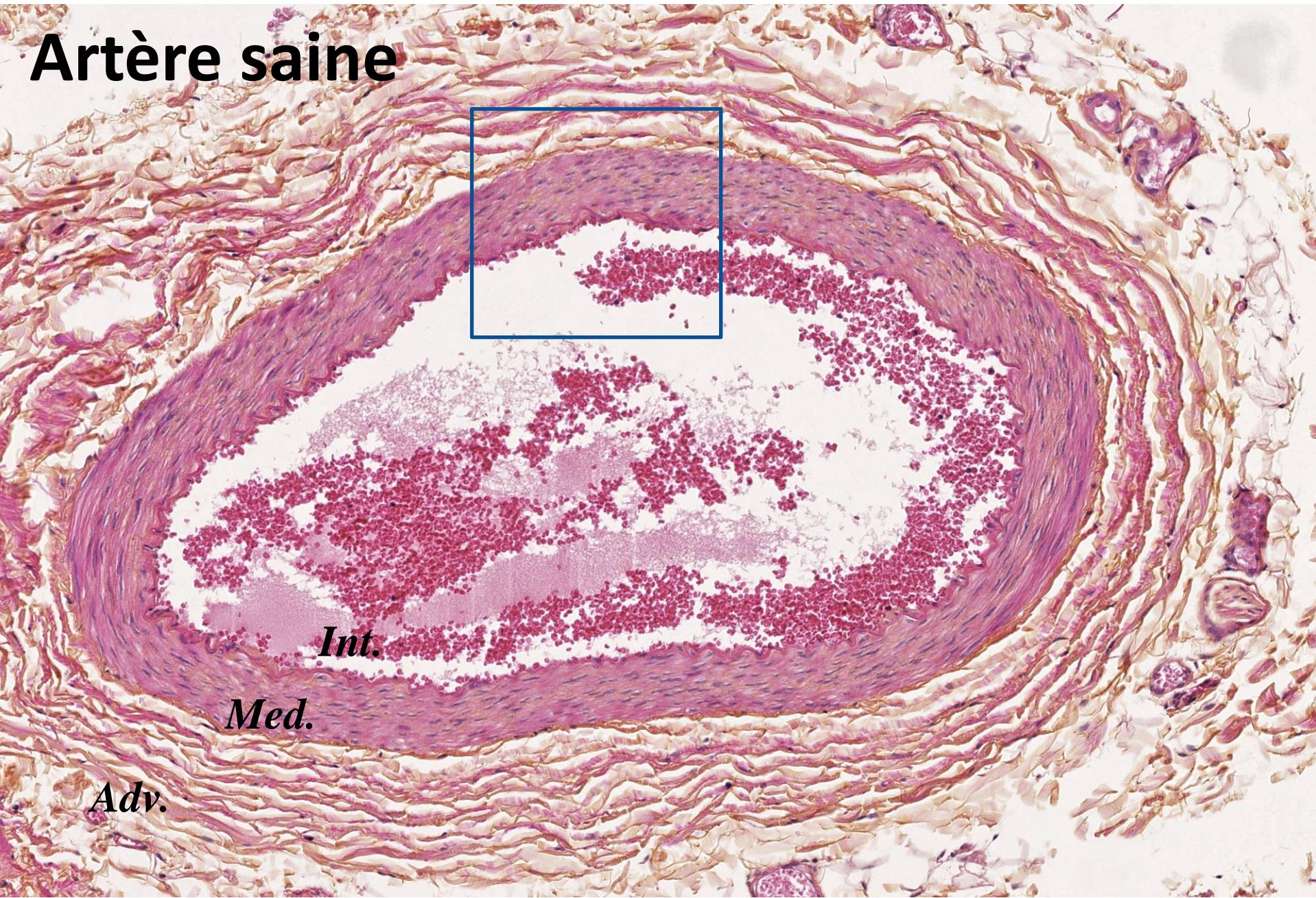
Céphalées, CM, AVC, NOIAA...

Artérite à cellules géantes

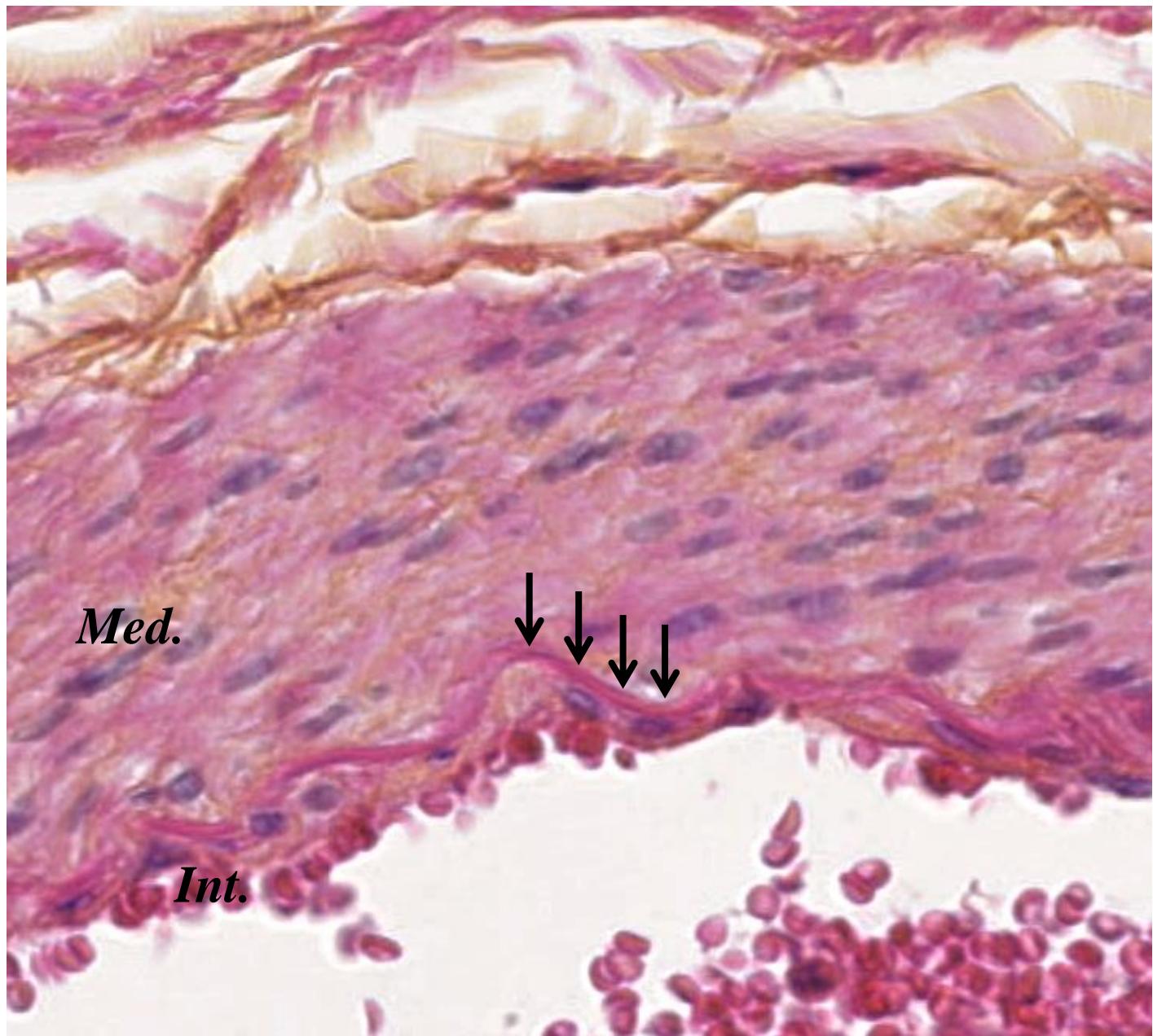
- > 50 ans
- Augmentation **CRP, VS, fibrinogène**
- « Preuve » de vascularite :
 - BAT +
 - Atteinte des gros vaisseaux en imagerie :
 - Angio-TDM, angio-IRM
 - PET-TDM
 - Echographie ?



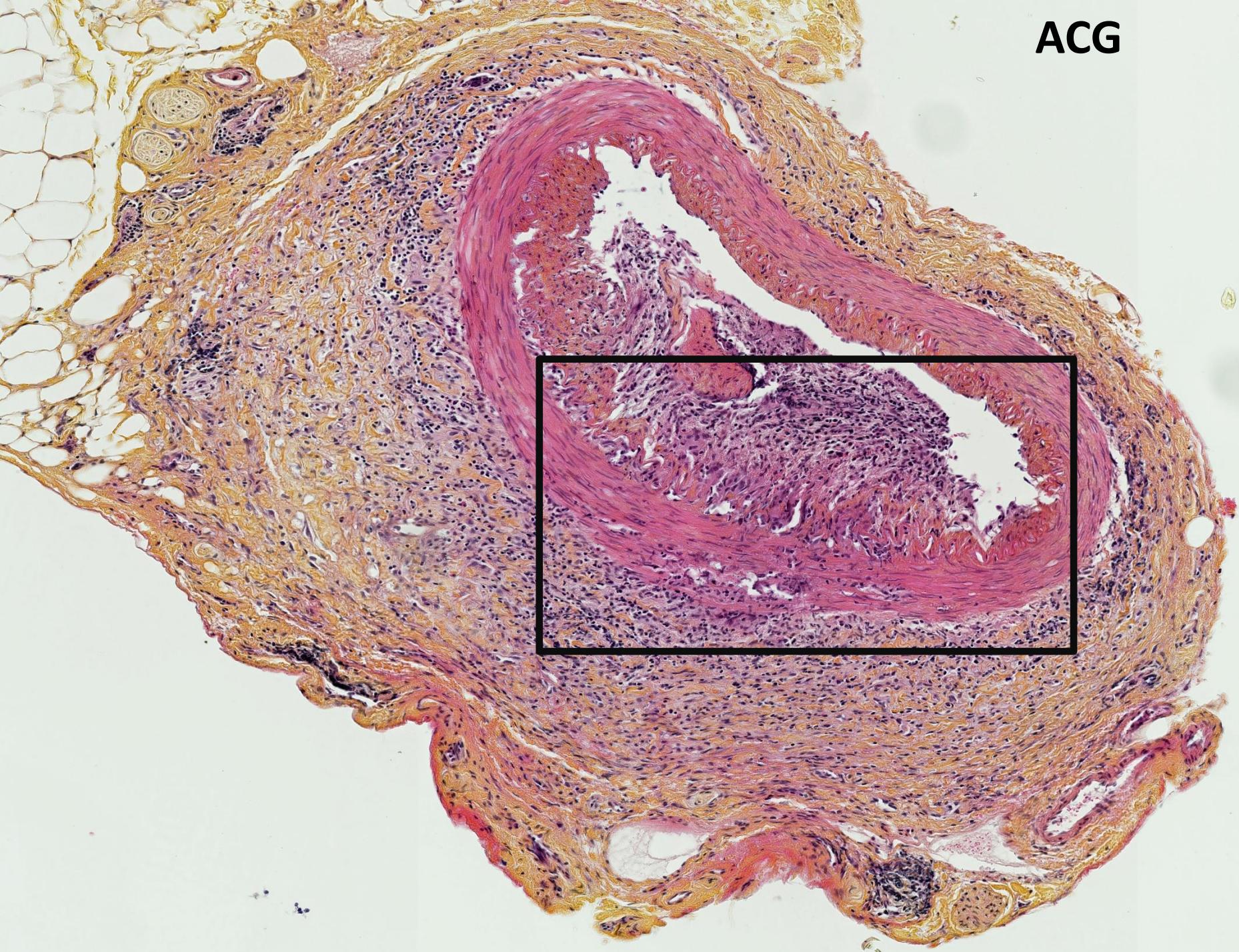
Artère saine



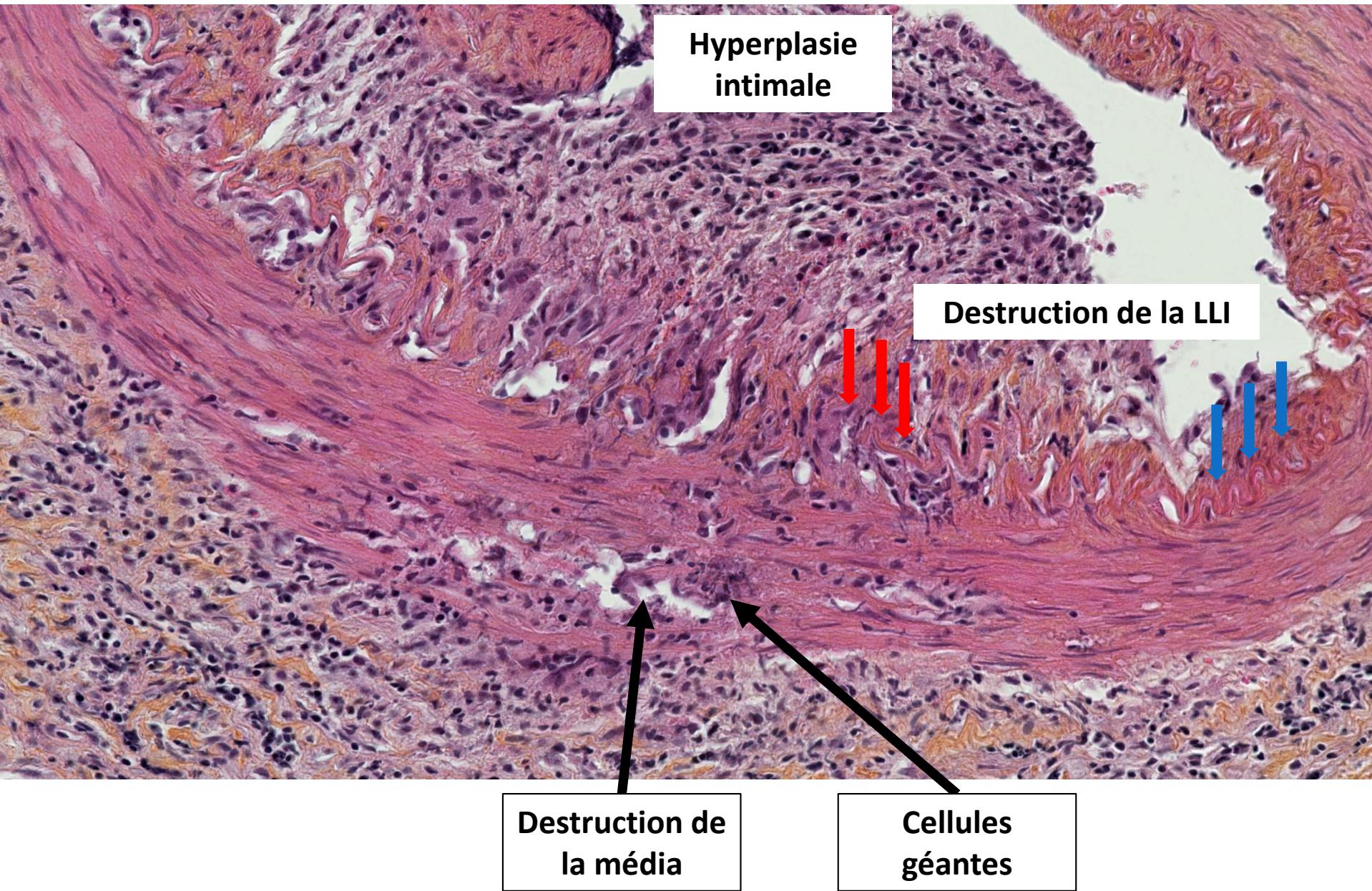
Artère saine



ACG



Infiltration par des cellules mononucléées (lymphocytes, macrophages)



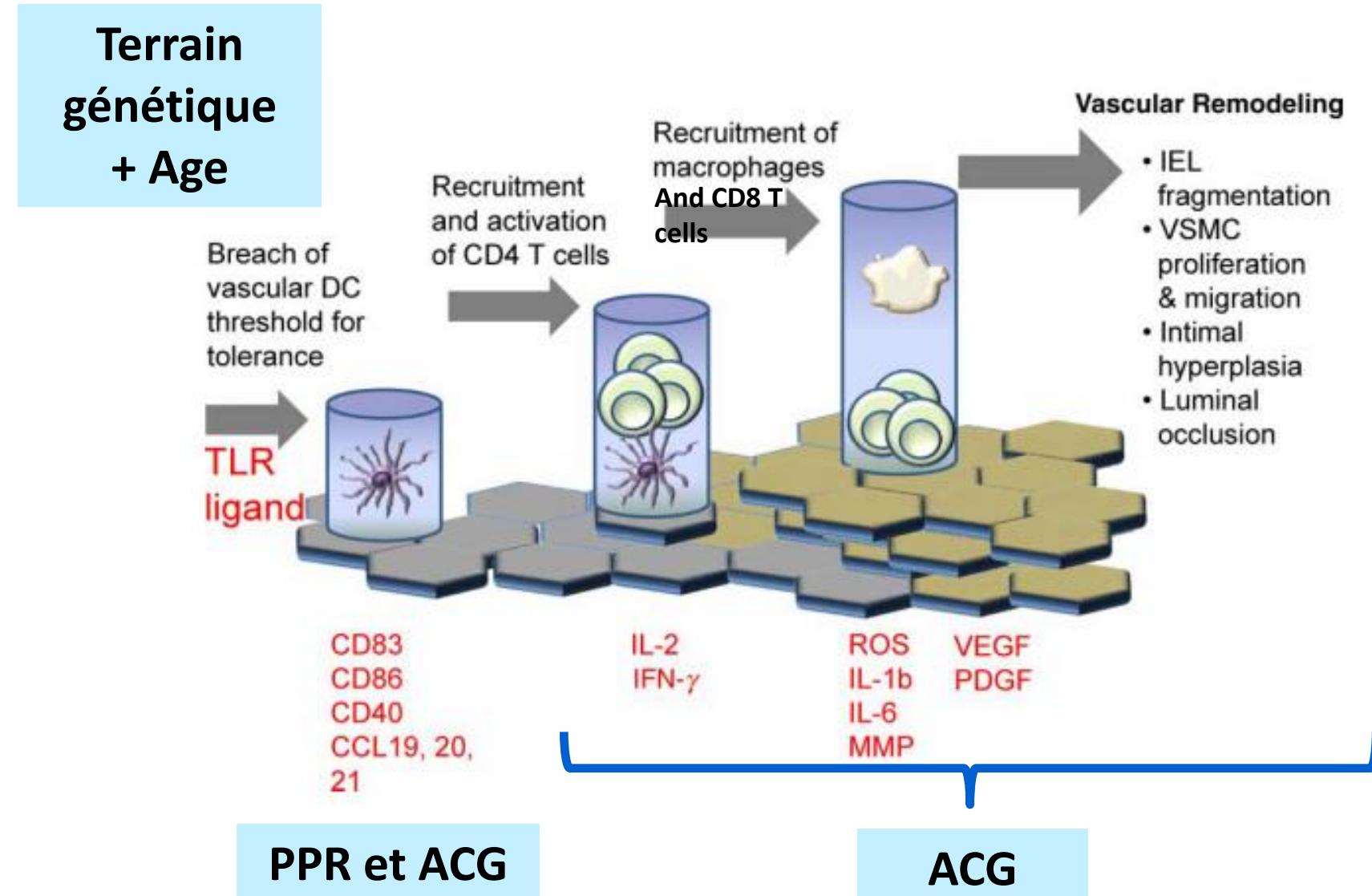


Terrain génétique

- Nombreux polymorphismes associés à un sur-risque d'ACG : *IL10, VEGF, NLRP1, IL33, TNF, CRH, ICAM, IL1RN, IL6, NOS2, eNOS, CD24, CCL2, promoter of CCL5, IL-17A, PTPN22*
 - Forte association entre ACG et HLA de classe II
 - HLA-DRB1*0401
 - HLA-DRB1*0404
 - HLA-DRB1*0408
- Confirmé par plusieurs études et un GWAS
60% des patients présentent un de ces allèles

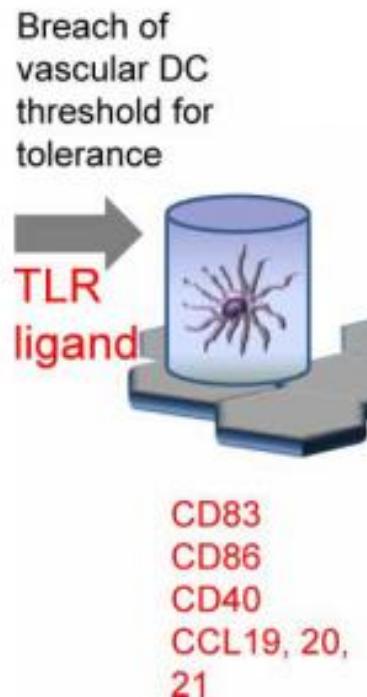
= LIEN entre ACG et immunité adaptative

Les 4 étapes du modèle physiopathologique de l'ACG

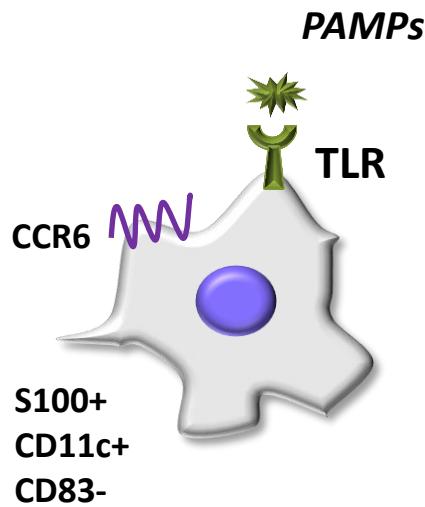


Les 4 étapes du modèle physiopathologique de l'ACG

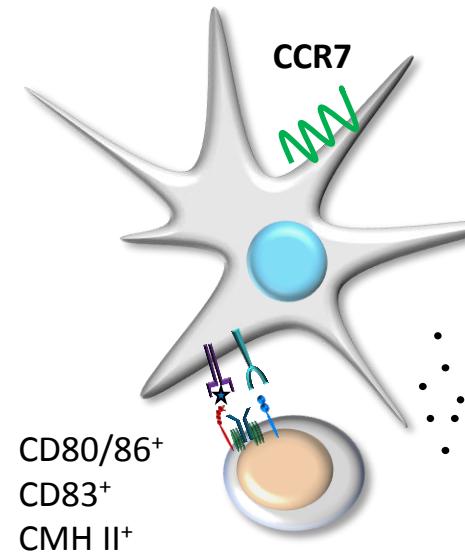
PHASE I



Cellules dendritiques de l'adventice



Artères saines
adventice



PPR et ACG
adventice et média

Les DC sont piégées
dans l'adventice au
cours de l'ACG

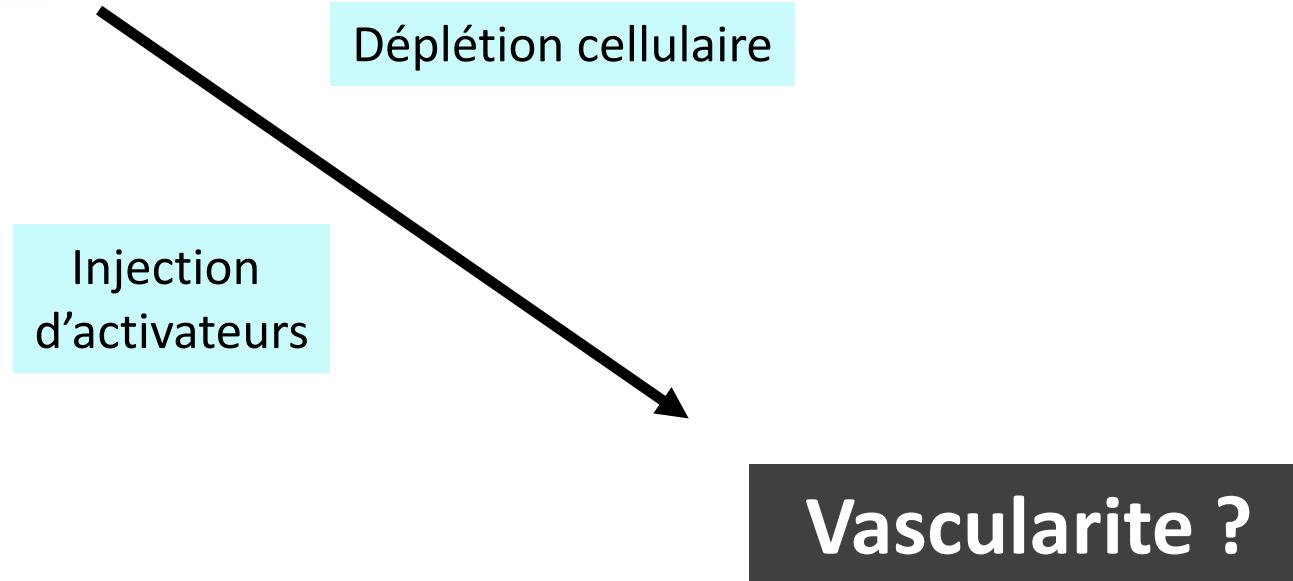
W.M Krupa et al. J Exp Med 2004
W.M Krupa et al. Am J of Pathol 2002

Souris SCID greffées avec des artères temporales humaines



Activation of Arterial Wall Dendritic Cells and Breakdown of Self-tolerance in Giant Cell Arteritis

Wei Ma-Krupa,^{1,2} Myung-Shin Jeon,^{1,2} Silvia Spoerl,^{1,2} Thomas F. Tedder,³
Jörg J. Goronzy,^{1,2} and Cornelia M. Weyand^{1,2}



Souris SCID greffées avec des artères temporales humaines



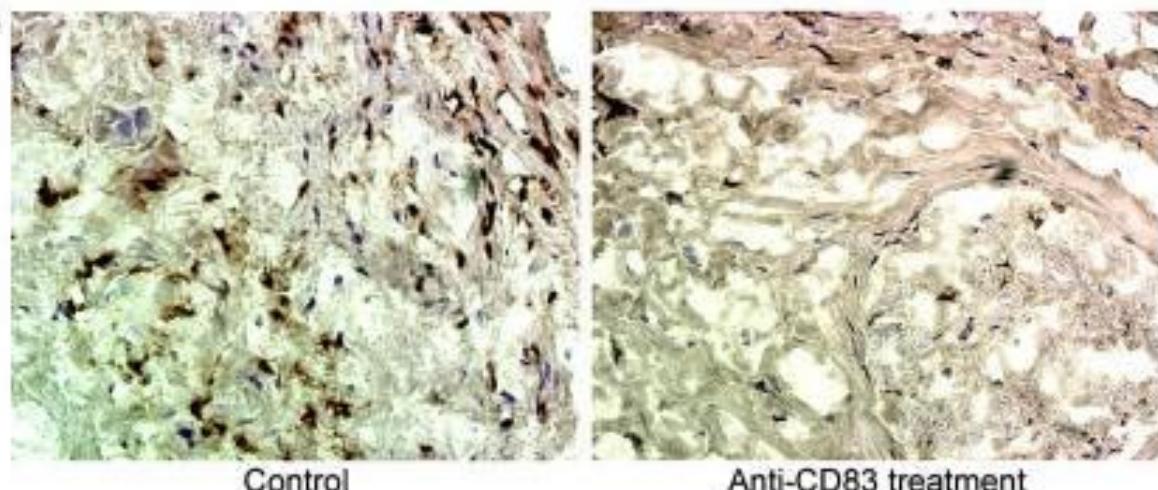
GCA

SCID

Déplétion en DC activées
(Ac anti CD83)

Diminution de la vascularité
↓ infiltration par les LT
↓ IFN- γ mRNA

A



Activation des DC avec des ligands de TLR

LPS

CFA

TNF

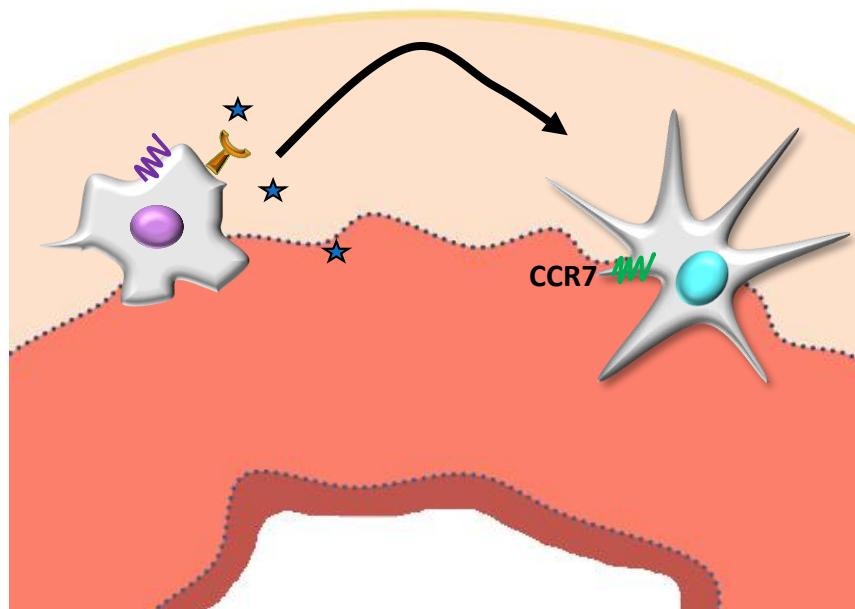


saine



SCID

Activation des DC
avec différents
activateurs (IV)



CFA → CCL19 et 21, IL-18

LPS → CCL18, 19 et 21, IL-18, CD83

TNF → CD83 et CCL21

Un agent injecté par voie sanguine peut activer les DCs

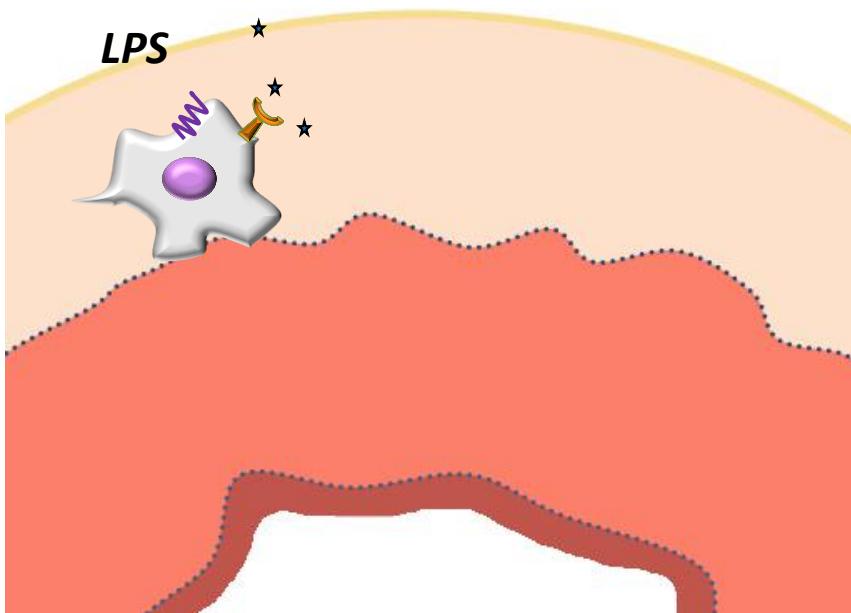
Activation des DC de l'adventice : rôle du LPS

