Immunosuppression in Atherosclerosis
Mobilizing the Opposition Within

Jörg J. Goronzy, MD; Cornelia M. Weyand, MD

Acute inflammation is short-lived and characterized by the recruitment of polymorphonuclear granulocytes followed by monocytes. Inflammation is self-amplifying and intensified through the sequential release of lipid mediators, cytokines, and chemokines. By 48 to 72 hours, granulocytes are replaced by lymphocytes. Despite the domino effect of a few inflammatory cells recruiting millions of others, inflammation is usually self-limited, resolving within days to weeks. If not, the response switches to chronic mode. Lymphocytes then dominate the force of inflammatory cells, and new rules dictate cell-cell communication, collateral damage, and repair. Most diseases categorized as chronic inflammatory disorders persist over decades, bringing a much greater time dimension to the inflammatory process. Although atherosclerosis is a quintessential chronic, persistent inflammatory disease, it differs from most others because its smoldering disease activity lasts for 30 to 70 years.

Obviously, the mechanisms of acute and chronic inflammation are distinct; granulocytes and lymphocytes utilize fundamentally different means to recognize danger and react. Neutrophils, which live only for hours and generate no memory of their engagement with harmful substances, sense microbial and nonmicrobial danger signals through inherited receptors that are similar in all hosts. Lymphocytes, however, are long-lived cells; some survive for decades and recirculate between lymphoid organs and peripheral tissues. Each lymphocyte has a unique receptor, generated by a gene rearrangement process, and the lymphocyte repertoire is tailored for each individual. When mobilized in immune responses, lymphocytes undergo clonal burst, differentiate into distinct types of effector cells, and “memorize” information about the antigen. Age, sex, infections, and environmental exposures mold the individual’s lymphocyte pool and essentially alter how each of us mounts chronic inflammatory responses. Varying clinical manifestations of chronic inflammatory disease are likely to reflect each patient’s unique immune repertoire.

Despite enormous progress in our understanding of the cellular players, receptors, and signaling pathways, we do not understand why an inflammatory response, meant to last for days to weeks, evolves into a persistent lesion. We do not know whether the signals, receptors, and mechanisms driving acute, lasting, but finally resolving inflammation are equally important for decades-old disease lesions. We are now beginning to examine the impact of time and tissue space dimensions in modulating immunocompetence and chronic inflammatory disease but depend on experimental models that are imperfect in recapitulating the longevity of human life or the dimensions of human arteries affected by atherosclerosis. Cellular and molecular immune activation profiles in long-standing inflammatory disorders appear to overlap with patterns in functional and self-limited antimicrobial host responses, giving hope that molecular rules are shared among different types of immunoinflammatory responses, including coronary atherosclerotic lesions in a 60-year-old patient.

Inflammatory reactions are programmed to resolve. Failure to do so leads to tissue injury and remodeling of the affected organ. Ironically, the inability to stop the host’s most powerful protective tool is potentially more harmful and may cause more pathology than the instigator ever would have. Duration of immunoinflammatory processes not only results in collateral damage and impaired organ function, it also exhausts the immune system’s reserve pool and eventually undermines the ability to regenerate and preserve immune competent cells.

Numerous endogenous antiinflammatory mediators cool down immune responses, including the cytokines interleukin (IL)-10 and transforming growth factor (TGF)-β and lipid mediators such as lipoxin and cyclopentenone prostaglandins. IL-10 is recognized as a potent antiinflammatory mediator in atherosclerosis. Recent work has pointed toward pathways of receptor-mediated macrophage suppression. CD200 ligation by a ligand expressed on activated immune cells signifies an internal mechanism of dampening macrophage activation in inflammatory responses. Additionally, the receptor d’origine nantis, also called the stem cell–derived tyrosine kinase...
receptor, mediates a negative signal when bound by its ligand macrophage–stimulating protein, a serum protein generated during the coagulation cascade. Macrophages and dendritic cells encountering apoptotic cells receive strong inhibitory signals that arrest their activation and maturation. CD36, a scavenger receptor abundantly expressed in atherosclerotic plaque, is instrumental in recognizing apoptotic bodies. Accordingly, Mer tyrosine kinase deficiency in knockout models causes inefficient clearance of apoptotic material and autoimmunity.

