BASIC SCIENCE SUMMER SCHOOL

SUNDAY 18 - THURSDAY 22 JUNE 2017 (5 DAYS), SOPHIA-ANTIPOLIS - FRANCE

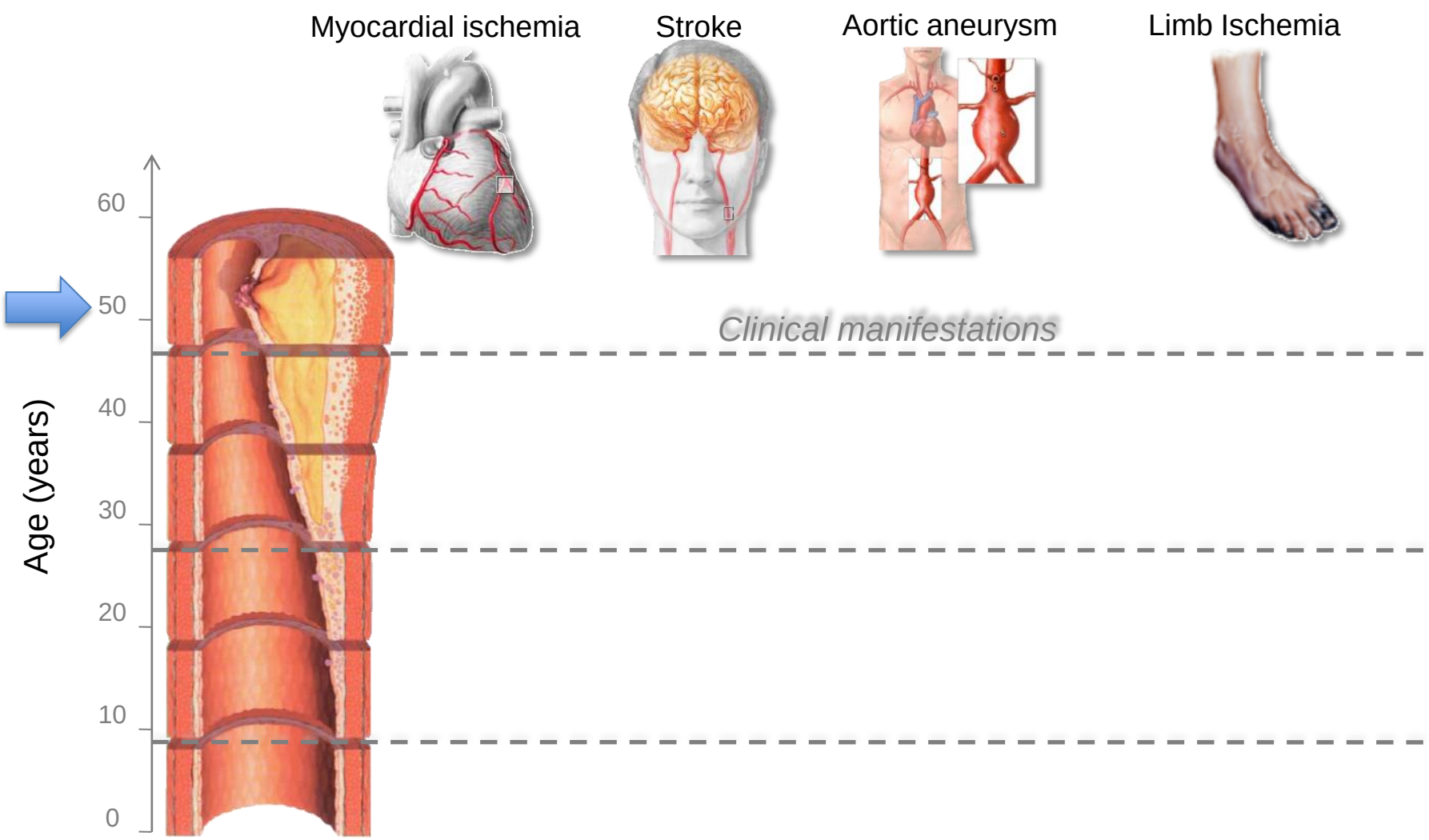
The roles of T and B cells

Giuseppina Caligiuri

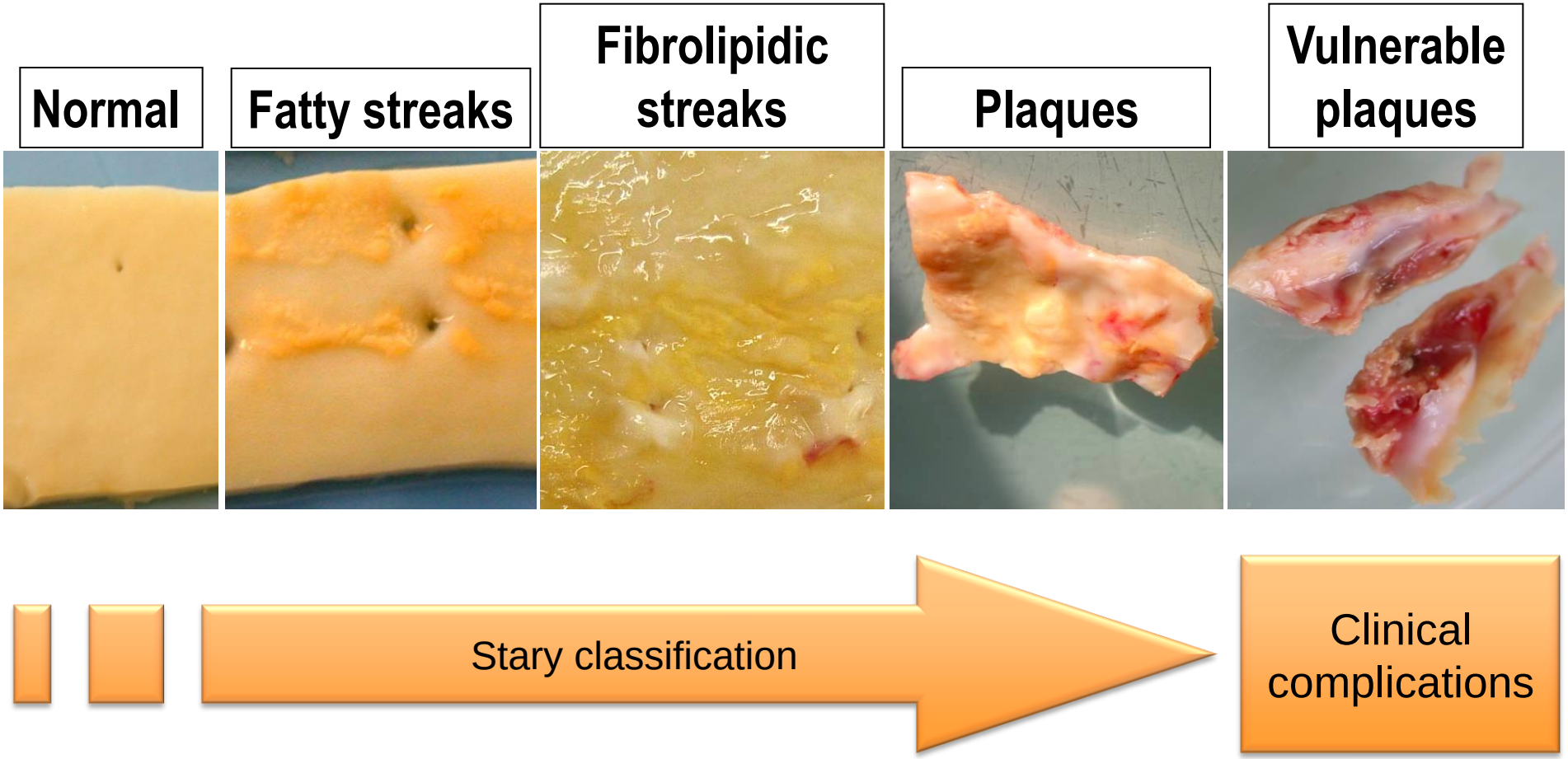
Laboratory for Vascular Translational Science

University Hospital Bichat, Paris, FR

Atherosclerotic diseases



Atherosclerosis



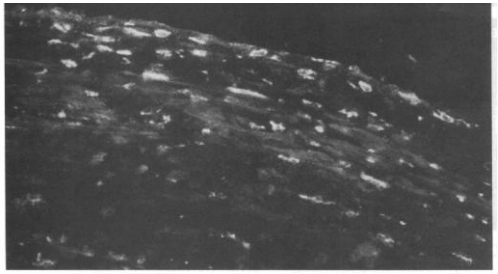
Expression of Class II Transplantation Antigen on Vascular Smooth Muscle Cells in Human Atherosclerosis

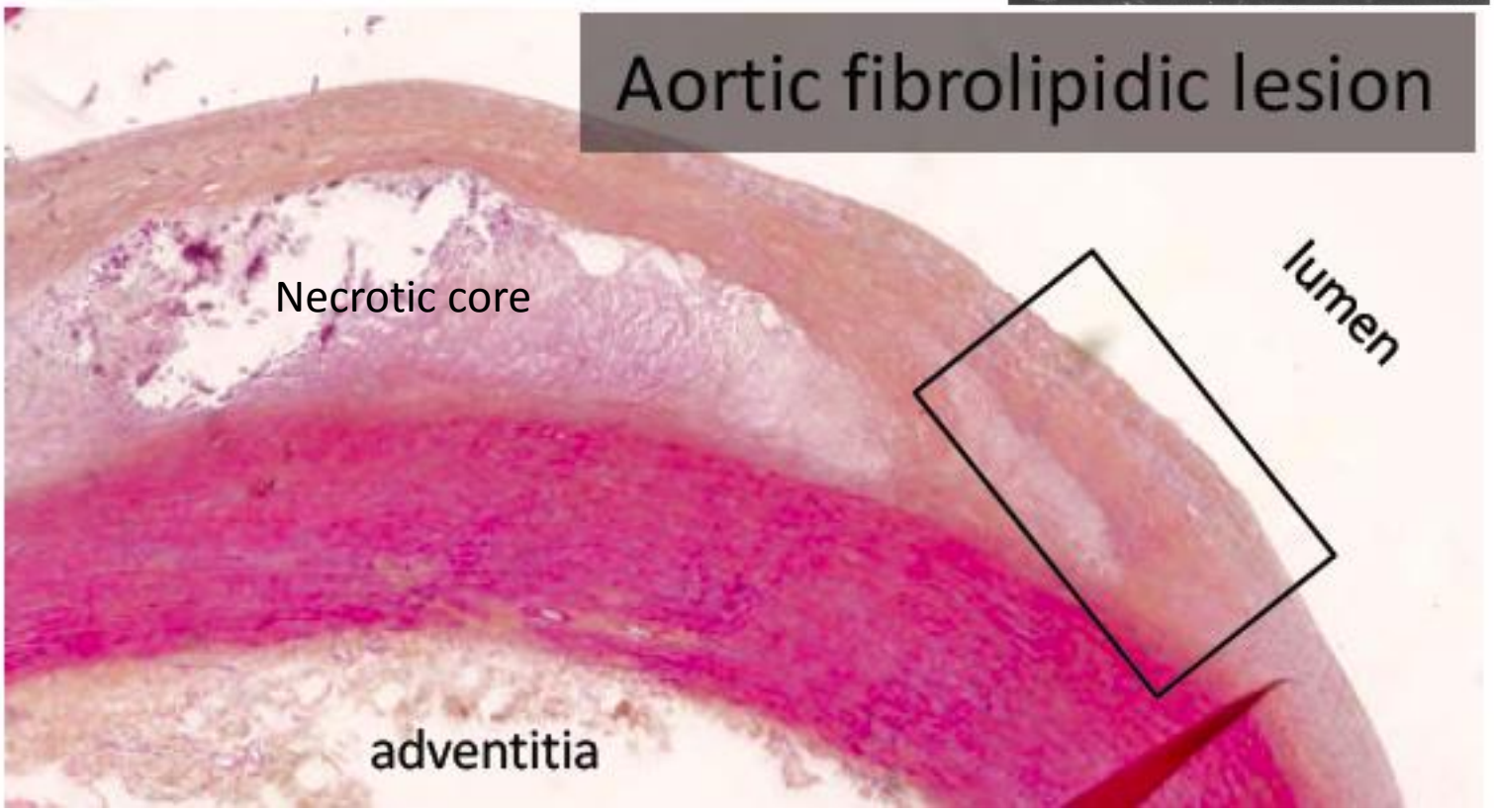
Lena Jonasson, Jan Holm, Omar Skalli, Giulio Gabbiani, and Göran K. Hansson

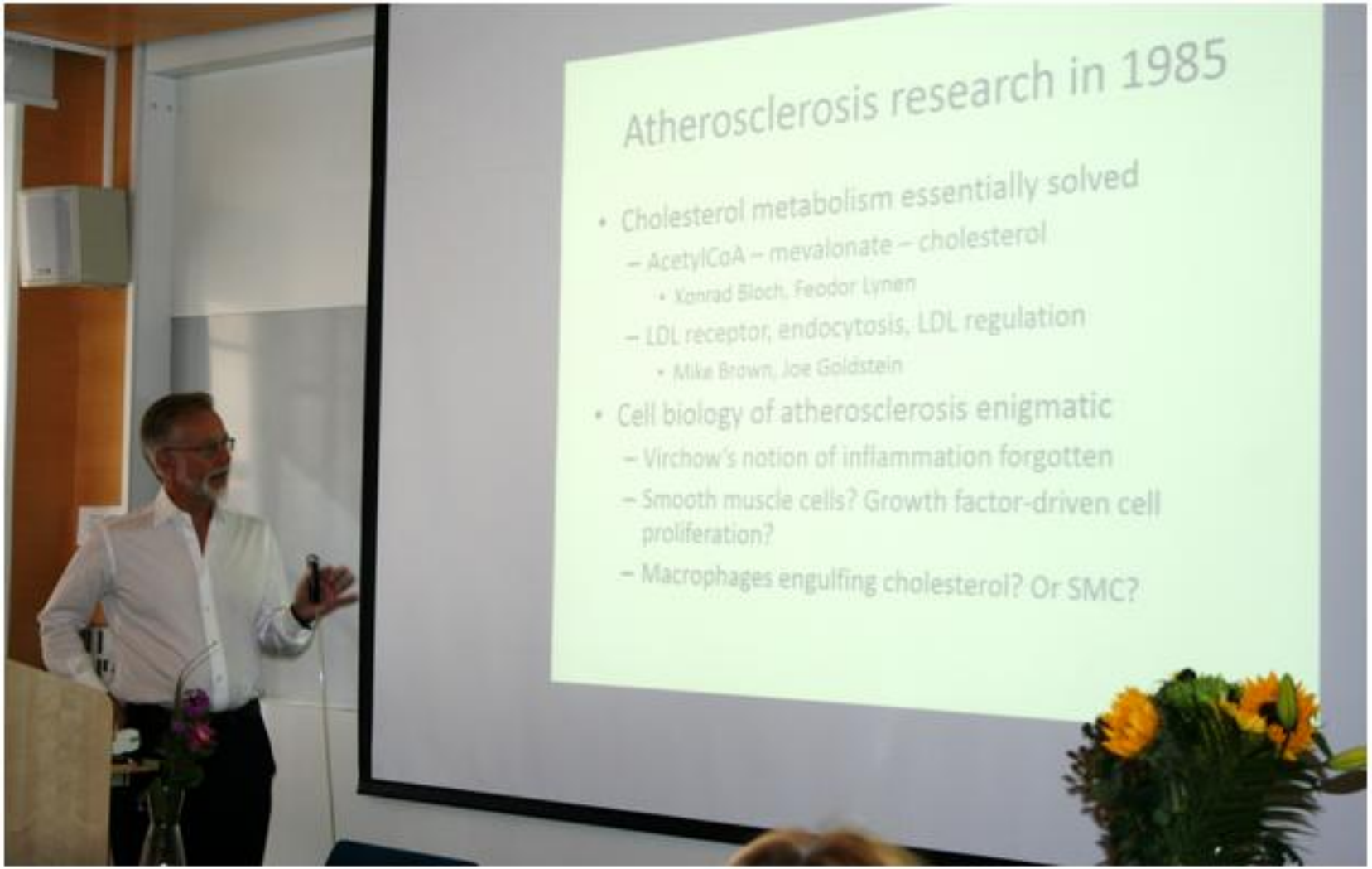
Arterial Biology Group, Departments of Histology and Medicine I, University of Göteborg, Sweden; Department of Surgery I, University of Göteborg, Sahlgrenska Hospital, Göteborg, Sweden; and Department of Pathology, University of Geneva, Switzerland