Artère saine



SCID

+ LPS à J6



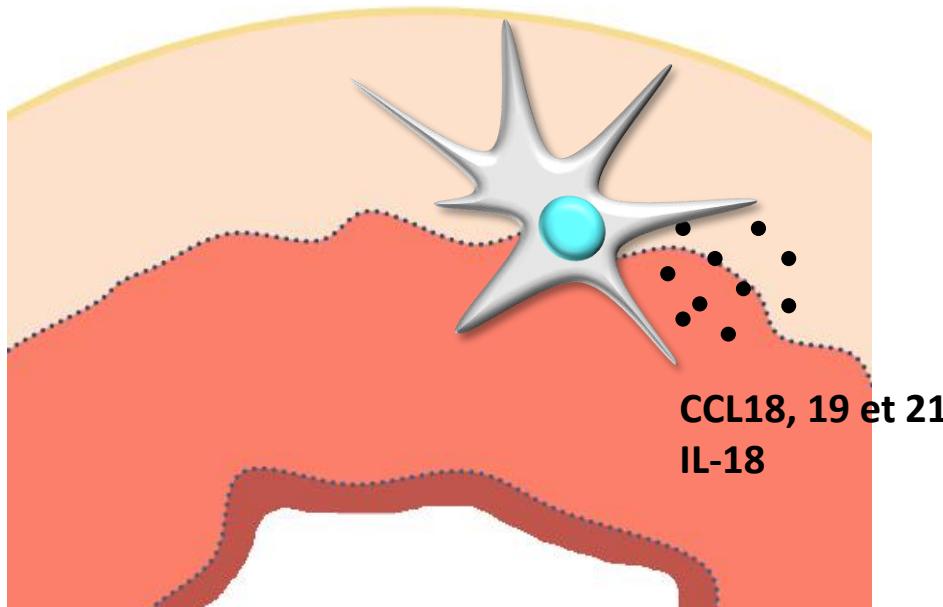
Activation des DC de l'adventice : rôle du LPS

Artère saine



SCID

+ LPS à J6



Activation des DC de l'adventice : rôle du LPS

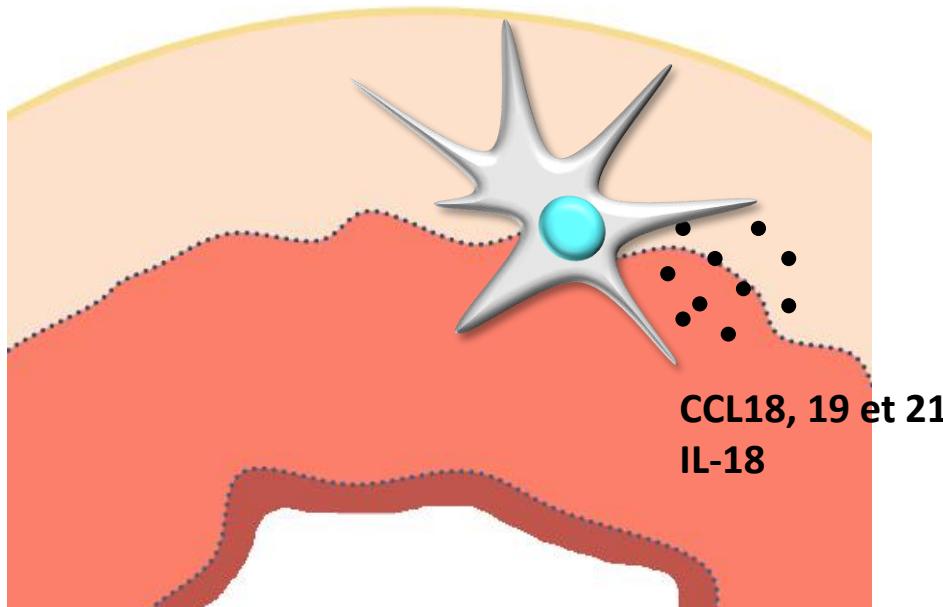
Artère saine



SCID

+ LPS à J6

+ LT CD4 humains
haplo-identiques
(HLA-DR B1*0401)
à J7



Activation des DC et recrutement des LT CD4

Artère saine

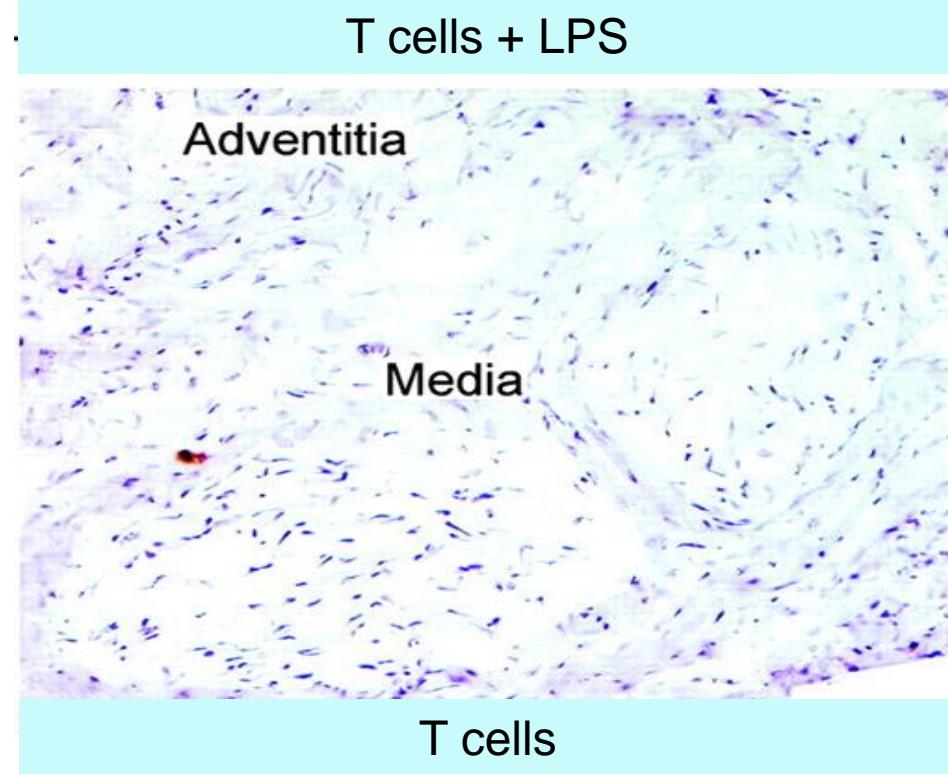
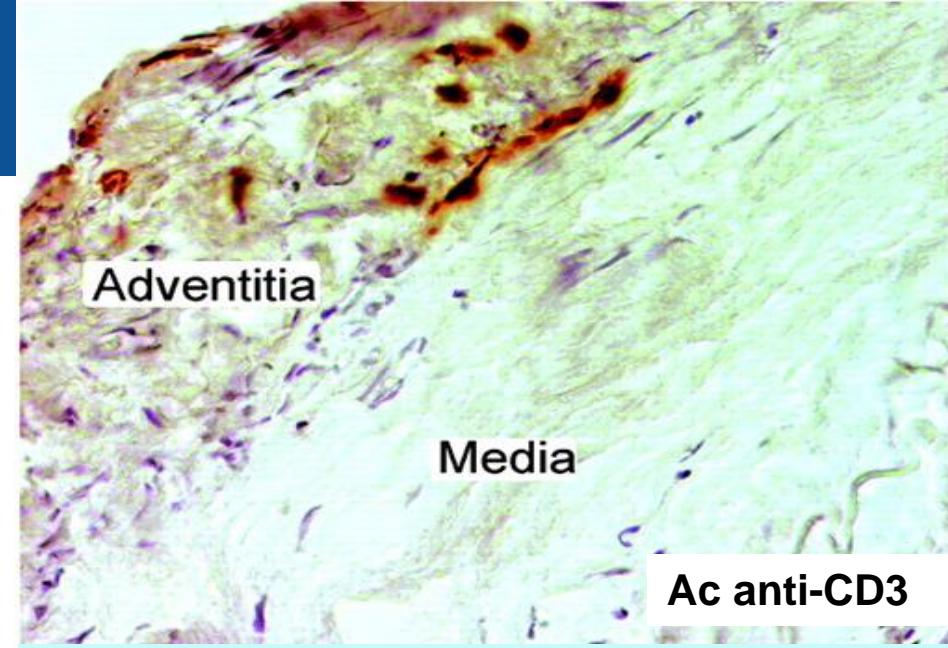


SCID

+ LPS à J6

+ LT CD4 humains
haplo-identiques
(HLA-DR B1*0401)
à J7

L'activation des DC est nécessaire au
recrutement des LT CD4⁺



Activation of Arterial Wall Dendritic Cells and Breakdown of Self-tolerance in Giant Cell Arteritis

Wei Ma-Krupa,^{1,2} Myung-Shin Jeon,^{1,2} Silvia Spoerl,^{1,2} Thomas F Tedder,³
Jörg J. Goronzy,^{1,2} and Cornelia M. Weyand^{1,2}



Saine/ACG/PPR + LT HLA-DR B1*0401



7 days

ACG et PPR : invasion de l'artère et production d'IFN-γ
Artère saine : artère saine intacte

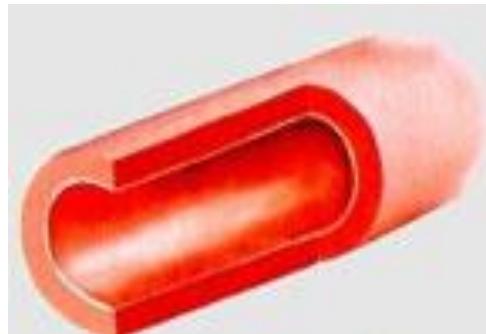
SCID

L'activation des DCs déclenche le recrutement des LT

Activation des DC : profil TLR

Activation of Arterial Wall Dendritic Cells and Breakdown of Self-tolerance in Giant Cell Arteritis

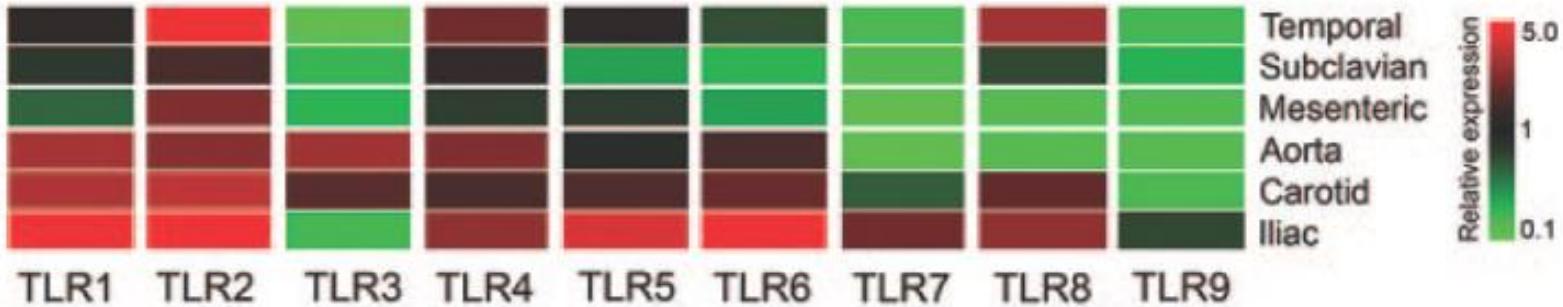
Wei Ma-Krupa,^{1,2} Myung-Shin Jeon,^{1,2} Silvia Spoerl,^{1,2} Thomas F Tedder,³
Jörg J. Goronzy,^{1,2} and Cornelia M. Weyand^{1,2}



Transcrits of **TLR2 & TLR4**

Activation des DC : profil TLR

A



Artère temporaire : TLR 2, 4 & 8

Profil proche : aorte & carotides

→ La cible des vascularites pourrait dépendre du profil TLR de chaque territoire vasculaire

Activation des DC : profil TLR

Activation of Arterial Wall Dendritic Cells and Breakdown of Self-tolerance in Giant Cell Arteritis

Wei Ma-Krupa,^{1,2} Myung-Shin Jeon,^{1,2} Silvia Spoerl,^{1,2} Thomas F Tedder,³
Jörg J. Goronzy,^{1,2} and Cornelia M. Weyand^{1,2}



Quel signal DANGER ?

Transcrits de TLR2 & TLR4

Est-ce que l'ACG est déclenchée par une infection ?

Case-control studies investigating the potential association between microbial agents and GCA.

Author, year [ref]	Number of TABs	Number of patients with biopsy-proven GCA	Methods used	Results of viral and bacterial DNA or proteins investigated								P
				HSV	VZV	EBV	CMV	HHV6	HPIV 1	PB 19	Cp	
Fest et al. [101]	174	38	Serology ISH	ND	ND	ND	+	ND	ND	ND	ND	0.03
Nordborg et al. [96]	10	10	IHC PCR	—	—	ND	ND	ND	ND	ND	ND	—
Gabriel et al. [102]	50	13	PCR	ND	ND	ND	—	ND	ND	+	ND	0.0013
Duhaut et al. [105]	305	159	Serology	—	ND	—	ND	ND	+	ND	ND	0.00005
Wagner et al. [39]	27	9 (14)*	IHC PCR	ND	ND	ND	ND	ND	ND	ND	+	ND
Haugeberg et al. [107]	20	12	Nested PCR	ND	ND	ND	ND	ND	ND	ND	—	—
Salvarani et al. [169]	83	31	Nested PCR	ND	ND	ND	ND	ND	ND	—	ND	—
Regan et al. [108]	180	79	PCR	ND	ND	ND	ND	ND	ND	ND	—	—
Helweg-Larsen et al. [97]	30	13	PCR	—	—	—	—	—	ND	—	—	—
Kennedy et al. [170]	22	15	ISH ISPCR	ND	—	ND	ND	ND	ND	ND	ND	—
Rodriguez-Pla et al. [98]	147	50	IHC PCR	—	—	—	—	—	ND	—	ND	—
Powers et al. [104]	39	24	HS PCR	+	ND	ND	ND	ND	ND	ND	ND	0.027
Alvarez et al. [99]	113	57	QPCR	ND	—	ND	ND	—	ND	+	ND	0.007
Cankovic et al. [103]	41	35	QPCR	—	ND	—	—	ND	ND	ND	ND	—
Cooper et al. [100]	104	37	PCR	—	—	—	—	—	ND	—	—	—
Njau et al. [106]	224	115	Nested PCR TETR-PCR Nested PCR	ND	ND	ND	ND	ND	ND	ND	—	—

*:number of TAB in patients with GCA. CP: *Chlamydia pneumoniae*; CMV: cytomegalovirus; EBV: Epstein-Barr virus; HHV6: human herpes virus 6; HPIV 1: human parainfluenzae 1 virus; HPV19: human papillomavirus B19; HSV: herpes simplex virus; ISH: *in situ* hybridization; IHC: immunohistochemical analysis; ND: not done; PCR: polymerase chain reaction, QPCR: quantitative real-time PCR, TETR-PCR: touchdown enzyme time-release PCR; TAB: temporal artery biopsy; VZV: varicella zoster virus.

—: no statistically significant difference between patients with biopsy-proven GCA and controls.

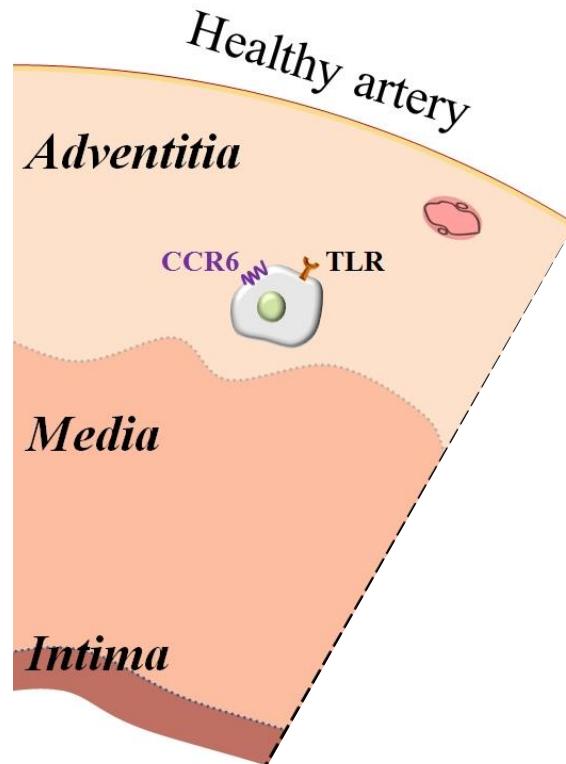
+: statistically significant difference between patients with biopsy-proven GCA and controls.

K-H. Ly et al. Autoimmunity Reviews 2010

Et VZV ???

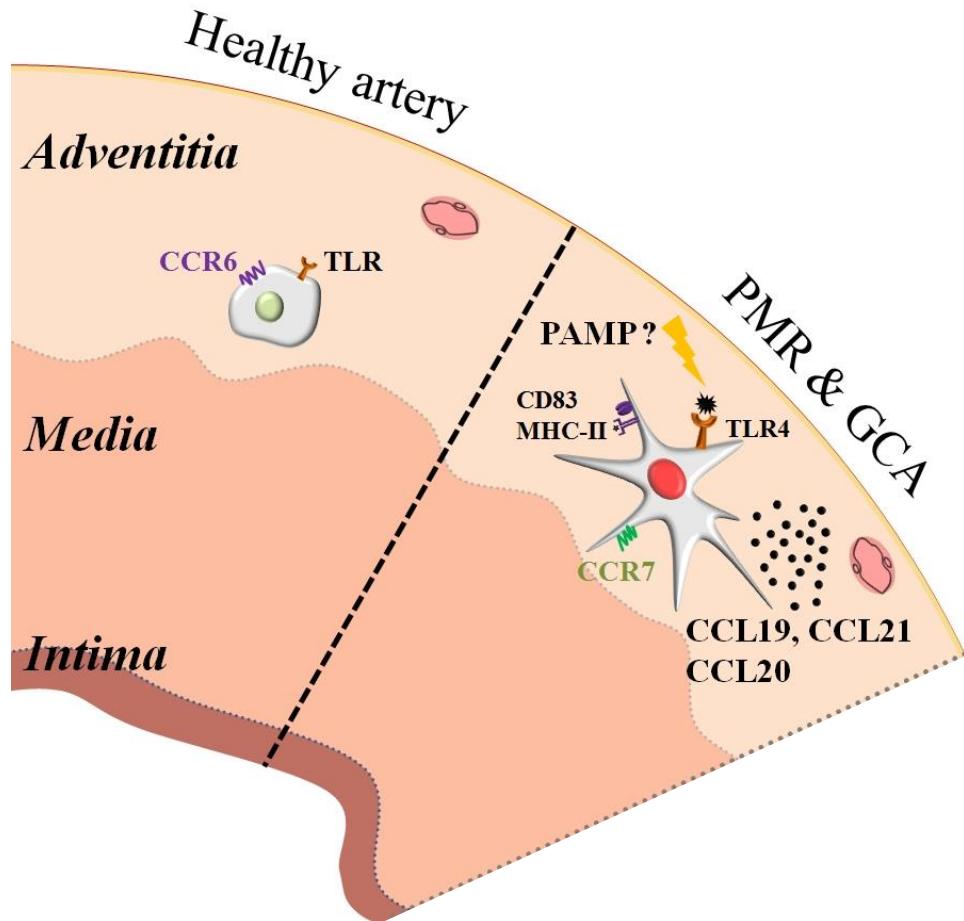
Nagel MA et al. JAMA Neurol 2015

PHASE 1



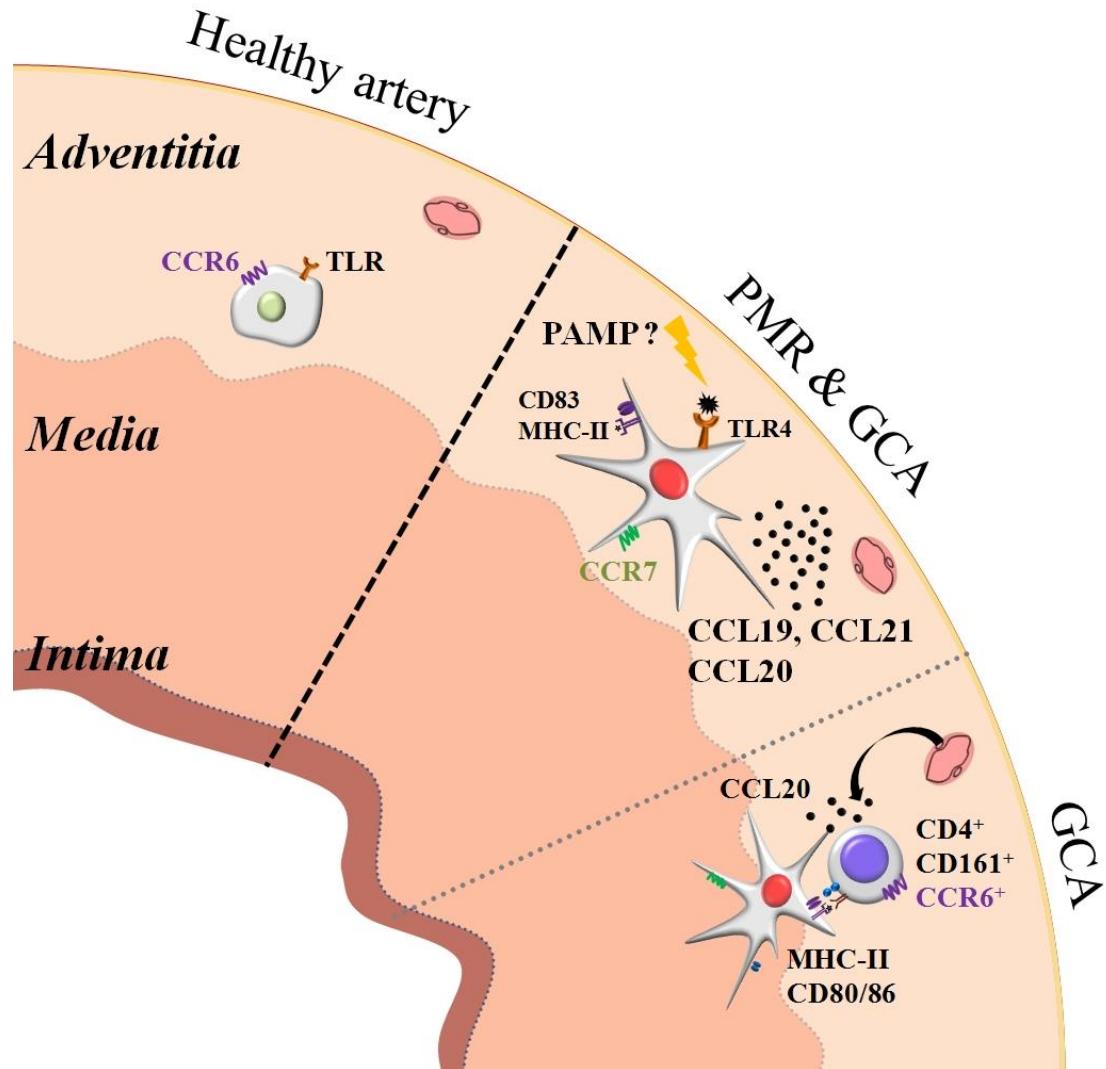
Krupa et al. *J Exp Med* 2000
Krupa et al. *Am J of Pathol* 2002

PHASE 1



Krupa et al. *J Exp Med* 2000
Krupa et al. *Am J of Pathol* 2002

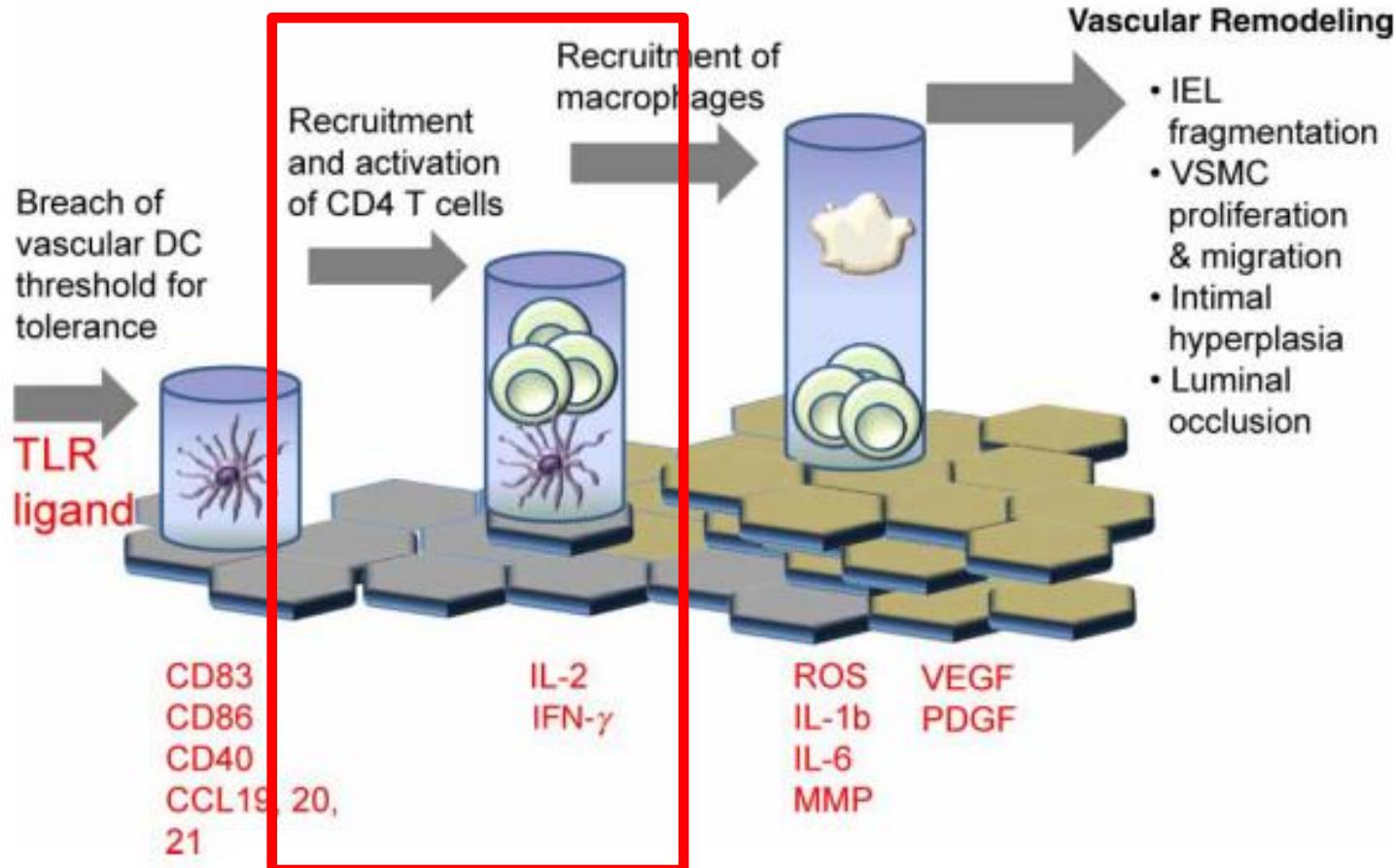
PHASE 1



Krupa et al. J Exp Med 2000

Krupa et al. Am J of Pathol 2002

Phase 2 : recrutement, activation et polarisation des LT CD4⁺



Souris SCID greffées avec des artères temporales humaines



GCA

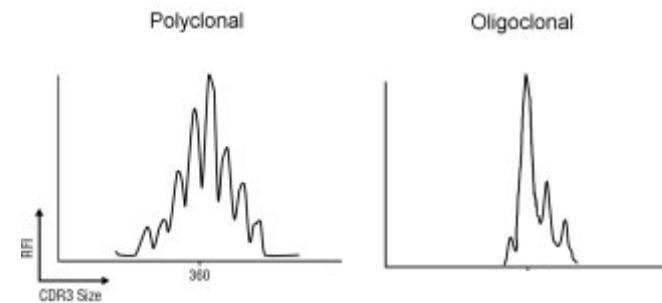
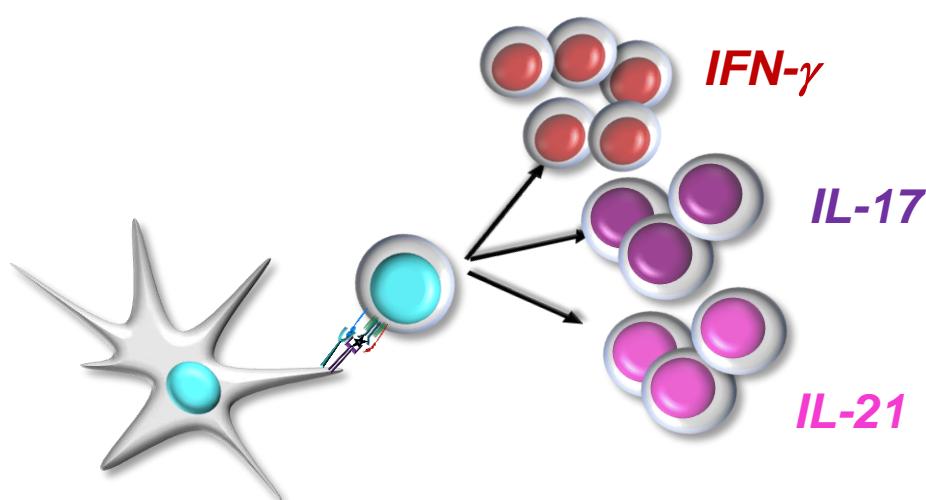
SCID

