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A snappy new concept for APS

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In this issue of Blood, Ağar and colleagues present data for a novel explanation of how an antigenic target on β2GPI, a central protein in the APS disease process, can become available for binding by antibodies.1

The antiphospholipid syndrome (APS), an autoimmune thrombophilic disorder, was recognized as a diagnostic entity by astute clinical observations of the coincidence of thrombosis and/or recurrent miscarriages with empirically derived clinical tests.2 These include immunoassays that were derived from the biologic false-positive syphilis test and blood tests that detect inhibitors of phospholipid-dependent coagulation reactions, known as lupus anticoagulant assays.

In retrospect, the “antiphospholipid” terminology is erroneous and reflects the initial belief that phospholipids themselves are the targets of the antibodies. This misconception—but not the name of the syndrome—was corrected approximately 20 years ago, when it was discovered that the actual target antigens are phospholipid-binding proteins, particularly β2-glycoprotein I (β2GPI), a relatively abundant plasma protein whose biologic function(s) has not been established.

X-ray crystallographic studies3,4 revealed that the protein, with its 5 homologous domains, has a J-shaped structure that is analogous to a fishhook with a “barb” consisting of a hydrophobic loop with surrounding positively charged residues near the carboxyterminus on domain V. This region allows the protein to bind bilayers containing anionic phospholipids via affinity for negatively charged polar heads and insertion of the loop within the hydrophobic middle of the bilayer (see figure). The binding of β2GPI to a phospholipid bilayer via the “barb” on domain V unsnaps this coiled protein into its open fishhook conformation, thereby exposing the epitope (see figure). The authors present a convincing body of evidence for this idea, including electron microscopic images, differential trypsin digestion profiles, surface plasmon resonance binding studies of the affinity of recombinant domains for each other, and functional studies that compare the anticoagulant effects of the conformations.

These results add a significant detail in our understanding of the APS disease process, which can be outlined as follows: β2GPI is present in plasma where it has been suggested to play a role in the clearance of apoptotic cells and microparticles. The circular protein then undergoes a conformational change when it comes into contact with membranes of cells that have entered the apoptotic program and express anionic phospholipids; domain V unsnaps from domain I and inserts into the bilayer and the fishhooks aggregate into disc-like clusters,5—a process that is probably required for the protein’s biologic role. In patients who have a genetic susceptibility for...
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PLDing a case for angiogenesis

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In this issue of Blood, Zhang et al identify the Src-PLD1-PKCγ axis as critically involved in the process that causes ROP, highlighting new potential targets for therapy.1

Vascular endothelial growth factor (VEGF) is acknowledged as the predominant regulator of angiogenesis; blockade of VEGF signaling is central in therapy for numerous cancers and the vascular retinopathies of diabetes, age-related macular degeneration, and retinopathy of prematurity (ROP). Although anti-VEGF monotherapy shows substantial results, in many cases it is becoming increasingly appreciated that combination therapy will be necessary.2 Tumors have demonstrated various degrees of intrinsic refractoriness or the development of treatment-related resistance. For age-related macular degeneration, expected to soon affect nearly 3 million people in the United States, the optimal therapy of long-term monthly intraocular injections of anti-VEGF agents will likely prove unsustainable for practical and clinical reasons. Thus, effective treatment will require the combination of anti-VEGF therapy with conventional chemotherapeutic agents, radiotherapy or phototherapy, or the targeting of multiple components of VEGF-activated processes.

The breadth of disease states in which VEGF-induced angiogenesis plays a central role correlates to a large and incompletely defined population of regulatory molecules of VEGF signaling, many likely to be tumor context-specific. The Zhang paper defines the players in a model of ROP and thus identifies potential new specific targets for therapy. In their report, Zhang and colleagues demonstrate that an intact VEGF-signaling axis—constituted by the sequential activation of Src, phospholipase 1 (PLD1), and protein kinase Cγ (PKCγ)—mediates the pathologic neovascularization seen in the oxygen-induced retinopathy model of ROP. This axis was delineated in vitro using chemical inhibitors (1-butanol and propranolol) and in vivo using intraocular administration of siRNAs specific to individual components of the pathway.

Previous work has identified the protein tyrosine kinase activity of Src as a regulator of both VEGF expression and of responses to VEGF stimulation.3 Zhang et al are the first to report activation of PLD1 by Src. Furthermore, they demonstrate that Src-dependent PLD1 activation is required for subsequent activation of PLCγ. The recent development of selective small molecule inhibitors that target Src and the demonstration that Src inhibition can attenuate chemoresistance of some solid tumors suggests a possible clinical use of Src inhibition in vascular retinopathy.

Investigation of the role of bioactive lipids in regulation of angiogenesis is a burgeoning area of research likely to result in a new class of therapeutic agents.4,5 Of particular topical interest are the bioactive lipids PLD1, phosphatidic acid (PA), lyso-phosphatidic acid (LPA), and sphingosine-1-phosphate (SIP). After activation by any of a variety of intracellular factors (including...
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