J. Clin. Invest.

0021-9738/85/07/0125/07 $1.00
Volume 76, July 1985, 125–131

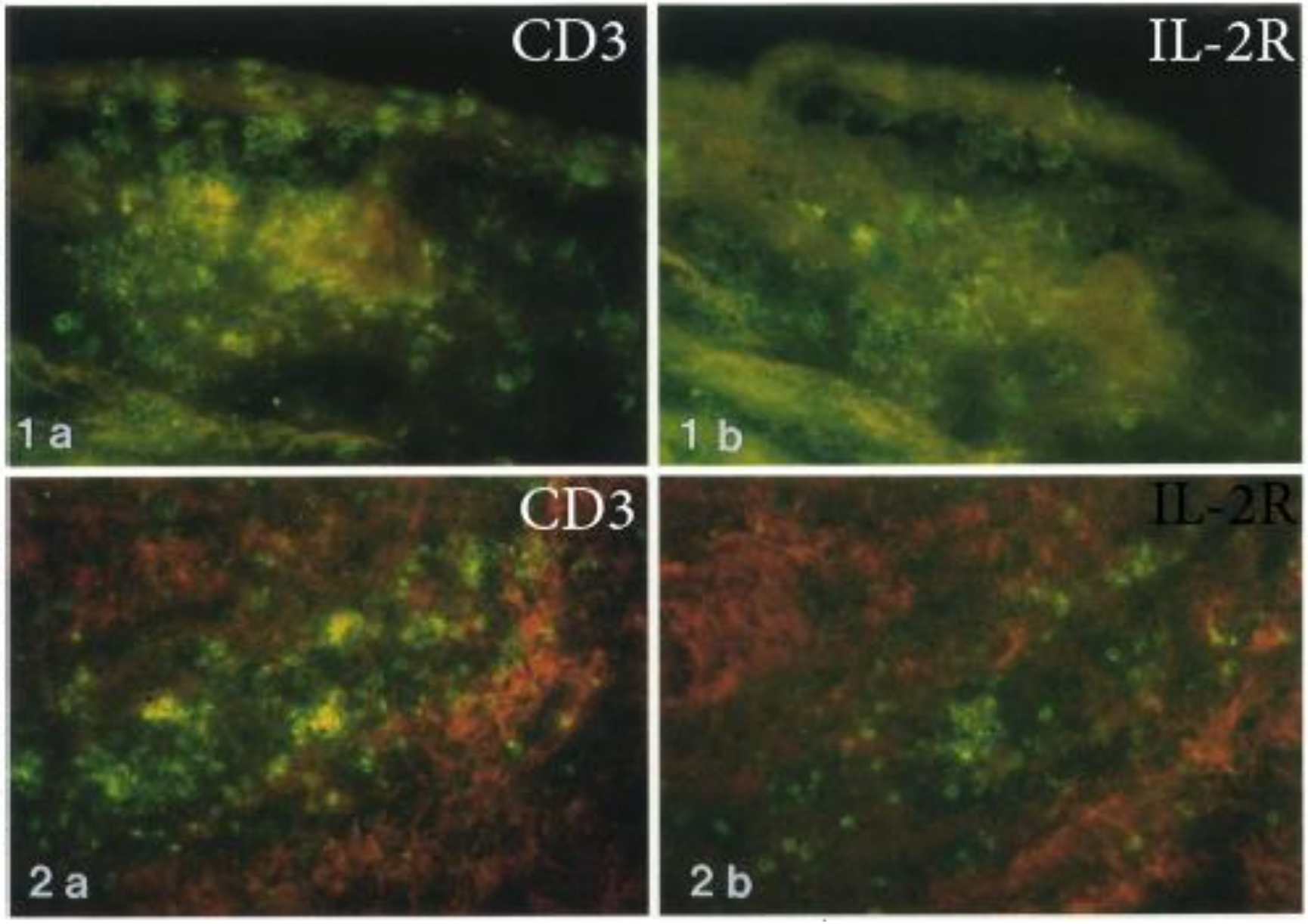




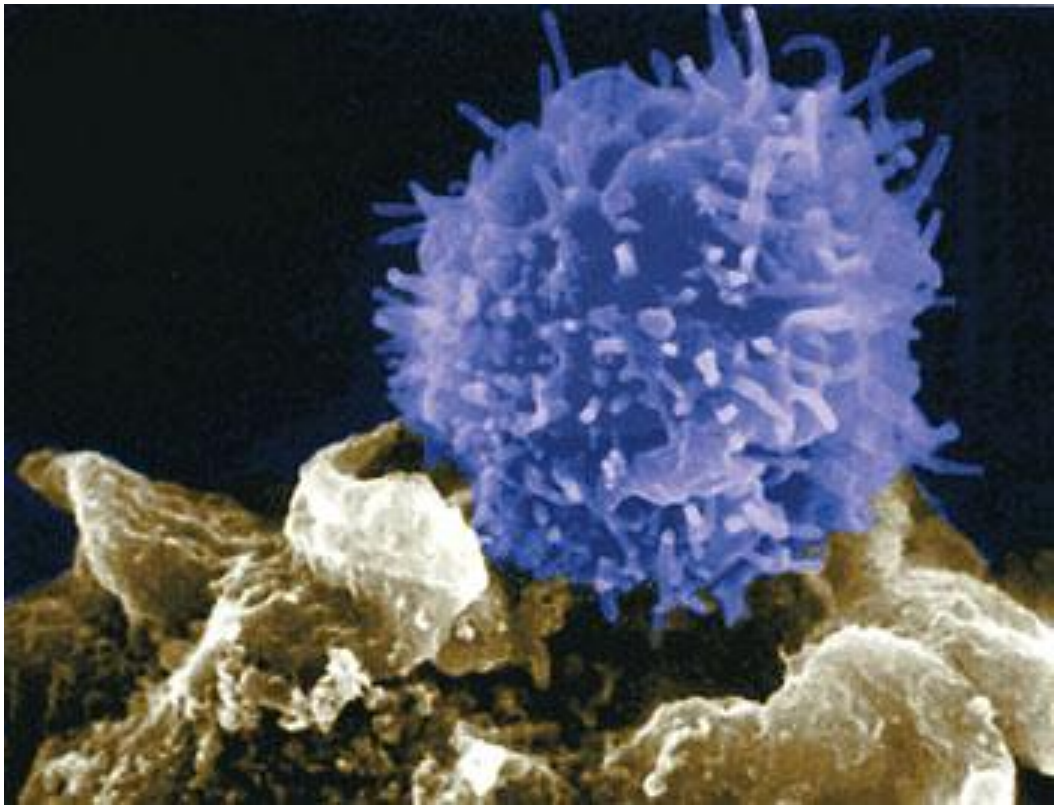


Göran K Hansson, CMM Karolinska Institutet, Stockholm, Sweden

Detection of Activated T Lymphocytes in the Human Atherosclerotic Plaque



T cells



Proc. Natl. Acad. Sci. USA
Vol. 92, pp. 3893–3897, April 1995
Medical Sciences

T lymphocytes from human atherosclerotic plaques recognize oxidized low density lipoprotein

(antigen/atherosclerosis/immune response/oxidation)

STEN STEMME*†, BEATA FABER*, JAN HOLM‡, OLOV WIKLUND§, JOSEPH L. WITZTUM¶, AND GÖRAN K. HANSSON*

ABSTRACT **Atherosclerosis, an underlying cause of myocardial infarction, stroke, and other cardiovascular diseases, consists of focal plaques characterized by cholesterol deposition, fibrosis, and inflammation. The presence of activated T lymphocytes and macrophages and high expression of HLA class II molecules are indicative of a local immunologic activation in the atherosclerotic plaque, but the antigen(s) involved has not yet been identified. We established T-cell clones from human atherosclerotic plaques using polyclonal mitogens as stimuli and exposed the clones to potential antigens in the presence of autologous monocytes as antigen-presenting cells. Four of the 27 CD4$^+$ clones responded to oxidized low density lipoprotein (oxLDL) by proliferation and cytokine secretion; this response was dependent on autologous antigen-presenting cells and restricted by HLA-DR. All clones that responded to oxLDL secreted interferon γ upon activation, but only one produced interleukin 4, suggesting that the response to oxLDL results in immune activation and inflammation but may not be a strong stimulus to antibody production. No significant response to oxLDL could be detected in CD4$^+$ T-cell clones derived from the peripheral blood of the same individuals. Together, the present data suggest that the inflammatory infiltrate in the atherosclerotic plaque is involved in a T-cell-dependent, autoimmune response to oxLDL.**

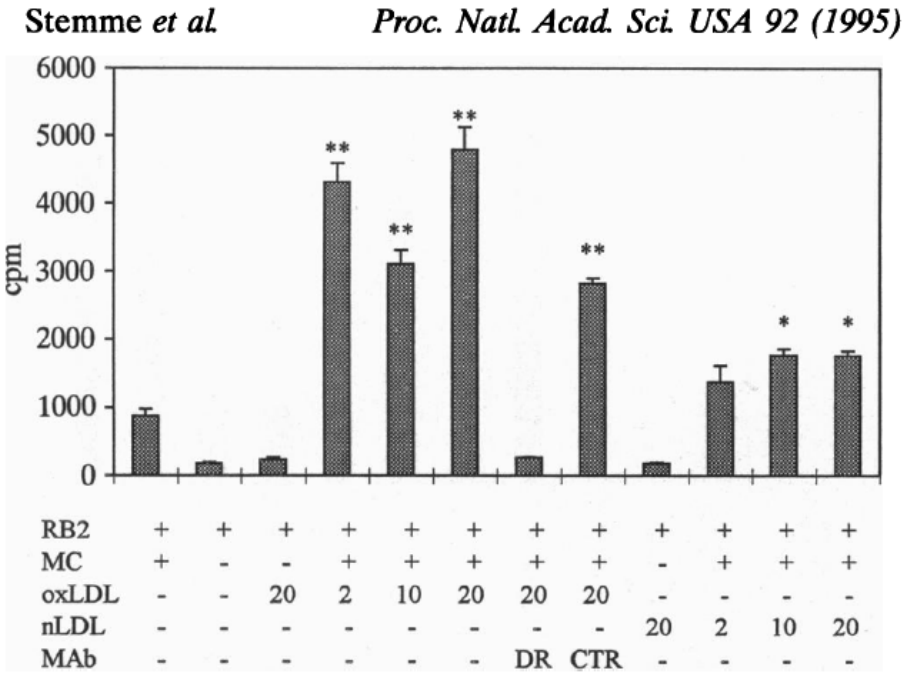


FIG. 1. Plaque T-cell clone RB2 was activated by oxLDL at concentrations of 2–20 μg/ml but not by nLDL. Activation was dependent on the presence of autologous PBMC (MC) and inhibited by the anti-HLA-DR monoclonal antibody L243 (DR)

Evidence for Antigen-Driven T-Cell Response in Unstable Angina

Giuseppina Caligiuri, MD, PhD; Gabrielle Paulsson, PhD; Antonino Nicoletti, PhD; Attilio Maseri, MD; Göran K. Hansson, MD, PhD

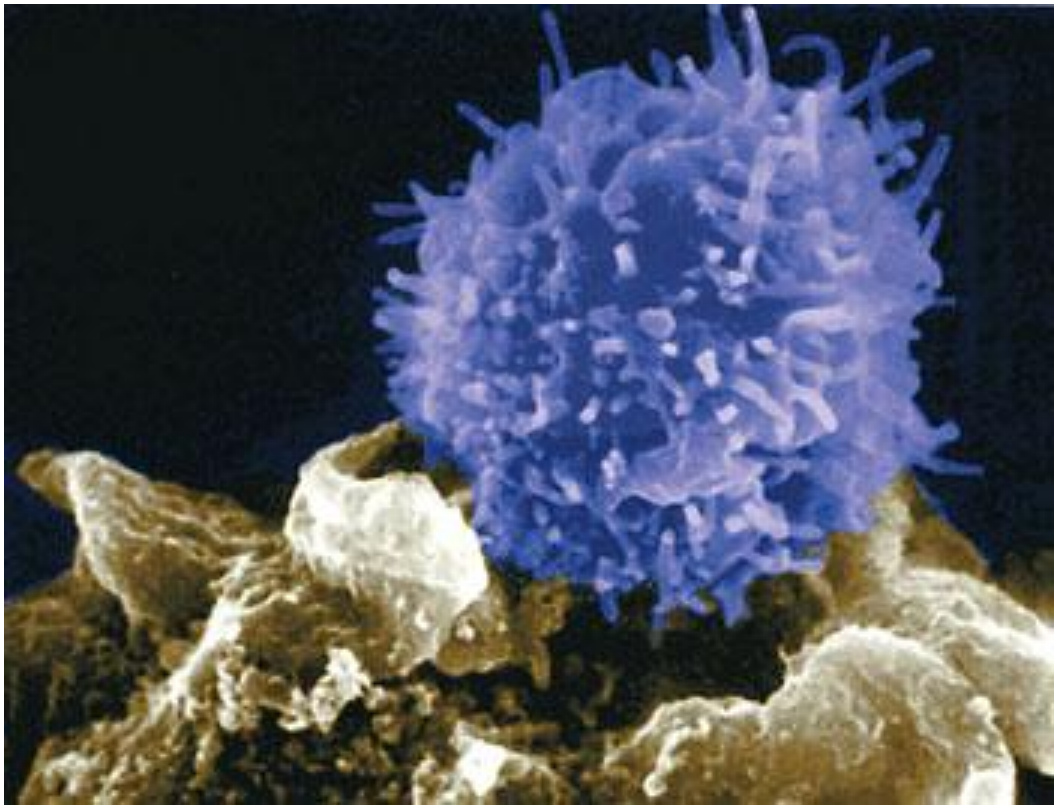
Background—Activation of T cells and macrophages has been associated with unstable angina (UA), but whether this reflects specific immune responses remains unclear.

Methods and Results—We analyzed the repertoire and the length of complementarity-determining region 3 of the T-cell receptor (TCR) β-chain variable (BV) gene segments of activated lymphocytes in 23 patients with UA, 13 patients with chronic stable angina (CSA), and 6 normal control subjects. We also tested the proliferation of systemic T cells in response to autologous coronary plaque proteins, oxidized LDL, and *Chlamydia pneumoniae* as candidate antigens, in vitro. The activated T cell–TCRBV repertoire was perturbed in 13 (57%) of 23 UA patients versus 3 (23%) of 13 CSA patients ($P=0.016$) and was restricted to 6 (28%) of 21 expanded TCRBV families; all were significantly higher in UA than in CSA patients. At least one monotypic or oligotypic activated TCRBV population was found in 15 (65%) of 23 UA patients and in 3 (23%) of 13 CSA patients ($P<0.001$). Finally, T cells from UA patients, but not from CSA patients or normal control subjects, proliferated in response to autologous proteins from coronary culprit lesions and/or to oxidized LDL.

Conclusions—Our findings suggest that the T-cell response observed in UA patients is antigen-driven and directed to antigens contained in the culprit coronary atherosclerotic plaques. (***Circulation*. 2000;102:1114-1119.**)

Key Words: angina ■ ischemia ■ prognosis ■ lymphocytes ■ antigens

T cells

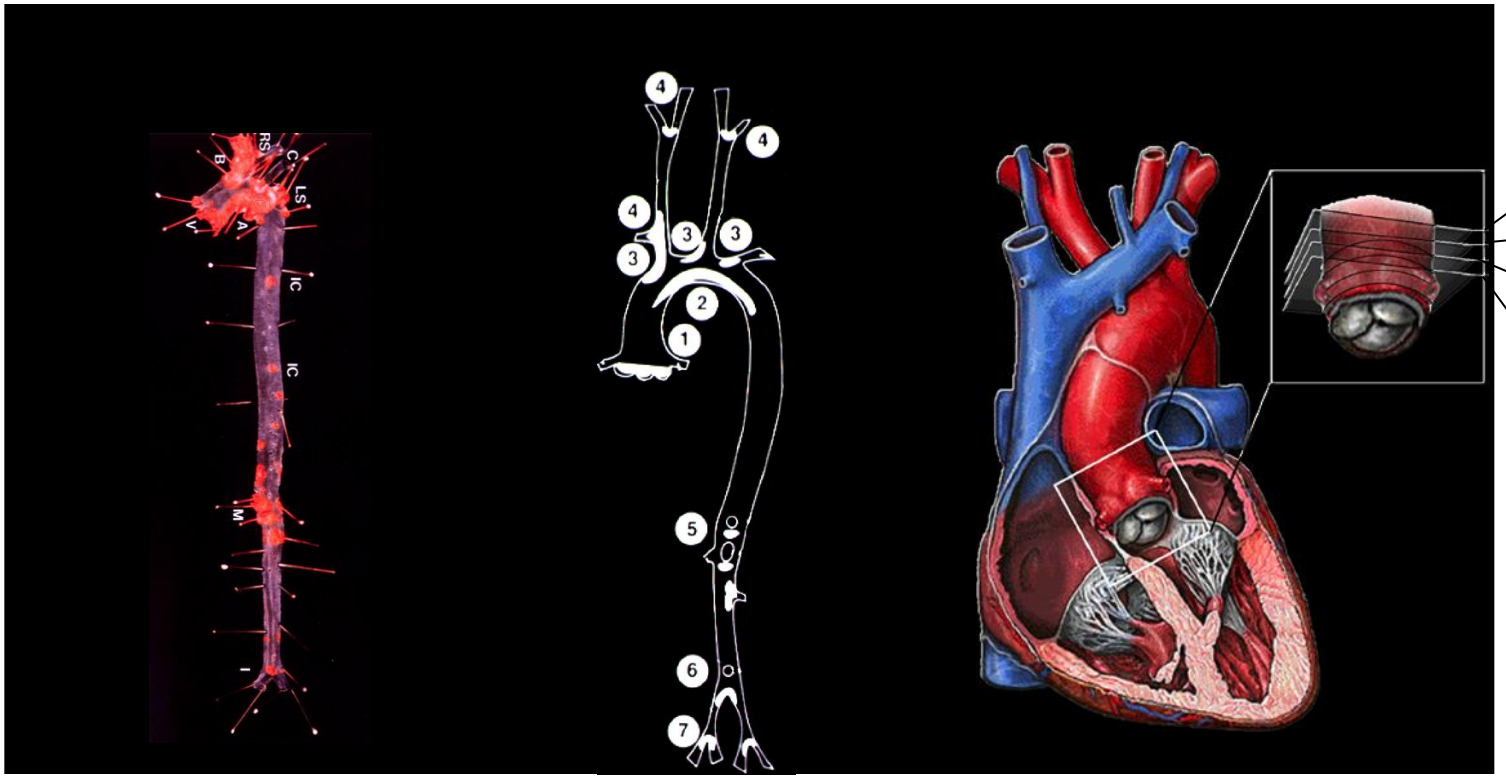


Role?

