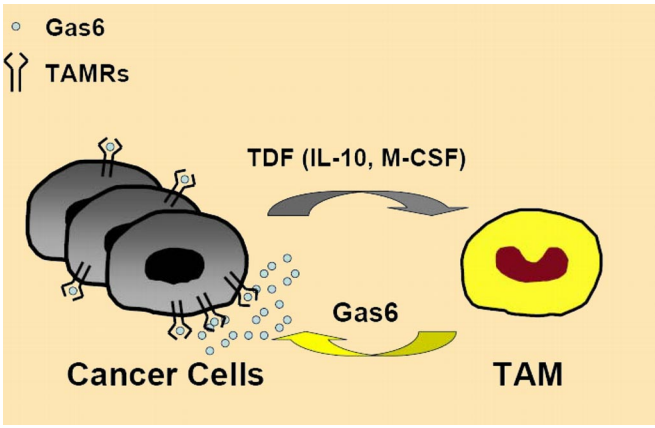

Tumor-derived factors (TDF), likely including IL-10 and M-CSF, educate TAM for Gas6 production to fuel cancer cell proliferation. TAMRs indicates TAM family of receptor tyrosine kinases; TAM, tumor-associated macrophages; M-CSF, macrophage colony-stimulating factor; and IL-10, interleukin-10.

an improved prognosis was observed in patients with renal cell carcinoma (RCC),[6] demonstrating the complexity of the system.

Loges et al provide new evidence on the regulation and significance of Gas6/Axl activity in cancer. Within the tumor microenvironment, tumor-associated macrophages acquire the capacity to express high levels of Gas6, suggesting that unidentified tumor-derived factor(s) contributes to their protumoral education.[7] Among the possible candidates, IL-10 and M-CSF were able to induce Gas6 upregulation in vitro. Interestingly, these factors promote M2 polarization of macrophages[7] and help shape the protumoral M2 phenotype (see figure) of tumor-associated macrophages.[7,8] It remains to be determined whether Gas6 represents a prototypical M2 marker and whether expression of high Gas6 levels is a feature of other tumor-associated myeloid cells, including myeloid-derived suppressor cells.[9]

Using different ectopic and orthotopic syngeneic tumor models, Loges et al demonstrate that inhibition of Gas6 does not influence accumulation of $CD45^+$ leukocytes, tumor-associated fibroblasts, angiogenesis, and coagulation, but it is limited to the selective inhibition of cancer cell proliferation. Further, the observed reduction in metastasis formation was secondary to reduced primary tumor growth. This scenario is rather surprising, as TAMRs are largely expressed by stromal components, including endothelial cells and leukocytes.[3]

The complex regulation of the Gas6 actions is highlighted by reports on its functional interplay with central regulators of inflammation and cell metabolism. Whereas activation of Gas6/Mer-dependent PI3K/akt pathway was reported to influence NF-κB activation,[3,10] alteration of the Von Hippel–Lindau tumor suppressor gene in clear cell renal cell carcinoma patients (ccRCC), a condition promoting a pseudohypoxic response and the consequent activation of the hypoxia-inducible factor-1 (HIF-1),[11] results in enhanced expression of Axl.[6] As tumor-associated macrophages accumulate in the hypoxic regions of a tumor[12] and activation of PI3K/Akt leads to HIF-1 activity,[13] it is tempting to speculate a possible link between the hypoxia/HIF-1 and Gas6/Axl pathways.

The contrasting information on the prognostic significance of Gas6/Axl in cancer, along with the multiple mechanisms involved in its regulation, suggest that the complexity of its biology is also contest- and tissue-specific. Although development of TAMRs antagonists may have therapeutic limitations, due to the possible induction of autoimmune disorders (eg, Lupus-like syndrome),[3] the cancer-restricted action of Gas6, along with its inertness on stroma cells, suggests that its therapeutic inhibition may well integrate strategies targeting cancer-related inflammation.

Conflict-of-interest disclosure: The author declares no competing financial interests. ■

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● ● ● PLATELETS & THROMBOPOIESIS

Comment on Kasirer-Friede et al, page 2274

If Virchow were to meet Newton

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In this issue of *Blood*, Kasirer-Friede and colleagues show that ADAP is a component of a signaling system triggered when blood flow pulls αIIbβ3 bound to fibrinogen. It serves to convert tension into a biochemical response that stabilizes platelet attachment by directing lamellipodia formation.