Déplétion en LT CD4⁺

Diminution de la vascularite
↓ IFN-γ mRNA

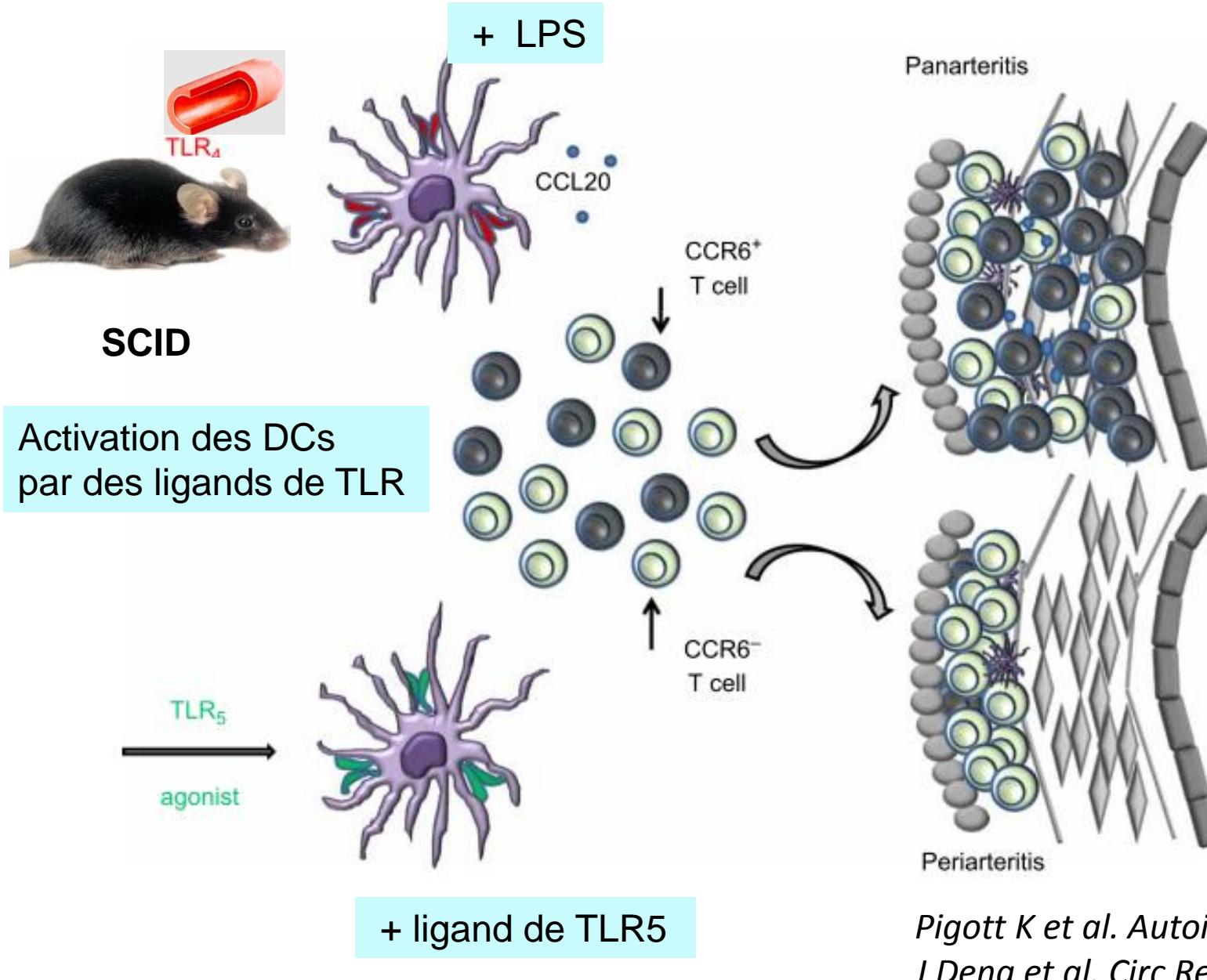
Phase 2: recrutement, activation et polarisation des LT CD4⁺

- Les LT CD4 sont **recrutés** dans la paroi vasculaire par les chémokines produites par les DC activées
- Les LT CD4 **prolifèrent** *in situ*
- **Même clones** entre artère temporale D et G
- Polarisation en lymphocytes **Th1** (+++), **Th17** et **Th21**
- Persistance des lymphocytes Th1 malgré la corticothérapie
- **Fort infiltrat en LT PD1⁺** et **déficit en PD-L1** sur les DCs artérielles



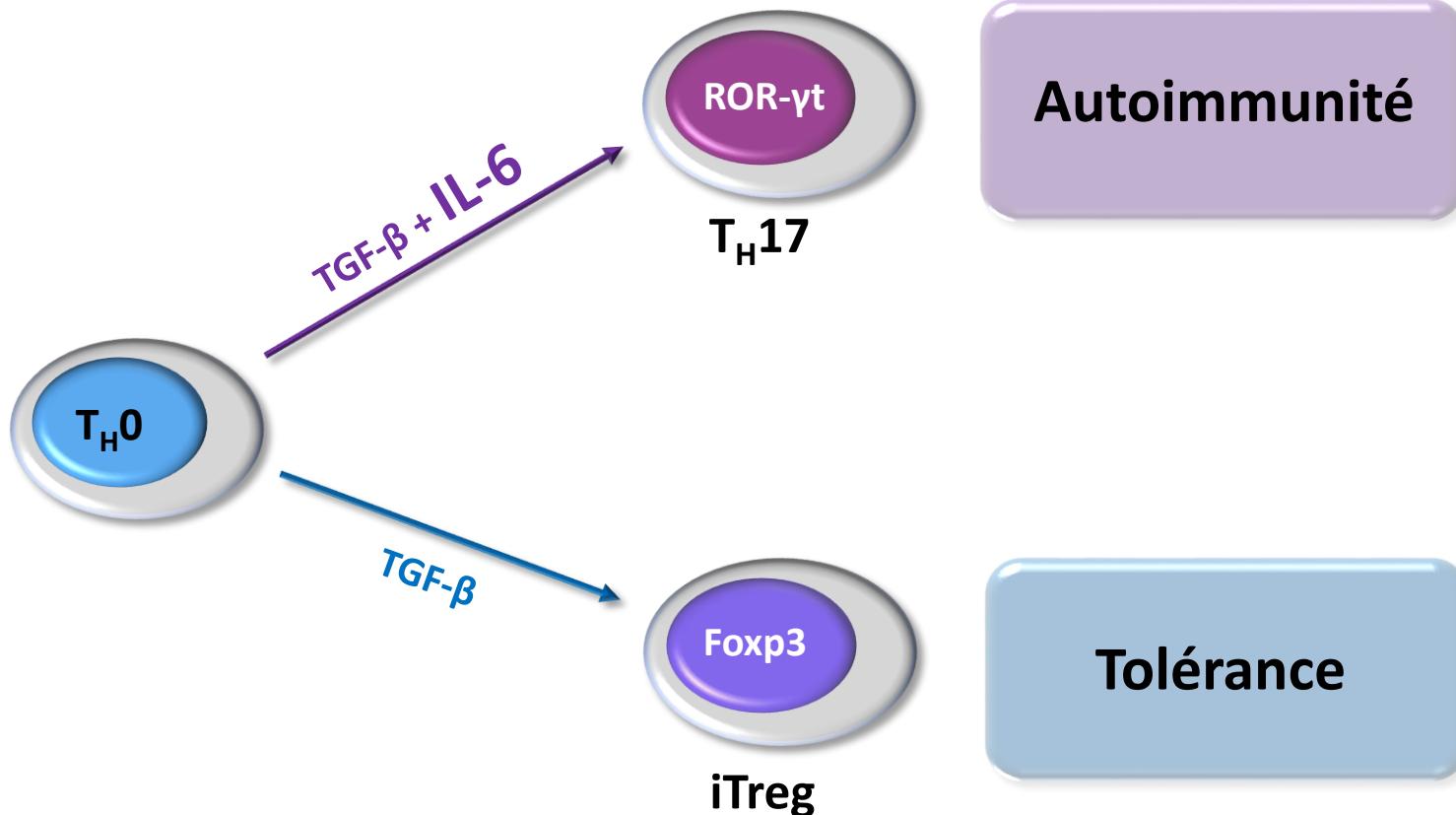
Deng J et al. Circulation 2010
Brack A et al. Mol Med 1997
Weyand C et al. J Exp Med 1994
Grunewald J et al. Arthritis Rheum 1994
Terrier et al. Arthritis Rheum 2011
Samson et al. Arthritis Rheum 2012
Zhang H et al. PNAS 2017

Le recrutement des LT CD4 dépend du signal danger



*Pigott K et al. Autoimmunity 2009
J Deng et al. Circ Res. 2009*

Polarisation des LT CD4+ : Balance Th17/Treg

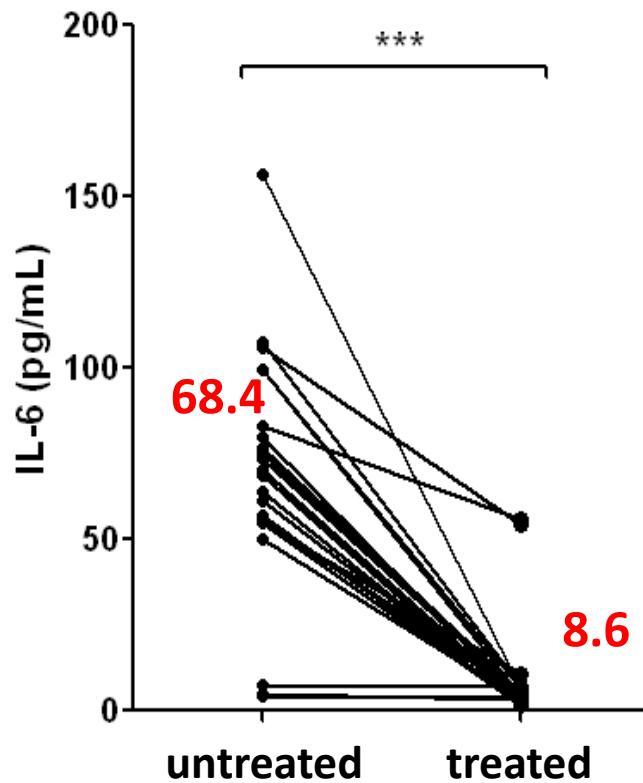
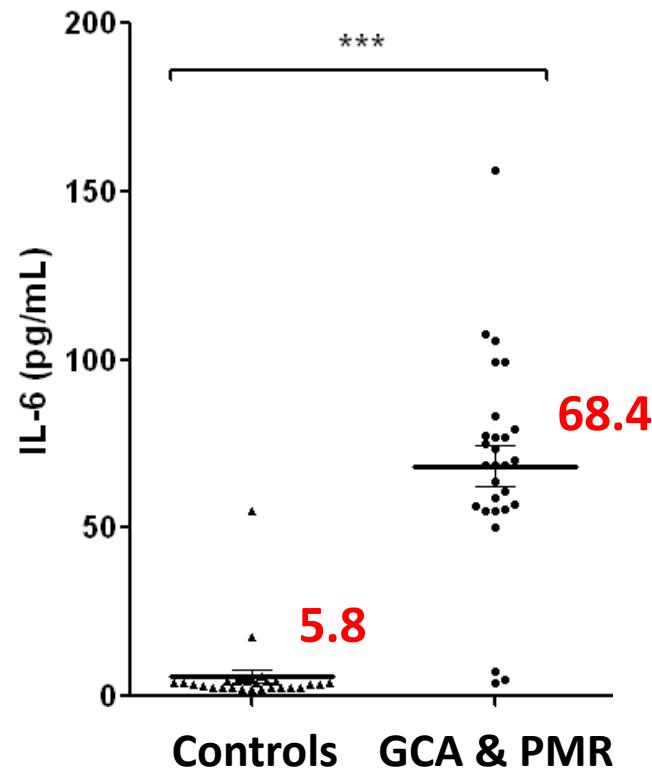


Th1 and Th17 Lymphocytes Expressing CD161 Are Implicated in Giant Cell Arteritis and Polymyalgia Rheumatica Pathogenesis

Maxime Samson,¹ Sylvain Audia,¹ Jennifer Fraszczak,² Malika Trad,² Paul Ornetti,³ Daniela Lakomy,³ Marion Ciudad,² Vanessa Leguy,³ Sabine Berthier,³ Julien Vinit,³ Patrick Manckoundia,³ Jean-Francis Maillefert,³ Jean-François Besancenot,³ Serge Aho-Glele,³ Nils Olivier Olsson,³ Bernard Lorcerie,³ Loïc Guillevin,⁴ Luc Mouthon,⁴ Philippe Saas,⁵ Andrew Bateman,⁶ Laurent Martin,³ Nona Janikashvili,² Nicolas Larmonier,⁷ and Bernard Bonnotte¹

Arthritis and Rheumatism 2012

IL-6 et GCA



La concentration
sérique en IL-6 est
corrélée à l'activité
de la maladie



Analyses sur le sang

34 patients non traités

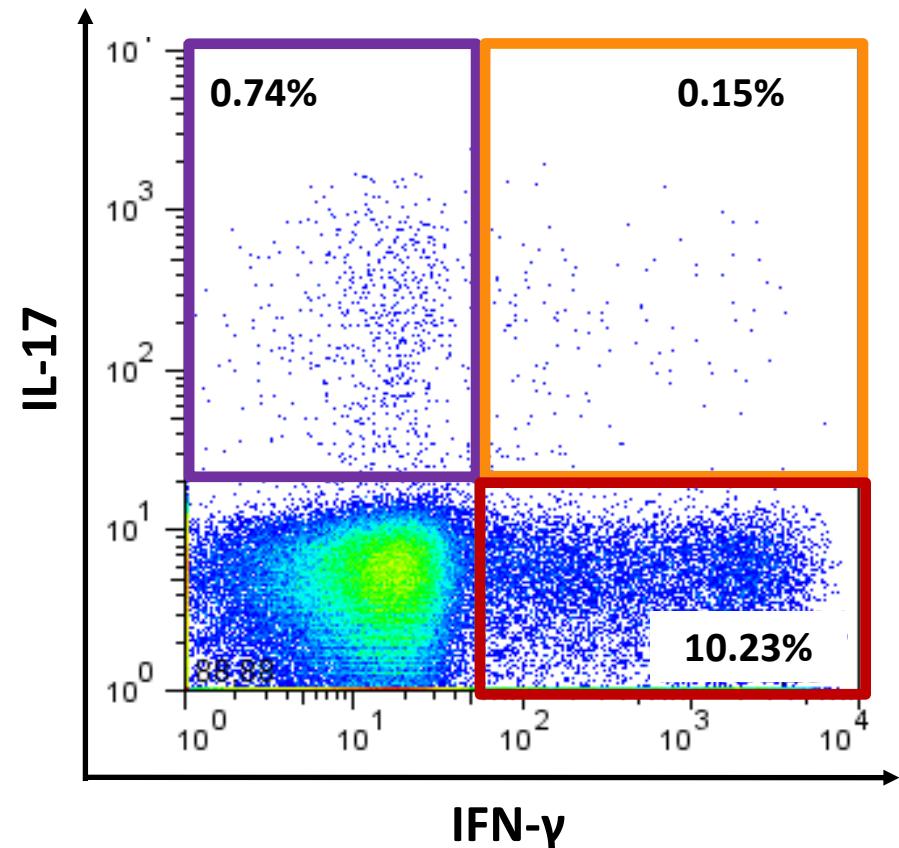
- ACG=22
- PPR=12

27 patients traités

31 volontaires sains

Cytométrie en flux

Th1 et Th17





Analyses sur le sang

34 patients non traités

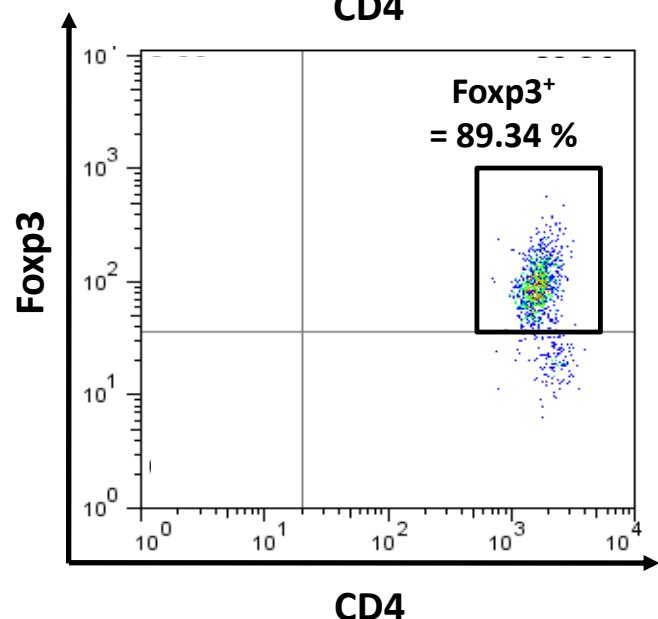
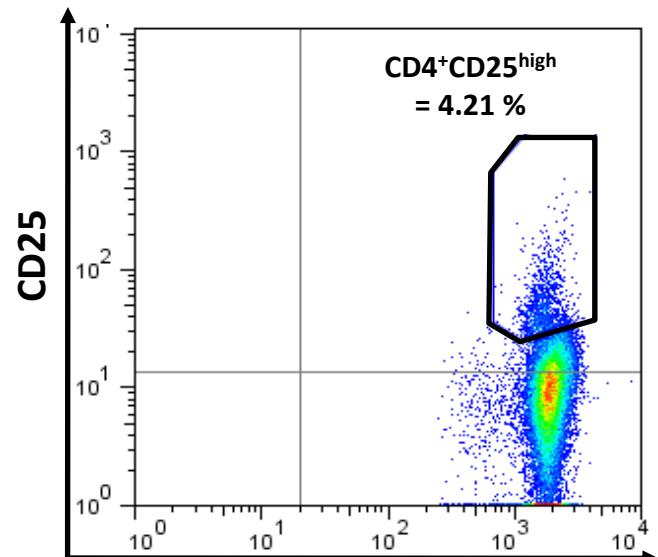
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Cytométrie en flux

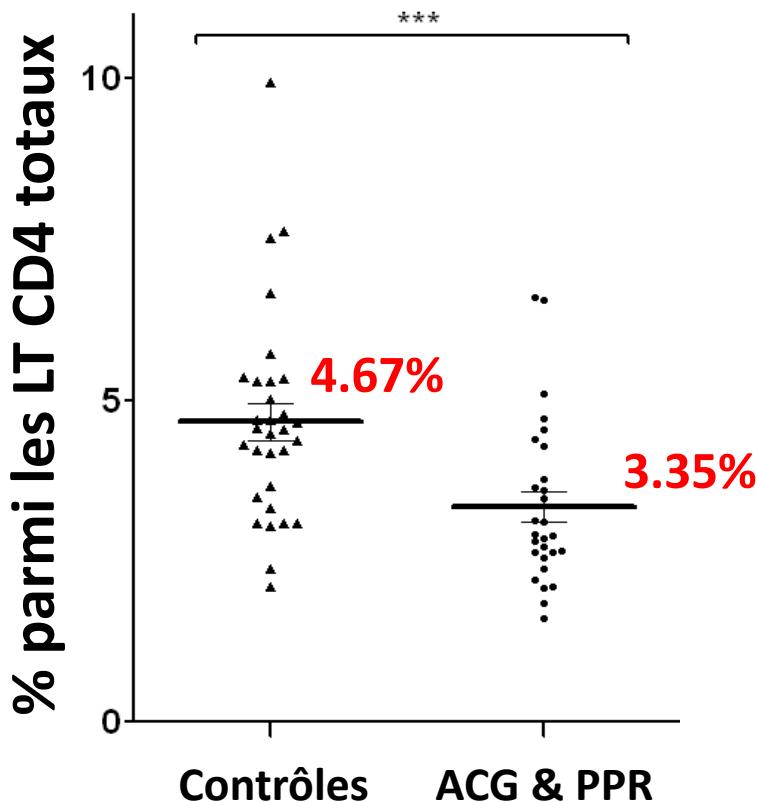
Treg



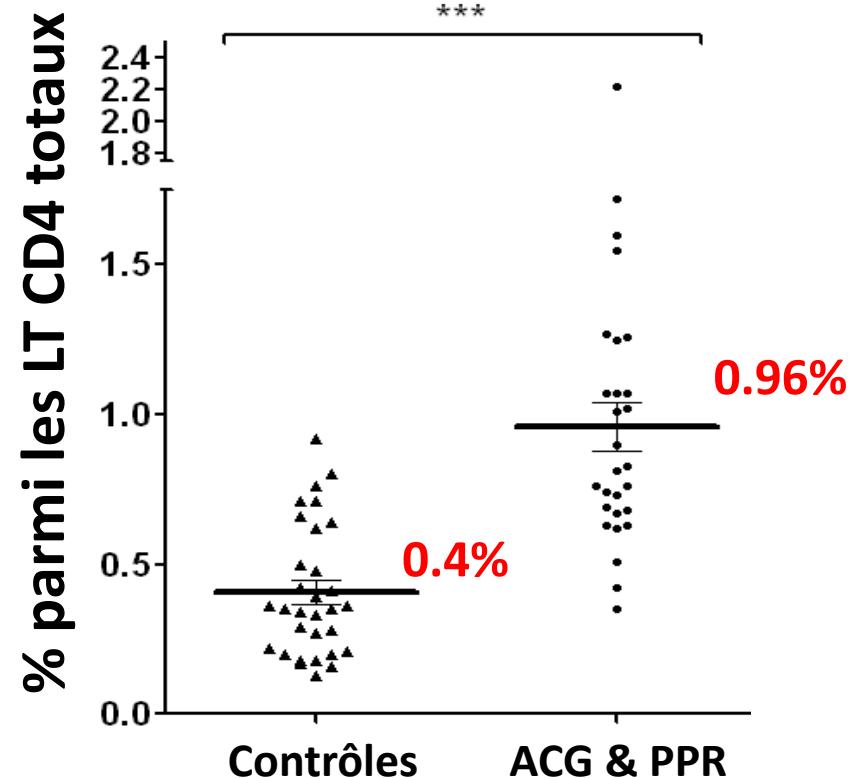


Déséquilibre de la balance Th17/Treg

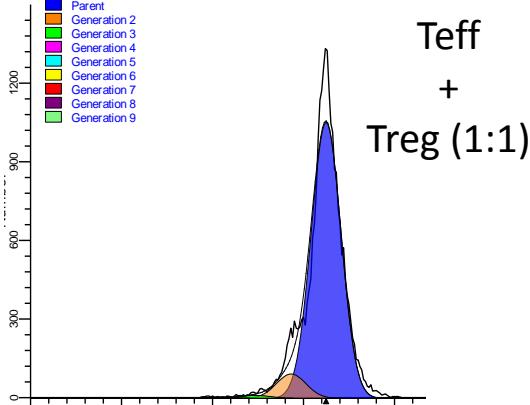
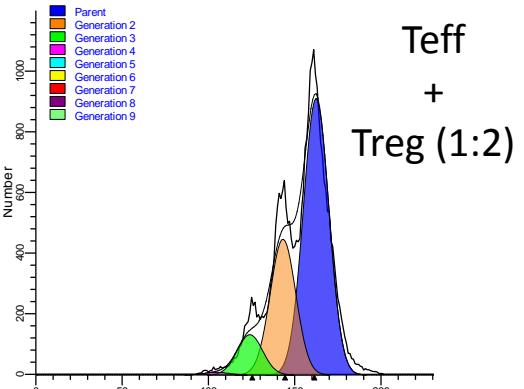
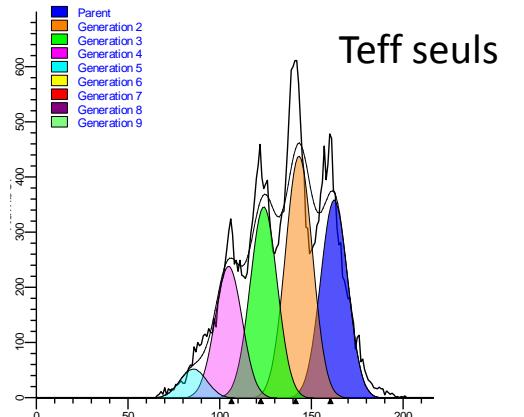
Diminution des Treg (CD4⁺ CD25^{high} Foxp3⁺)



Augmentation des Th17 (CD4⁺ IL-17⁺)



Analyse fonctionnelle des Treg



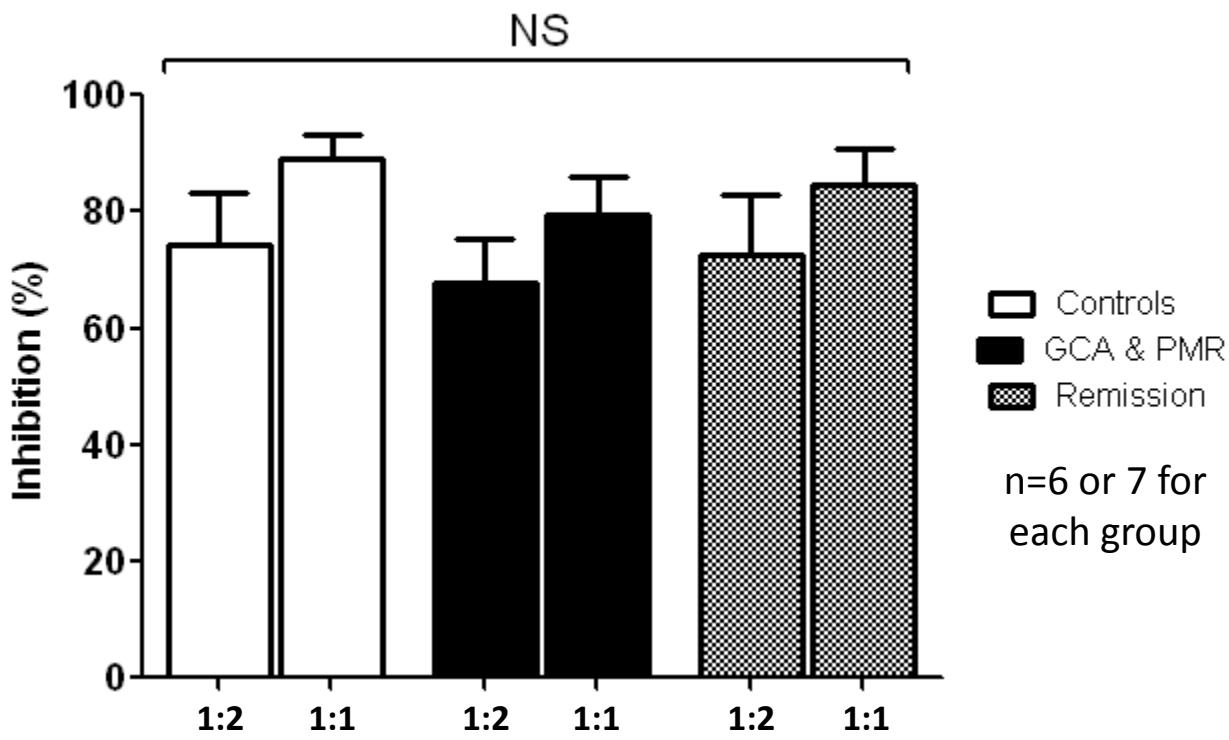
Co-culture (96 heures)