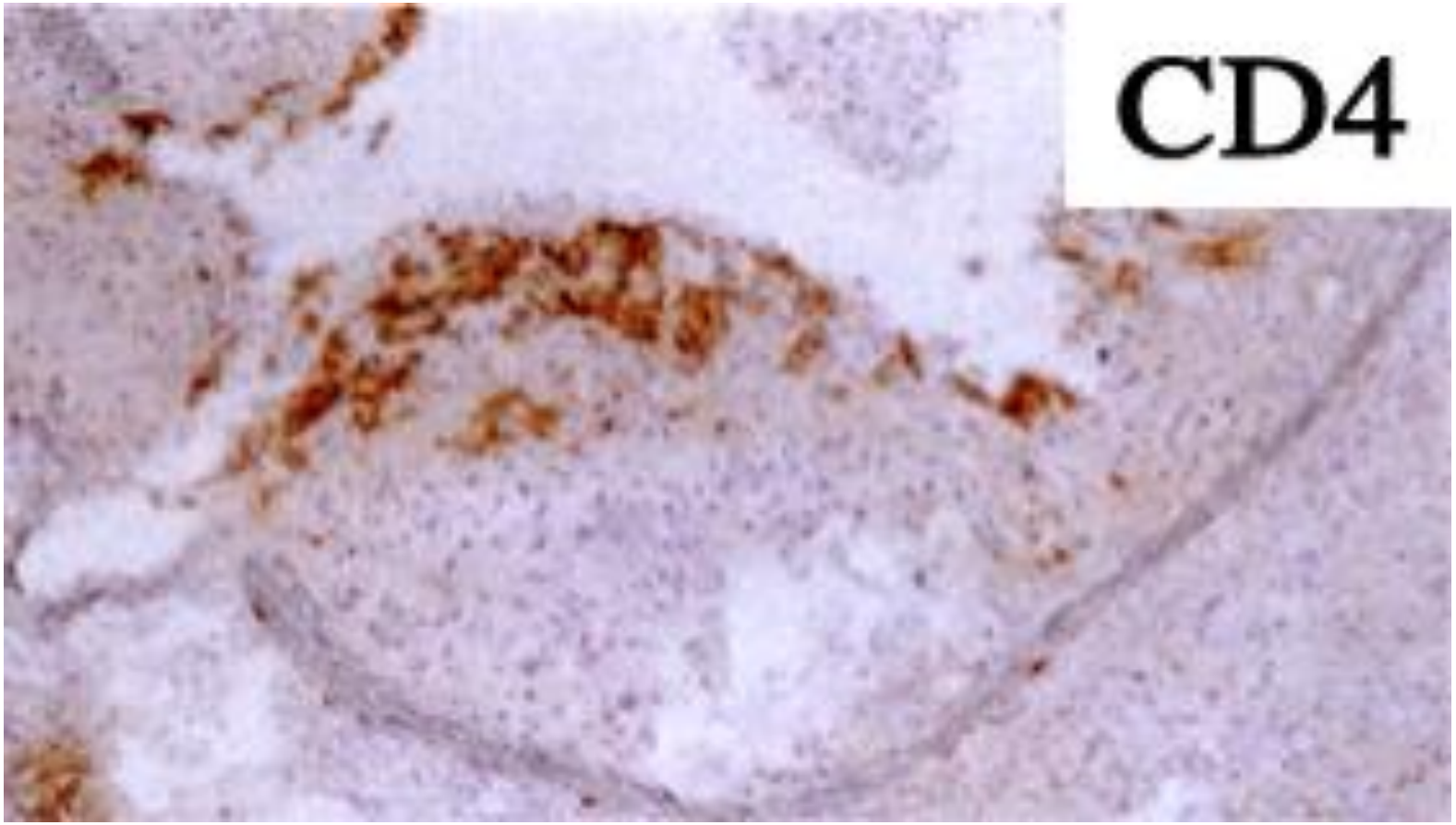
Mechanistic studies require a suitable experimental model

The Apo(lipoprotein) E° mouse

The ApoE° mouse model



Y. Nakashima, et al. *Arterioscler. Thromb.* 1994. 14 133-140



T cell

From Zhou et al, Am J Pathol 1996

How to assess the role of lymphocytes in atherosclerosis?

SCID

ApoE KO

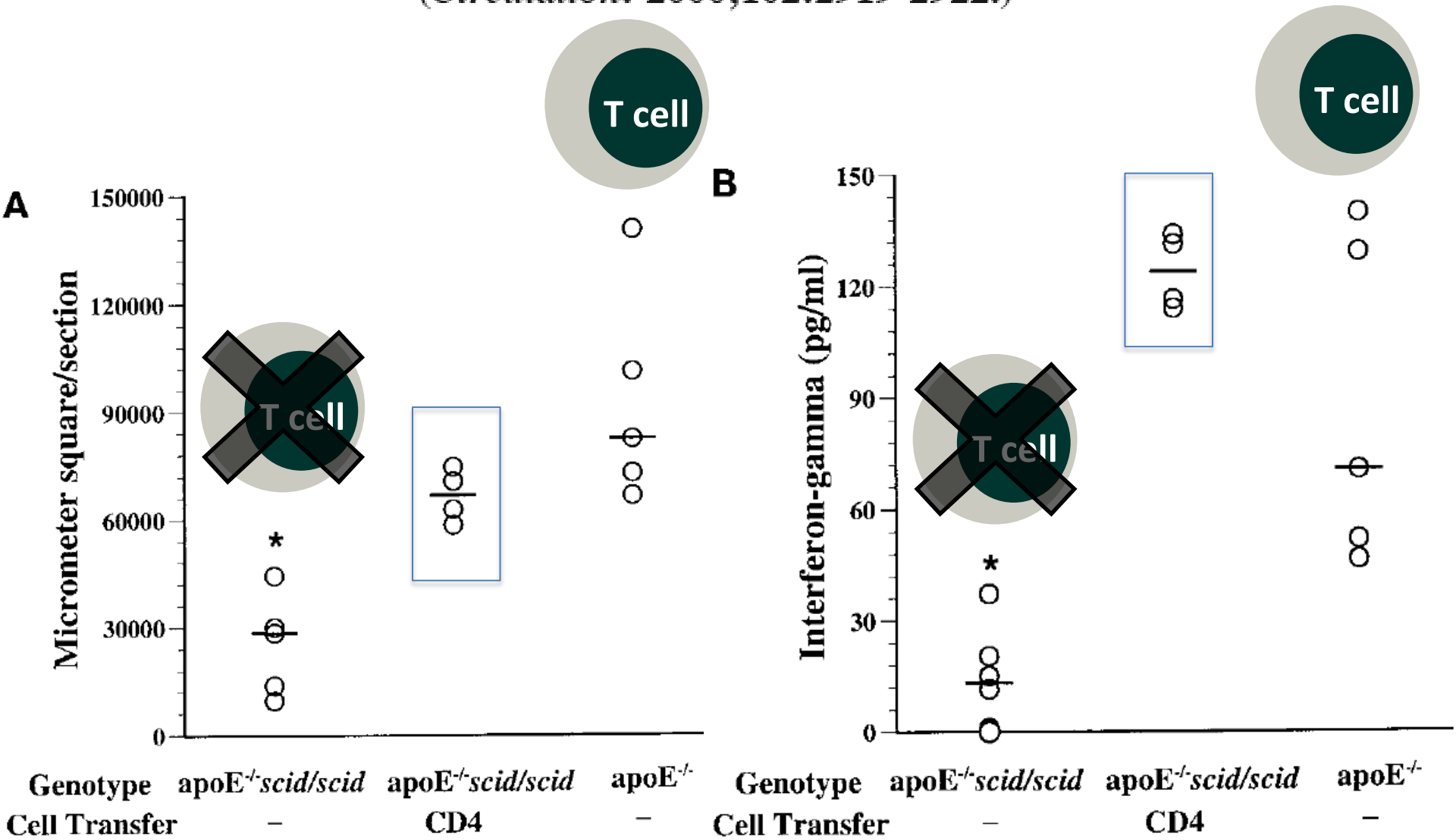
Chimeric mice:

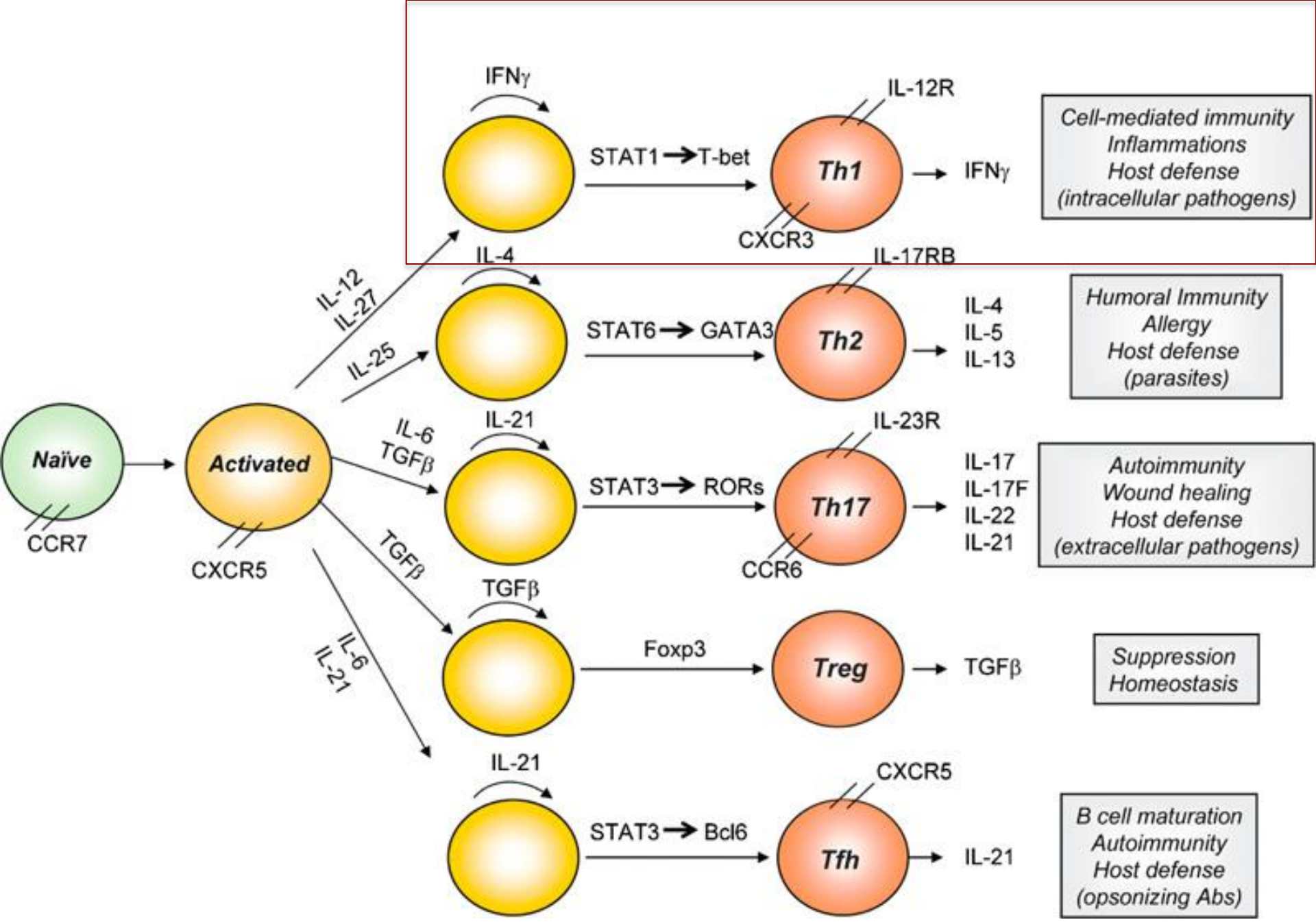
- Hypercholesterolemic
- Absence of lymphocytes

Transfer of CD4$^+$ T Cells Aggravates Atherosclerosis in Immunodeficient Apolipoprotein E Knockout Mice

Xinghua Zhou, MD, PhD; Antonino Nicoletti, PhD; Rima Elhage, PhD; Göran K. Hansson, MD, PhD

(*Circulation*. 2000;102:2919-2922.)





Cellular & Molecular Immunology (2010) 7, 190–197

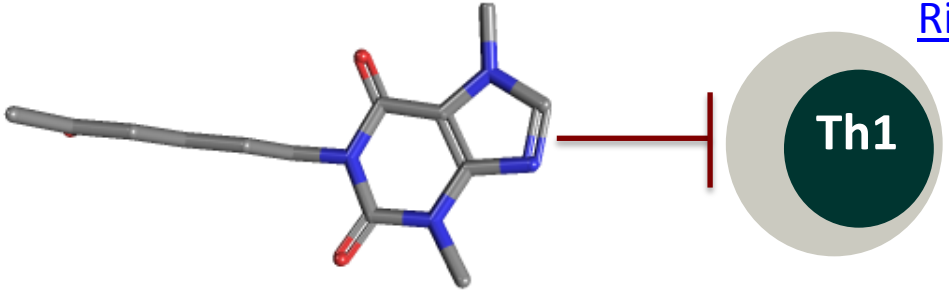
In Vivo Downregulation of T Helper Cell 1 Immune Responses Reduces Atherogenesis in Apolipoprotein E–Knockout Mice

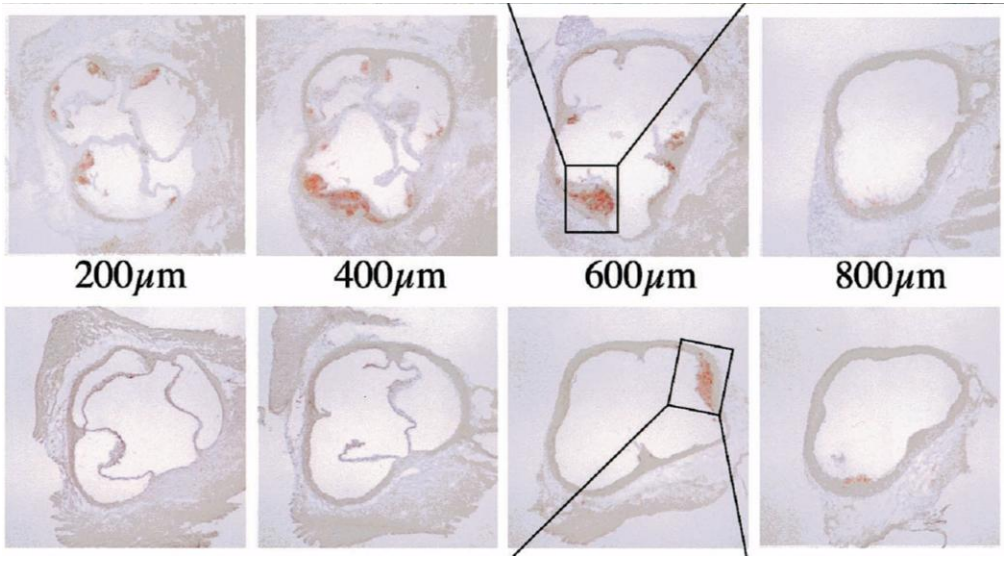
E. Laurat, MD; B. Poirier, PhD; E. Tupin, MSc; G. Caligiuri, MD, PhD; G.K. Hansson, MD, PhD; J. Bariéty, MD, PhD; A. Nicoletti, PhD

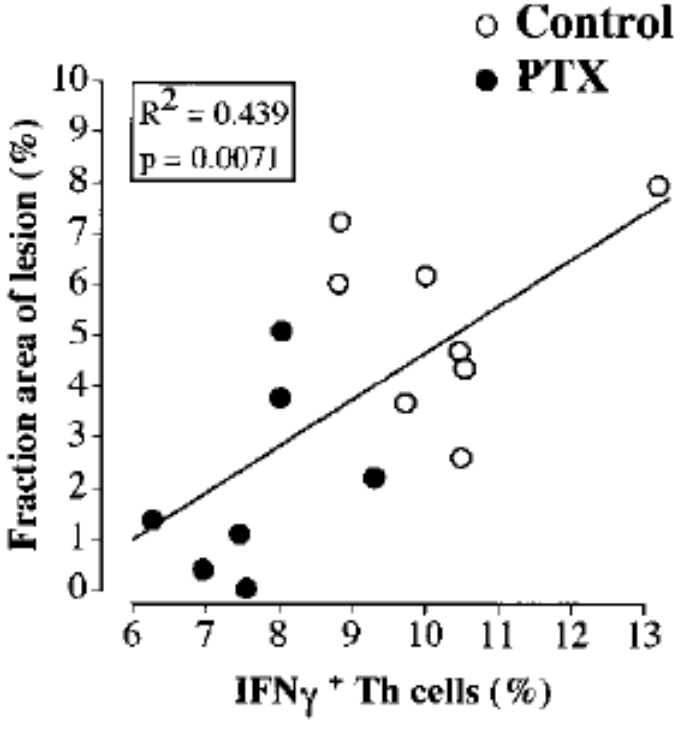
(*Circulation*. 2001;104:197-202.)

Pentoxifylline (Trental, inhibitor of phosphodiesterase) blocks Th1 cell development