T cells, the main drivers of relentless inflammation and key players in the atherosclerotic lesion, are enormously powerful, with single cells mediating adaptive immune responses. This unparalleled efficacy necessitates an equally robust machinery to inhibit T-cell activity. Human T-cell subsets express killer immunoglobulin-like receptors, which have long cytoplasmic tails with 2 immunoreceptor tyrosine–based inhibitory motifs. On ligation, the immunoreceptor tyrosine–based inhibitory motifs are phosphorylated, leading to activation of SHP-1. SHP-1 dephosphorylates multiple signaling molecules, including ZAP70, LAT, SLP-76, PLC-γ, and the guanine nucleotide exchange factor Vav-1. Dephosphorylation of Vav-1 has profound suppressive effects, including inhibition of cytoskeletal rearrangement. CD4 T cells from patients with acute coronary syndrome (ACS) express a spectrum of inhibitory KIRs yet fail to utilize these receptors to prevent T-cell activation. Contrarily, in these patients, KIRs with short cytoplasmic tails amplify T-cell receptor signaling and facilitate T-cell cytotoxicity toward endothelial cells. In essence, physiological mechanisms aimed at resolving inflammation may be defective in atherosclerotic plaque. Most molecules providing negative signals to T cells and macrophages remain unexplored as potential targets in atherosclerotic disease, yet a sophisticated antiinflammatory pathway network offers many potential immunosuppressive therapy targets.

Concepts of suppressor T cells entered the immunologic landscape in the 1970s but fell out of favor when attempts to elucidate the underlying molecular mechanisms failed. In the mid-1980s, through the work of Shimon Sakaguchi, a new type of regulatory T cell (Treg), CD4+CD25+ T cells, entered the stage and soon attracted deep interest. CD4+CD25+ T cells constitutively express the surface receptors CTLA-4 and GITR and use the transcription factor Foxp3. They are naturally occurring suppressor cells, possibly arising as a separate thymic lineage. Activated by self-antigen or non–self-antigen, they suppress T cells in an antigen-nonspecific manner. Cell membrane contact is necessary for this effect. CD4+CD25+ Tregs are centrally involved in maintaining self-tolerance and suppress aberrant or excessive immune responses. Not unexpectedly, they also impede cancer surveillance and disrupt effective antitumor immunity. Given their role in T-cell–mediated immunity, CD4+CD25+ T cells are now proposed as a panacea of immunomodulation. Evidence shows that they control the development of atherosclerosis in mice. Increasing their number or improving their function may effectively treat autoimmunity and help to establish transplantation tolerance. Attenuation of Tregs may emerge as a means of cancer immunotherapy. CD4+CD25+ T cells are far from the only type of regulatory T cells. TGF-β-producing Th3 cells, IL-10–producing Tr1 cells, and CD8+CD28− T cells have all been described as exhibiting effective immunomodulatory effects.

The use of CD4+CD25+ T cells for therapeutic purposes is a major quest in immunotherapeutics today. Although adoptive cell therapy with suppressive CD4+CD25+ T cells has been envisaged as a novel strategy for fighting inflammatory disease, 2 reports in this issue of Circulation point toward the possibility that immunomodulatory interventions may function through the internal mobilization of CD4+CD25+ T cells.

In an elegant, appealing study, van Puijvelde and colleagues have induced antigen-specific immune tolerance resulting in effective suppression of atherosclerosis. Building on the concept that oxidized low-density lipoprotein (oxLDL) is critical for atherosclerotic lesion induction and progression, the authors set out to modulate anti-oxLDL immunity through vaccination by orally administering the antigen. Feeding mice 30 μg of oxLDL 4 times, they mitigated plaque formation in the carotid arteries and aortas of LDL receptor knockout mice on a Western diet. Oral antigen challenge even slowed progression of established disease. On induction of oral tolerance, frequencies of CD4+CD25+ T cells in the spleen increased from 0.8% to 1.6% and in mesenteric lymph nodes from 3% to 5.6%. CD25, Foxp3 and CTLA-4 mRNA increased in atherosclerotic plaques, suggesting Treg enrichment at the inflamed site.

Using an antigen-nonspecific approach, Steffens and colleagues present persuasive results from experiments in which they administered nonmitogenic antibodies targeting the CD3 molecule. This study builds on reports that anti–CD3-specific antibodies given short term to nonobese diabetic mice induce long-term remission of diabetes by stimulating TGF-β–producing CD4+CD25+ Tregs. Anti-CD3 treatment reduced atherosclerotic lesion size in the aortas of LDL receptor knockout mice fed a high-cholesterol diet. Injecting nonmitogenic anti-CD3 also slowed progression of established lesions. Surprisingly, this antiinflammatory effect left the plaque macrophage–T-cell infiltrate and the smooth muscle cell scaffold unaffected. CD3–directed therapy markedly decreased IFN-γ, TNF-α, and IL-10 production by lymph node cells and boosted their release of TGF-β. Although the treatment failed to increase CD4+CD25+ Treg proportions, Foxp3 mRNA increased by 2-fold in the spleen and blood. Although lesional macrophage, T-cell, and smooth muscle cell frequencies were maintained, the apoptotic cell rate in the atherosclerotic lesions increased, raising the intriguing question of which cell populations were undergoing apoptosis.

