El Dorado Found: Calreticulin Mutations in JAK2 V617F-Negative MPNs


Activation of the JAK-STAT pathway is a cardinal feature of the Philadelphia chromosome-negative myeloproliferative neoplasms (MPNs). Dysregulation of this signaling axis can be attributed to the somatic JAK2 V617F mutation in ~98 percent of patients with polycythemia vera (PV), and 50 to 60 percent of patients with essential thrombocythemia (ET) or primary myelofibrosis (PMF). Less commonly, JAK-STAT activation may result from JAK2 exon 12 mutations in 1 to 2 percent of PV cases or mutation of the thrombopoietin receptor, MPL, in 1 to 5 percent of ET and 5 to 10 percent of PMF cases. Rarely, JAK-STAT activation is due to mutations in negative regulators of JAK2, including SH2B3 (LUK) and CBL. Although mutations in tumor suppressors or components of the epigenetic and spliceosome machinery (e.g., TET2, ASXL1, EZH2, DNMT3A, IDH1/2, U2AF1, SRSF2, SFPQ1, TP53) are found in the chronic or blast phase of MPNs, these mutations do not account for the constitutive JAK-STAT signaling observed in the 30 to 40 percent of MPN patients who are JAK2- or MPL-mutant-negative. With eight years passing since the discovery of JAK2 V617F, the question of whether another highly recurrent mutation would ever be found in MPN patients began to assume the mythical narrative of El Dorado, the Lost City of Gold.

Recently, however, two groups of investigators laid claim to the discovery of this elusive treasure. Dr. Jyoti Nangalia and colleagues from the laboratory of Dr. Tony Green at Cambridge, and Dr. Thorsten Klampfl and colleagues from the laboratory of Dr. Robert Kralovic in Vienna, in conjunction with teams of collaborators, used whole-exome sequencing to identify somatic mutations in the calreticulin gene (CALR) in the majority of ET and PMF patients with nonmutated JAK2. From the results of sequencing samples from large numbers of patients with malignant diseases, two important findings emerged: 1) CALR mutations are predominantly found in ET, PMF, and the MDS/MPN overlap disorder, refractory anemia with ring sideroblasts (RARS-T), and they are rare or absent in other myeloid or lymphoid neoplasms or solid tumors; and 2) CALR mutations are mutually exclusive of JAK2 and MPL mutations. In the study by Dr. Klampfl and colleagues, mutated CALR was found in 67 percent of ET and 88 percent of PMF without JAK2 or MPL mutations. In a similarly designed analysis by Dr. Nangalia and colleagues, 71 percent of ET, 56 percent of PMF, and 86 percent of post-ET MF patients exhibited CALR mutations. The latter study surveyed the genomic landscape of MPNs and found that the median number of somatic mutations in patients with PV, ET, and PMF was 6.5, 6.5, and 13, respectively. The finding of a more complex genotype in myelofibrosis is consistent with its heterogeneous and more aggressive clinical course compared with PV and ET.
The Changing Practice Landscape: What is ASH Doing to Help?

As a researcher, I count myself among many interested in ASH’s efforts to continue the fight for medical research funding. However, as a physician, I am equally concerned about appropriately incorporating new discoveries into my clinical practice, providing high-quality, evidence-based care to my patients, and ensuring that my concerns as a practicing hematologist are heard during this time of tumultuous change in our health-care system. As 2014 ushered in changes in board certification requirements, the launch of health exchanges, and revised reimbursement policies linked more closely to performance, I was reminded of how ASH helps its members provide quality care to patients while navigating through a transforming practice environment.