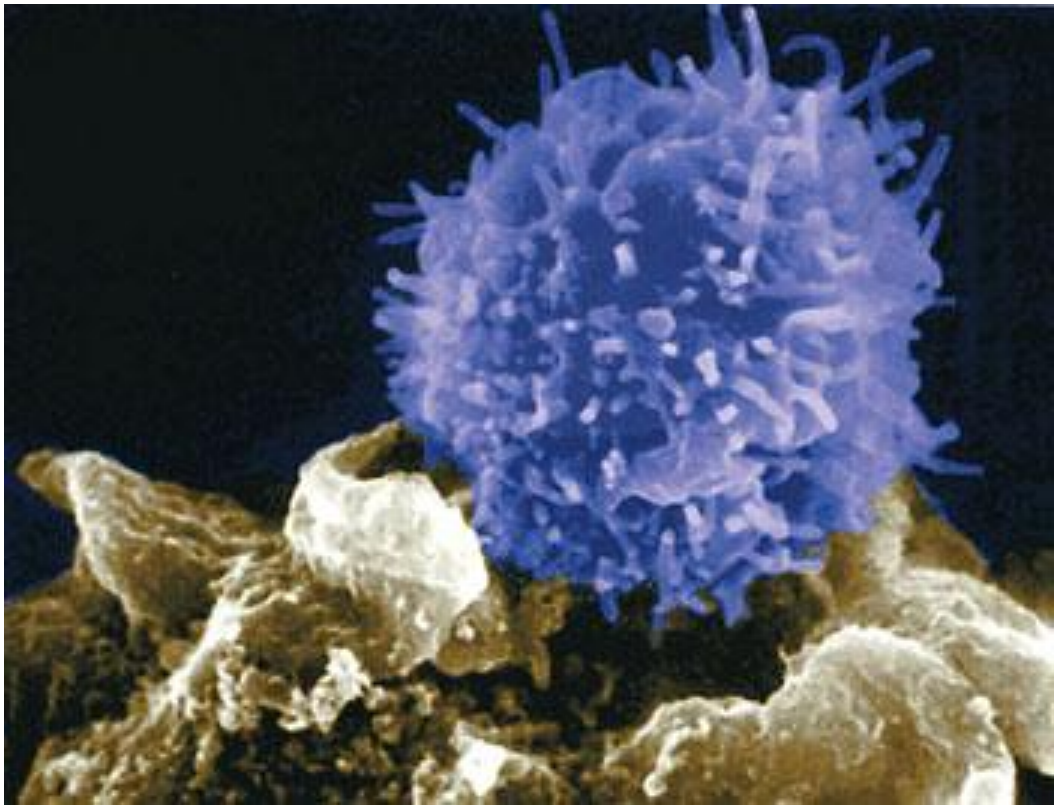
Rieckmann P et al, J Neuroimmunol 1996



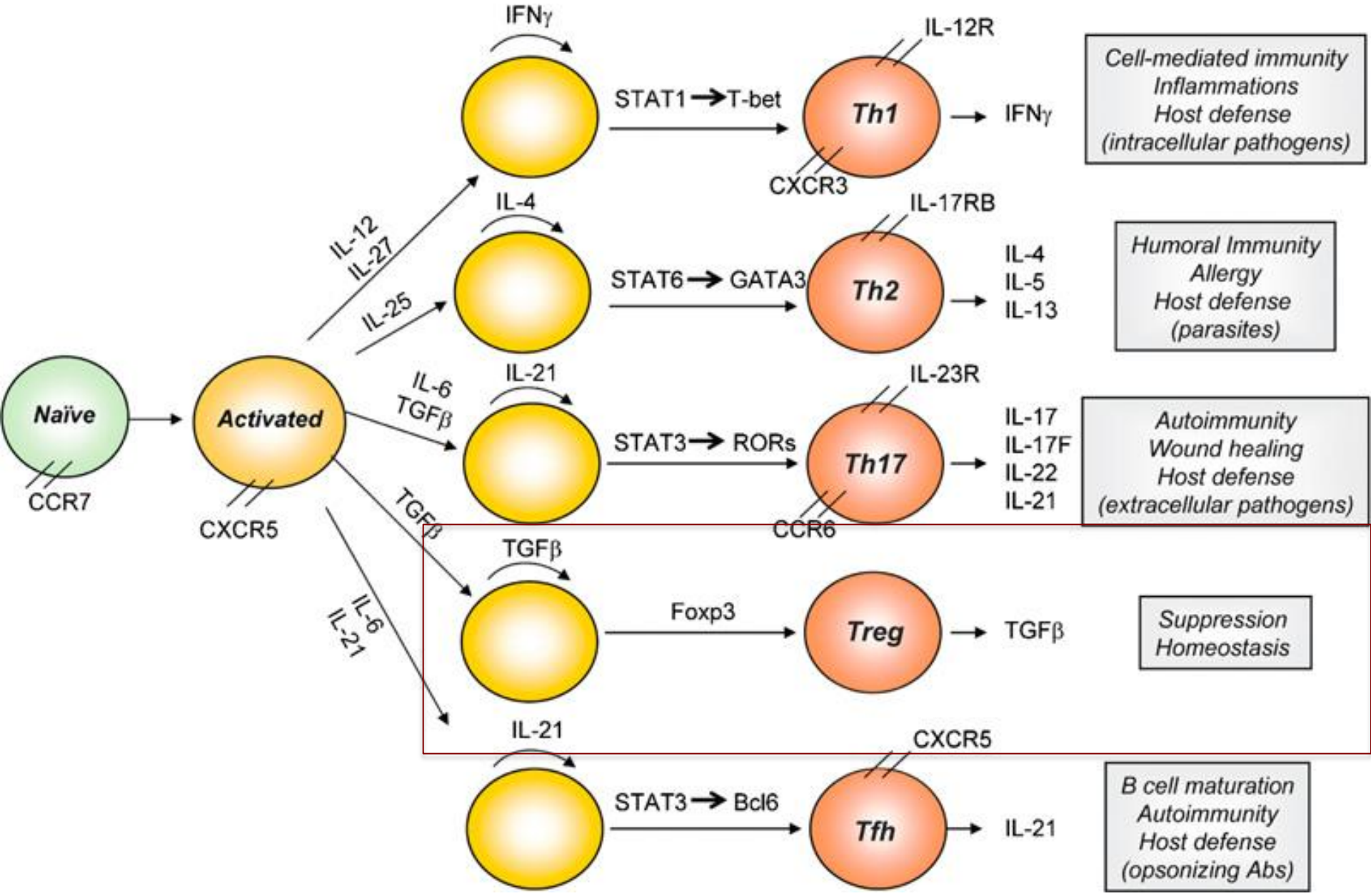




T cells



Deleterious role?

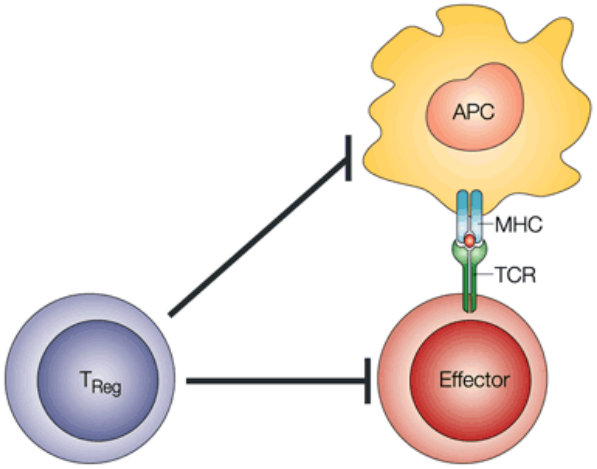


Cellular & Molecular Immunology (2010) 7, 190–197

Immunity, Vol. 12, 431–440, April, 2000, Copyright ©2000 by Cell Press

B7/CD28 Costimulation Is Essential for the Homeostasis of the CD4$^+$CD25$^+$ Immunoregulatory T Cells that Control Autoimmune Diabetes

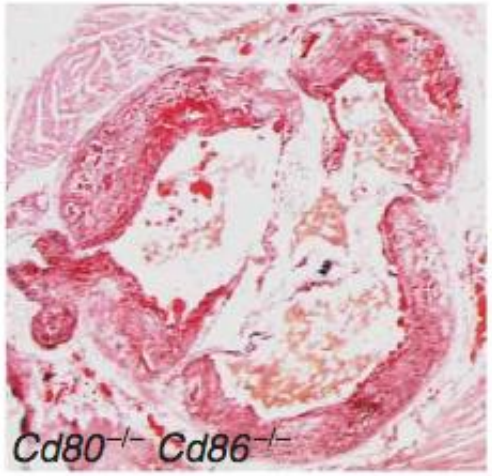
Benoît Salomon,* Deborah J. Lenschow,* Lesley Rhee,* Neda Ashourian,* Bhagarith Singh,† Arlene Sharpe,‡ and Jeffrey A. Bluestone*§

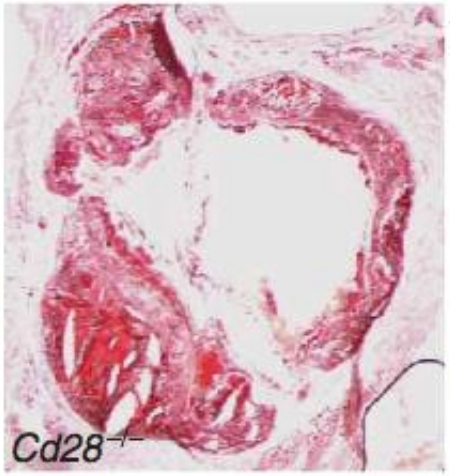


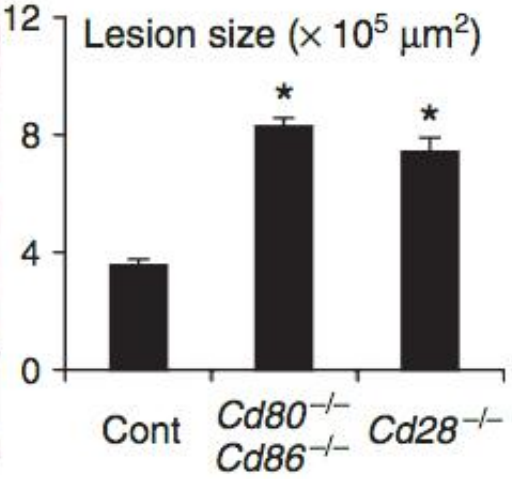
Natural regulatory T cells control the development of atherosclerosis in mice

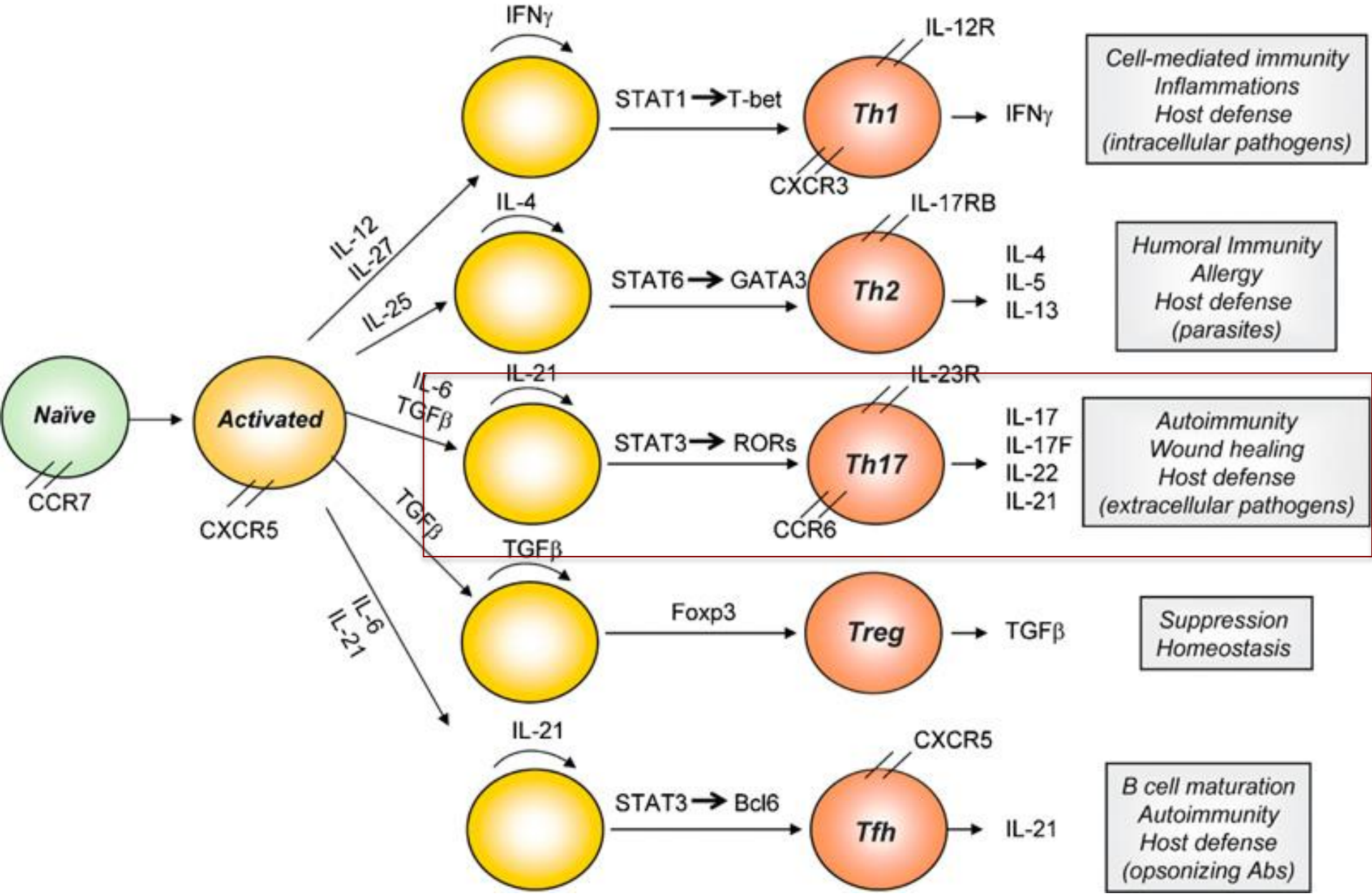
Hafid Ait-Oufella[1], Benoît L Salomon[2], Stéphane Potteaux[1], Anna-Karin L Robertson[3], Pierre Gourdy[4], Joffrey Zoll[1], Régine Merval[1], Bruno Esposito[1], José L Cohen[2], Sylvain Fisson[2], Richard A Flavell[5], Göran K Hansson[3], David Klatzmann[2], Alain Tedgui[1] & Ziad Mallat[1]