Activation par billes anti-CD2, -CD3, CD28

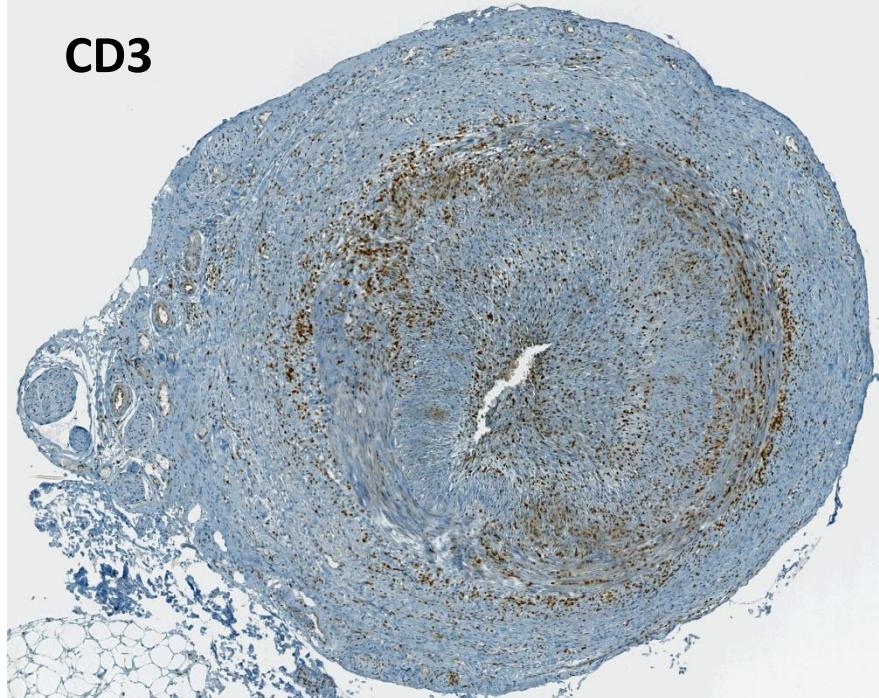
Proliferation index = Cell Trace Violet

Teff = CD4⁺CD25^{-/low}

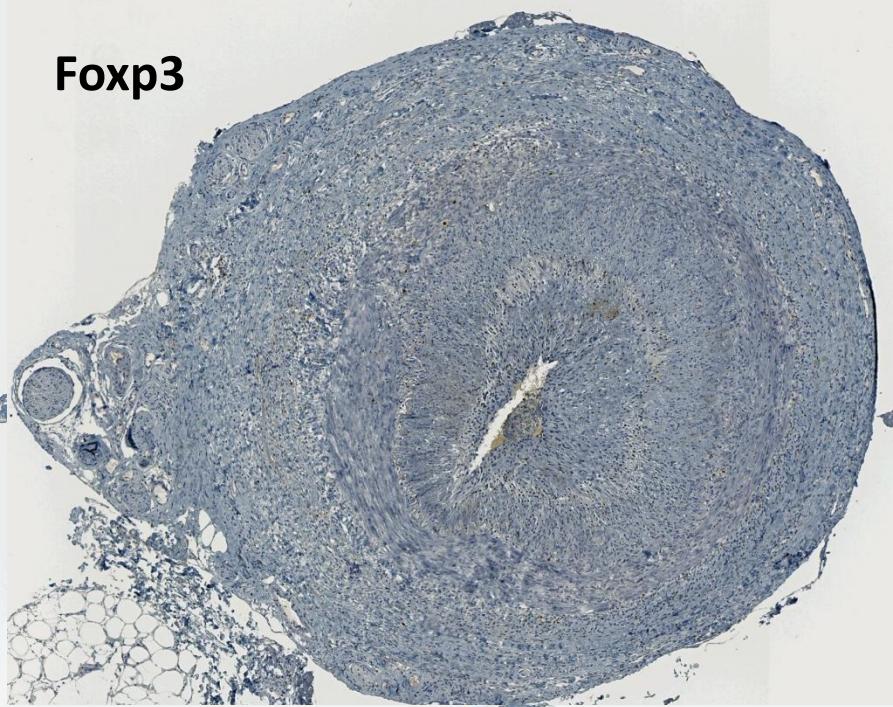
Treg = CD4⁺CD25^{high}



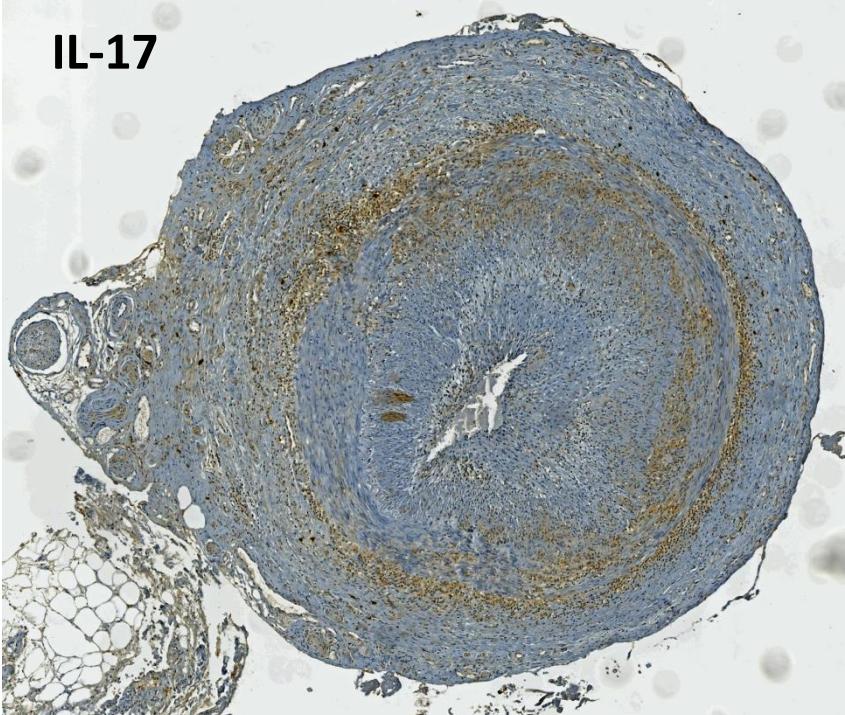
CD3



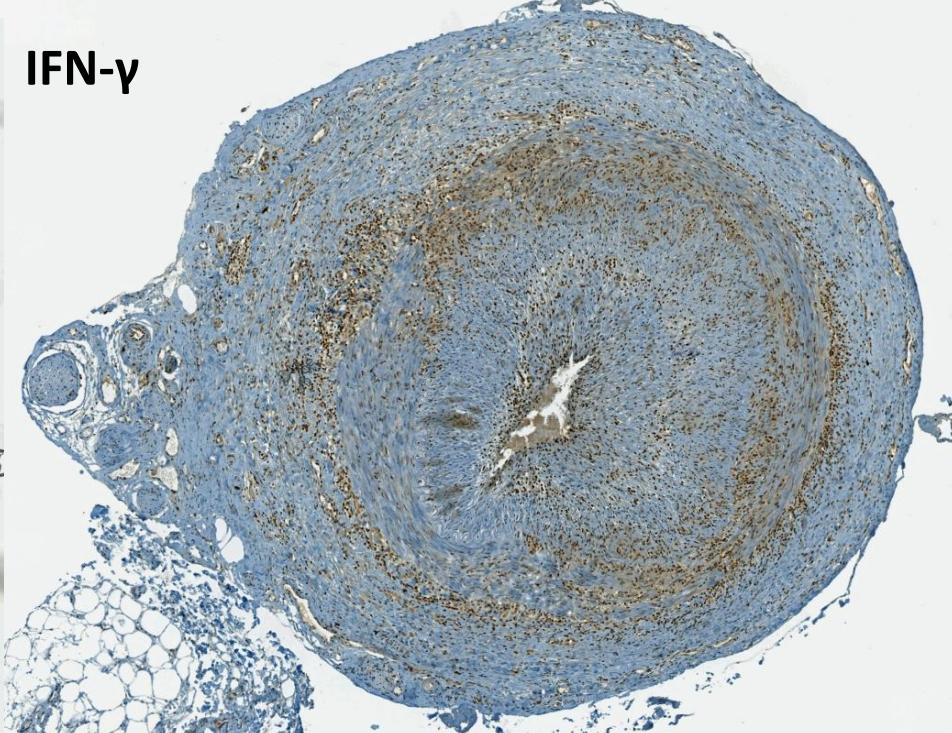
Foxp3



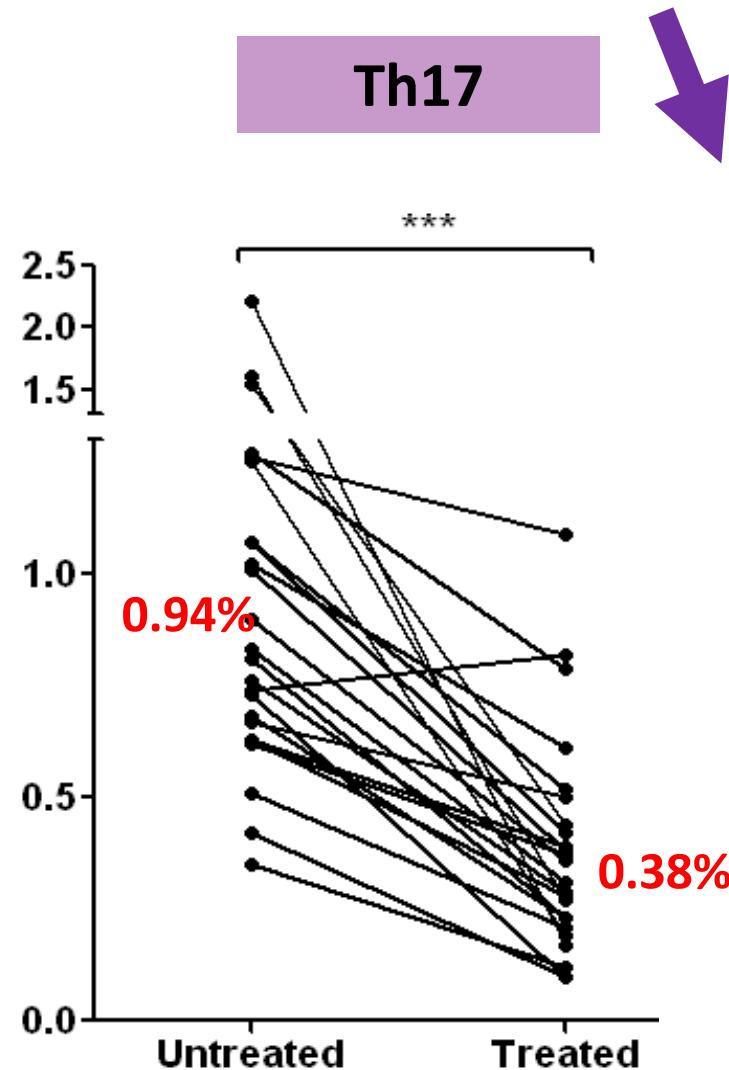
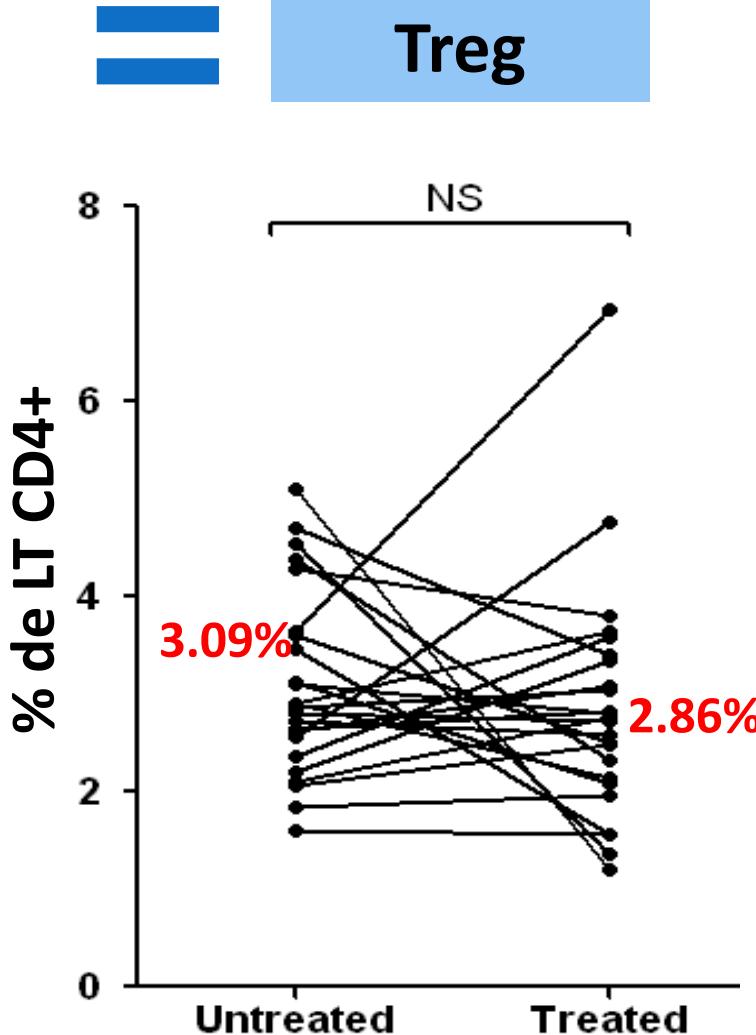
IL-17



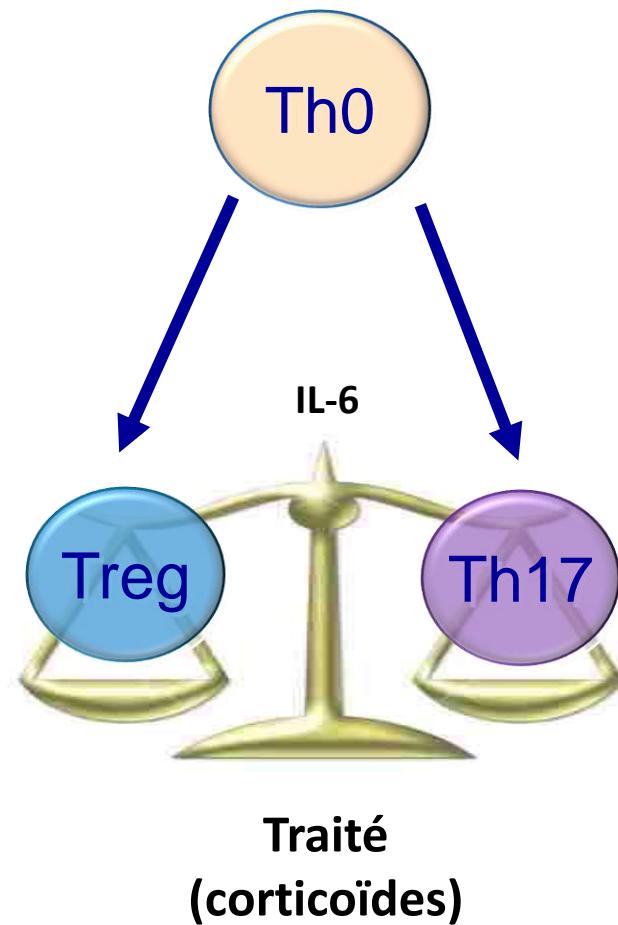
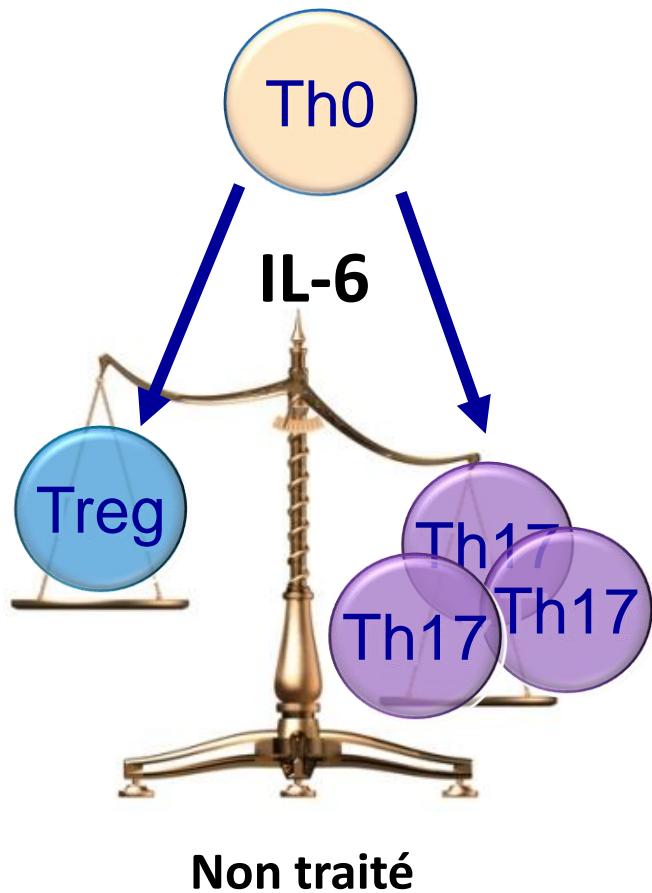
IFN- γ



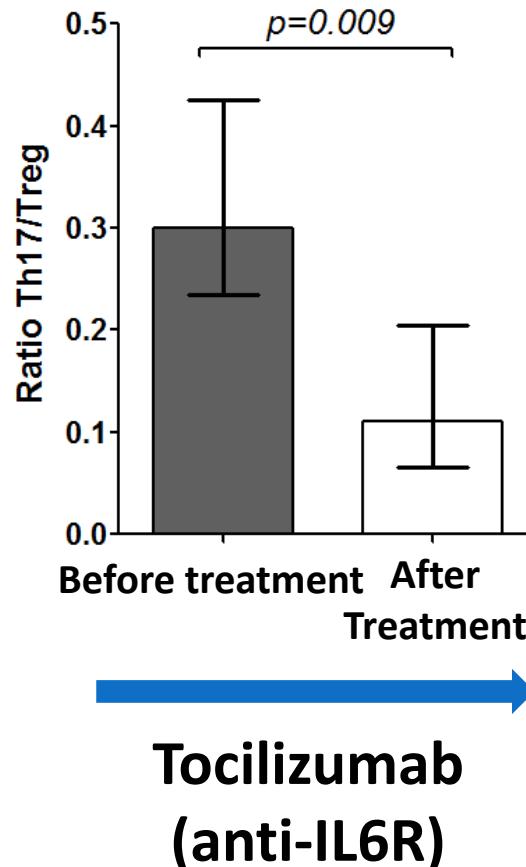
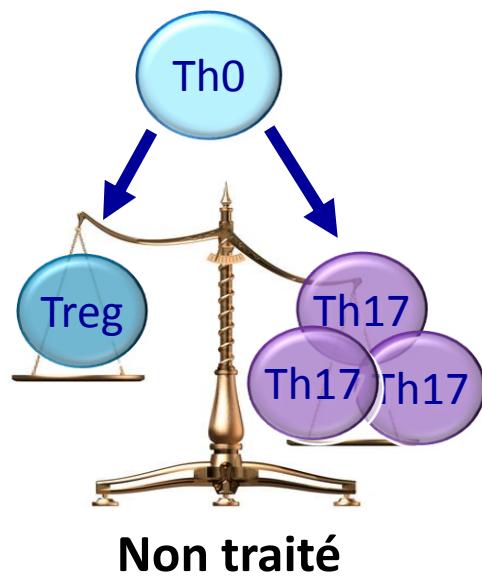
Effet des corticoïdes



La balance Th17/Treg est partiellement corrigée par la corticothérapie

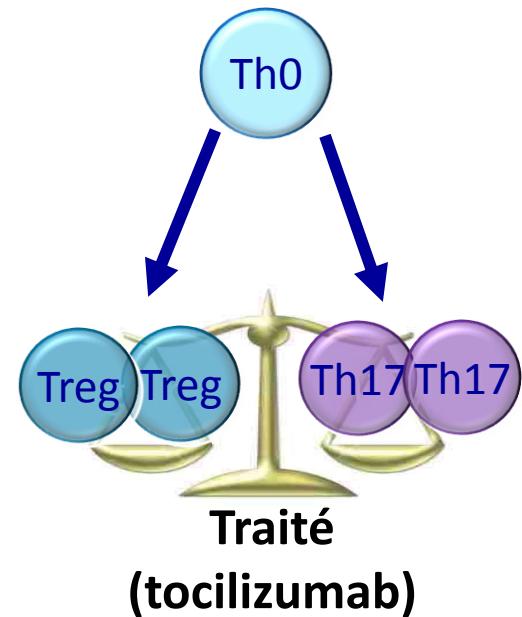


La balance Th17/Treg est corrigée par le blocage de l'IL-6 (tocilizumab)

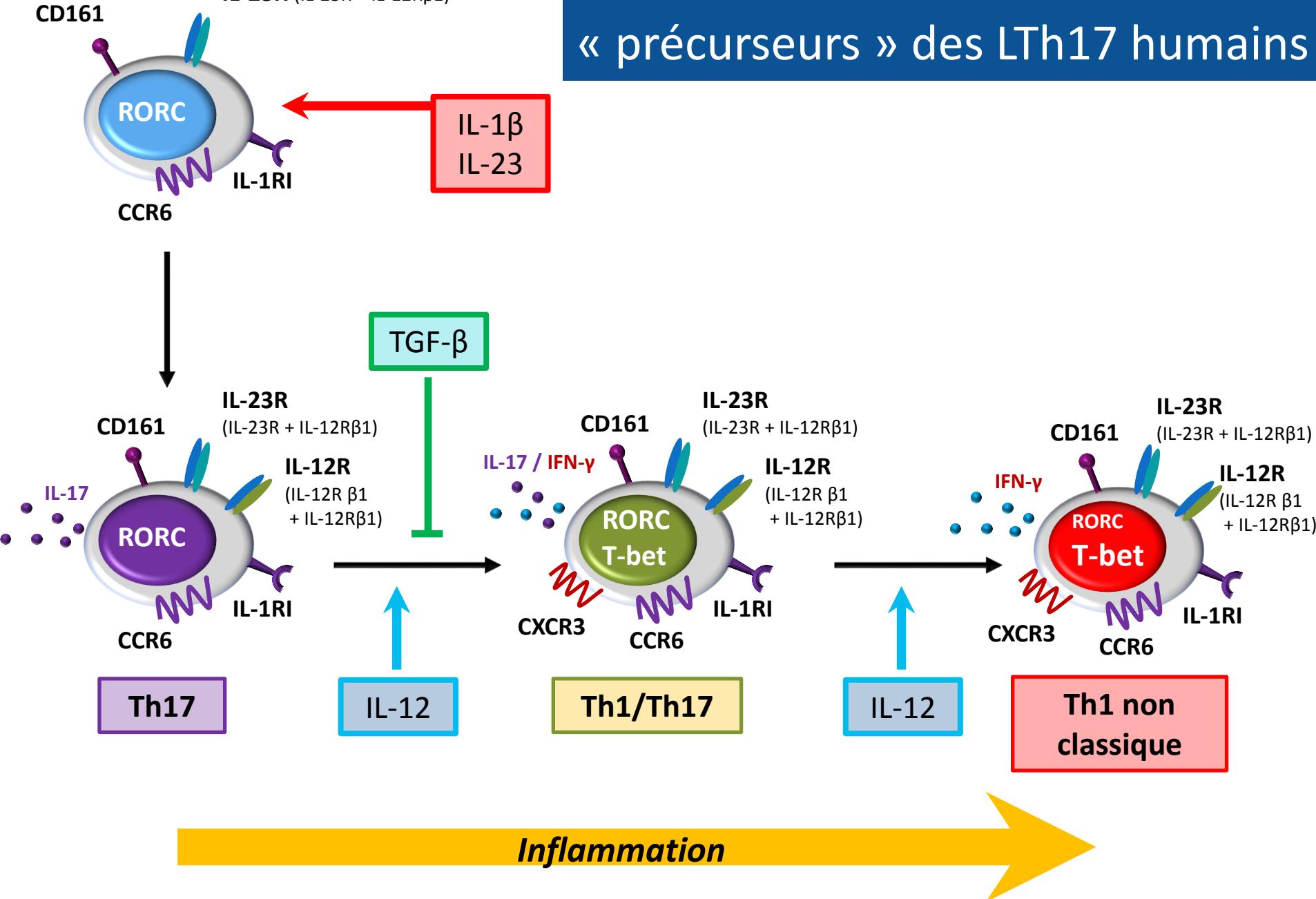


Polyarthrite rhumatoïde
Samson M et al. Arthritis Rheum 2012
Pesce B et al. Clin and Exp Immunol 2013
Thiolat A et al. Arthritis Rheum 2014
Samson M et al. Arthritis Rheum 2014

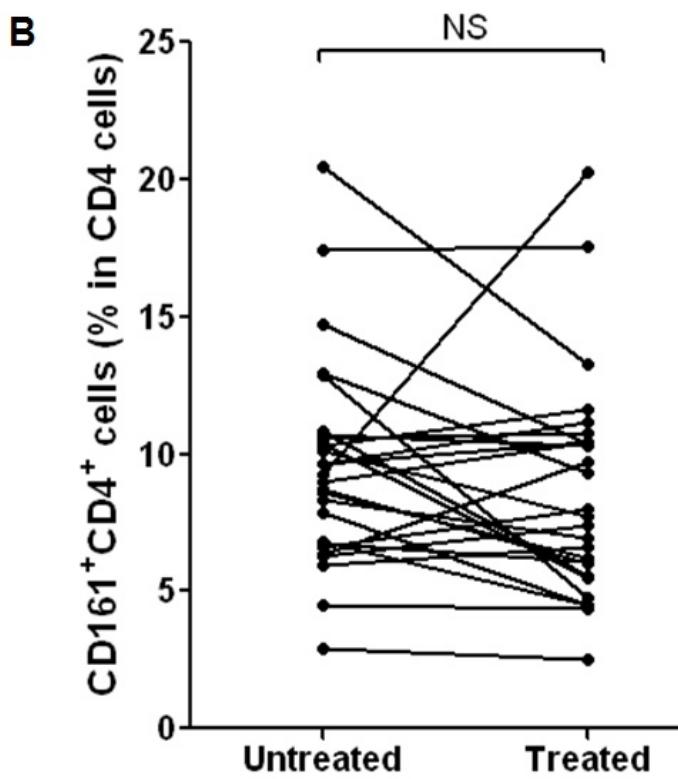
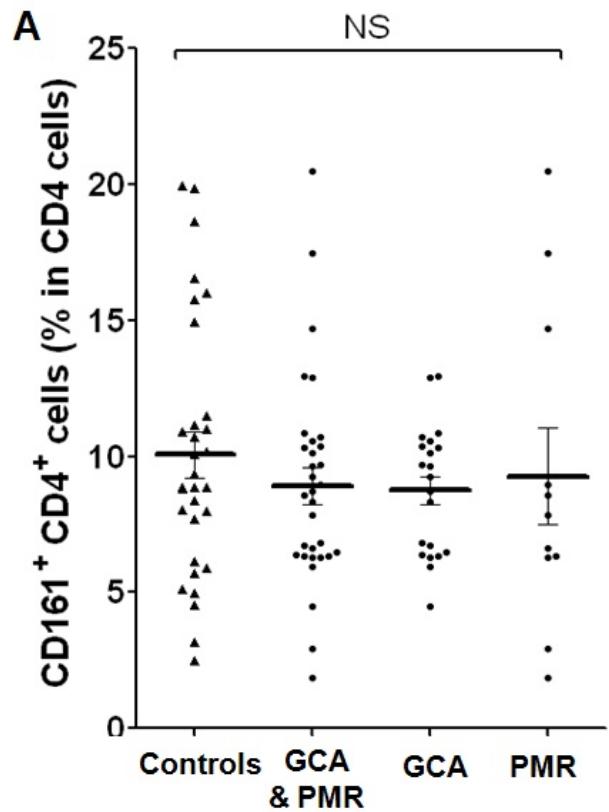
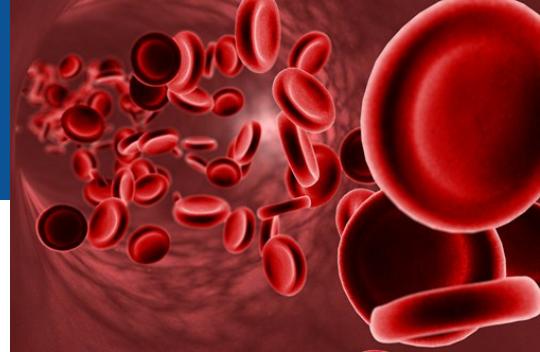
ACG
Miyabe C et al. ARD 2017



LT CD4⁺CD161⁺ « précurseurs » des LTh17 humains

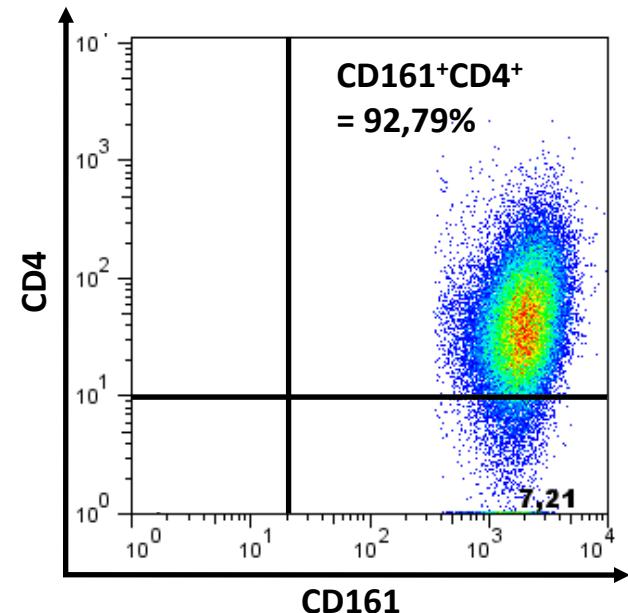
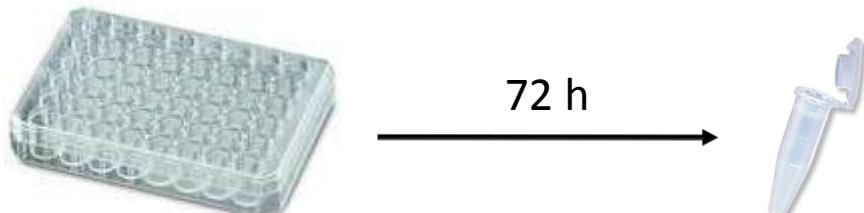
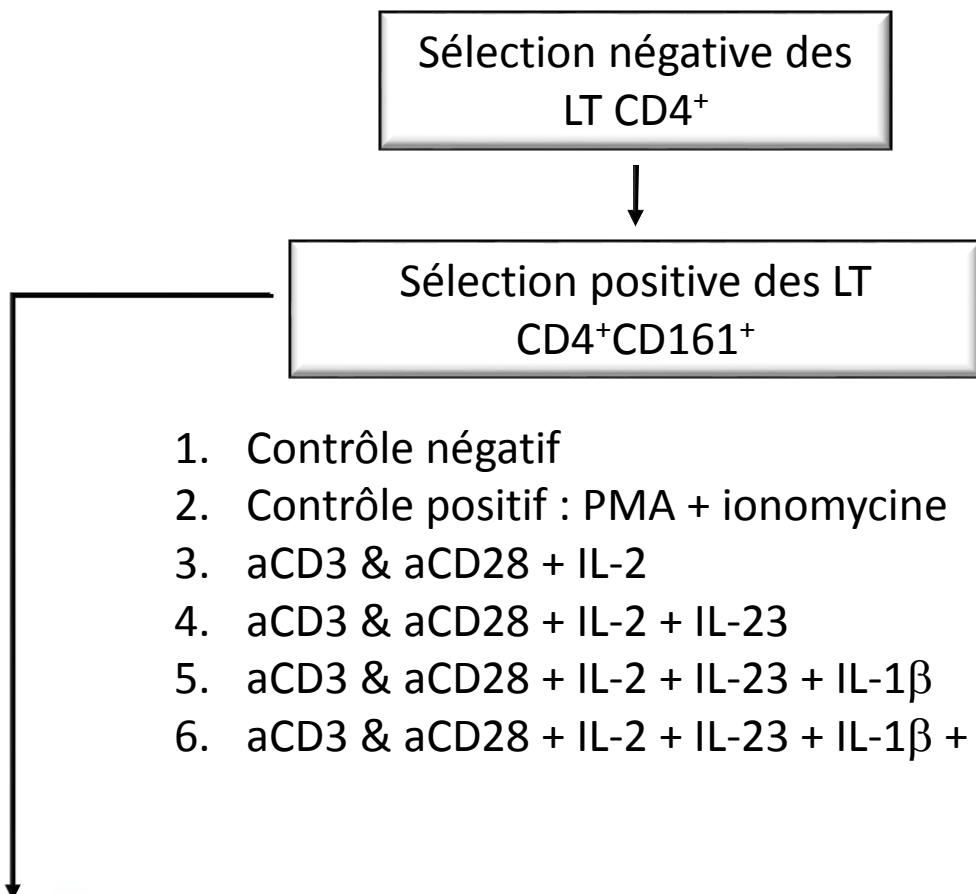


