also under development for the treatment of human cancer, may have an impact on the surface binding of MMP-9.

An interesting technical achievement by Redondo-Muñoz and colleagues is the very high efficiency of siRNA transfection of primary CLL B cells with little induction of apoptosis. Transfection of primary CLL B cells is technically challenging and typically observed low transfection frequencies have been a major obstacle in CLL research. The transfection strategy used by the authors, if reproducible by other researchers, may be a significant step forward.

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

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HEMOSTASIS

Comment on Stegenga et al, page 82

Glucose, insulin, coagulation: endotoxin as exohormone

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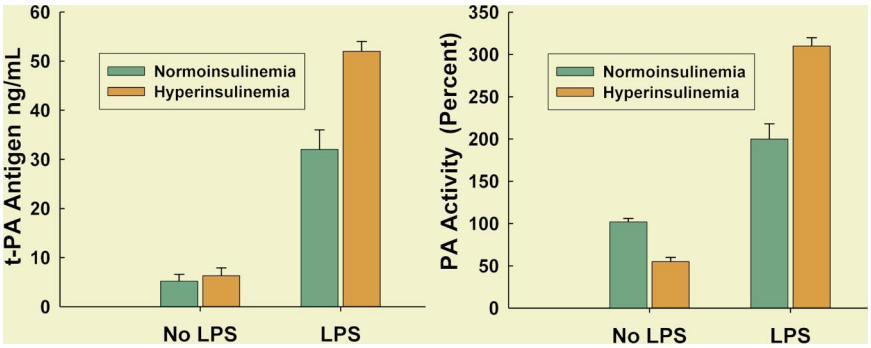
In this issue of *Blood*, Stegenga and colleagues at the University of Amsterdam report an elegant series of studies probing the independent effects of glucose and insulin on endogenous inflammatory and coagulant responses in a well-characterized model of acute inflammation—bolus endotoxin challenge in human volunteers.

Diabetes mellitus affects more than 170 million people worldwide, or 2.8% of the population of the planet; its prevalence is expected to rise to 4.4% by 2030.[1] The morbidity of diabetes is linked to a spectrum of procoagulant and inflammatory derangements. Conversely, abnormalities of glucose regulation are common to a number of acute and chronic inflammatory disorders.[2] By clamping plasma levels of glucose or insulin at normal or elevated levels, Stegenga and colleagues demonstrate that glucose and insulin differentially regulate neutrophil and coagulant response to endotoxin challenge. Hyperglycemia alone inhibits the release of neutrophil elastase and enhances levels of thrombin: antithrombin (TAT) complexes, whereas hyperinsulinemia reduces plasminogen activator (PA) activity and increases both the level and the activity of plasminogen activator inhibitor type 1 (PAI-1). Neither significantly alter circulating levels of key cytokines whose release is induced by endotoxin, nor do they affect neutrophil numbers or markers of endothelial-cell activation. The effects are specific to the regulation of coagulation, and both promote a net procoagulant state.



Circulating levels of t-PA antigen and PA activity in endotoxemic volunteers were both increased by hyperinsulinemia, suggesting that increased insulin primes for an augmented response to endotoxin.

These are important studies, but their interpretation is complicated. Endocrine interactions are dynamic: elevated or depressed levels of one hormone evoke a response that modulates levels by altering the release of that hormone, or by inducing the release or inhibition of a counterregulatory hormone. These dynamic interactions were inhibited in this study, both by the clamp technique and by the pretreatment of study subjects with somatostatin to inhibit endogenous insulin release. Nonetheless, 3 key conclusions emerge.

First, hyperglycemia alone supports a procoagulant state by increasing levels of soluble tissue factor and TAT complexes. This observation provides a further rationale for maintaining strict euglycemia in critically ill patients.[2]

Second, insulin alone, independent of endotoxin, supports a procoagulant state through the induction of PAI antigen and activity, and inhibition of plasminogen activator activity. Thus, exogenous insulin may prove insufficient to reverse the procoagulant state associated with hyperglycemia.

Third, and perhaps most intriguing, is the insight that arises from the characterization of the interaction of an endogenous hormone, insulin, with what might legitimately be considered an exogenous hormone, endotoxin. When data from the current report are compiled with those from the authors' earlier studies in volunteers who had not received endotoxin, it becomes apparent that endotoxin, itself a potent inducer of a procoagulant and proinflammatory state, synergizes with insulin to further increase levels of t-PA antigen and plasminogen activator activity[3] (see figure). Endotoxemia is common in critical illness[4] in which derangements of glucose-insulin homeostasis are particularly prominent,[2] and insights such as those provided by Stegenga et al underline the challenges of modulating the hormonal milieu in a complex and vulnerable patient population.

Scientific experimentation is inherently reductionist, and imperfect in its capacity to encompass the nuances of diseases that arise within the complexity and intrinsic variability of whole organisms. Nonetheless,

thoughtfully applied models such as that reported by Stegenga et al provide the opportunity to tease systematic insights out of the chaos of adaptive biology. And in this initiative, they can offer valuable perspectives on the capacity of living organisms to respond to the vicissitudes of a frequently hostile world, and on the challenges we face in trying to treat the consequences.