VOLUME 12 | NUMBER 2 | FEBRUARY 2006 NATURE MEDICINE







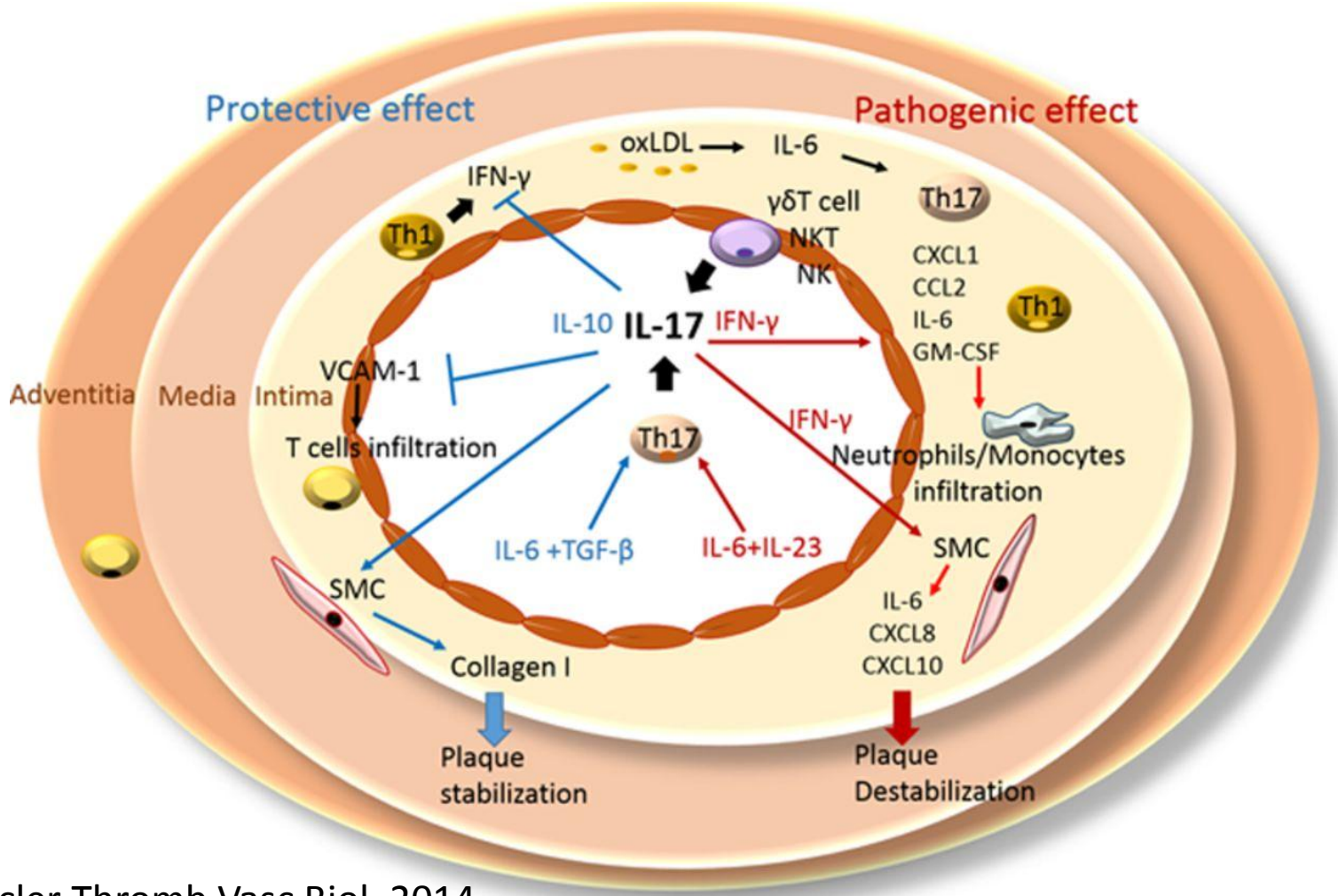


Cellular & Molecular Immunology (2010) 7, 190–197

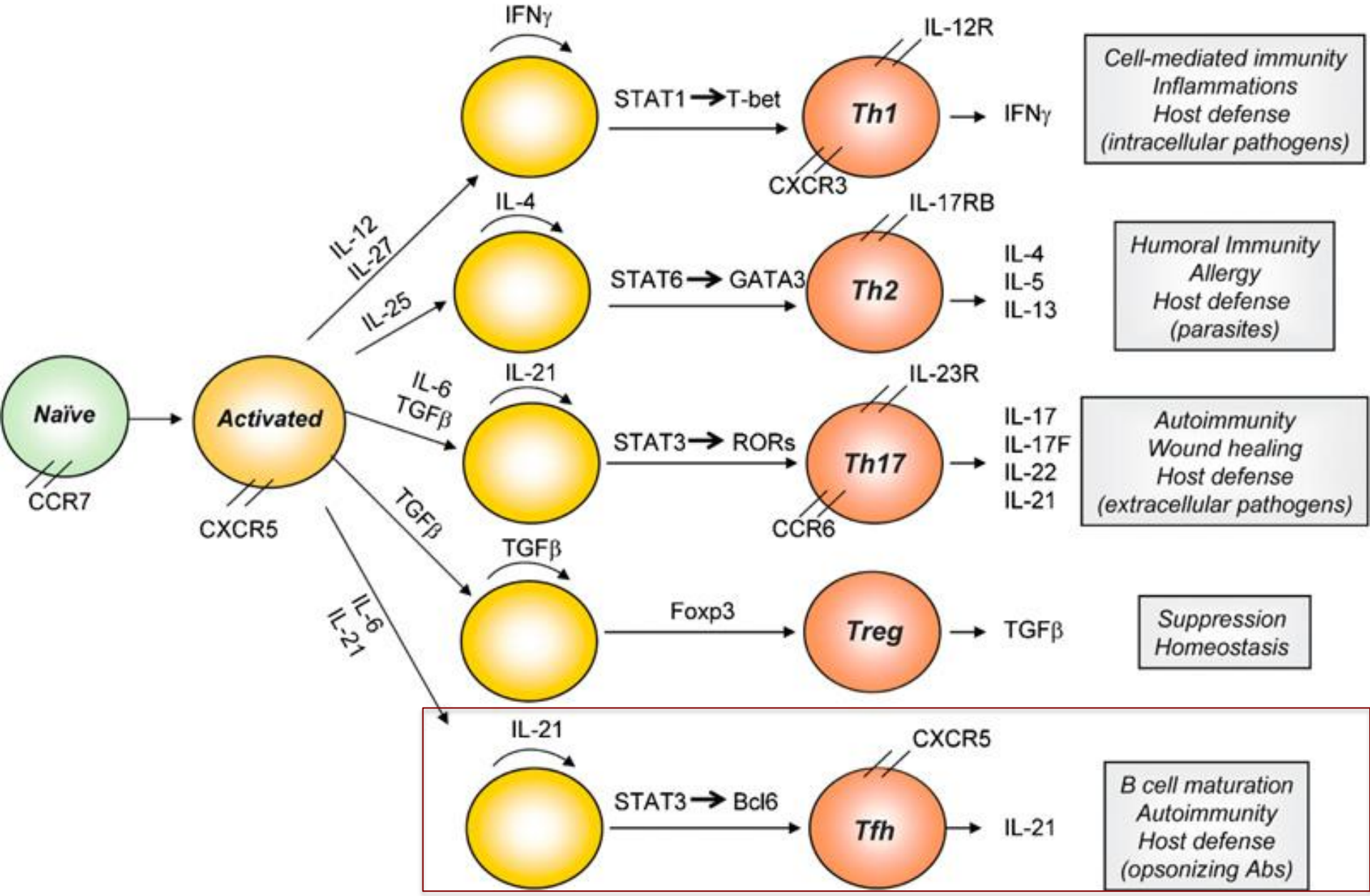
IL-17 and Th17 Cells in Atherosclerosis

Subtle and Contextual Roles

Soraya Taleb, Alain Tedgui, Ziad Mallat



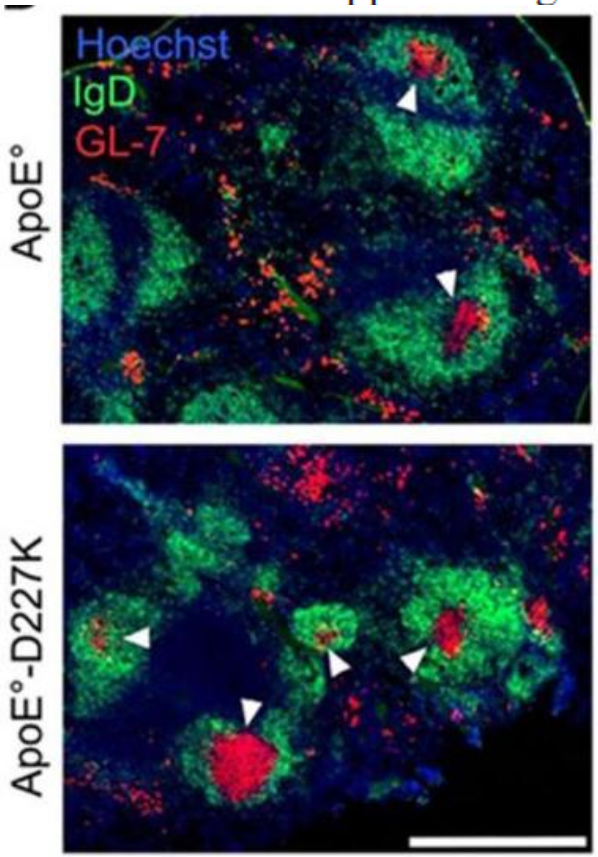
Arterioscler Thromb Vasc Biol. 2014

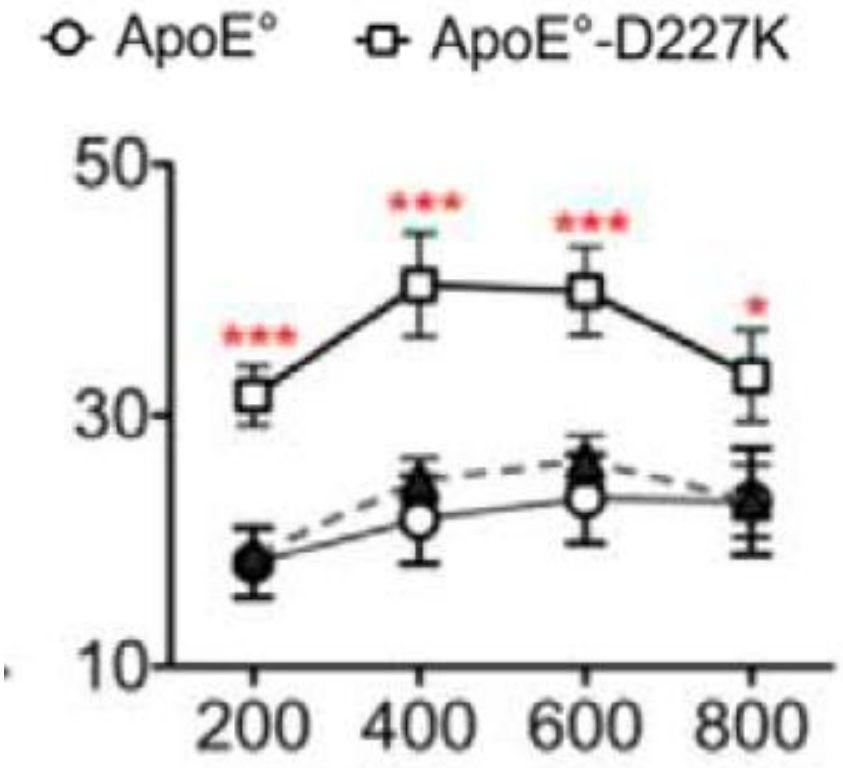


Cellular & Molecular Immunology (2010) 7, 190–197

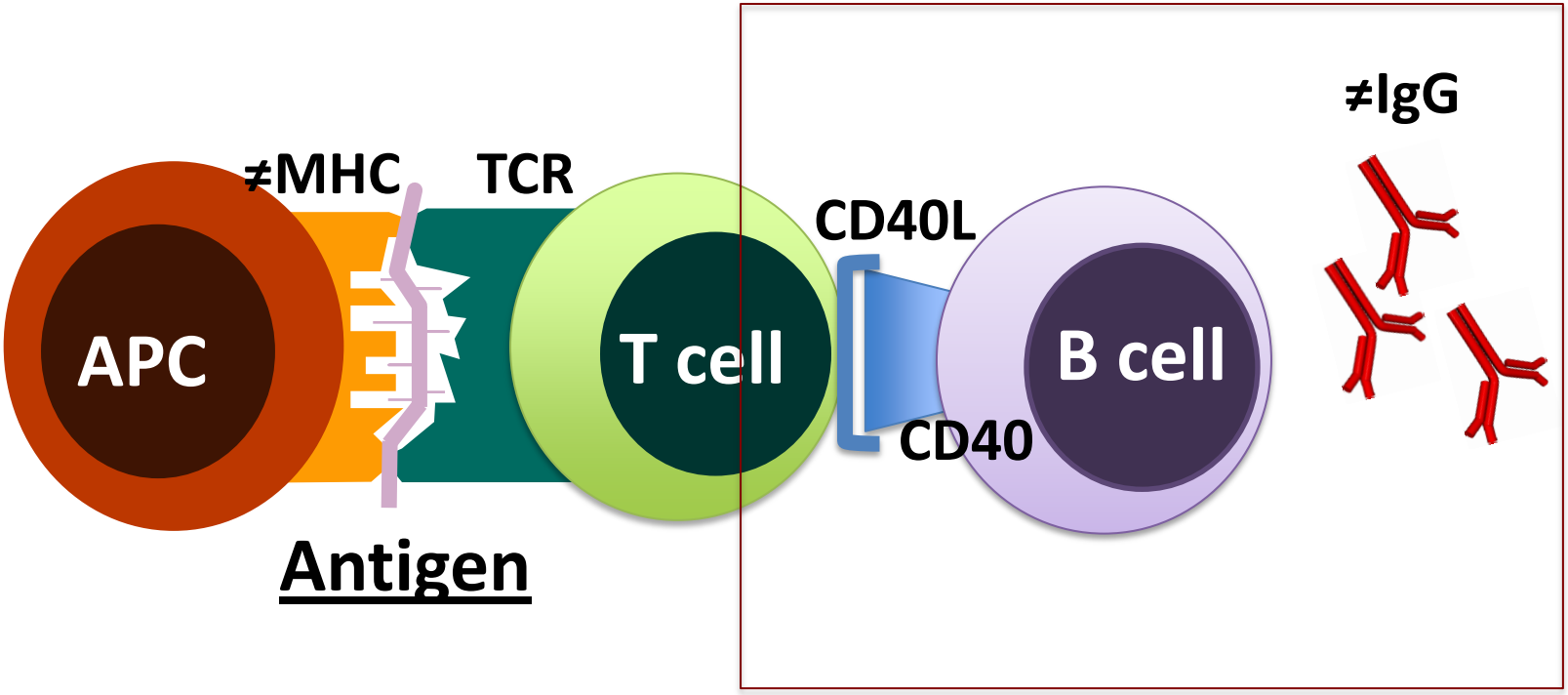
Control of the T Follicular Helper–Germinal Center B-Cell Axis by CD8⁺ Regulatory T Cells Limits Atherosclerosis and Tertiary Lymphoid Organ Development

Marc Clement, PhD; Kevin Guedj, PhD; Francesco Andreata, MSc; Marion Morvan, MSc; Laetitia Bey, MSc; Jamila Khallou-Laschet, PhD; Anh-Thu Gaston, MSc; Sandrine Delbosc, PhD; Jean-Marc Alsac, MD, PhD; Patrick Bruneval, MD, PhD; Catherine Deschildre, MSc; Marie Le Borgne, PhD; Yves Castier, MD, PhD; Hye-Jung Kim, PhD; Harvey Cantor, MD, PhD; Jean-Baptiste Michel, MD, PhD; Giuseppina Caligiuri, MD, PhD; Antonino Nicoletti, PhD





Adaptive immune response



Atherosclerosis, 38 (1981) 211—216

DEMONSTRATION OF IMMUNOGLOBULIN IN THE NEIGHBOURHOOD OF ADVANCED ATHEROSCLEROTIC PLAQUES

D. PARUMS and M.J. MITCHINSON

Department of Pathology, University of Cambridge, Tennis Court Road, Cambridge (Great Britain)

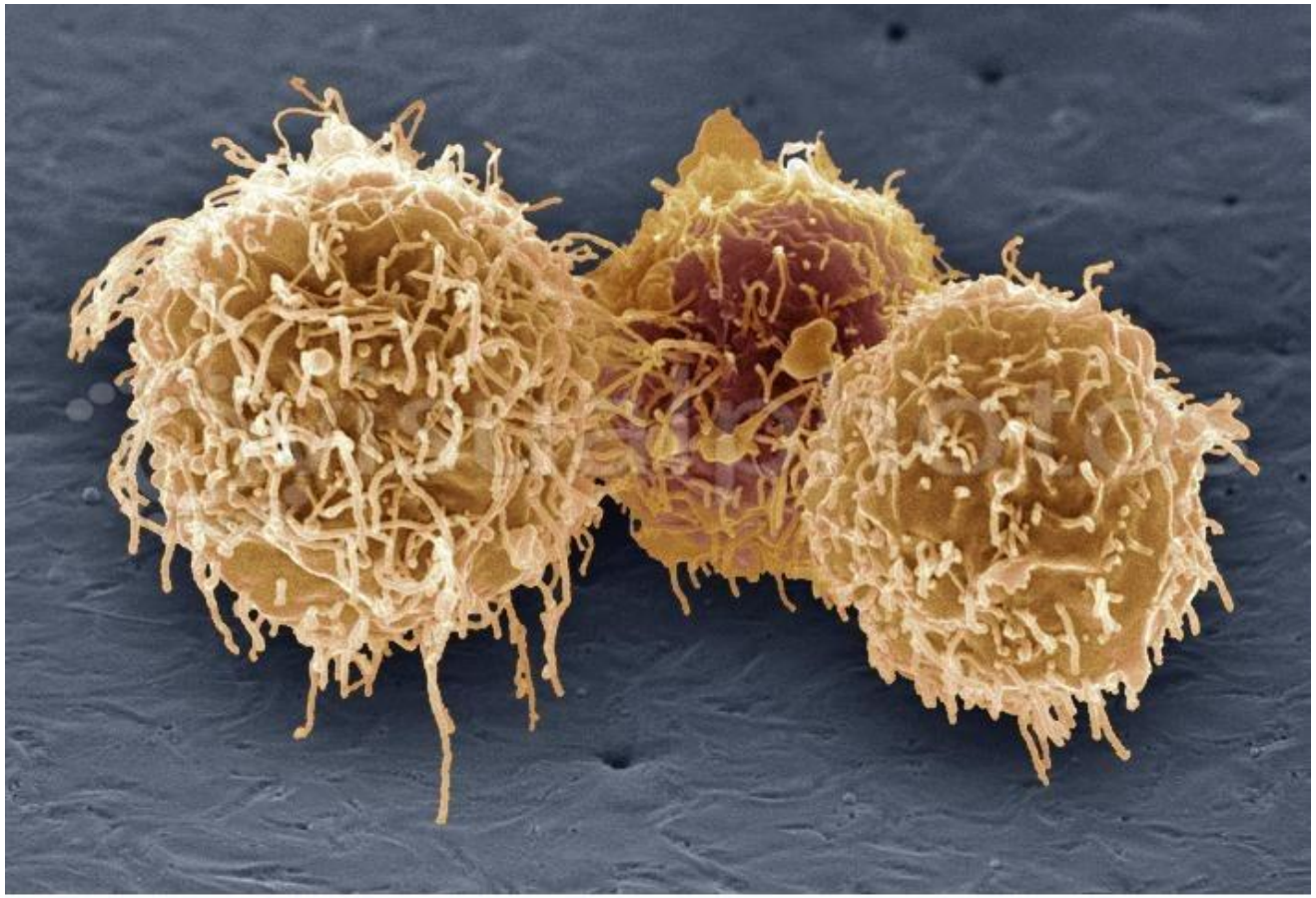
Summary

A necropsy survey of inflammatory cellular infiltration associated with atherosclerosis showed that the degree of inflammation correlated with the severity of the atherosclerotic plaque and was greater in the presence of medial disruption.

Sections were stained for IgG and IgM using the immunoperoxidase technique. Advanced plaques with medial disruption showed staining of the cytoplasm of plasma cells in the adventitia with IgG and IgM antisera. IgG was also demonstrated in the atheromatous material.

The immunoglobulin production may have been stimulated by antigens formed in the advanced lesion. Such an allergic reaction might explain some of the complications of advanced atherosclerosis.

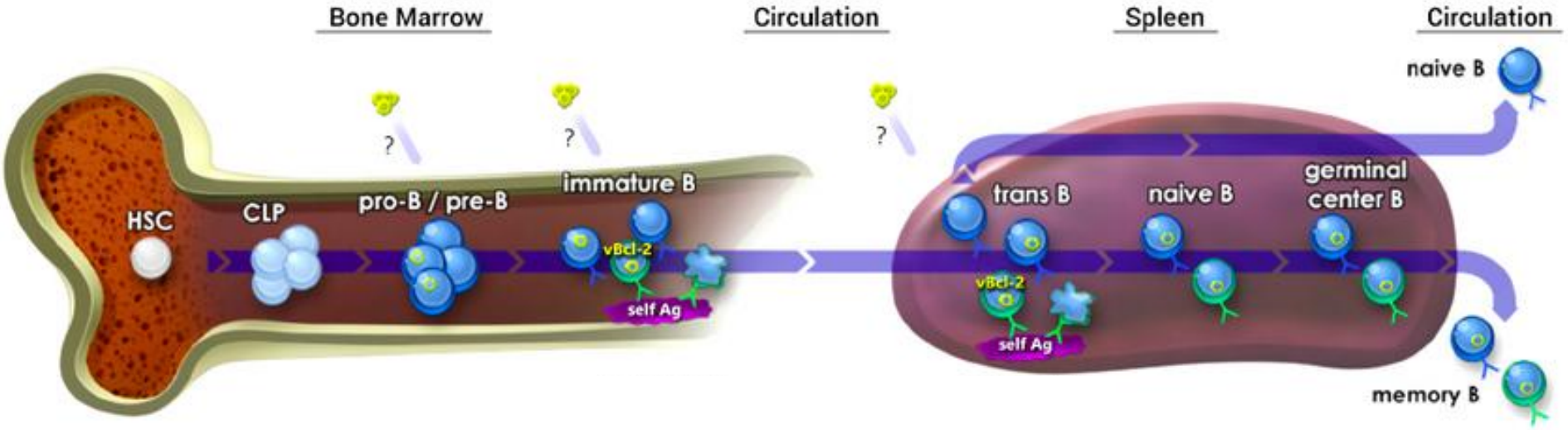
B cells



F0010059 [RF] (c) www.visualphotos.com

Role?