Etude des LT CD4⁺CD161⁺ sanguins

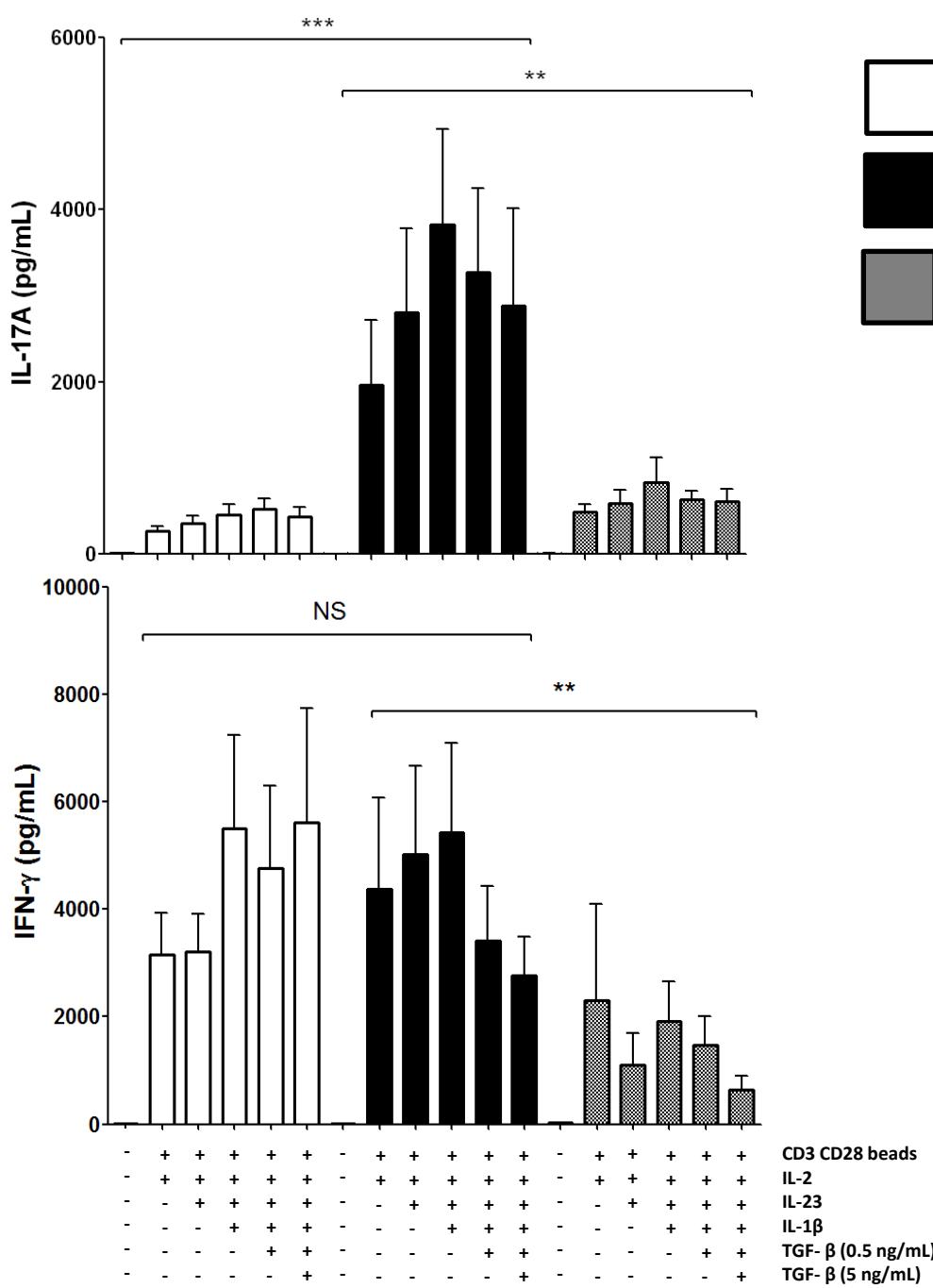


Pas de différence
concernant le % de
LT CD4⁺CD161⁺
circulants

Isolation des LT CD4⁺CD161⁺ circulants



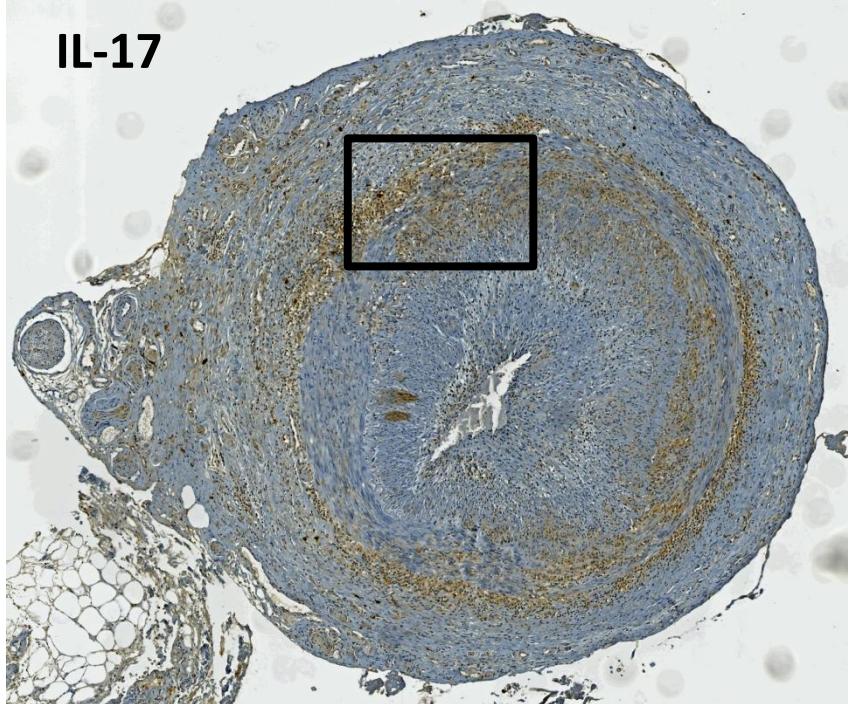
Surnageants :
IL-17A, IFN-γ, TNF-α
(Luminex)



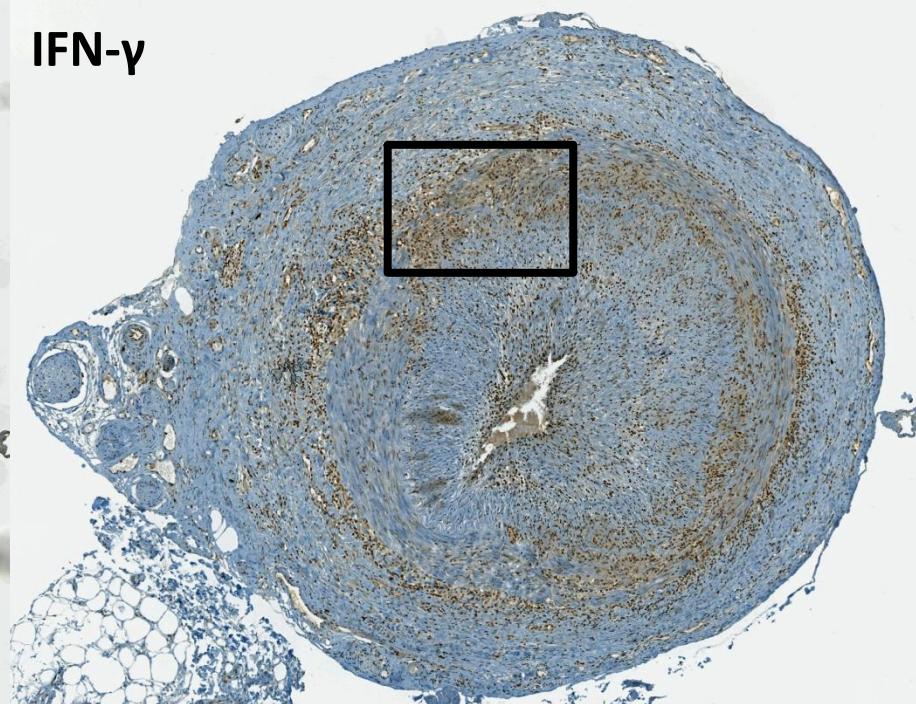
Les LT CD4 $^{+}$ CD161 $^{+}$ des patients produisent **plus d'IL-17** que ceux des contrôles et des patients traités

Les LT CD4 $^{+}$ CD161 $^{+}$ peuvent aussi se **polariser en Th1** (IFN- γ)

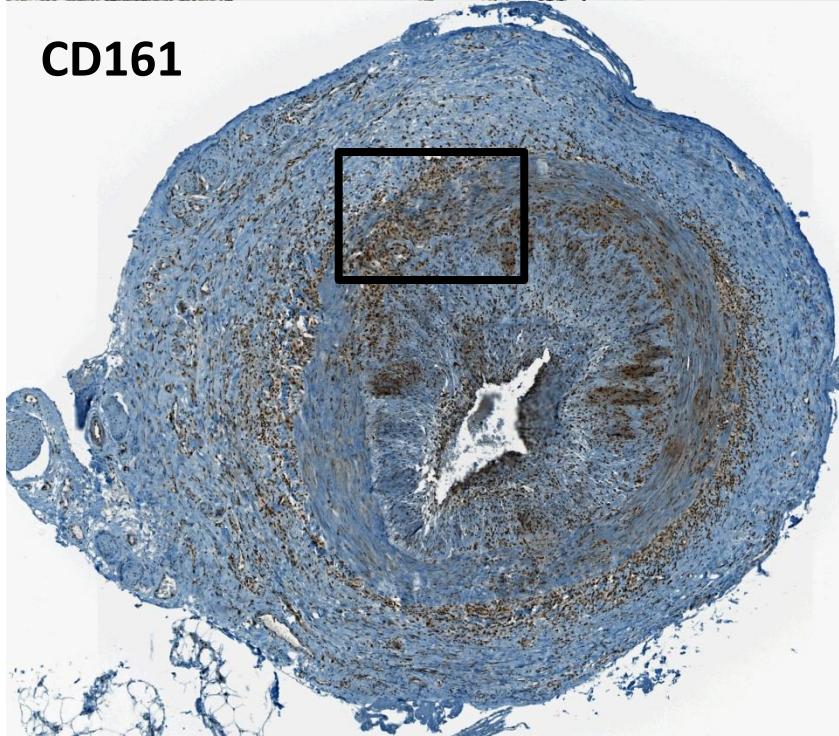
IL-17



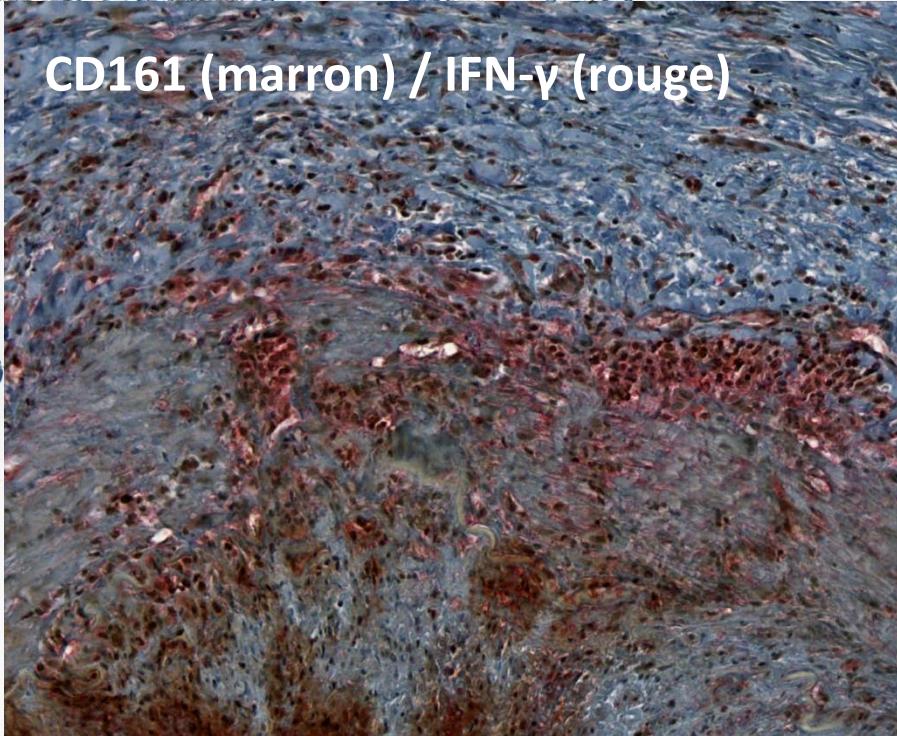
IFN- γ

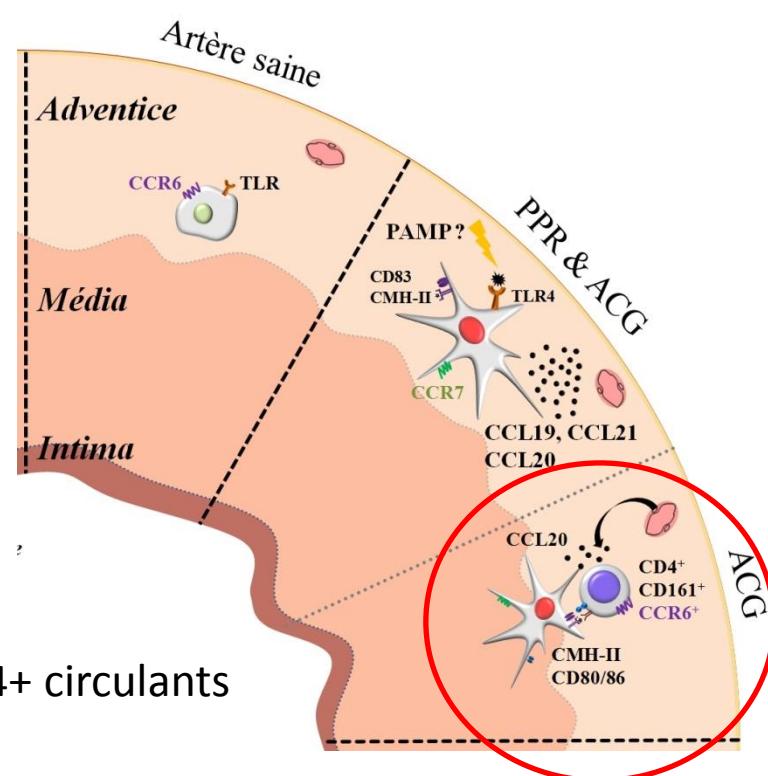
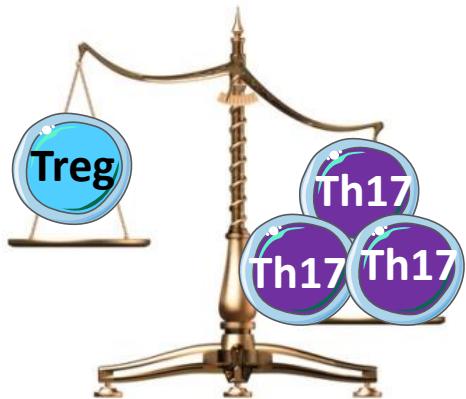


CD161

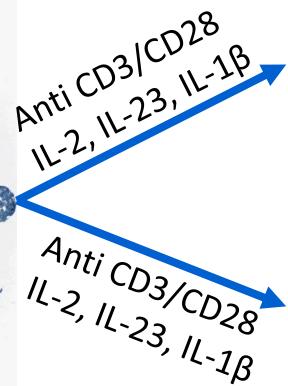
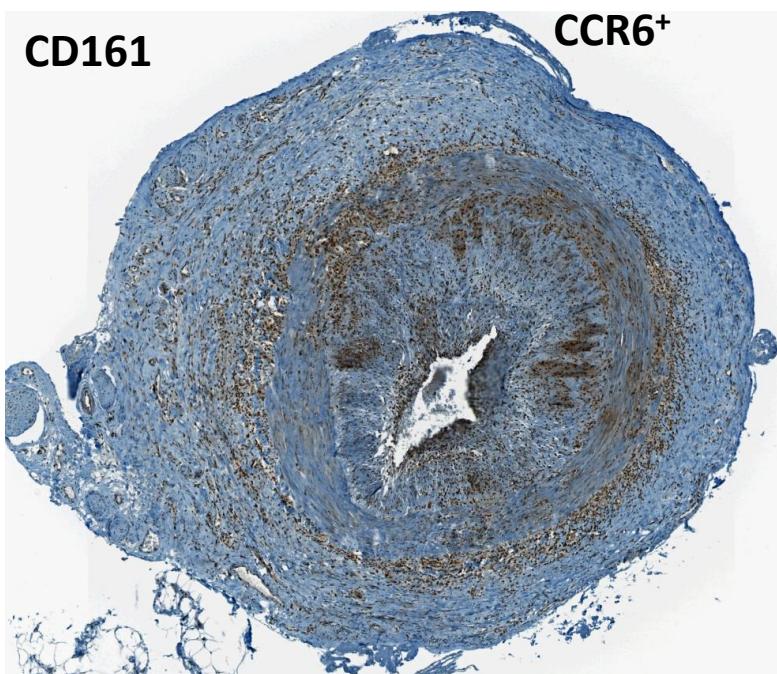


CD161 (marron) / IFN- γ (rouge)





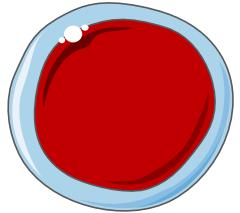
LT CD4⁺CD161⁺
10% des LT CD4+ circulants
CCR6⁺



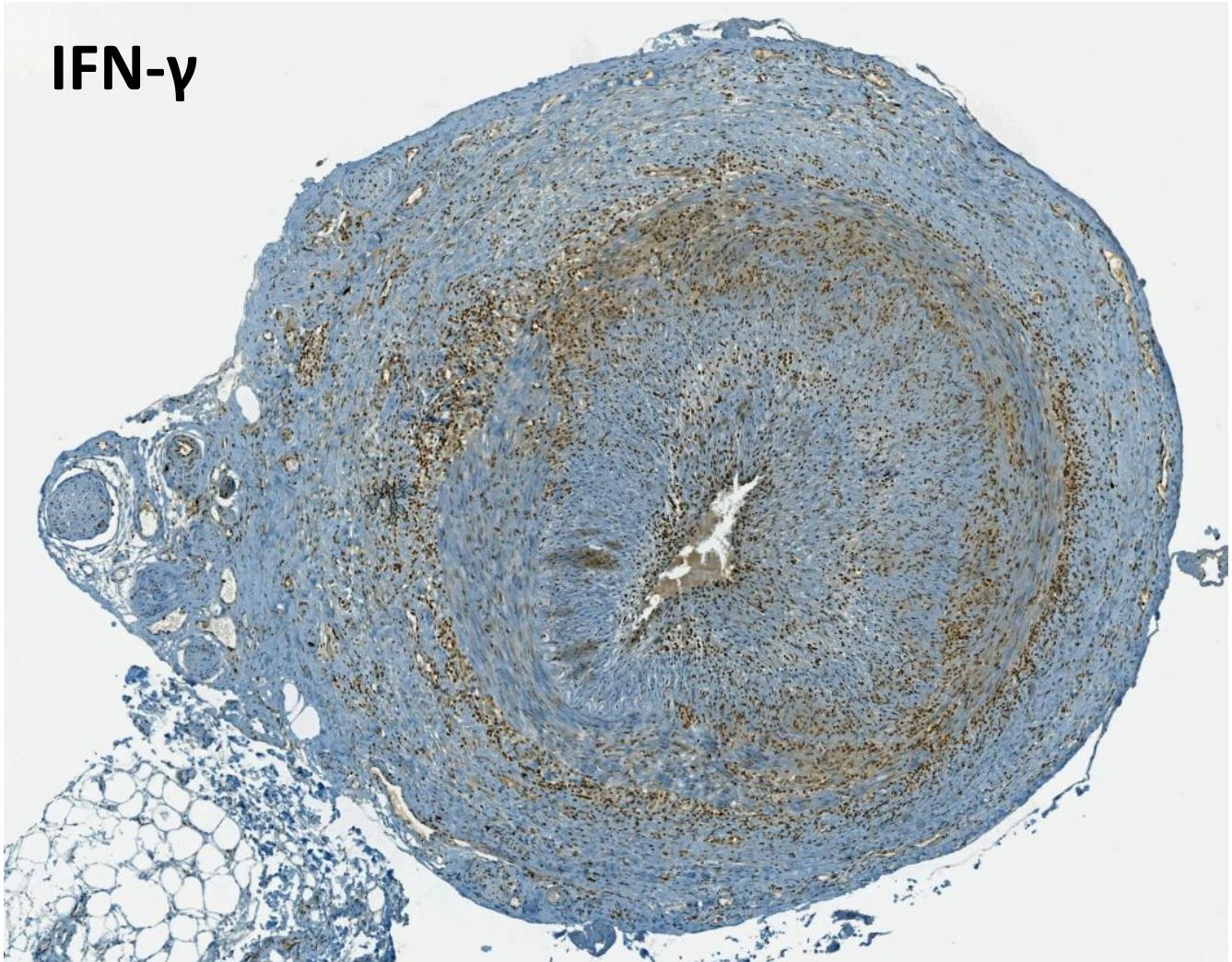
Polarisation Th17
↑ entre patients/contrôles
↓ après traitement

Polarisation Th1
≈ entre patients/contrôles
↓ (faible) après traitement

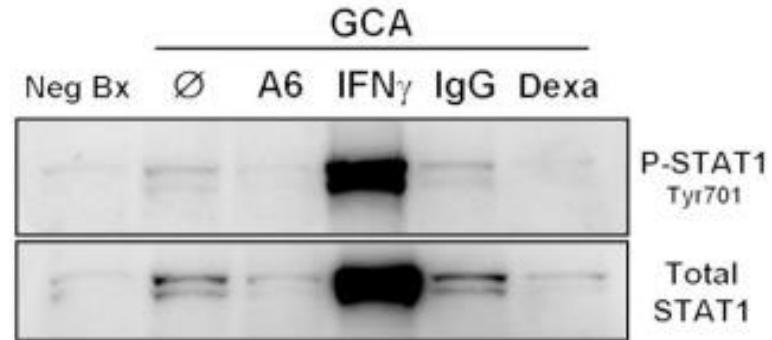
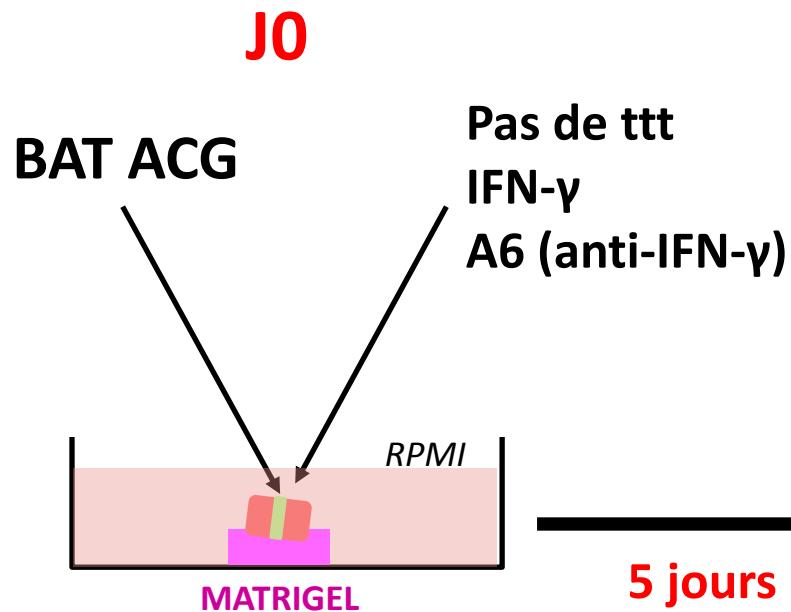
Quel est le rôle de l'IFN- γ ?



Th1 cells



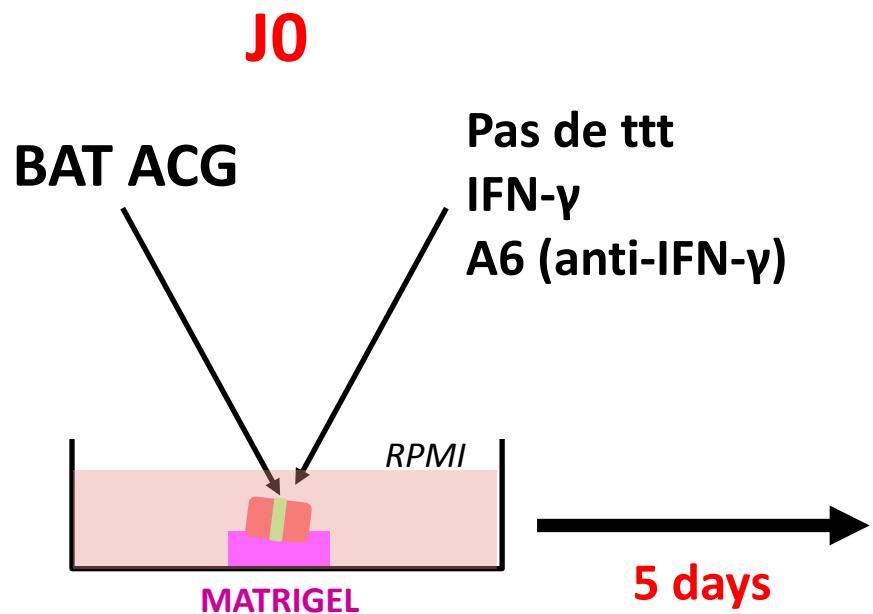
Le modèle du MATRIGEL®



- surnageants
- biopsie (TRIZOL → PCR)
- confocale



The modèle du MATRIGEL®

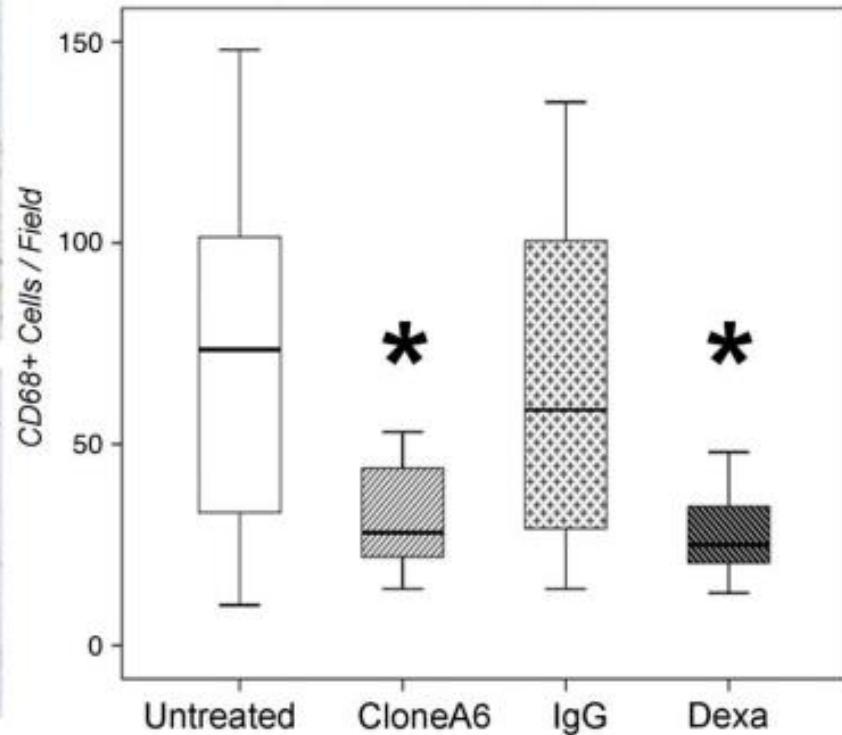
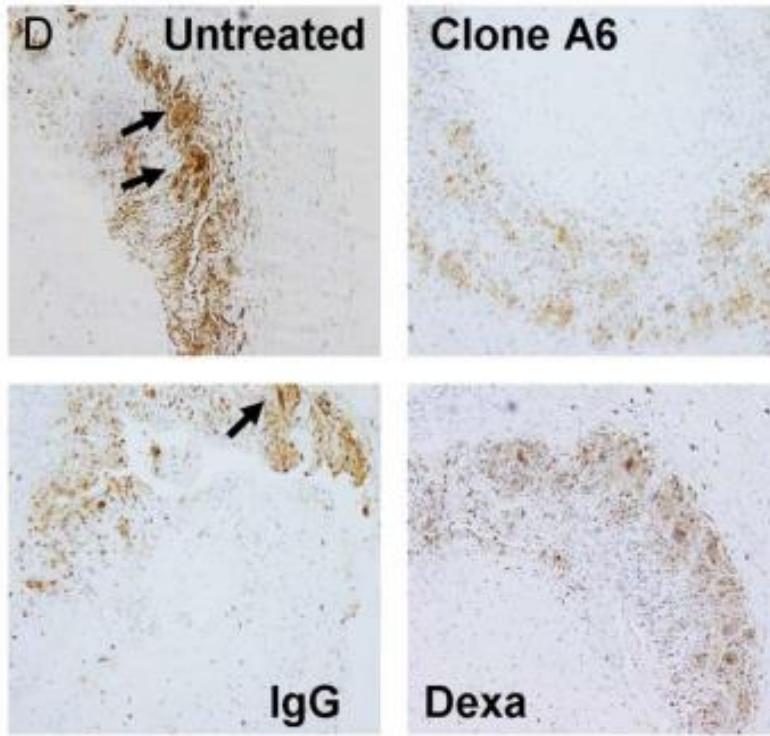


IFN- γ entraîne :

- production de **CXCL9, -10 et -11** (ligands de CXCR3) par les CMLV
- production de **CCL2** par les CMLV

Role de l'IFN- γ ?