Both studies provide strong evidence that Tregs may be potent therapeutic targets, capable of disrupting early and chronic stages of atherosclerosis. Possibly, increased functional competence of Tregs accompanies other interventions that cool down plaque inflammation, and even pharmacological approaches, such as statin therapy, may call on the natural suppressive arm of the immune system. Selectively inducing nonresponsiveness to an antigen through vaccinating with a modified version of the antigen or by choosing the optimal
The route of antigen delivery is obviously appealing because nonselective immunosuppression is avoided. However, attempts to induce tolerance in human autoimmune diseases by oral antigenic challenge have been disappointing.

Mobilizing physiological antiinflammatory mechanisms and cell-based therapies are timely and may allow profound therapeutic effects but could have limitations. In the studies by Steffens et al.10 and van Puijvelde et al.9 CD4⁺CD25⁺ T cells were only partially effective. Lesion size decreased, but lesions did not clear, nor was the vessel wall repaired. This could be an inherent problem of using CD4⁺CD25⁺ Tregs. While suppressing the activity of proinflammatory T cells, Tregs may impair pathways urgently needed for orchestrating debris clearance and healing. Indeed, in the many animal models studied for the potential therapeutic benefits of manipulating CD4⁺CD25⁺ Tregs, only colitis can be cured. All other conditions are at best improved.

Clinical utility of Tregs in any autoimmune syndrome will depend on their survival in vivo, their ability to infiltrate relevant tissue niches, and their functionality in distinguishing between restorative and destructive immune responses. CD4⁺CD25⁺ T cells have been implicated in suppressing antitumor responses6 and certainly are capable of impeding antimicrobial immune responses. As much as inhibiting inflammation may be clinically desirable, the dangers of unwanted immune suppression are clear. This may be more relevant in nonspecific approaches, such as anti-CD3 target therapies. Steffens et al.10 demonstrate profound declines in the T-cell counts of treated animals. The data are in line with reports that in patients with newly diagnosed diabetes, responsiveness to anti-CD3 treatment was associated with a decline in the CD4/CD8 ratio, suggesting systemic and persistent immunosuppression.13

A significant challenge for translating results from experimental models to clinical application comes from the duration and complexity of the atherosclerotic disease process itself.14 Atherosclerosis persists for decades, a time frame that cannot be mimicked in rodent models. Cellular players, communication platforms, receptor-ligand pairs, and processes are dynamic and adaptive. Thus, suppressing early stages of atherosclerosis, interfering with persistent disease, or, more ambitiously, stopping immune processes involved in plaque instability and ACS will be distinct in humans (Figure). Preventing naive CD4 T-cell priming is different from blocking memory CD4 T-cell restimulation. Furthermore, the unstable human plaque is clearly distinct from an antigen-recognition and priming site; highly differentiated CD4 T–effector cells characterize inflamed atherosclerotic plaque. Plaque-residing CD4 effector T cells are senescent and utilize regulatory receptors not even present on naive T cells.15 Patients with ACS have CD4 T-cell populations that are powerful cytotoxic effectors,16 killing endothelial cells17 and triggering death receptors on vascular smooth muscle cells.18 Some effector pathways, for example, TNF-related apoptosis-inducing ligand (TRAIL)-mediated vascular smooth muscle cells apoptosis, are reminiscent of antitumor immune responses. Such highly differentiated T cells may be resistant to the suppressive effects of CD4⁺CD25⁺ T cells, such as shown in joint inflammation.19 Last but not least, the patient’s age must be considered. Thymic T-cell production is exhausted by the fifth decade of life, forcing the system into alternative modes of T-cell generation, which determine the immune system in the elderly and are partially responsible for a proinflammatory state. Whether CD4⁺CD25⁺ T cells have any effect inhibiting end-differentiated CD4 T effector cells in the tissue of a 60-year-old individual is unknown. These
considerations, however, do not detract from the intellectual beauty and tremendous therapeutic potential of the concept that unwanted inflammation can be treated by mobilizing the internal opposition within the immune system.

Acknowledgments

We thank Dr Sergey Pryshchep for preparing the figure and Tamela Yeargin for editorial support.

Sources of Funding

This work was funded in part by grants from the National Institutes of Health (RO1-HL-63919, RO1-EY-11916, and RO1-AG-15043).

Disclosures

None.

References


Key Words: Editorials • atherosclerosis • immune system • inflammation • lymphocytes • plaque • T-lymphocytes, regulatory
Immunosuppression in Atherosclerosis: Mobilizing the Opposition Within
Jörg J. Goronzy and Cornelia M. Weyand

Circulation. 2006;114:1901-1904
doi: 10.1161/CIRCULATIONAHA.106.656751
Circulation is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 2006 American Heart Association, Inc. All rights reserved.
Print ISSN: 0009-7322. Online ISSN: 1524-4539

The online version of this article, along with updated information and services, is located on the
World Wide Web at:
http://circ.ahajournals.org/content/114/18/1901

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Circulation can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to Circulation is online at:
http://circ.ahajournals.org/subscriptions/