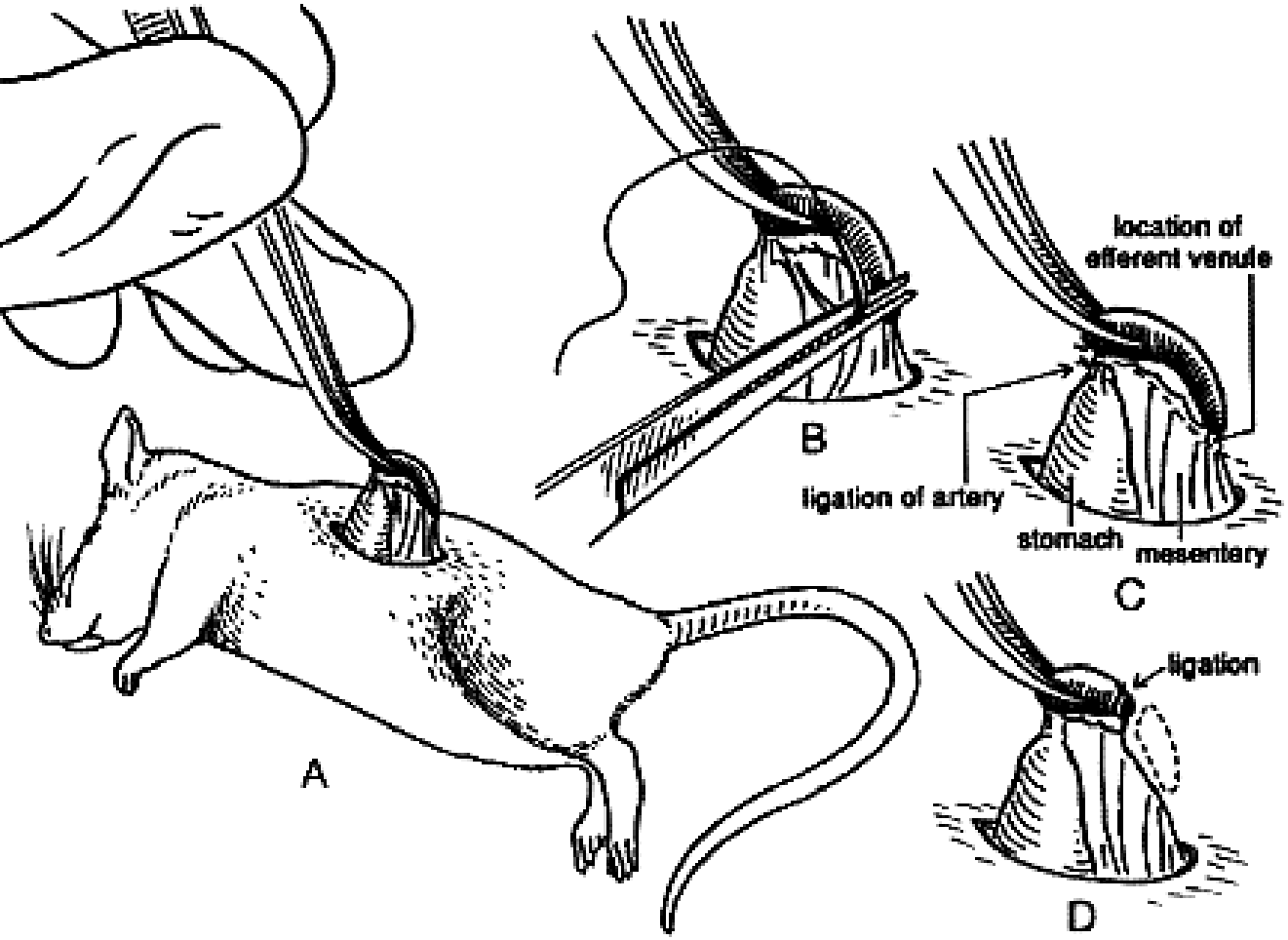
The spleen is a major reservoir of mature B cells

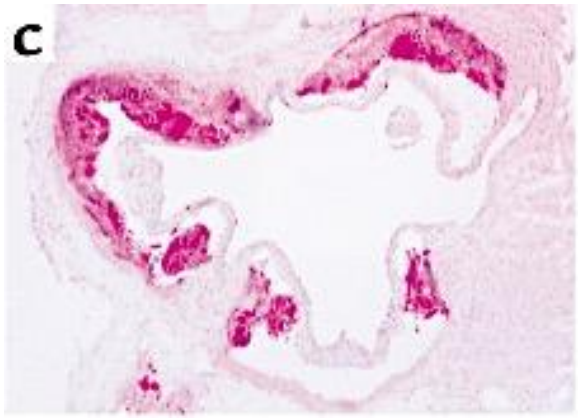


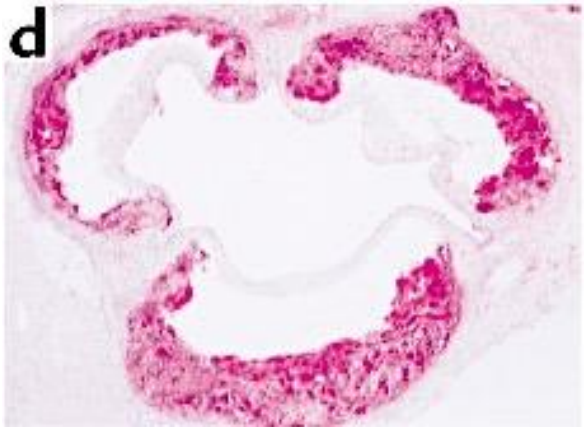
Protective immunity against atherosclerosis carried by B cells of hypercholesterolemic mice

J. Clin. Invest. **109**:745–753 (2002)

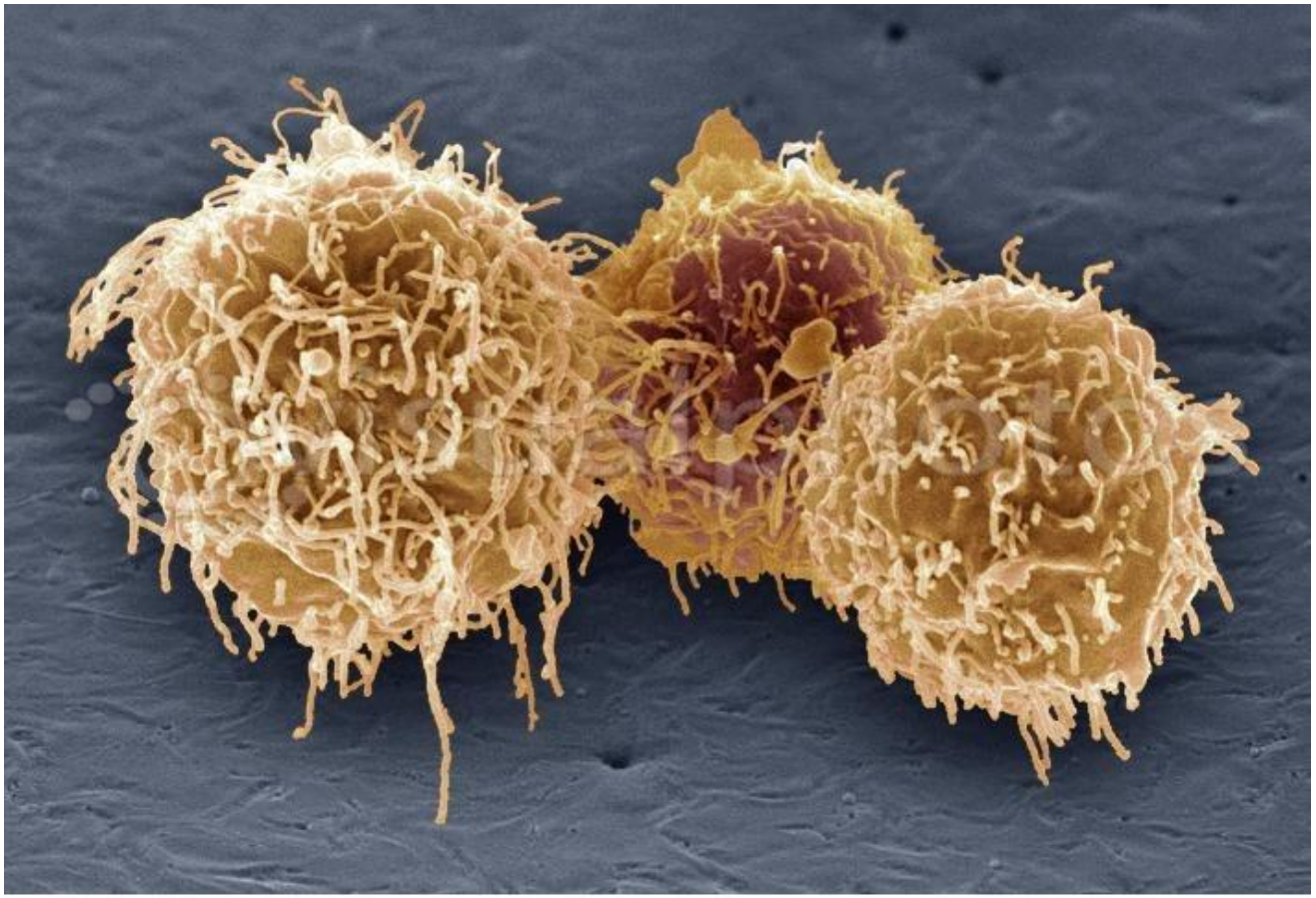
Giuseppina Caligiuri,[1] Antonino Nicoletti,[1,2] Bruno Poirier,[2] and Göran K. Hansson[1]







B cells



F0010059 [RF] (c) www.visualphotos.com

Protective role?

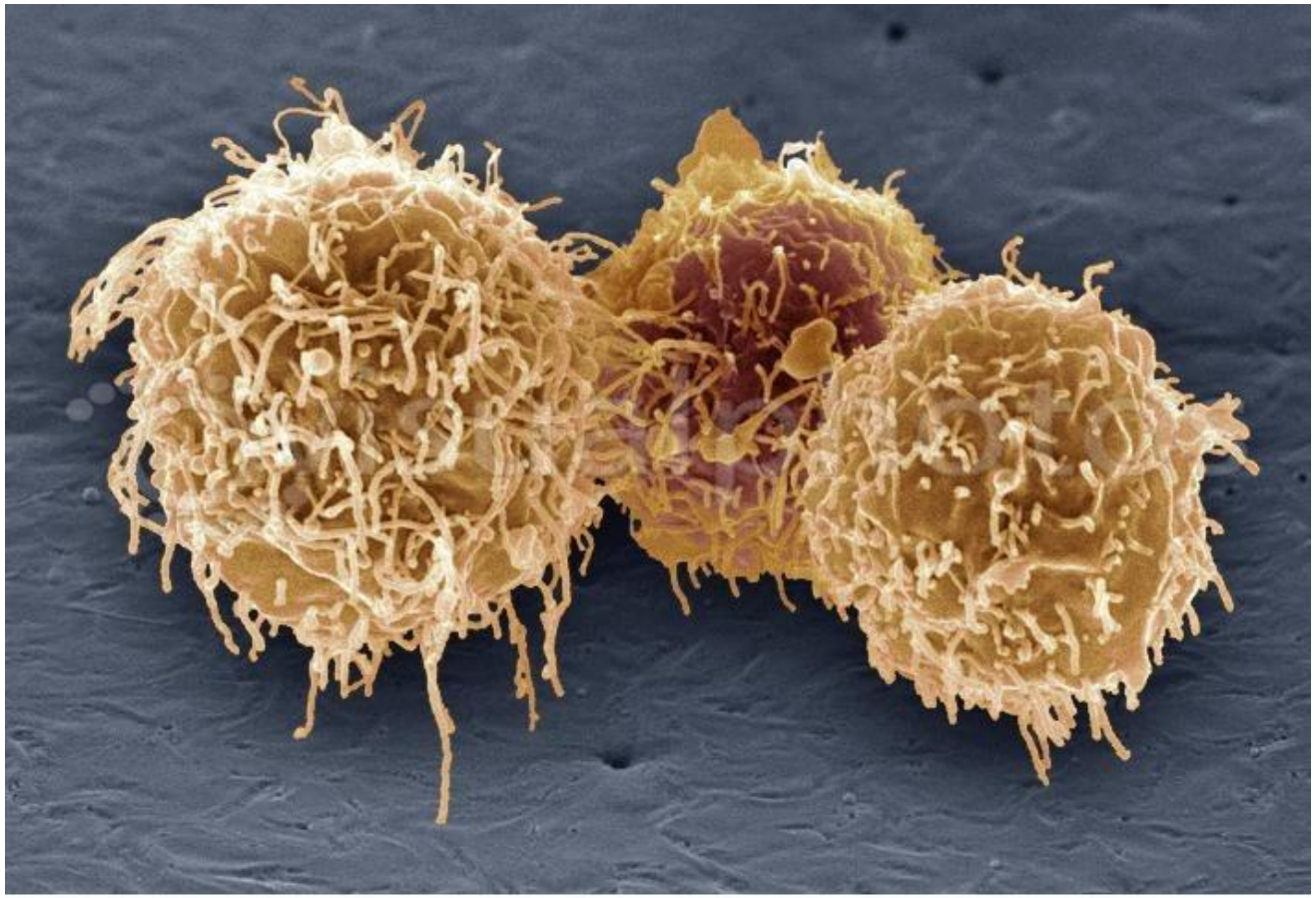
B cell depletion reduces the development of atherosclerosis in mice

J. Exp. Med. Vol. 207 No. 8 1579-1587

Hafid Ait-Oufella,[1,2] Olivier Herbin,[1] Jean-David Bouaziz,[3,4] Christoph J. Binder,[5,6] Catherine Uyttenhove,[7,8] Ludivine Laurans,[1] Soraya Taleb,[1] Emily Van Vré,[1] Bruno Esposito,[1] José Vilar,[1] Jérôme Sirvent,[1] Jacques Van Snick,[7,8] Alain Tedgui,[1] Thomas F. Tedder,[3] and Ziad Mallat[1,9]

B cell depletion significantly reduces the burden of several immune-mediated diseases. However, B cell activation has been until now associated with a protection against atherosclerosis, suggesting that B cell–depleting therapies would enhance cardiovascular risk. We unexpectedly show that mature B cell depletion using a CD20-specific monoclonal antibody induces a significant reduction of atherosclerosis in various mouse models of the disease. This treatment preserves the production of natural and potentially protective anti–oxidized low-density lipoprotein (oxLDL) IgM autoantibodies over IgG type anti-oxLDL antibodies, and markedly reduces pathogenic T cell activation. B cell depletion diminished T cell–derived IFN-γ secretion and enhanced production of IL-17; neutralization of the latter abrogated CD20 antibody–mediated atheroprotection. These results challenge the current paradigm that B cell activation plays an overall protective role in atherogenesis and identify new antiatherogenic strategies based on B cell modulation.

B cells



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Deleterious role?

BAFF Receptor Deficiency Reduces the Development of Atherosclerosis in Mice—Brief Report

Andrew P. Sage, Dimitrios Tsiantoulas, Lauren Baker, James Harrison, Leanne Masters, Deirdre Murphy, Celine Loinard, Christoph J. Binder, Ziad Mallat

Objective—The goal of this study was to assess the role of B-cell activating factor (BAFF) receptor in B-cell regulation of atherosclerosis.

Methods and Results—Male LDL receptor-deficient mice (*Ldlr*$^{-/-}$) were lethally irradiated and reconstituted with either wild type or BAFF receptor (BAFF-R)–deficient bone marrow. After 4 weeks of recovery, mice were put on a high-fat diet for 6 or 8 weeks. BAFF-R deficiency in bone marrow cells led to a marked reduction of conventional mature B2 cells but did not affect the B1a cell subtype. This was associated with a significant reduction of dendritic cell activation and T-cell proliferation along with a reduction of IgG antibodies against malondialdehyde-modified low-density lipoprotein. In contrast, serum IgM type antibodies were preserved. Interestingly, BAFF-R deficiency was associated with a significant reduction in atherosclerotic lesion development and reduced numbers of plaque T cells. Selective BAFF-R deficiency on B cells led to a similar reduction in lesion size and T-cell infiltration but in contrast did not affect dendritic cell activation.

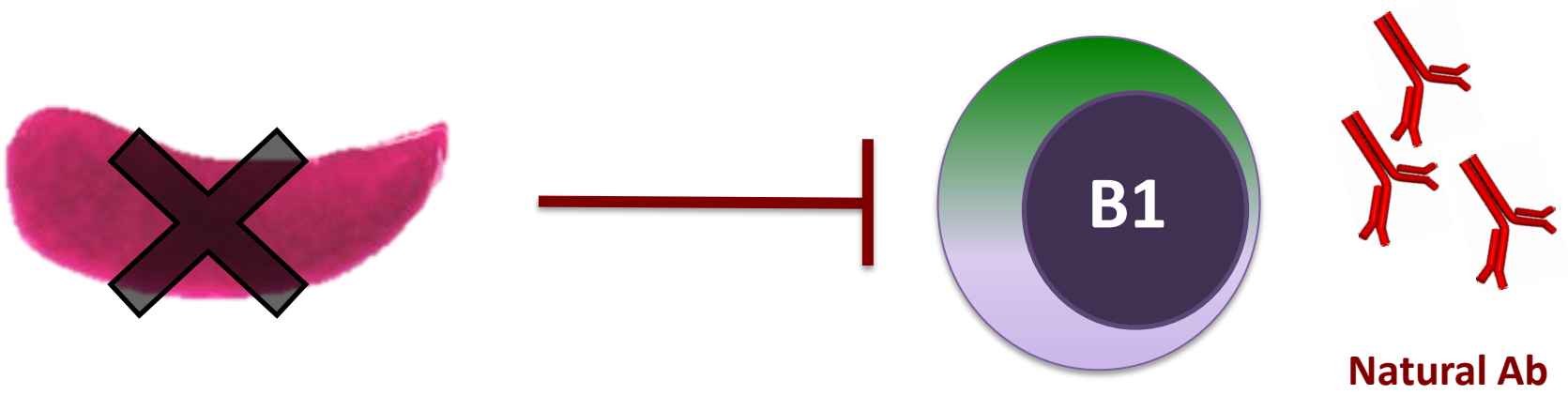
Conclusion—BAFF-R deficiency in mice selectively alters mature B2 cell-dependent cellular and humoral immune responses and limits the development of atherosclerosis. **(*Arterioscler Thromb Vasc Biol*. 2012;32:1573–1576.)**

B1 cell pool function needs the presence of the spleen

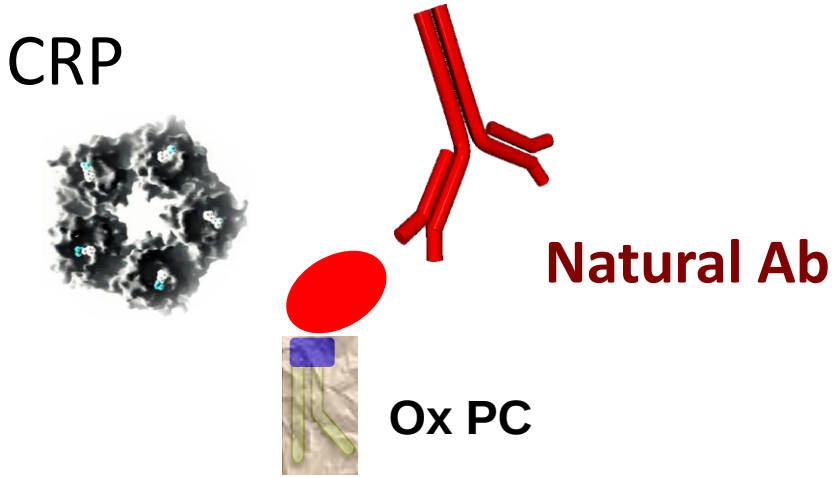
B-1a B Cells that Link the Innate and Adaptive Immune Responses Are Lacking in the Absence of the Spleen

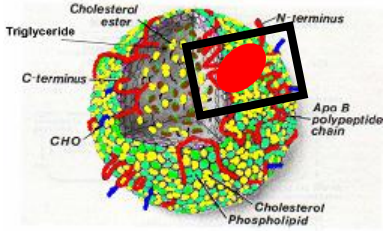
Hedda Wardemann, Thomas Boehm, Neil Dear, and Rita Carsetti

J. Exp. Med. © The Rockefeller University Press • (
Volume 195, Number 6, March 18, 2002 771–780

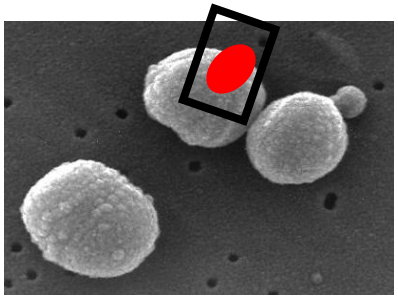


Natural antibodies and CRP bind to the same epitope: oxidized Phosphorylcholine

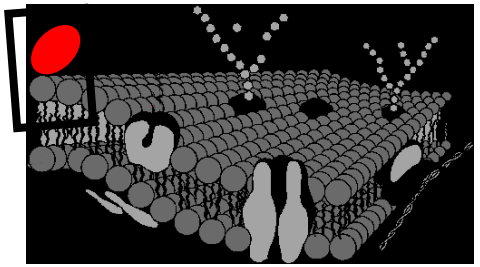




oxLDL



Wall of Gram⁺ bacteria



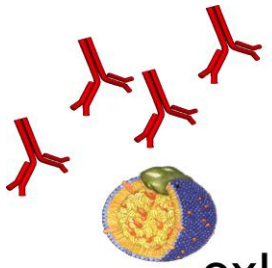
Cytoplasmic membrane
Apoptotic cells

(Binder et al - Nat Med 2003)