Platelets are mechanical devices. Frictional forces generated by flowing blood induce an adhesive couple by altering the conformation of the extracellular domain of platelet glycoprotein (Gp) Ibα and by exposing the A1 domain of its ligand von Willebrand factor

(VWF). A catch bond is formed that tightens as the platelet is pulled by the flowing blood. This bond triggers microvascular hemostasis and acute atherothrombosis. It is capable of withstanding vessel wall shearing forces that sweep away all other biochemical reactions, such as those leading to leukocyte attachment and fibrin deposition. The bond slips, slowed platelets roll or flip as new bonds form, catch, and slip; and stable attachment finally develops through platelet GpVI binding collagen and activated αIIβ3 binding VWF and fibrinogen.[1] Shear-induced binding of VWF to GpIbα is sufficient for αIIbβ3 activation, and it is noteworthy that Kasirer-Friede and colleagues previously reported that this response is modulated by adhesion and degranulation promoting adapter protein (ADAP) under flow conditions found in arterioles and stenotic arteries (a shear rate of 1500/sec).[2]

Mechanotransduction is the cellular process by which physical forces are sensed and converted into biochemical signals.[3] Molecular mechanisms of mechanotransduction are poorly understood but perhaps best worked out for integrins. Integrins transduce forces only after they are ligand-bound.[3] Integrin-mediated mechanotransduction triggers cellular responses aimed at balancing the force applied, whether the force is shear, strain, or stretch.[4] In this regard, the integrin is used by a cell to permit compliance with Newton's third law that "for every action, there is an equal and opposite reaction." Forces applied to cells bound to matriceal proteins at focal contacts or focal adhesions are transduced from the integrin's extracellular ligand-binding domain to its β chain's cytoplasmic domain connected to the cytoskeleton. Integrin-transduced forces cause cytoskeletal deformations and actomyosin-driven cytoskeletal remodeling that generate an opposing force, which in a platelet is the force used to maintain adhesion in the face of flowing blood. The biochemical mechanisms by which these responses occur are vague but are thought to be allosteric, reversible, and bidirectional. It is also hypothesized that mechanical forces accelerate chemical reactions, spatial arrangements, and colocalizations that are diffusion-limited under static conditions.[5]

The article by Kasirer-Friede et al identifies several of the allosteric interactions that operate to enhance mouse platelet adhesion under shear conditions.[6] The hypothesis that ADAP regulates the actin cytoskeleton under hydrodynamic forces focuses on integrin-mediated mechanotransduction because of previous work showing that shear stress modulates human αIIbβ3-mediated adherence through a mechanism that involves β3/cytoskeletal interactions and SLP-76/ADAP colocalization[7]; that integrin-induced outside-in signaling in static T cells utilizes the ADAP partner SLP-76[8]; and that ADAP, because it mediates "inside-out" signaling to αIIbβ3,[2] is likely to operate allosterically and therefore bidirectionally.[5] By using normal and ADAP deleted mice to examine in vivo carotid thrombosis and ex vivo platelet deposition onto fibrinogen in a flow chamber perfused with blood at a shear rate of 500/sec (recapitulating flow conditions in large arteries and arterioles), these investigators prove that ADAP fine-tunes αIIbβ3-mediated adhesion. ADAP is a component of a cytoskeletal and kinase-mediated signaling cascade that stimulates increased surface contact points through the generation of lamellipodia, a mechanism as teleologically sound as protecting a tent from a windstorm by adding support cables while spreading, flattening, and tethering the tent to the ground with hundreds of additional spikes.

What is the clinical significance of these results? At this time probably distant, as ADAP functions under both physiologic and pathologic shear conditions, suggesting that an ADAP inhibitor would have antihemostatic effects separate from any antithrombotic activity. In addition, because ADAP is a protein found in all hematopoietic cell lineages, its inhibition would carry the risk of immune suppression, as demonstrated by children who suffer severe bleeding and recurrent infections from a point mutation in kindlin-3 that disrupts its integrin-activating function.[9]

Nonetheless, it is my view that this study is more important than its translational potential because it excites the process of blending clinical medicine with mechanical engineering (ie, bioengineering) within our community of hematologists. Virchow's conceptual triad of the pathogenesis of thrombosis (perturbations in blood, blood vessel, and/or blood flow) has done far better than simply withstanding the test of time. It has provided a conceptual framework for discoveries inconceivable 150 years ago. Considering that Virchow identified "stasis" as a cause of thrombosis, he probably would have scoffed at the idea that "fluidity" can also cause thrombosis. But today, after more than a century of technological developments represented by work like that of Kasirer-Friede et al, one can imagine that Professor Virchow, if given the chance, would be delighted—and enlightened enough—to aggressively seek a collaboration with Sir Isaac Newton.

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● ● ● THROMBOSIS & HEMOSTASIS

Comment on Rand et al, page 2292

A novel antiphospholipid antibody agent?

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In this issue of *Blood*, Rand and colleagues have shown that in vitro hydroxychloroquine can reverse antiphospholipid antibody–mediated disruption of the annexin VA crystalline shield that protects phospholipid bilayers from taking part in coagulation reactions.[1] The protective effect of hydroxychloroquine against antiphospholipid antibody–induced damage was seen on both cultured endothelial cells and a syncytialized trophoblast cell line.