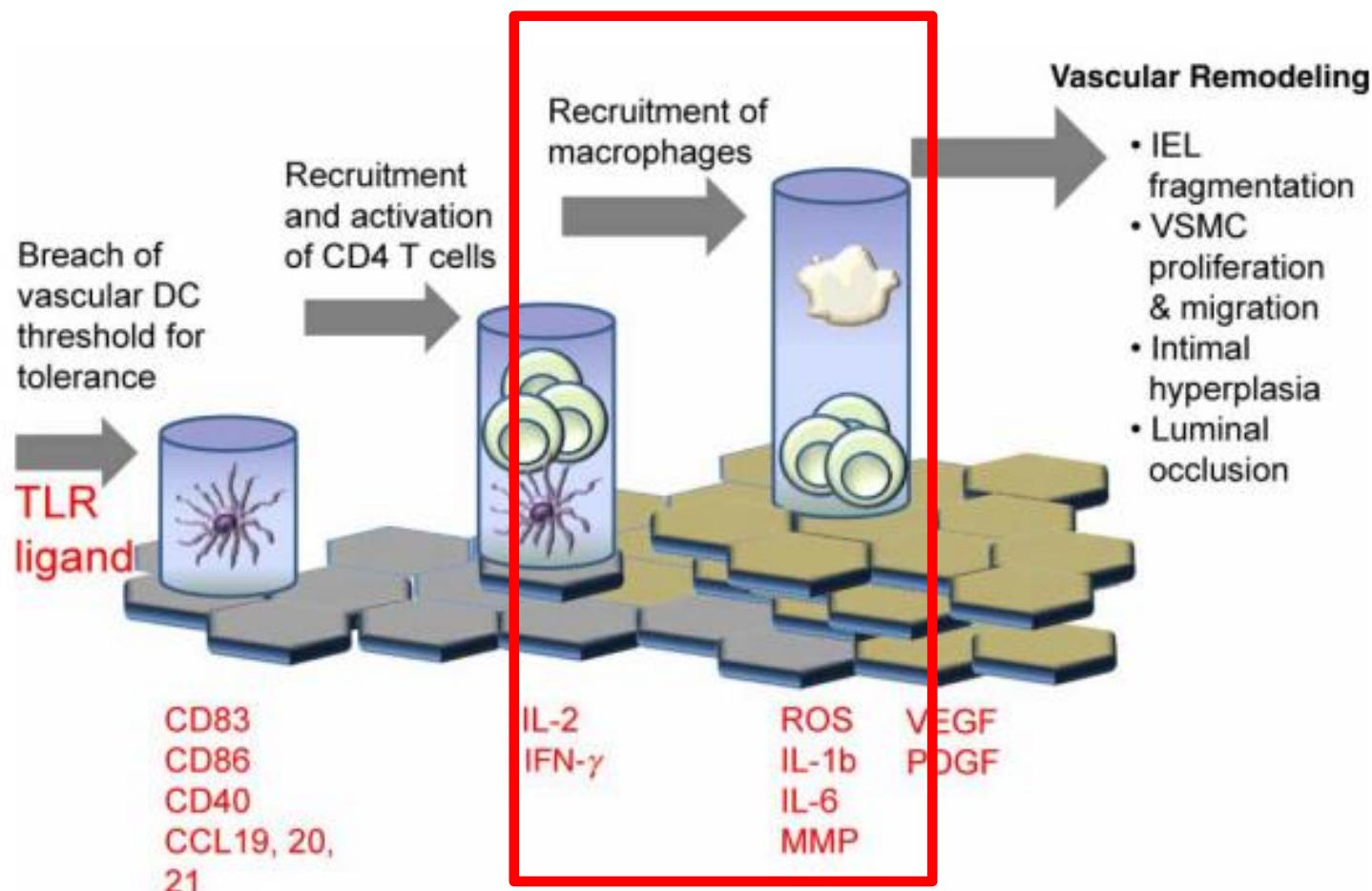
Marquage CD68



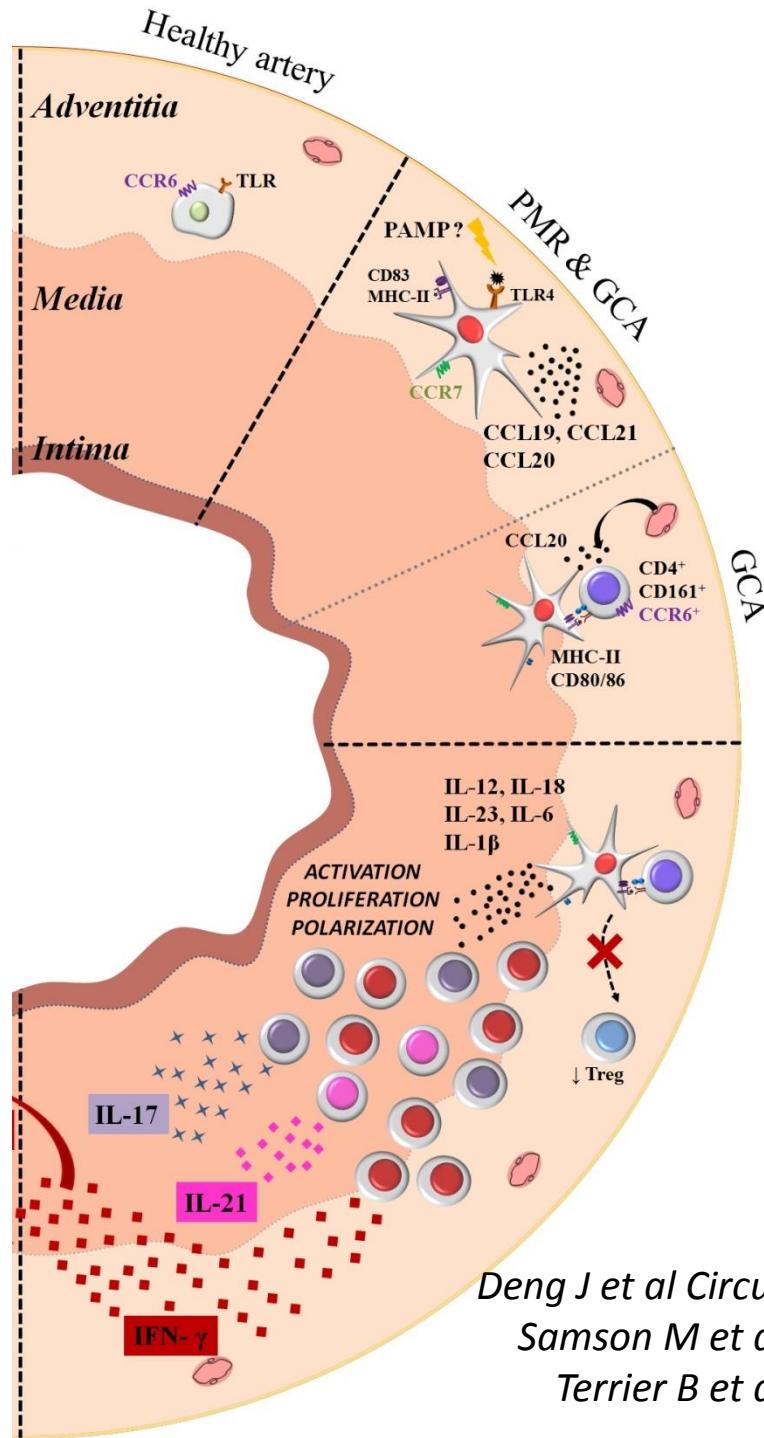
IFN- γ conduit au recrutement des macrophages CD68+
(interaction entre CCL2/CCR2)

Le modèle physiopathologique de l'ACG

PHASE III



IFN- γ

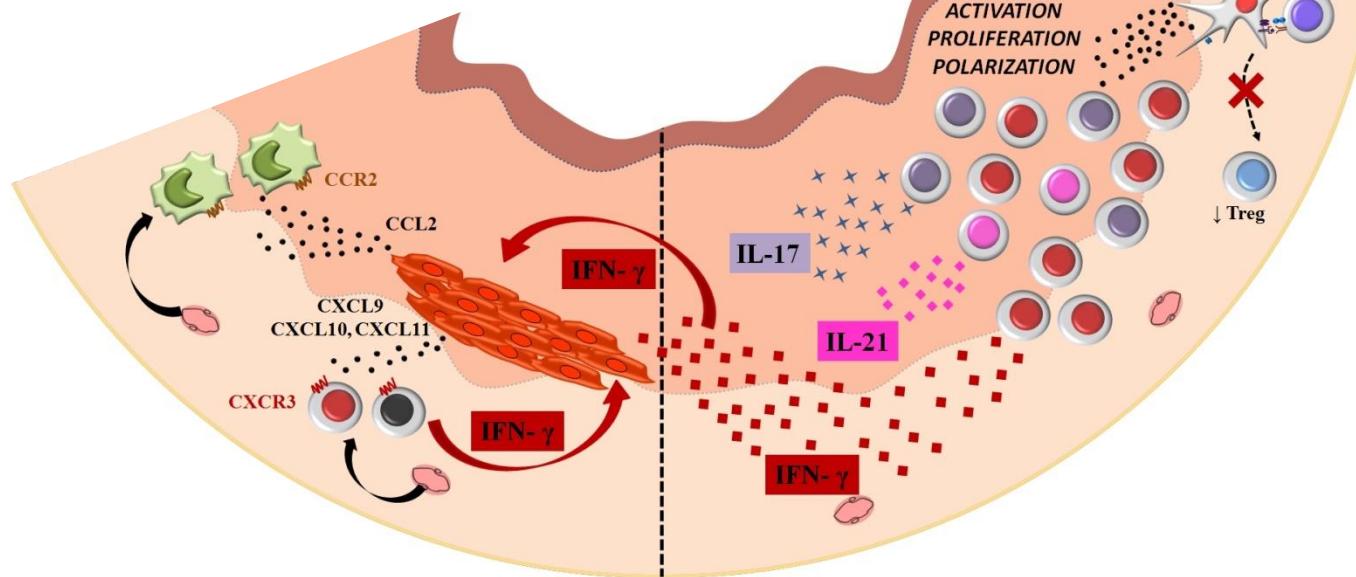
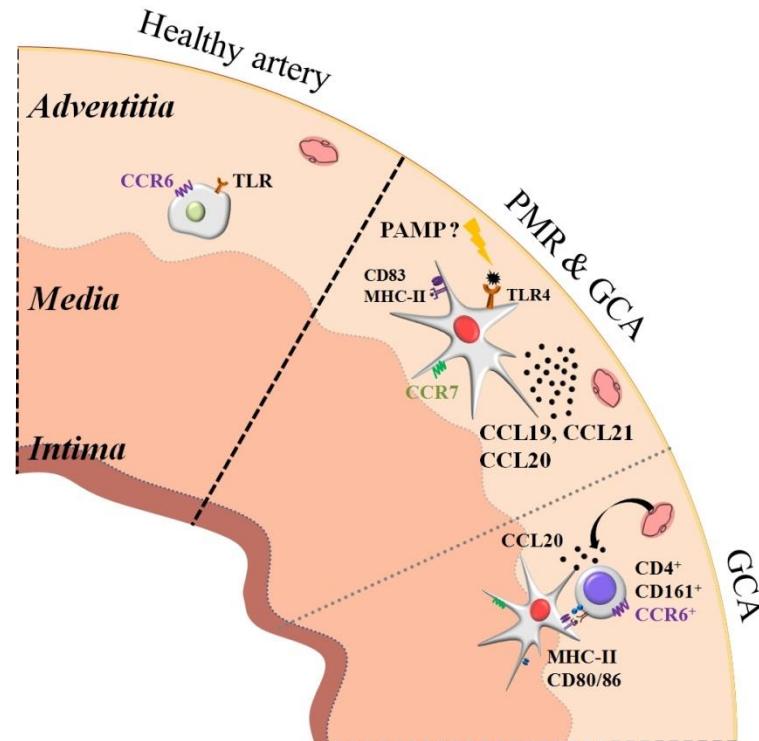


Deng J et al Circulation 2011
 Samson M et al A&R 2012
 Terrier B et al A&R 2012

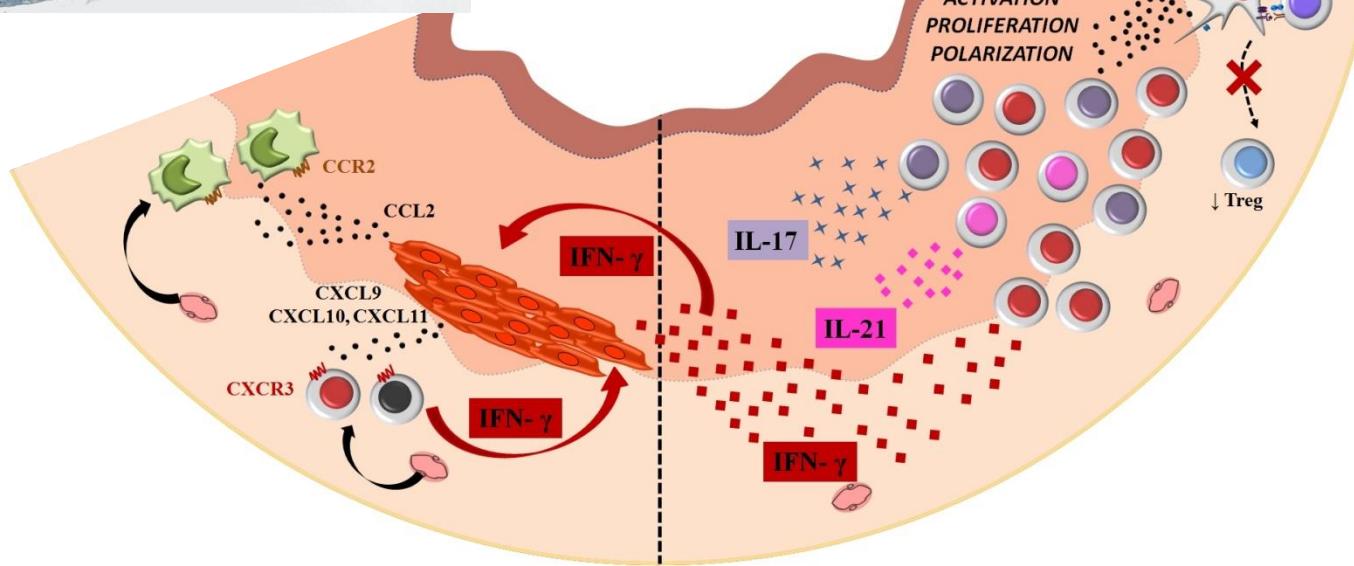
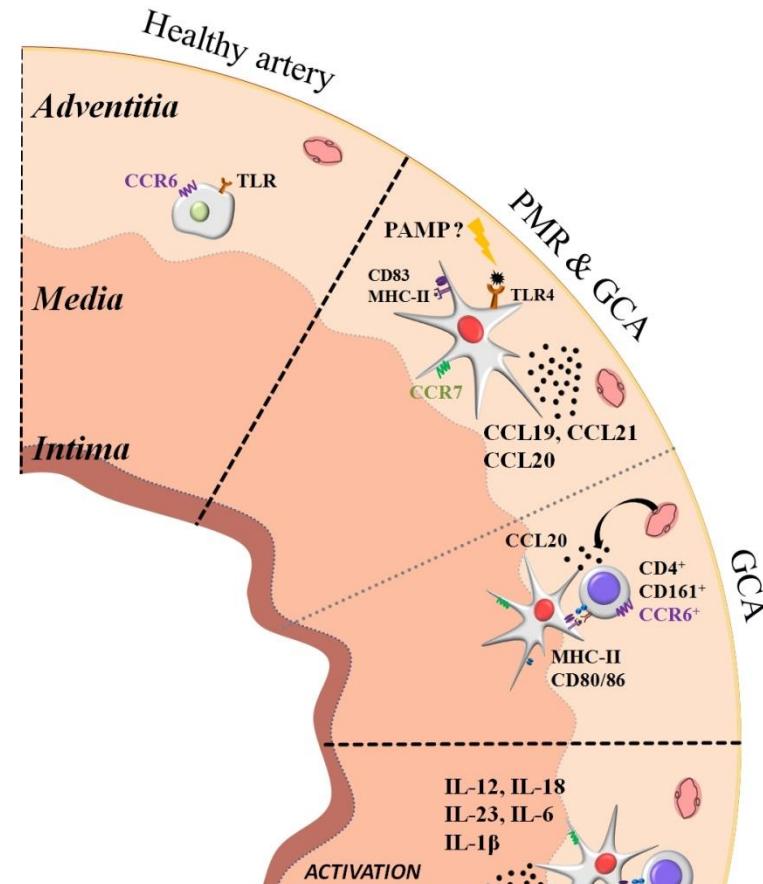
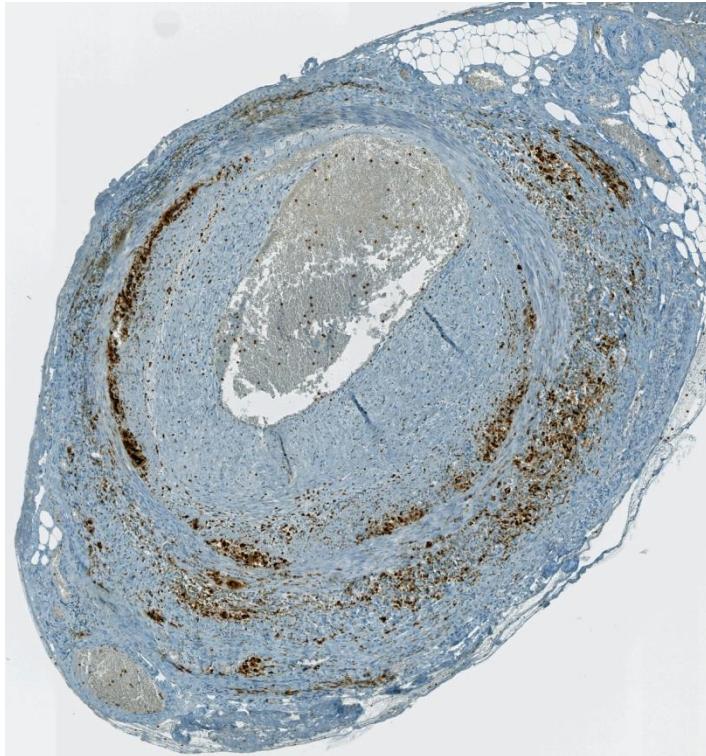
IFN- γ induit la production de CCL2 par les CMLV

CCL2 = ligand de CCR2

Rôle important des CMLV



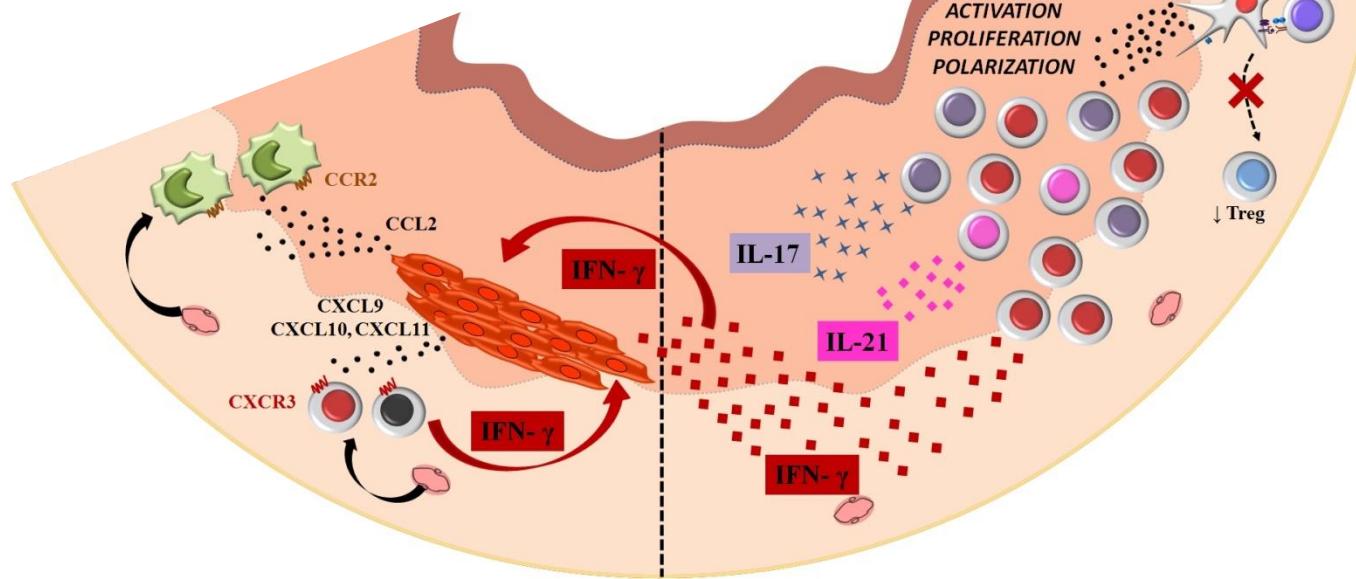
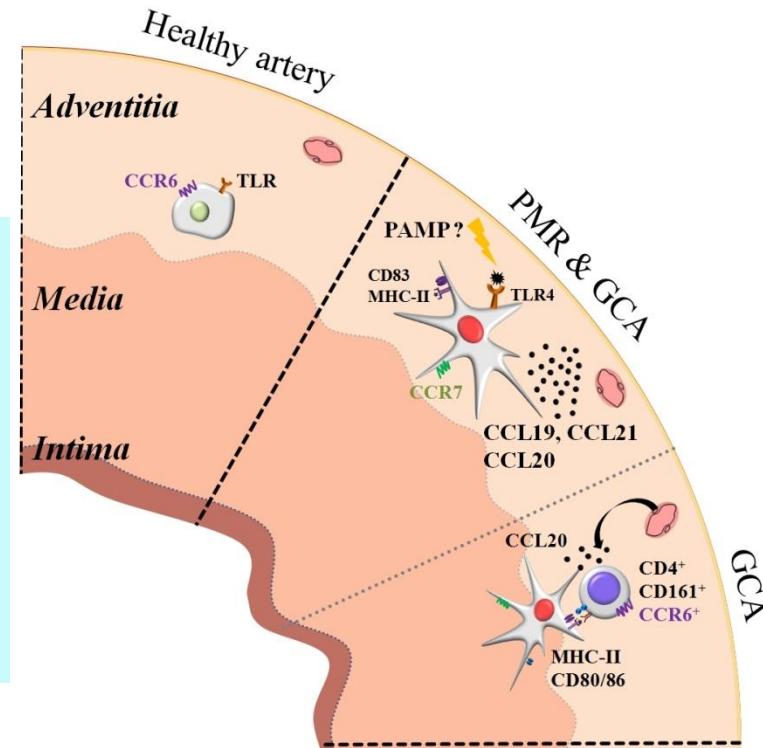
CCR2 = monocytes



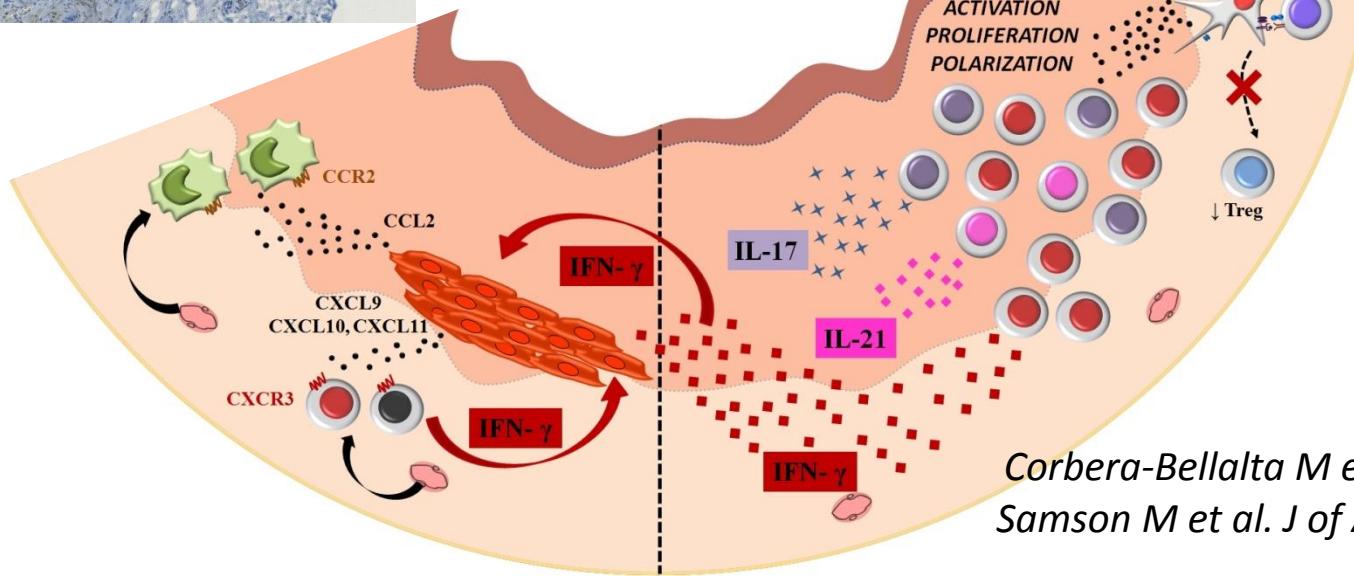
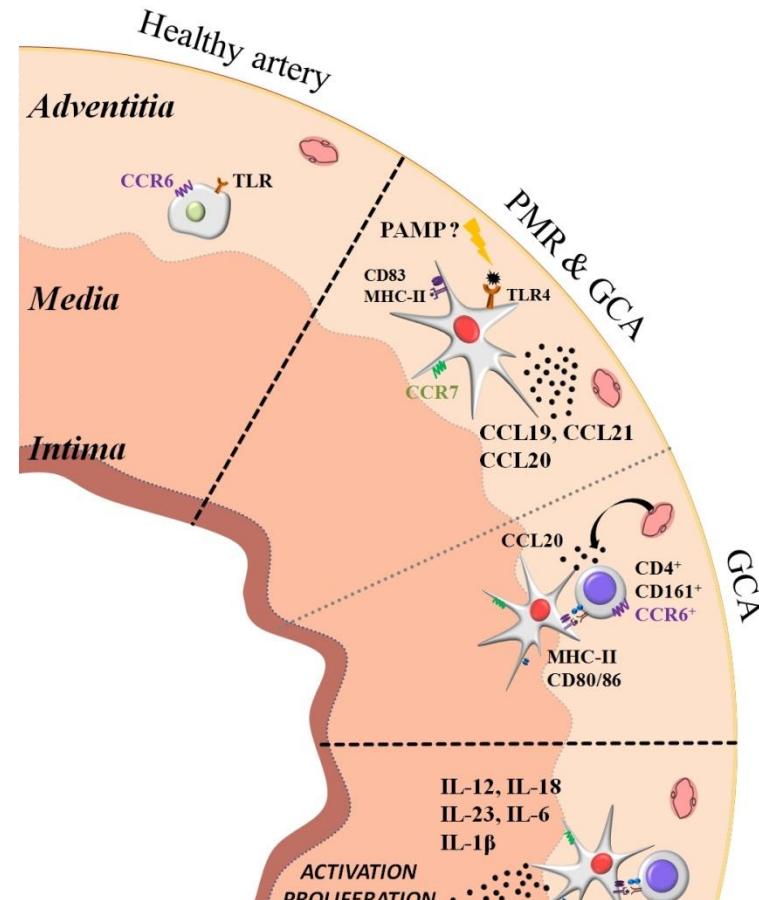
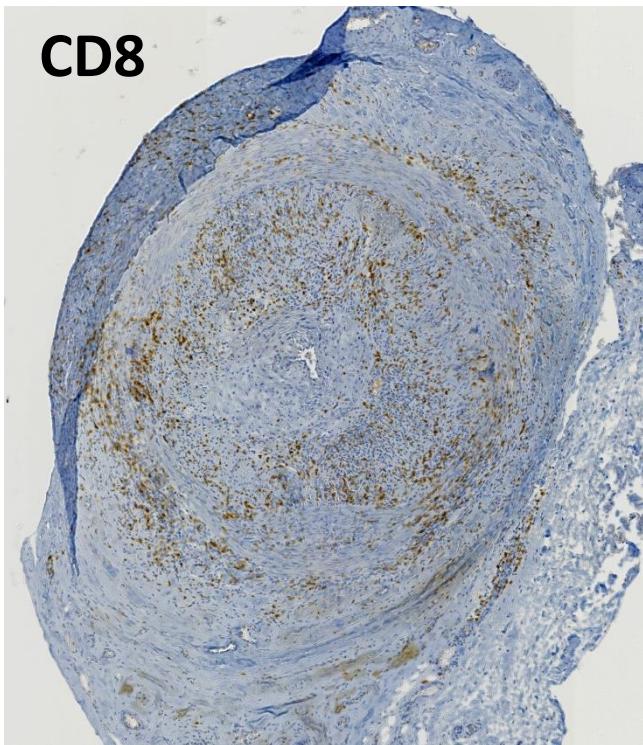
IFN- γ induit la production de CCL2 par les CMLV

CCL2 = ligand de CCR2

CXCL9,-10,-11 = ligands de CXCR3

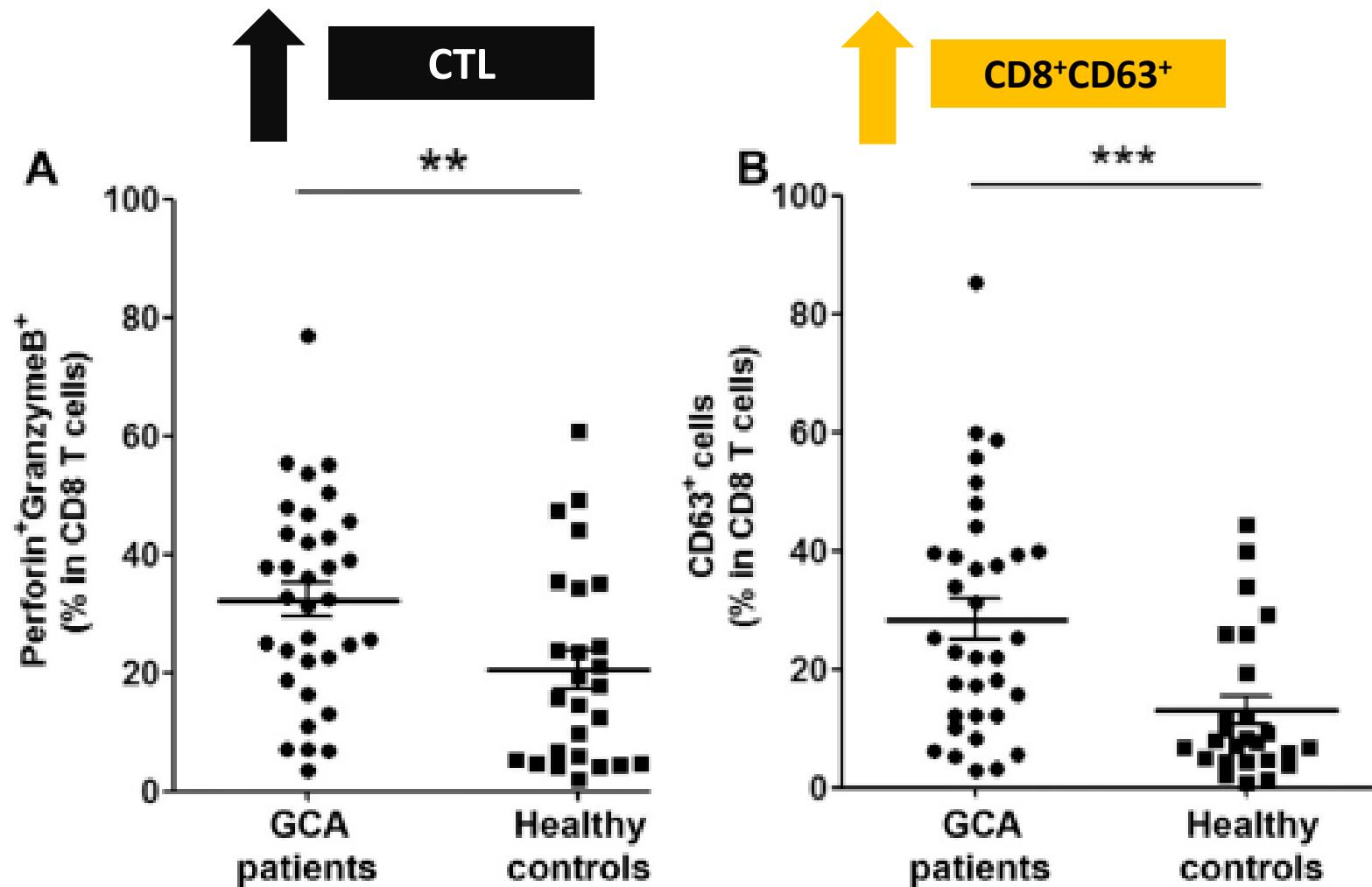


CXCR3 = Th1 et LT CD8⁺



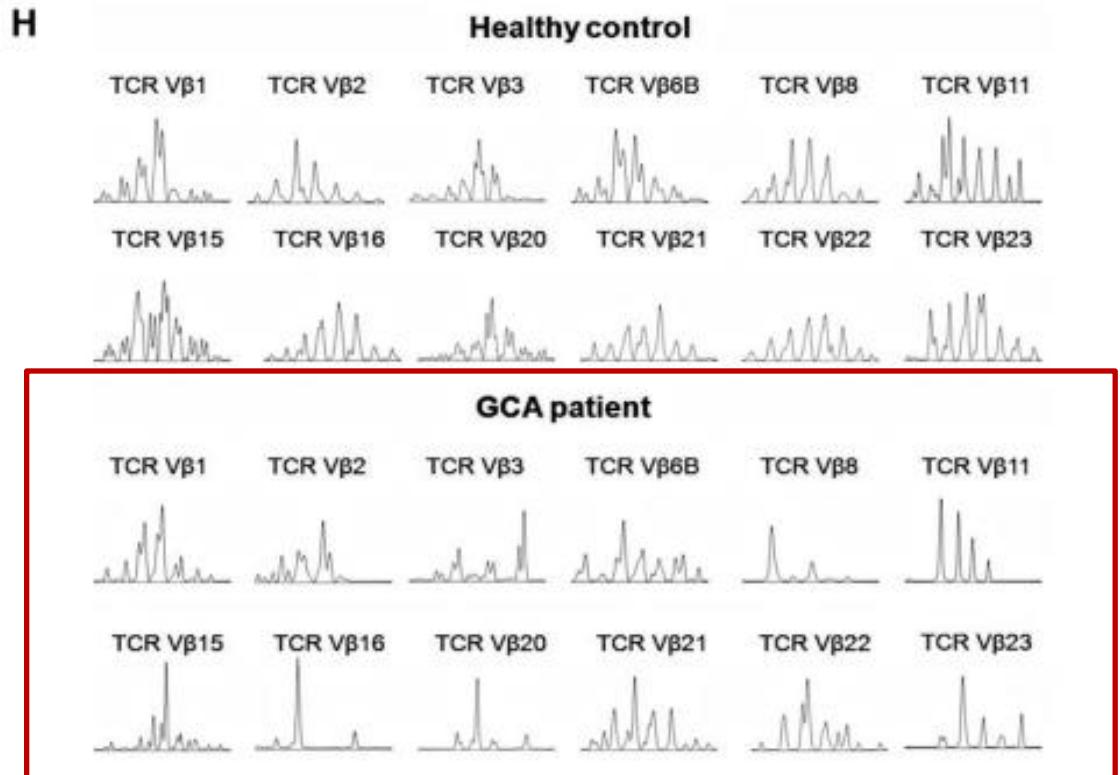
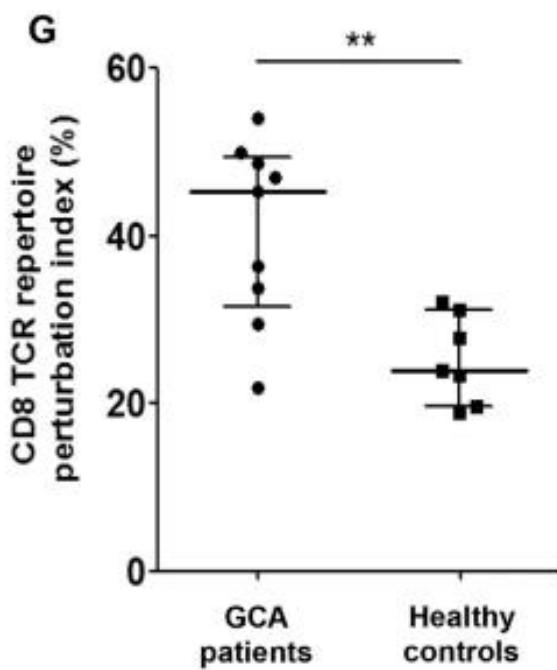
Corbera-Bellalta M et al. ARD 2015
Samson M et al. J of Autoimm 2016