Protective immunity against atherosclerosis carried by B cells of hypercholesterolemic mice

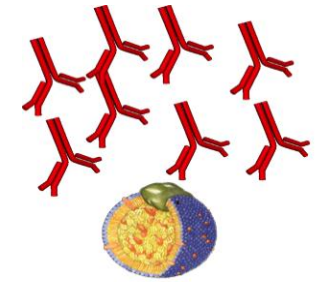
J. Clin. Invest. **109**:745–753 (2002)

Giuseppina Caligiuri,[1] Antonino Nicoletti,[1,2] Bruno Poirier,[2] and Göran K. Hansson[1]

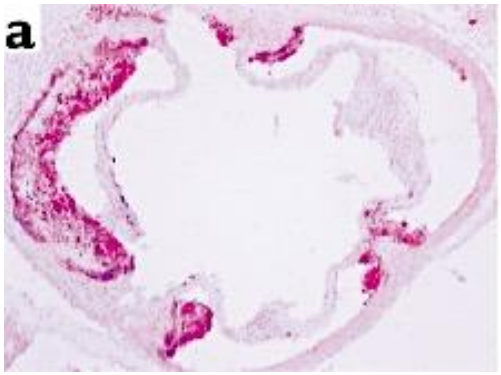


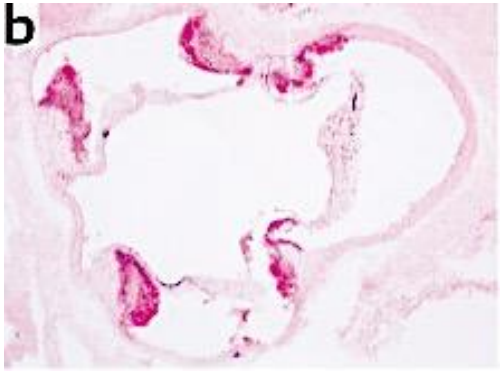
oxLDL

Spleen cells



oxLDL





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Phosphorylcholine-Targeting Immunization Reduces Atherosclerosis

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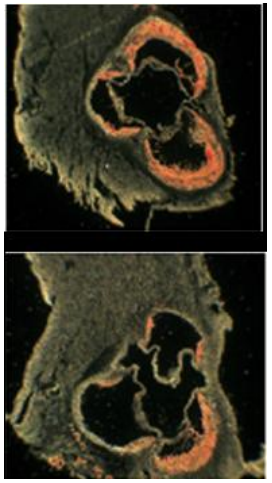
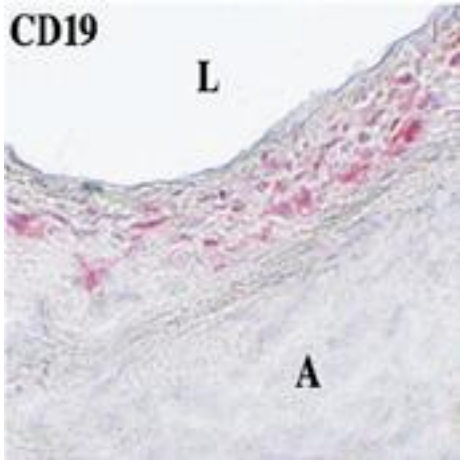
Anh-Thu Gaston, BSC,* Sandrine Delignat, BSC,* Chantal Mandet, BSC,†
Heinz V. Kohler, MD, PHD,‡ Srini V. Kaveri, DVM, PHD,* Antonino Nicoletti, PHD*

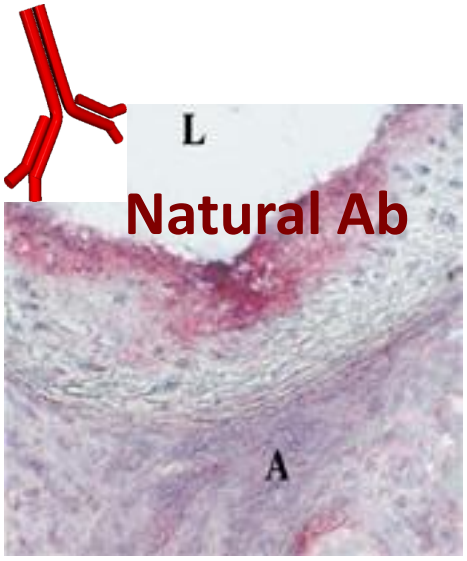
Paris, France; and Lexington, Kentucky

control

PC vaccined



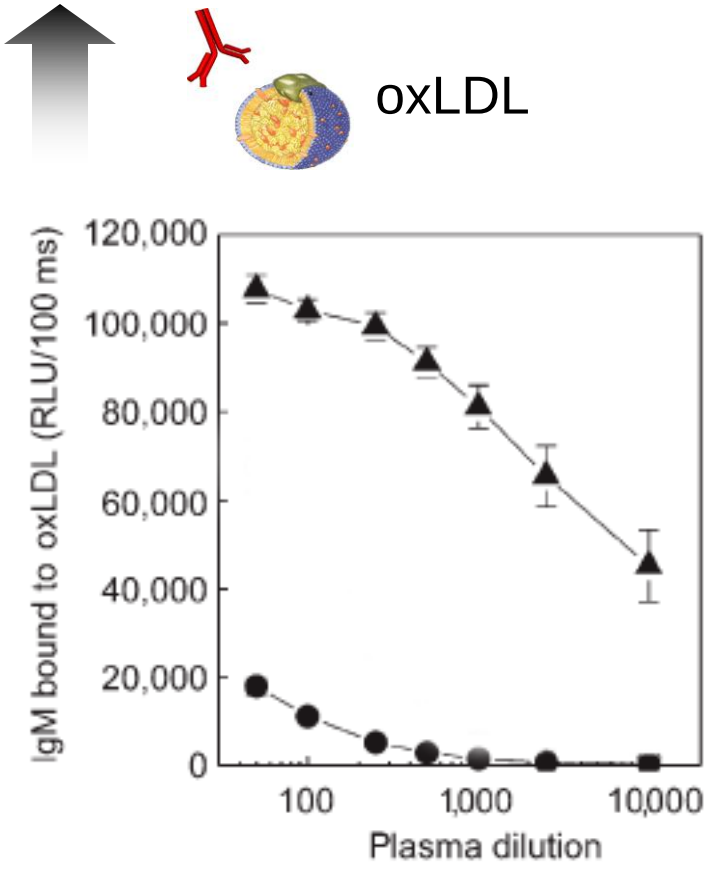


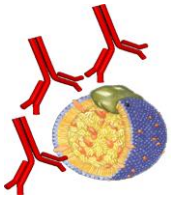


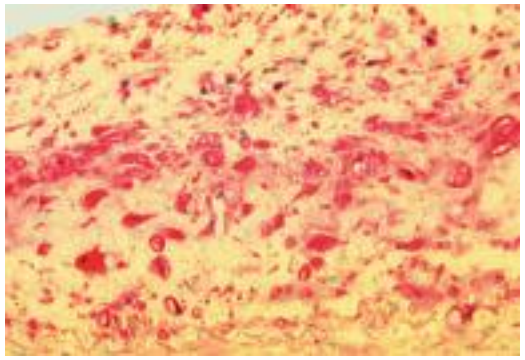
Pneumococcal vaccination decreases atherosclerotic lesion formation: molecular mimicry between *Streptococcus pneumoniae* and oxidized LDL

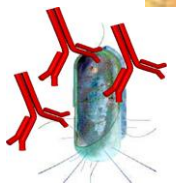
Nat Med 2003

Christoph J Binder[1,4], Sohvi Hörkkö[1,3,4], Asheesh Dewan[1,4], Mi-Kyung Chang[1], Emily P Kieu[1], Carl S Goodyear[2], Peter X Shaw[1], Wulf Palinski[1], Joseph L Witztum[1] & Gregg J Silverman[2]









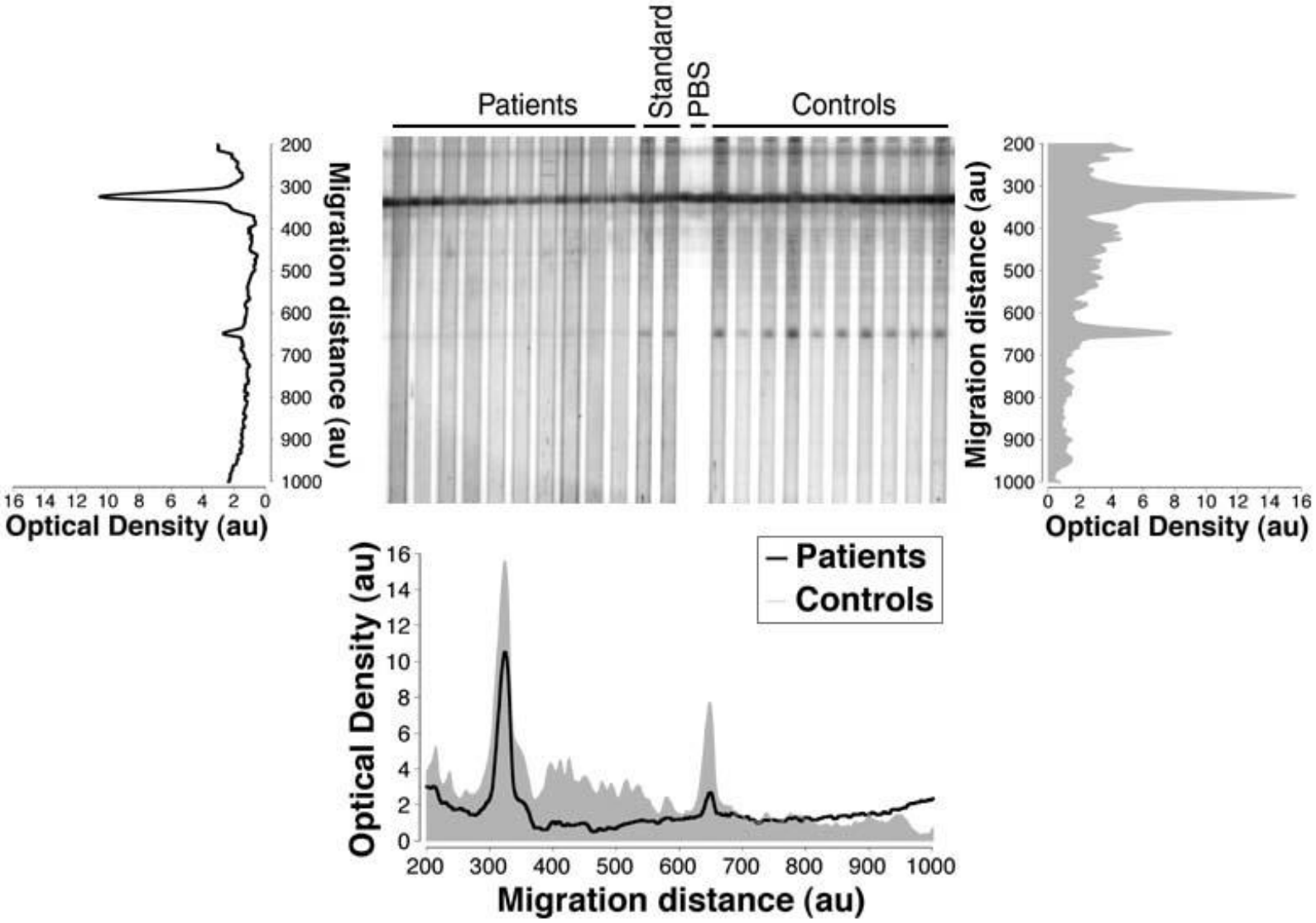
S pneumoniae

Atherosclerosis-associated antibodies

oxLDL, HSP, *CP*…

Evidence for antigen restriction?

The antibodies of atherosclerotic patients do not recognize a specific antigen

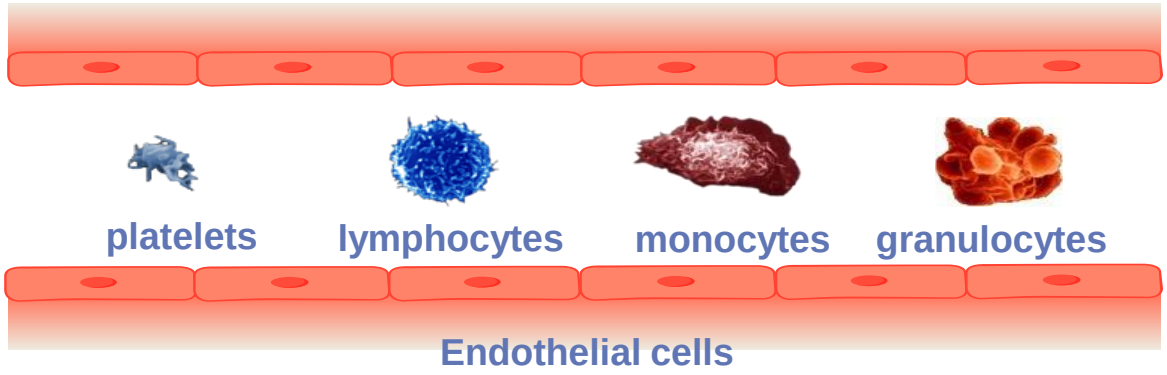


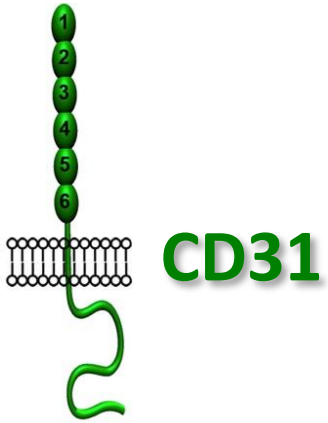
Caligiuri et al. Lab Invest 2003;83:939

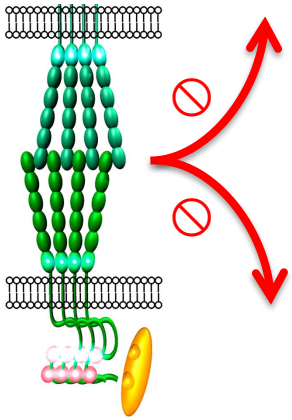
Role of T and B lymphocytes in atherosclerosis

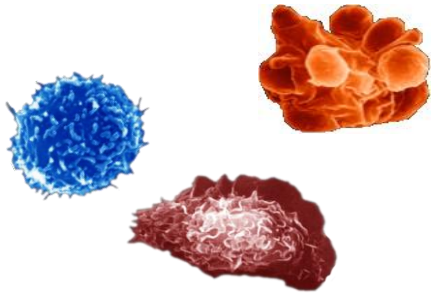
- T and B cell responses are associated with atherogenesis from the initial through to the advanced stages of the disease
- No true specific antigen
- Role not univocal: as all biologic “responses” they should be beneficial but can drive mad
- Immune control is the key