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

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IMMUNOBIOLOGY

Comment on Liu et al, page 120

Thymocyte-fate decisions: bittersweet new flavor

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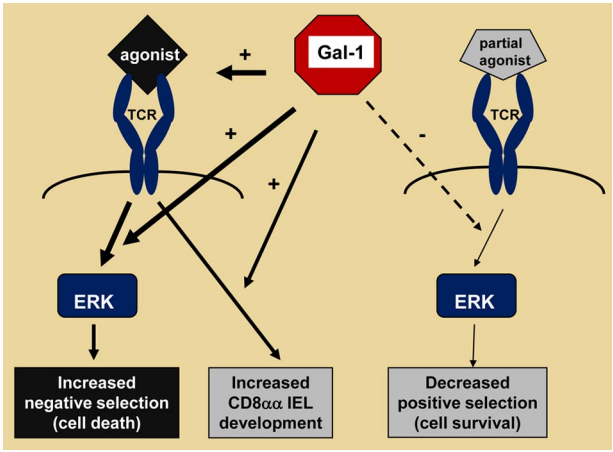
In this issue of Blood, Liu and colleagues demonstrate a novel mechanism for regulation of thymocyte-fate decisions by which a sugar-binding protein, galectin-1, enforces death while opposing survival of developing thymocytes.

The immune system's central tolerance is shaped in part in the thymus during T-cell development. Negative selection removes overtly autoreactive thymocytes, while positive selection promotes survival and development of self-tolerant T cells. Ligand binding to the T-cell receptor (TCR) controls both cell-fate decisions.[1] High-affinity agonists are thought to induce thymocyte death during negative selection, or to promote the generation of regulatory T cells such as the CD8αα intestinal intraepithelial lymphocytes (IELs).[1,2] On the other hand, partial agonists with low or moderate affinities trigger positive selection and thymocyte differentiation into T-helper or cytotoxic lineages.[1] Liu and colleagues add a new flavor to this mechanism by demonstrating that a particular lectin, galectin-1 (gal-1), contributes to discriminating TCR-controlled fate decisions by promoting the association of the TCR and its ligand. This regulatory mechanism enforces agonist-driven signals leading to negative selection, while opposing partial agonist-induced thymocyte survival.



gal-1 is a modulator of thymocyte-fate decisions with opposing activities. By enforcing agonist ligand binding to the TCR, gal-1 promotes fast and transient activation of ERK and negative selection of autoreactive thymocytes, while permitting the generation of regulatory CD8αα IELs. In contrast, gal-1 expression antagonizes ERK signaling in thymocytes undergoing partial agonist-driven positive selection.

gal-1 is a member of a family of β-galactose binding proteins that, by cross-linking cell-surface receptors, influence fate decisions in a variety of cells and tissues.[3,4] Relevant to T-cell ontogeny, thymic cortical epithelial cells that present antigens to thymocytes also express gal-1.[3] This suggests a role for gal-1 in thymocyte-fate decisions. Liu and coauthors have investigated this question by using 2 distinct MHC class I–restricted, TCR-transgenic mouse models bearing a targeted deletion of gal-1. Ablating gal-1 markedly increased positive selection in these mouse models, while negative selection was decreased. Augmented positive selection correlated with a higher number of functional CD8 T cells in the spleens of gal-$1^{-/-}$ TCR-transgenic animals. Interestingly, Liu and colleagues found a higher degree of ERK activation in the post-selection thymocytes of gal-$1^{-/-}$ mice. This result might explain the augmented positive selection observed in the absence of gal-1. Indeed, the intracellular kinase ERK is required for transducing TCR-triggered thymocyte-fate decision signals, and its distinct kinetics of activation represent a specific signature for positive and negative selection.[1] From their findings, the authors conclude that endogenous gal-1 opposes partial agonist-driven positive selection of conventional CD8 T cells by lowering the degree of ERK activation. It will be interesting to document, in the future, whether gal-1 modulates the kinetics of TCR partial agonist-driven ERK activation, as well as other TCR-linked signaling molecules such as the ERK-related kinase, JNK.

Nonetheless, Liu and colleagues demonstrate that by enhancing the association of agonist ligands to the TCR, gal-1 increases the magnitude of the fast and transient ERK activation signature that is characteristic for negative selection. Thus, by enforcing agonist-driven signal transduction, gal-1 promotes the removal of potentially autoreactive thymocytes from the repertoire. Interestingly, the authors found that gal-1 also promotes the selection of CD8αα IELs. These regulatory T cells preferentially migrate to the gut after positive selection on TCR agonists instead of partial agonists.[2] Surprisingly, the authors observed a significant increase of CD8αα IELs in the absence of gal-1 and in the context of TCR partial agonists. This might reflect a means by which gal-1 can influence the control of selection of T cells bearing autoreactive potential. However, additional work needs to be done to determine whether CD8αα IELs can be selected by partial agonists, or whether these ligands simply promote better peripheral homing of CD8αα IELs in the absence of gal-1. Nonetheless, Liu and colleagues highlight a novel mechanism for gal-1 contribution to central tolerance via shaping the reservoir of CD8 T cells. This might lead to novel therapeutic strategies in which the addition, or specific blocking, of gal-1 could be used as a unique tool to fine-tune the development of T cells with