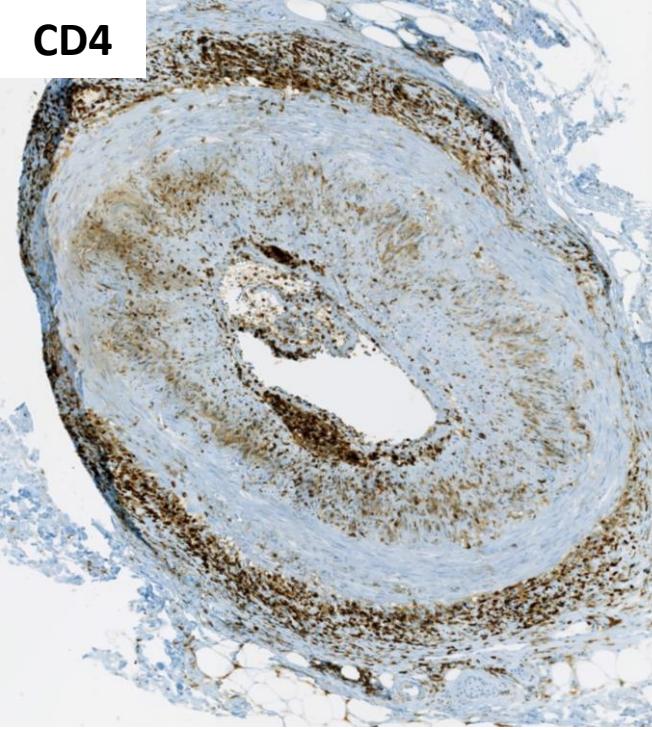
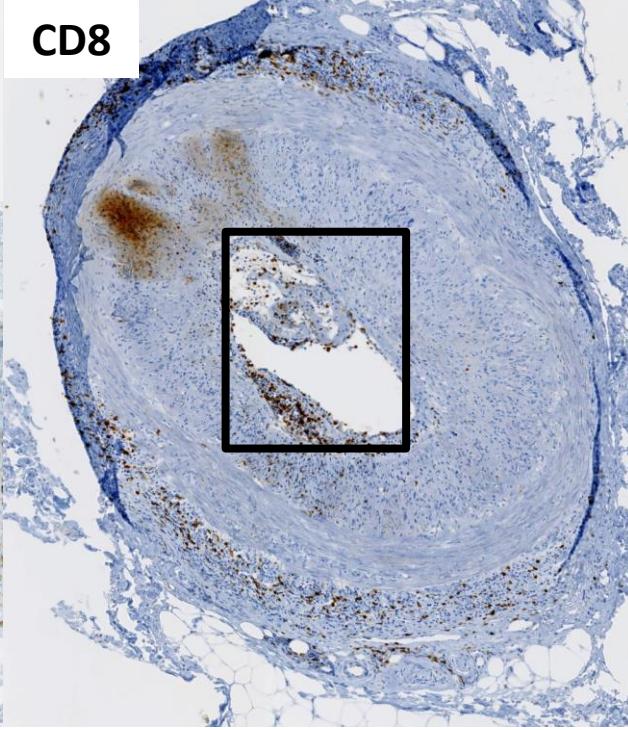
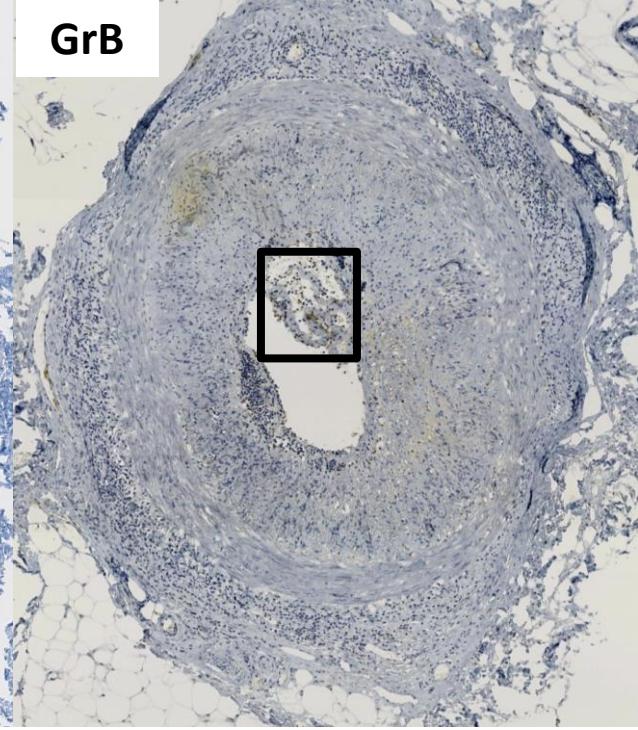
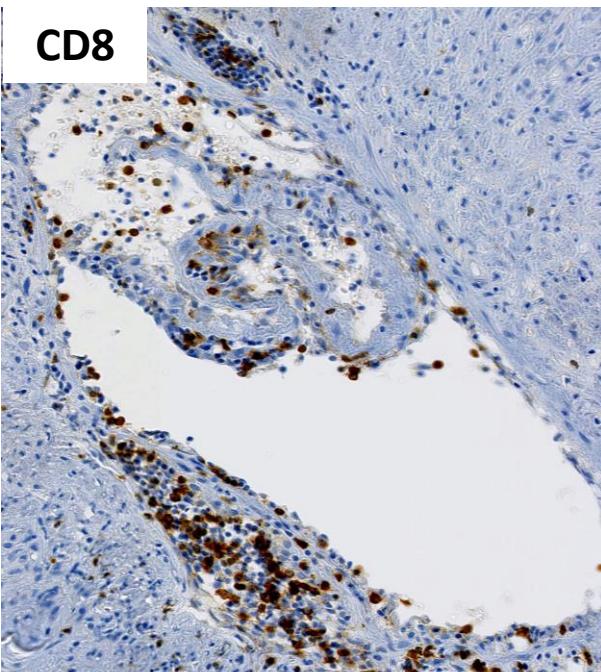
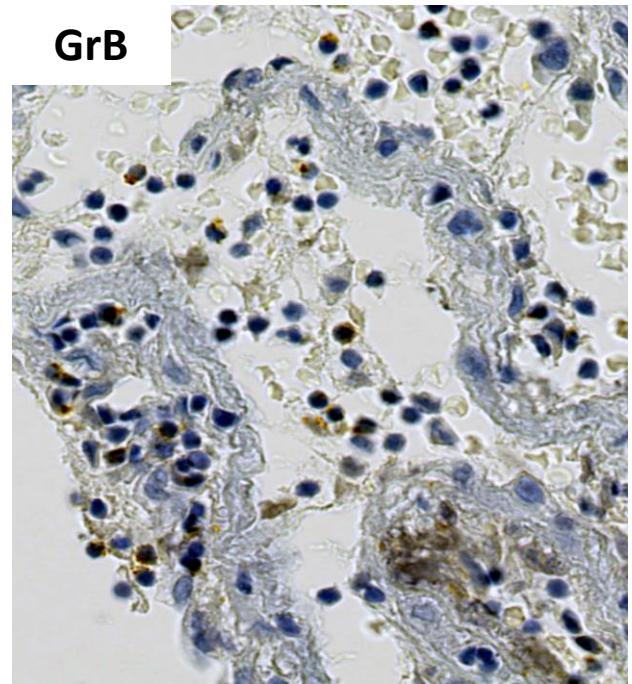
LT CD8 au cours de l'ACG



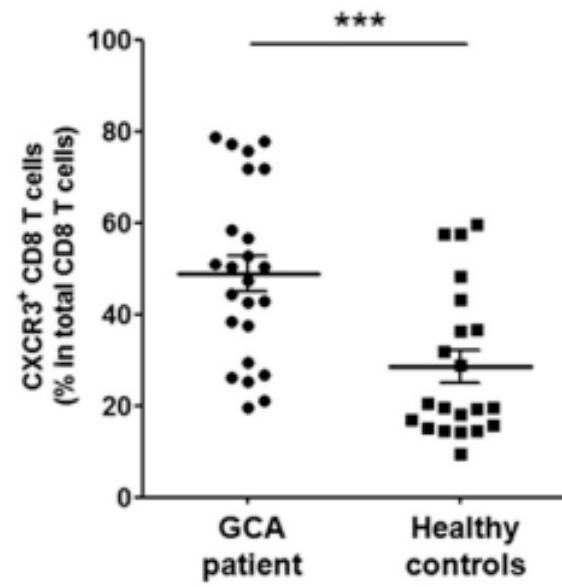
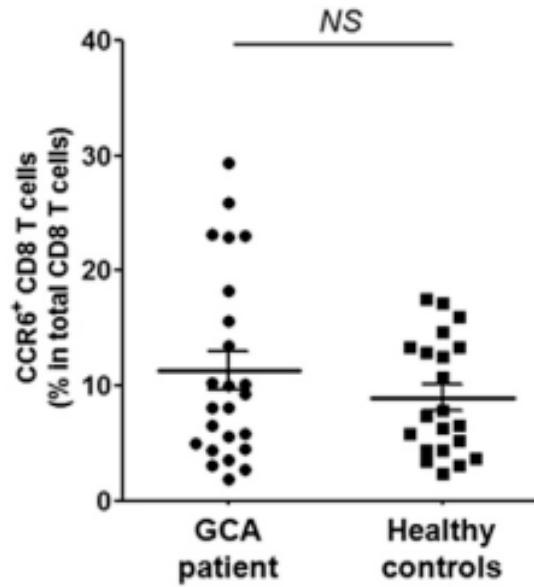
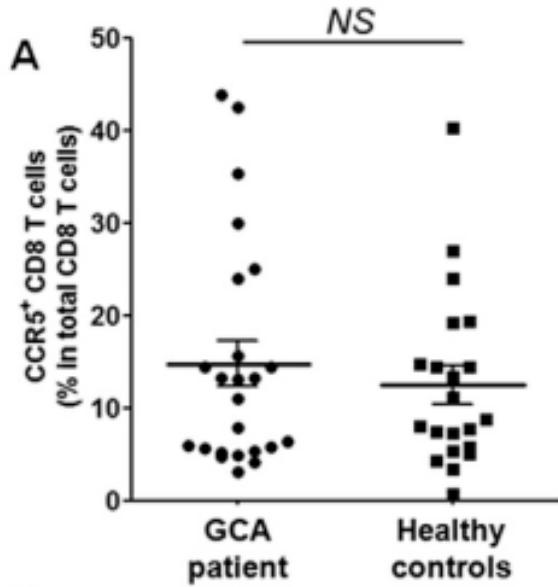
LT CD8 au cours de l'ACG

Le répertoire des LT CD8 est restreint au cours de l'ACG (stimulation oligoclonale)



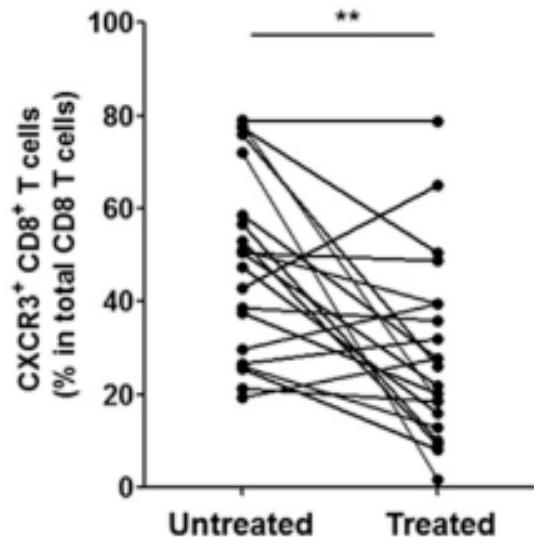
CD4**CD8****GrB****HES****CD8****GrB**

LT CD8 au cours de l'ACG

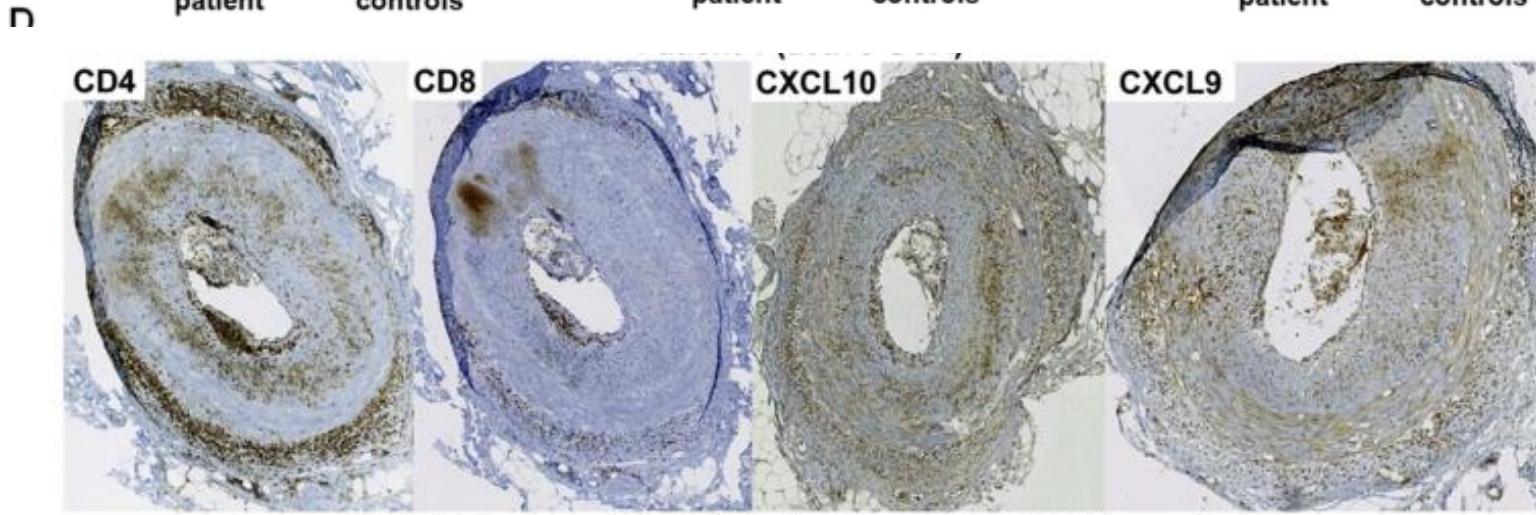
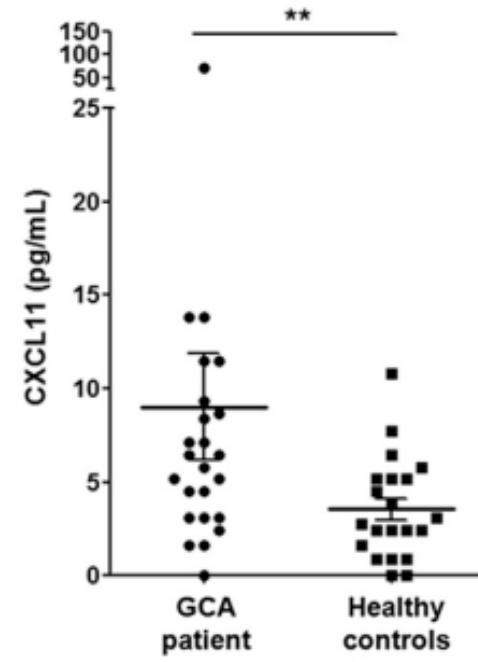
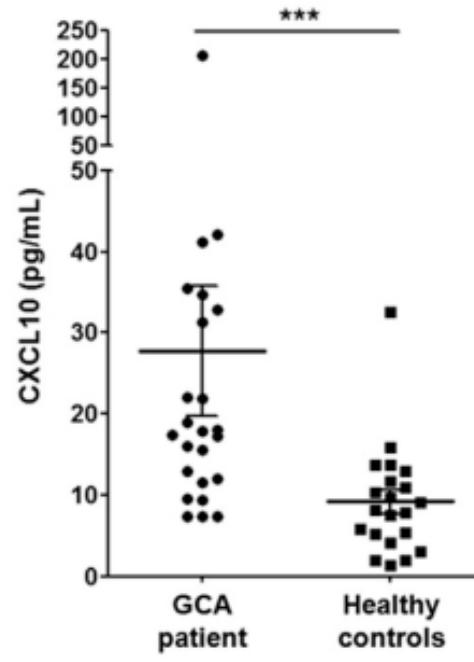
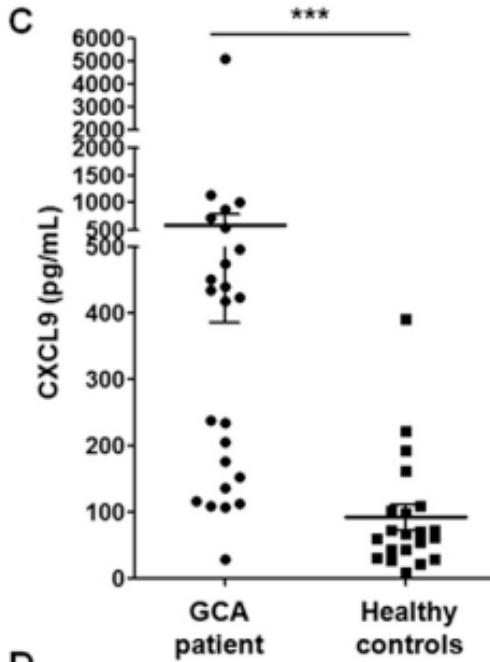


Les LT CD8 expriment surtout **CXCR3**

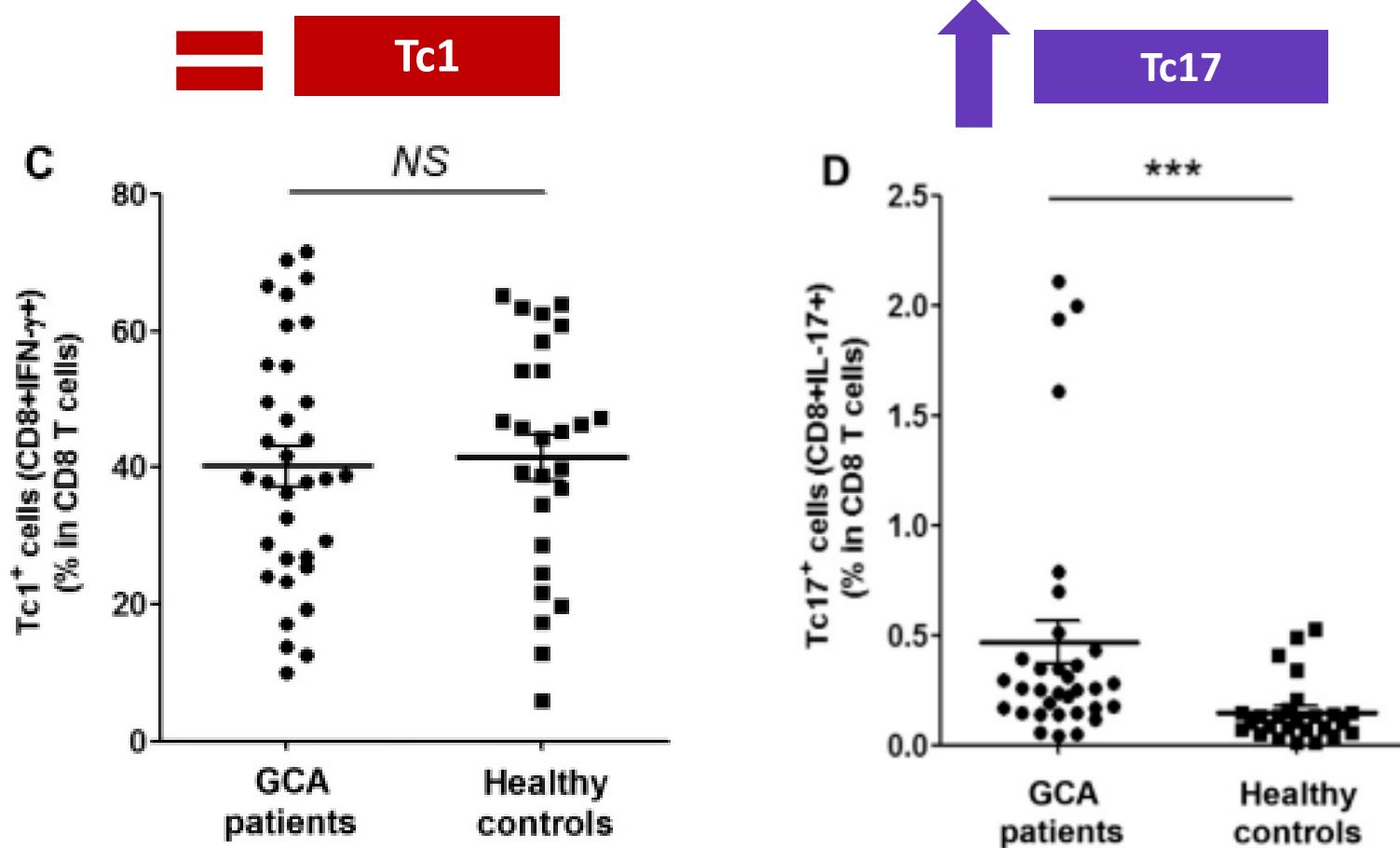
L'expression de CXCR3 est augmentée au cours de l'ACG et diminue après traitement par CS



LT CD8 au cours de l'ACG

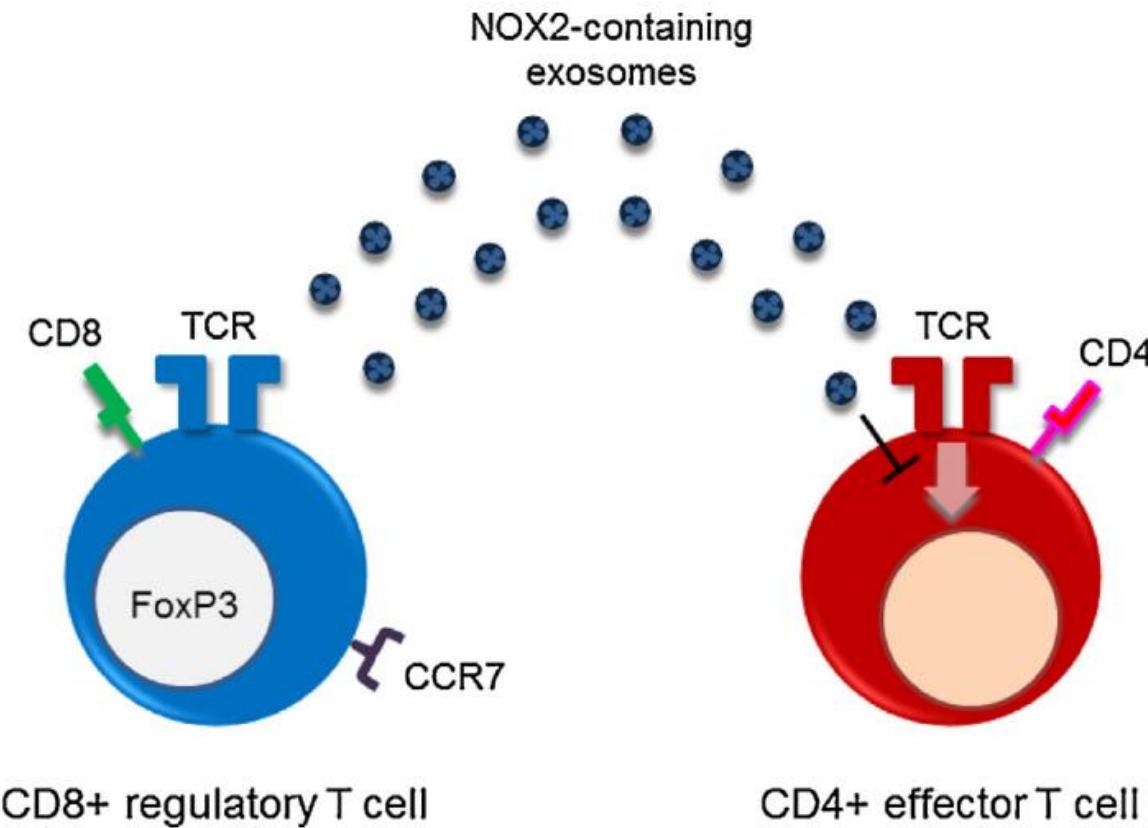


LT CD8 AU COURS DE L'ACG



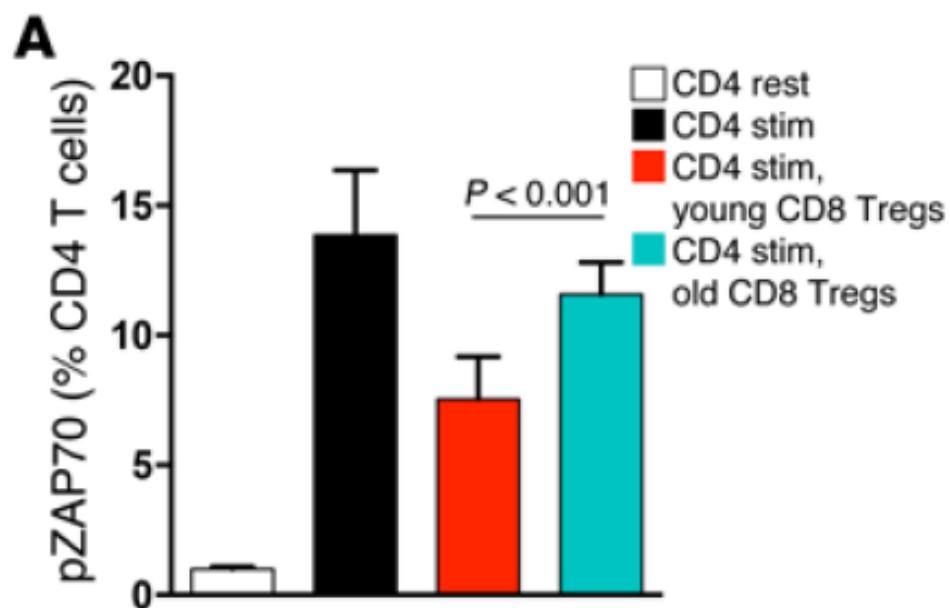
Implication des LT CD8reg au cours de l'ACG

CD8 reg (CD8⁺CCR7⁺FoxP3⁺)



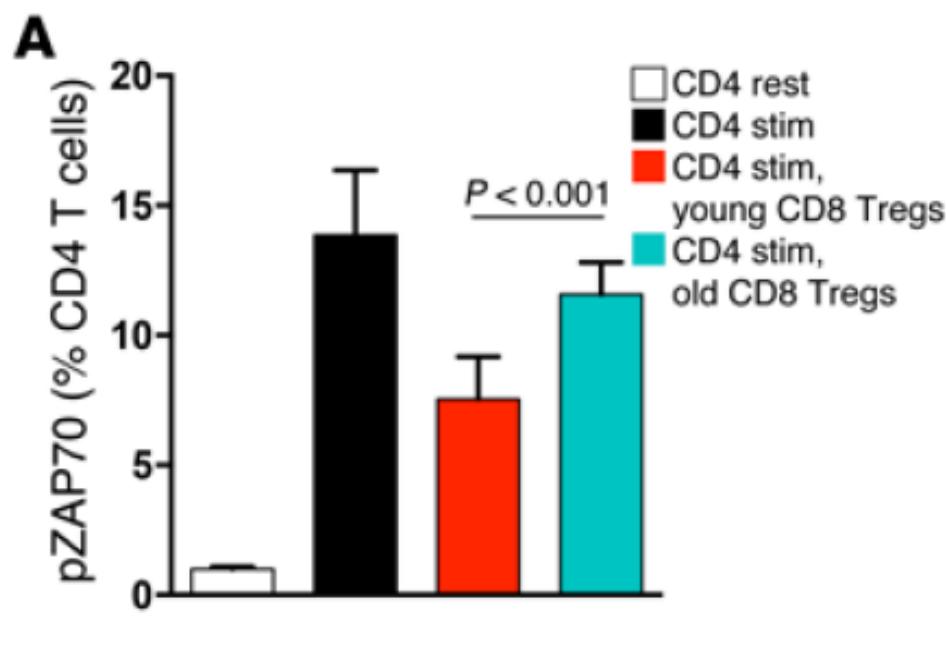
Implication des LT CD8reg au cours de l'ACG