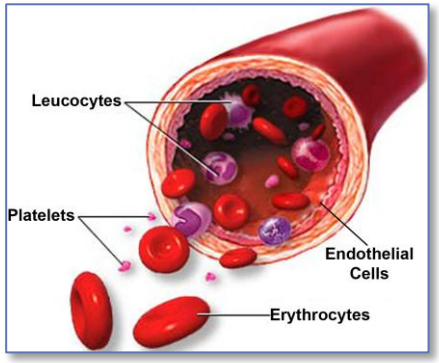
CD31: cell specificity and function





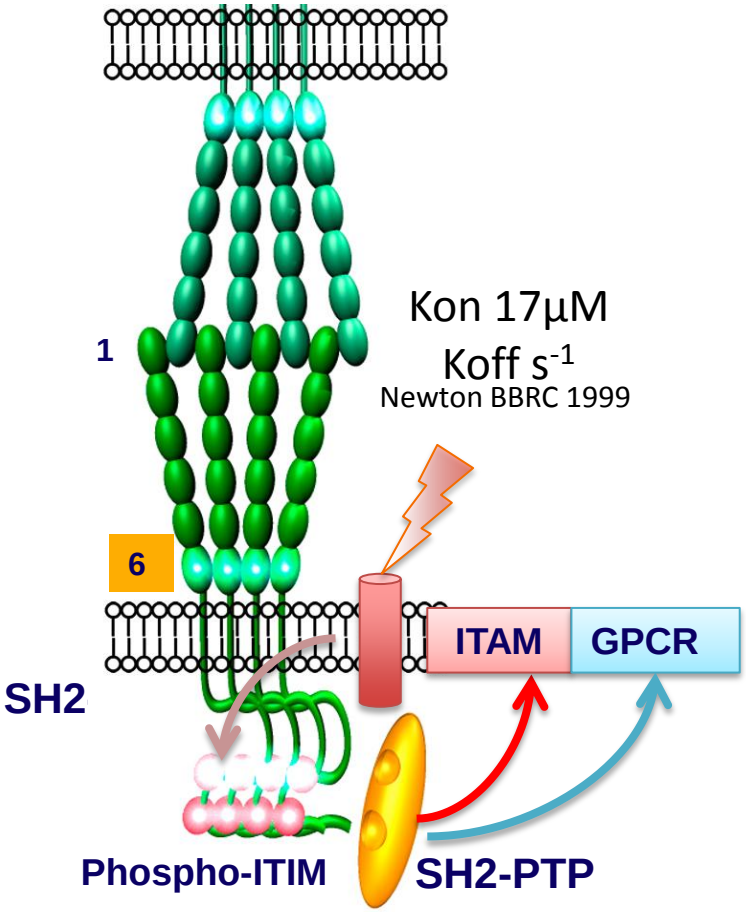


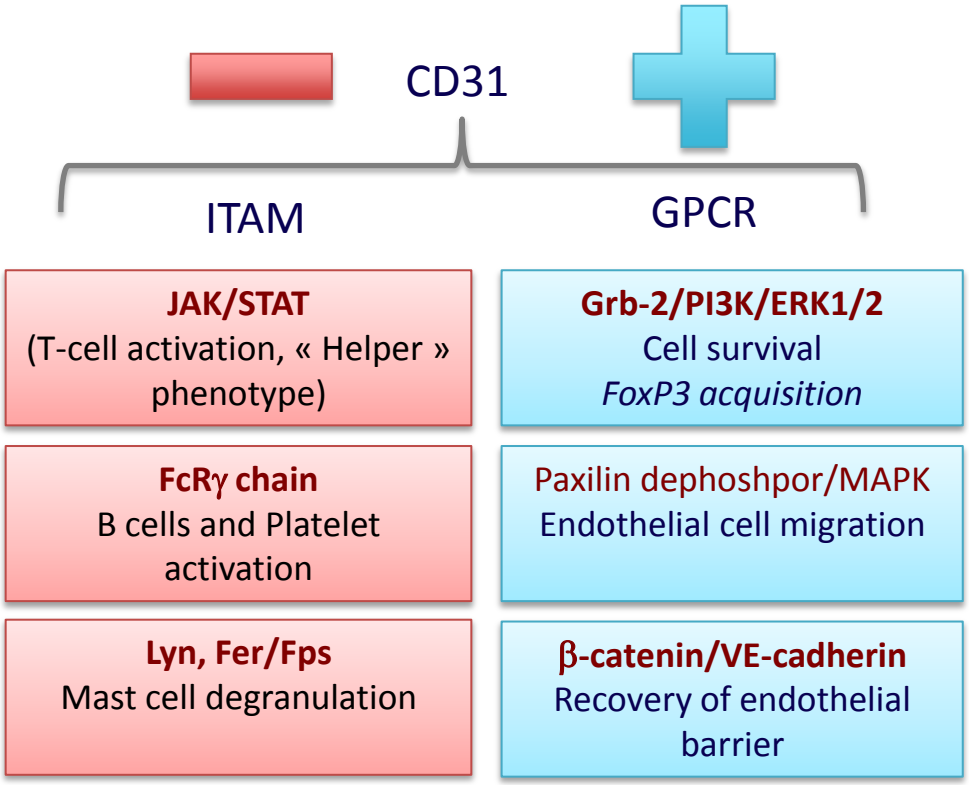




Brown et al. Nature 2002

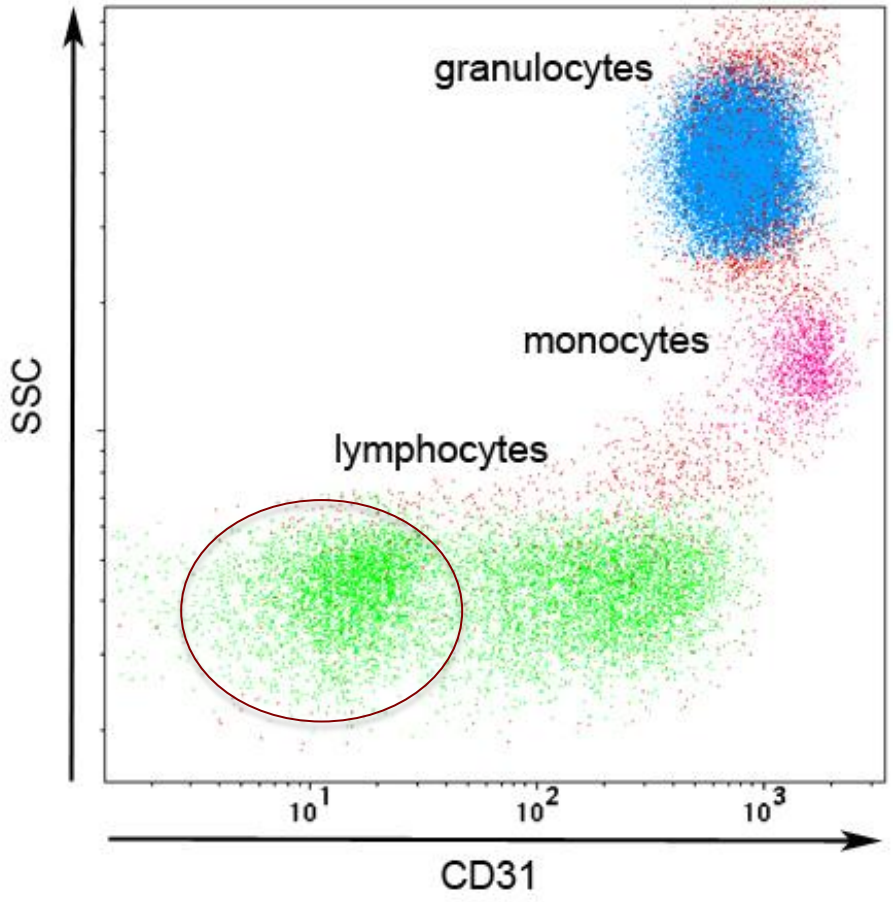
CD31: homophilic co-signaling receptor





Reviewed in Marelli-Berg et al J Cell Sci 2013

CD31 is lost by some lymphocytes



The % of blood CD31"negative" lymphocytes is higher in mice and patients with severe atherosclerosis

Caligiuri et al, ATVB 2005

Caligiuri et al, ATVB 2006

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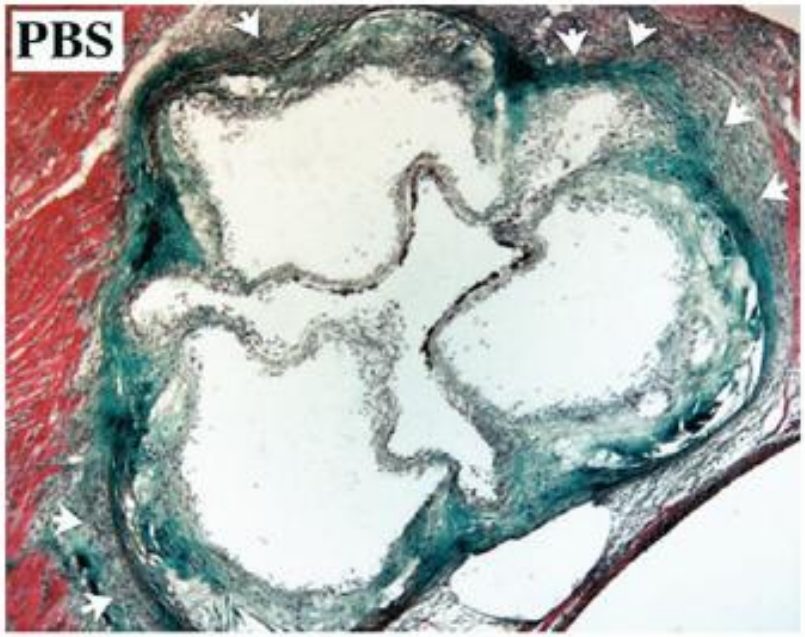
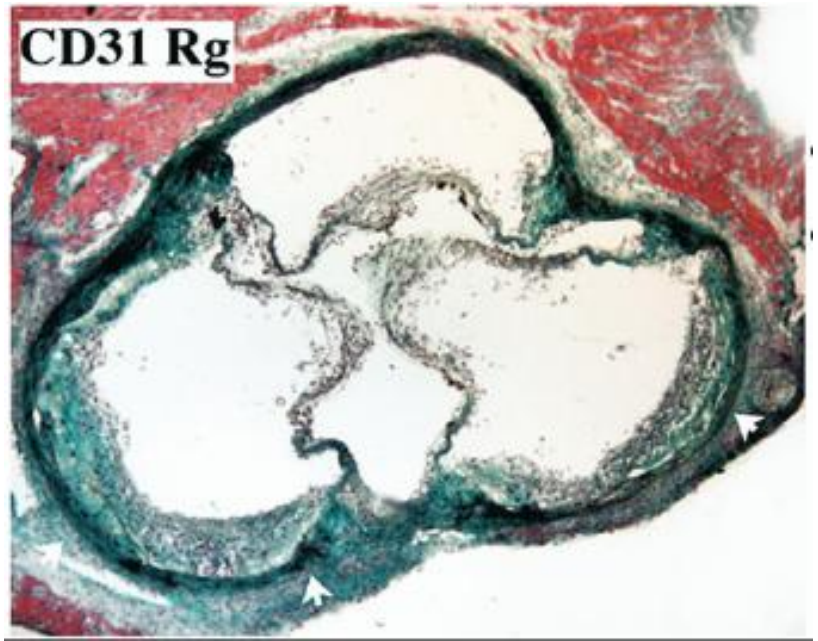
Atheroprotective Effect of CD31 Receptor Globulin Through Enrichment of Circulating Regulatory T-Cells

Emilie Groyer, MSc,* Antonino Nicoletti, PhD,* Hafid Ait-Oufella, MD,†
Jamila Khallou-Laschet, PhD,* Aditi Varthaman, MSc,* Anh-Thu Gaston, BSc,*
Olivier Thaunat, MD,* Srini V. Kaveri, DVM, PhD,* Radek Blatny, MSc,‡ Hannes Stockinger, PhD,‡
Ziad Mallat, MD, PhD,† Giuseppina Caligiuri, MD, PhD*





CD31 is a key coinhibitory receptor in the development of immunogenic dendritic cells

Marc Clement[a], Giulia Fornasa[a], Kevin Guedj[a], Sanae Ben Mkaddem[b], Anh-Thu Gaston[a], Jamila Khallou-Laschet[a], Marion Morvan[a], Antonino Nicoletti[a], and Giuseppina Caligiuri[a,1]

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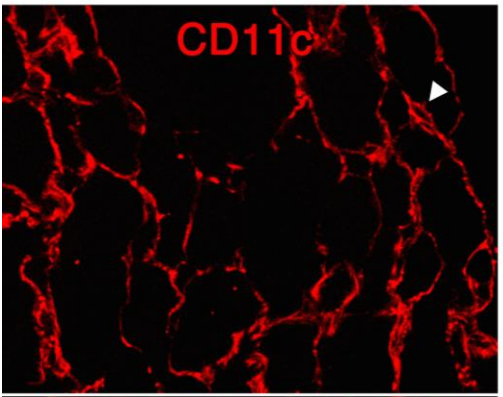
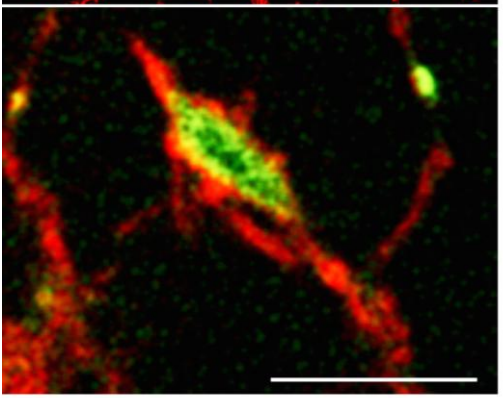
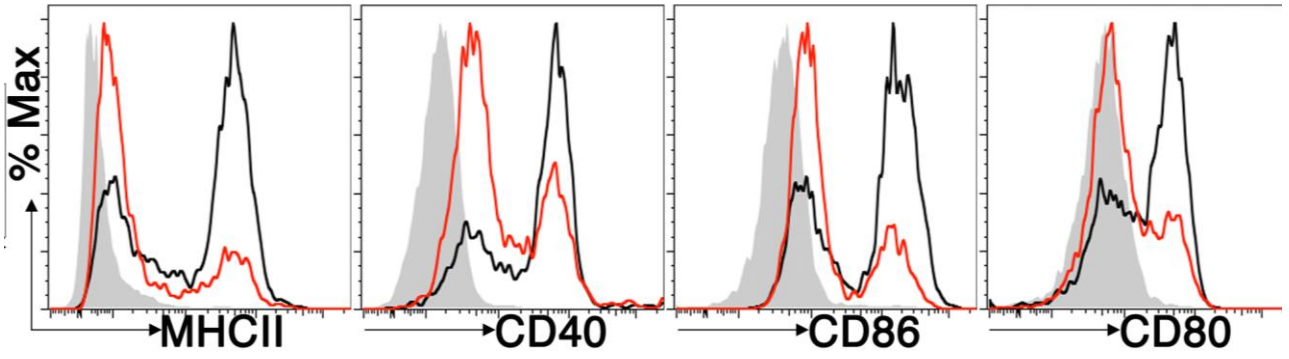
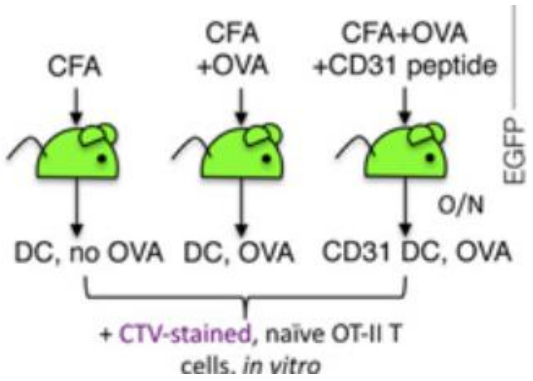
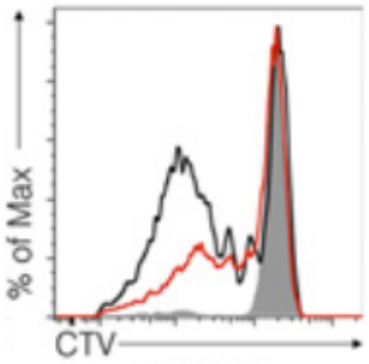
Edited by Ira Mellman, Genentech, Inc., South San Francisco, CA, and approved February 10, 2014 (received for review August 6, 2013)

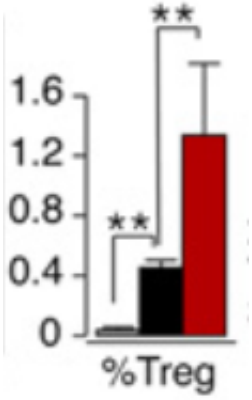


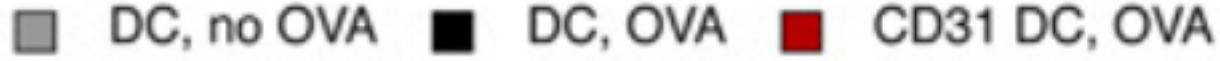












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Upholding the T cell immune-regulatory function of CD31 inhibits the formation of T/B immunological synapses *in vitro* and attenuates the development of experimental autoimmune arthritis *in vivo*

Marc Clement [a, b, c], Giulia Fornasa [a, b, c], Stéphane Loyau [a, b, c], Marion Morvan [a, b, c], Francesco Andreata [a, b, c], Kevin Guedj [a, b, c], Jamila Khallou-Laschet [a, b, c], Paola Larghi [d], Delphine Le Roux [e, f, g], Georges Bismuth [e, f, g], Gilles Chiocchia [h, i, j], Claire Hivroz [d], Debra K. Newman [k], Antonino Nicoletti [a, b, c], Giuseppina Caligiuri [a, b, c, *]

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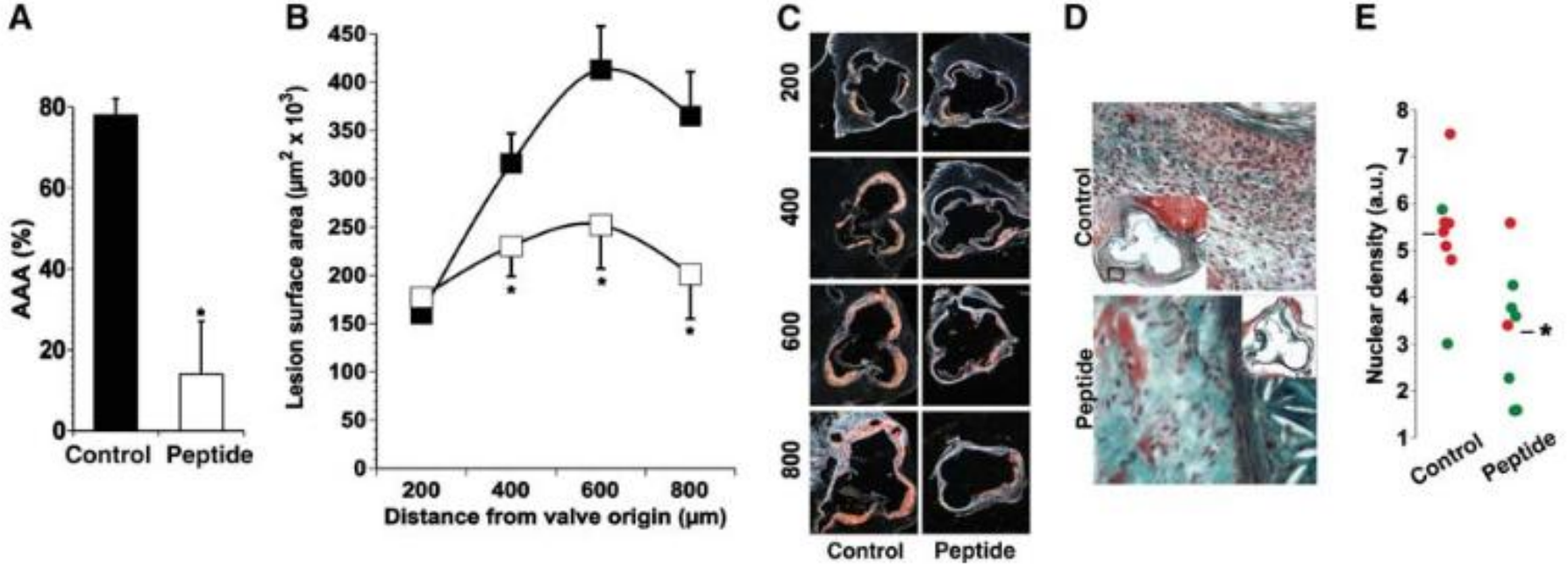
A CD31-derived peptide prevents angiotensin II-induced atherosclerosis progression and aneurysm formation

Giulia Fornasa[1], Marc Clement[1], Emilie Groyer[1], Anh-Thu Gaston[1], Jamila Khallou-Laschet[1,2], Marion Morvan[1], Kevin Guedj[1], Srini V. Kaveri[3], Alain Tedgui[4], Jean-Baptiste Michel[1], Antonino Nicoletti[1,2], and Giuseppina Caligiuri[1]*



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Atherosclerosis & Vascular Biology
ESC Working Group

SECTION III
Pathogenesis of atherosclerosis

Take home message

- T and B cells play both a beneficial and deleterious role in atherosclerosis
- The critical issue does not reside in the antigenic specificity but rather in the appropriateness of the response
- Appropriate T and B cell response require a fine regulation provided by an orchestra of co-stimulatory and co-inhibitory immune receptors
- CD31, a co-inhibitory immune receptor, plays a critical role in the homeostasis of the circulation

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