Le vieillissement induit une altération fonctionnelle des CD8reg

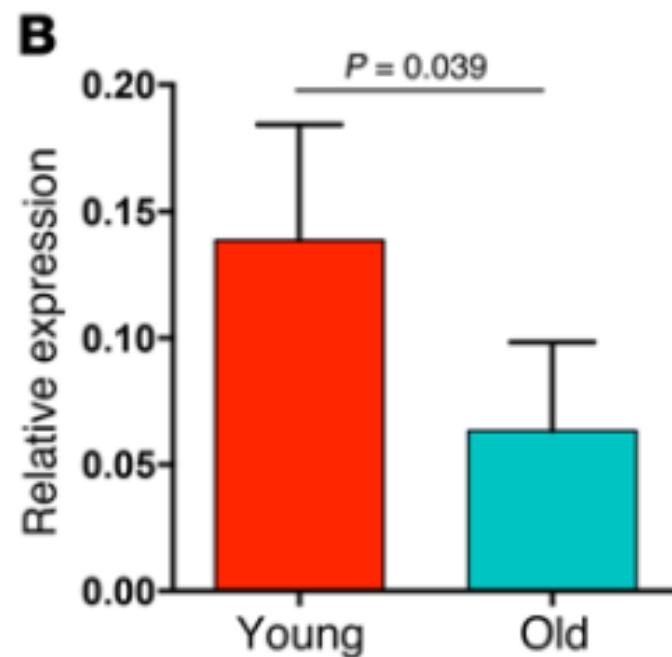


Implication des LT CD8reg au cours de l'ACG

Le vieillissement induit une altération fonctionnelle des CD8reg

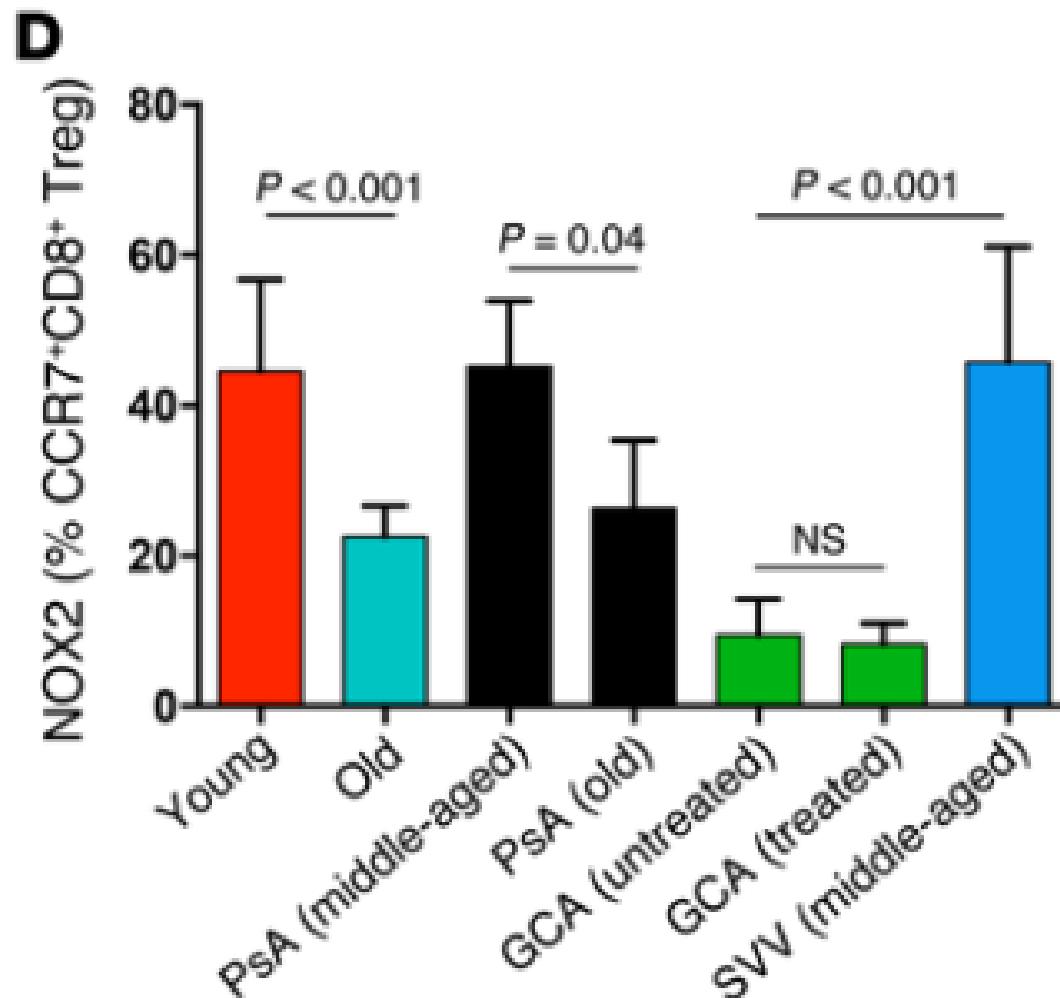


Cause = déficit en NOX2



Implication des LT CD8reg au cours de l'ACG

Vieillissement prématué des CD8reg au cours de l'ACG ?

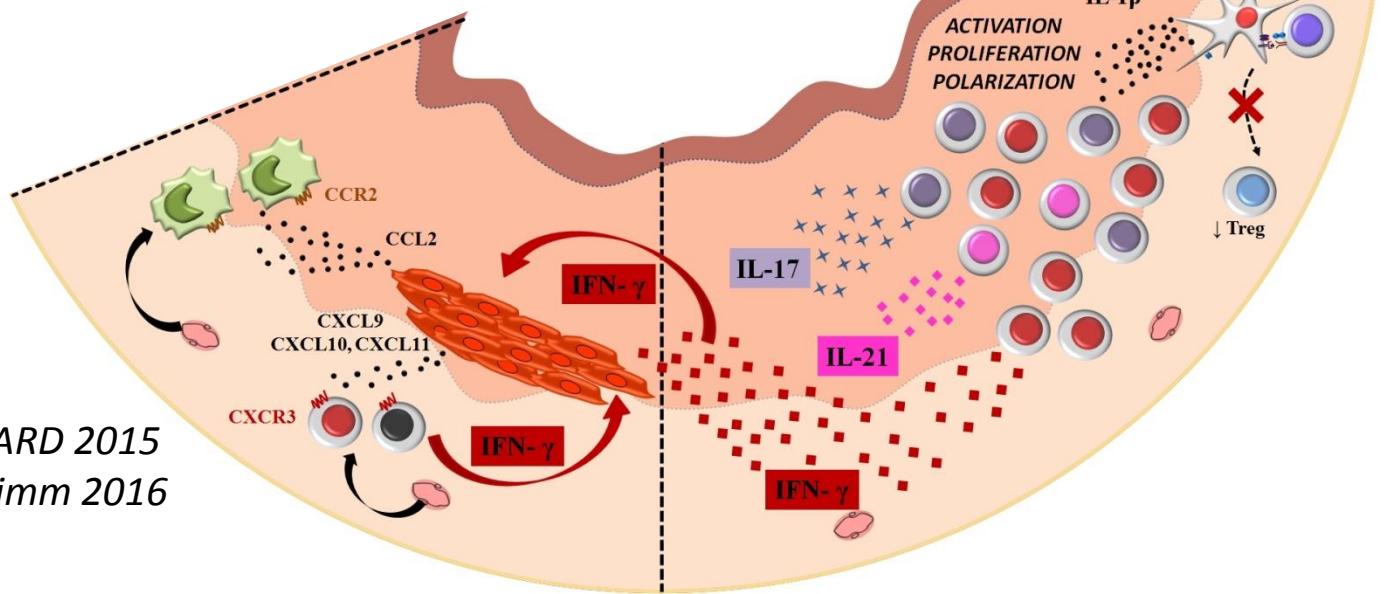
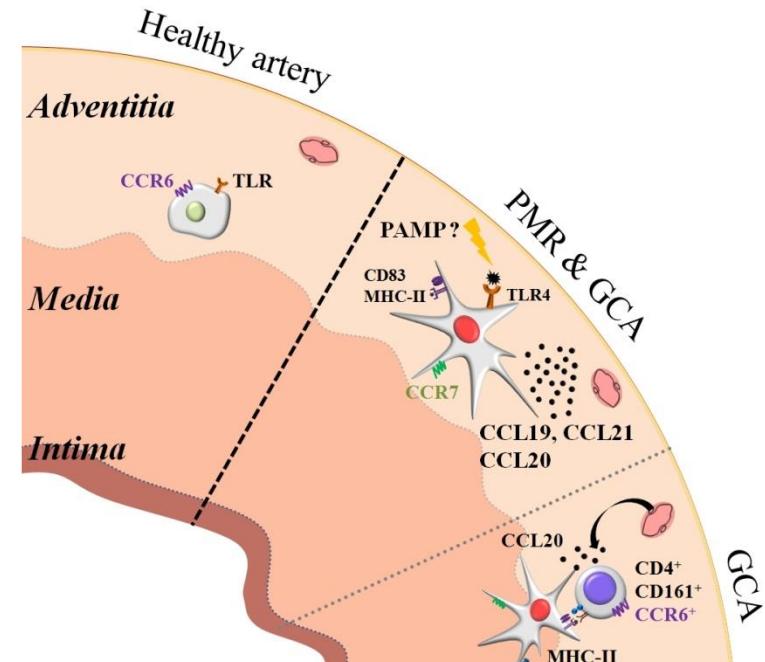


Recrutement des monocytes

Différenciation en macrophages et cellules géantes

Recrutement des LT CD8

- ↑ CTL
- ↑ Tc1 et Tc17
- ↓ fonction des CD8 reg (Nox2)



Corbera-Bellalta M et al. ARD 2015
Samson M et al. J of Autoimm 2016
Wen Z et al JCI 2016

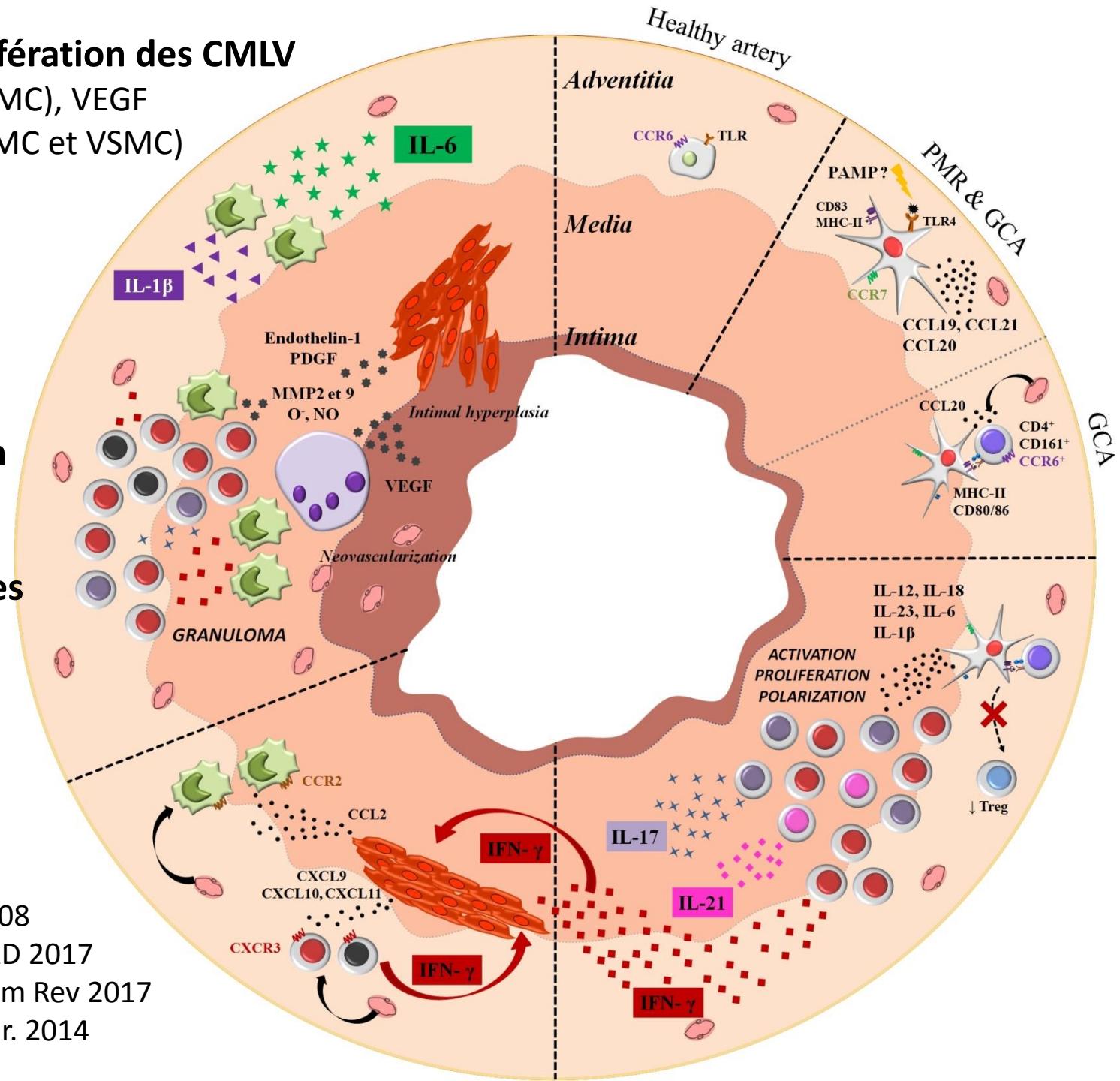
Migration et prolifération des CMLV

- PDGF (PBMC et VSMC), VEGF
- endotheline-1 (PBMC et VSMC)

↓
Neointima

↓
Sténose/occlusion

↓
Signes ischémiques



Lozano E et al. ARD 2008

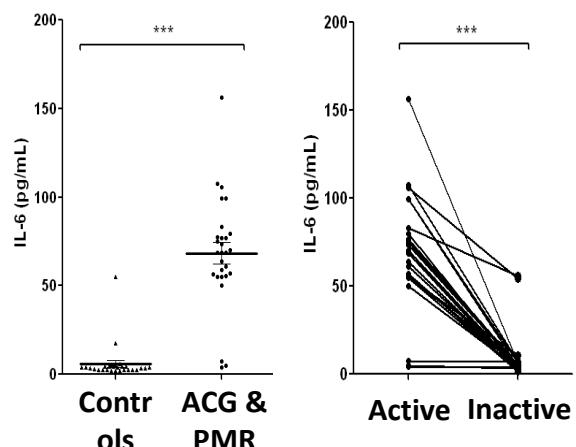
Planas Rigol E et al. ARD 2017

Regent A et al. Autoimm Rev 2017

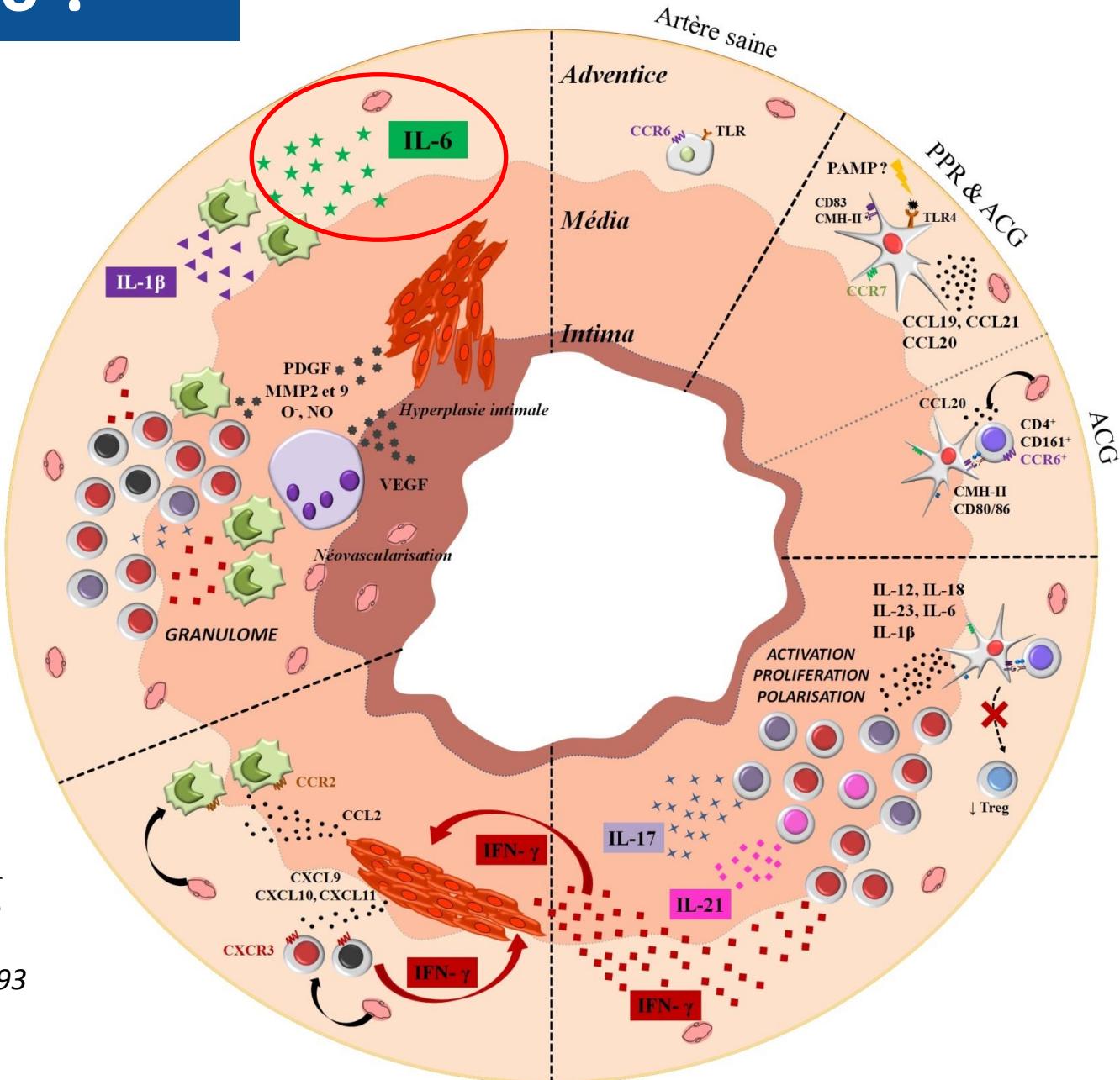
Ly KH Arthritis Res Ther. 2014

Cibler l'IL-6 ?

Corrélation IL-6 sérique et activité clinique



Roche N.E. et al. Arthritis Rheum 1993
Samson et al. Arthritis Rheum 2012



Amplification et recrutement des leucocytes

Espigol-Frigolé G et al. Science Signal 2016

\uparrow SAA

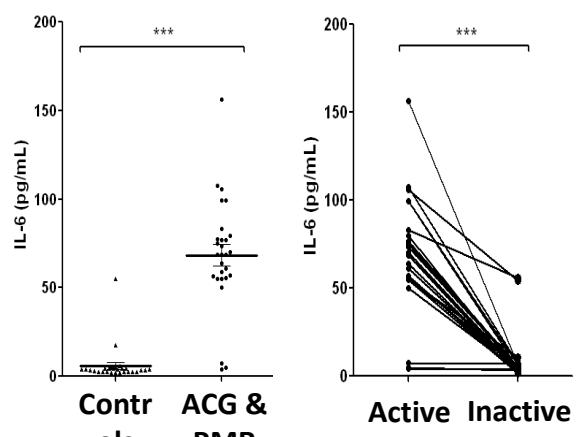
O'Neill L et al. A&R 2015

Athérosclérose

Johnson BD et al. Circulation 2004

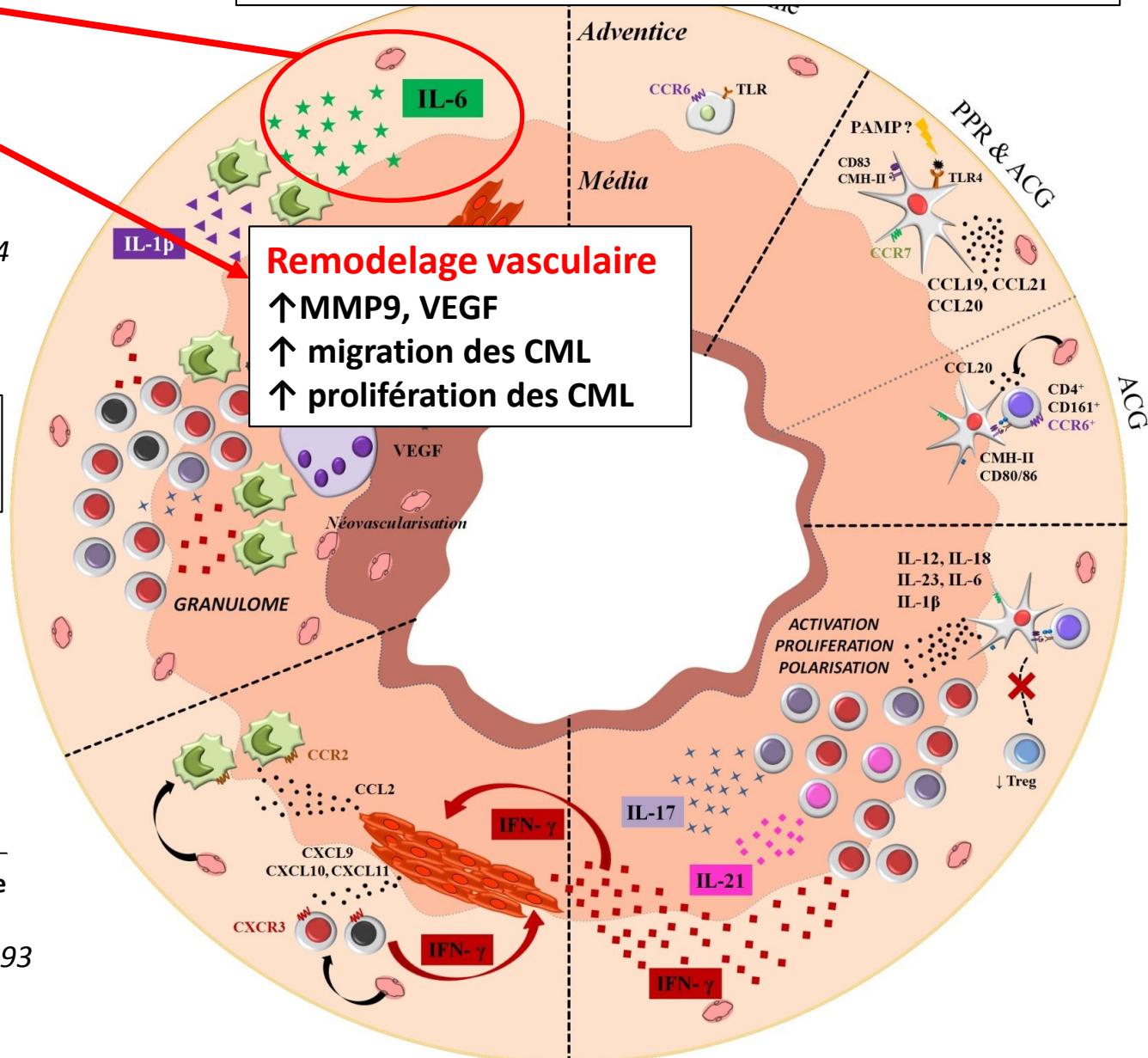
Lewis KE et al. Circulation 2004

Corrélation IL-6 sérique
et activité clinique



Remodelage vasculaire

- \uparrow MMP9, VEGF
- \uparrow migration des CML
- \uparrow prolifération des CML



Roche N.E. et al. Arthritis Rheum 1993

Samson et al. Arthritis Rheum 2012

Amplification et recrutement des leucocytes

Espigol-Frigolé G et al. Science Signal 2016

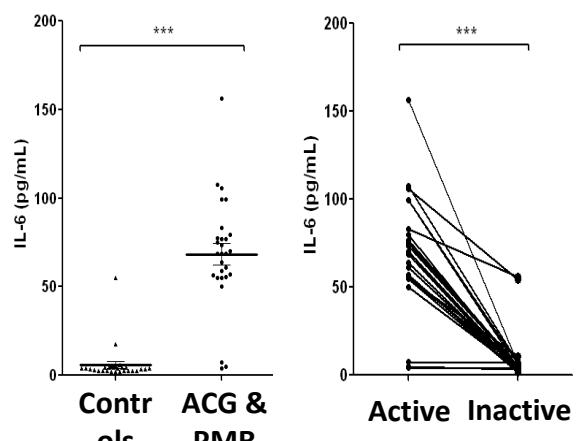
\uparrow SAA

O'Neill L et al. A&R 2015

Athérosclérose

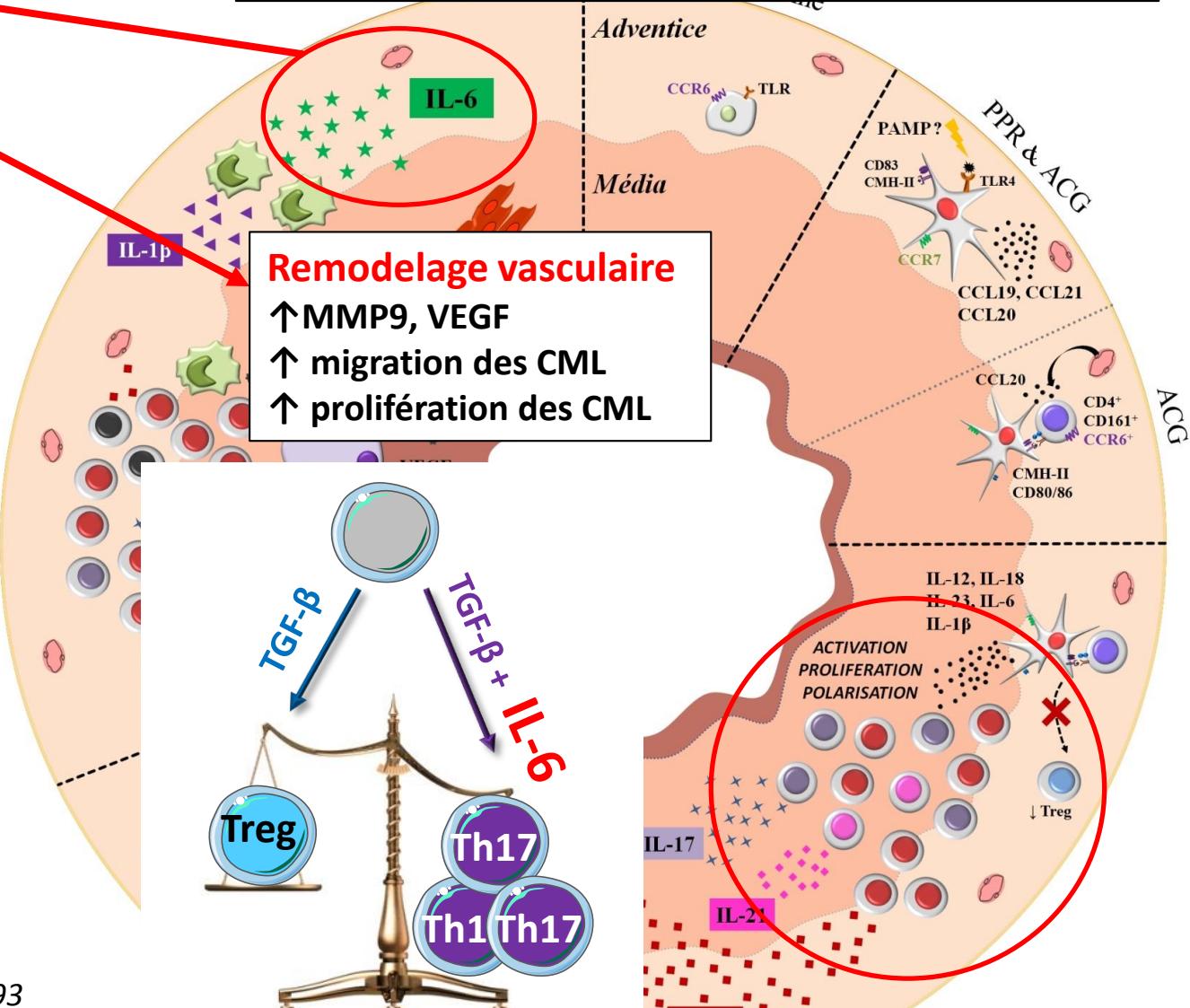
Johnson BD et al. Circulation 2004
Lewis KE et al. Circulation 2004

Corrélation IL-6 sérique
et activité clinique



Roche N.E. et al. Arthritis Rheum 1993
Samson et al. Arthritis Rheum 2012

Bettelli E et al. Nature 2006
Samson M et al. Arthritis Rheum 2012



Polarisation
des LT CD4+

Tocilizumab for induction and maintenance of remission in giant cell arteritis: a phase 2, randomised, double-blind, placebo-controlled trial

Peter M Villiger*, Sabine Adler*, Stefan Kuchen, Felix Wermelinger, Diana Dan, Veronika Fiege, Lukas Bütkofer, Michael Seitz, Stephan Reichenbach

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ESTABLISHED IN 1812

JULY 27, 2017

VOL. 377 NO. 4

Trial of Tocilizumab in Giant-Cell Arteritis

J.H. Stone, K. Tuckwell, S. Dimonaco, M. Klearman, M. Aringer, D. Blockmans, E. Brouwer, M.C. Cid, B. Dasgupta, J. Rech, C. Salvarani, G. Schett, H. Schulze-Koops, R. Spiera, S.H. Unizony, and N. Collinson

Autres voies impliquées dans la physiopathologie de l'ACG

- **Lymphocytes B et réponse immunitaire à médiation humorale**
(auto-anticorps, IL-33, organes lymphoïdes tertiaires)
- **Vieillissement immunitaire**
- **Voie NOTCH (via VEGF et JAGGED1)**
- **Neutrophiles**
- **Certaines sous-populations de monocytes**

CHU de Dijon

Médecine Interne et Immunologie Clinique

Médecine Interne et Maladies Systémiques

Rhumatologie

Médecine Interne Gériatrique

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Hôpital Belle Isle Metz

Hospital Clinic, Barcelona

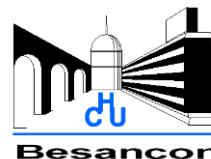
Dr M.C. Cid

Vasculitis Research Unit - IDIBAPS

Hospital Clínic, University of Barcelona



Centre Hospitalier Universitaire Dijon



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Dr S. AUDIA

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M. CIUDAD

M. THEBAULT

A. GAUTHERON

M. TRAD

C. GERARD

Anatomie et Cytologie Pathologiques

Pr L. MARTIN, Dr B. TOURNIER

Epidémiologie

Dr AHO, Dr DEVILLIERS

Plateforme du cytométrie , IFR100

A. SEQUEIRA , A. HAMMANN, S. MONIER