The ASH Academy is a good place to start, with its online platform that aids practitioners in maintaining certification and claiming Continuing Medical Education credits. The ASH PQR/SRS is an electronic tool that helps members submit quality measures to Medicare’s Physician Quality Reporting System for incentive payments. ASH provides several resources to help practitioners address commonly encountered clinical problems, including the Consultative Hematology Course held prior to State-of-the-Art Symposia (SAS) and during the annual meeting. The course focuses on non-malignant hematology and uses case-based presentations and interactive discussions to convey the information. The Highlights of ASH® meetings held annually in six North American cities and two international locations (March 29-30, in Asia; April 25-26, in Latin America) and SAS (September 12-13, in Chicago; and October 10-11, in Washington, DC) presented by world-renowned experts, are both small, focused meetings that help clinicians learn about evolving therapies and the latest treatment options along with clinical implications of basic research. In addition to ASH’s ever-popular pocket guides, a new publication, How I Treat: A Compendium for the Practicing Hematologist, flew off the shelves at the ASH annual meeting. The Compendium features 33 “How I Treat” articles published in Blood from 2010 through 2012 and updated for this special Compendium. Finally, the ASH Practice Partnership, a group of ASH members representing all types of practice settings, provides the Society with feedback both on the state of practice across the country and on the needs of practitioners. The Committee on Practice, in conjunction with the Committee on Government Affairs, spearheads advocacy efforts of importance to all practitioners, including mitigating drug shortages, championing insurance coverage parity for cancer treatments, and lobbying for physician payment reform.

These are some of the Society’s resources, which are all supported by the ASH Foundation and aimed at assisting the practicing hematologist. While we are operating in a period of uncertainty, be assured that ASH has your back!

Linda F. Burns MD
ASH and the European Hematology Association (EHA) have selected 20 early-career hematologists to participate in the 2014 Translational Research Training in Hematology (TRTH) program. (See names below.) Now in its fifth year, TRTH provides junior researchers from around the world with an unique, yearlong training and mentoring experience with the goal of fostering the next generation of global leaders in translational research. The TRTH program begins with a weeklong course held March 15–21 at a learning center near Milan, Italy, followed by a meeting in conjunction the EHA Annual Congress in June, and culminating in a gathering held coincident with the ASH annual meeting in December where trainees present the status of their research. To learn more about the TRTH program, visit www.hematology.org/Awards/TRTH/2632.aspx.

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Tiffany Chang, MD, University of California, San Francisco, San Francisco, CA, United States

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Richard Green, PhD, UMC Utrecht, Utrecht, Netherlands
Hubert Tsui, MD, PhD, University of Toronto, Toronto, Canada

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ASH Wants to Recognize Your Mentor
Deadline to Submit Nominations is April 4

Don’t miss the opportunity to recognize someone who has been instrumental in guiding and supporting your career development. The ASH Mentor Award was created to recognize outstanding mentors in the hematology community, including adult or pediatric hematologists, academic or community practitioners, basic or clinical researchers, hematopathologists, transfusion medicine specialists, and hematologists working in industry or government.

Nominations will be accepted throughout the year; nominations submitted after the April 4 deadline will be considered for recognition in 2015. Visit www.hematology.org/Awards/Mentor2505.aspx for more information and for instructions on how to nominate your mentor.

ASH Seeks Members to Develop a New Medical Educator Program

Are you passionate about medical education in hematology? Supporting hematology education for medical students, residents, and fellows; providing continuing medical education for practicing physicians; and creating new career options for hematologists are important goals for ASH. The Committee on Training is soliciting nominations for inaugural directors of a proposed program to contribute to the professional development of hematology educators, the ASH Medical Educators Institute.

ASH expects to select up to four individuals who will be charged with the initial conception and, pending approval from the ASH Executive Committee, implementation of the program. Nominees should possess an MD or equivalent medical degree and should have formal experience in medical education, a network of colleagues in both hematology and medical education, and experience in the development of new programs.

Self-nominations are encouraged. Nomination packets should include a personal statement conveying ideas about development of the institute that incorporates the applicant’s relevant past experiences, a CV, and one to two letters of recommendation from colleagues who can attest both to the expertise of the applicant and to the applicant’s past contributions to program development. More information on the proposed program and on the application and selection processes is available at www.hematology.org/Training/12311.aspx.

The deadline to submit applications is March 31. Please email application material to training@hematology.org. Contact the ASH Training Department at 202-776-0544 or training@hematology.org with any questions.

Submit an Abstract to the First ASH Meeting on Lymphoma Biology

The deadline to submit an abstract is April 29.* Go to www.hematology.org/Meetings/Lymphoma-Biology-Meeting/11507.aspx for more information.

Save the date for the first ASH Meeting on Lymphoma Biology scheduled for August 10-13, 2014, in Colorado Springs, CO. The keynote speaker is Klaus Rajewsky, MD, of The Max Delbrück Center for Molecular Medicine, in Berlin, Germany. Dr. Rajewsky will discuss immune regulation and cancer. At the meeting, attendees will learn about the current state of lymphoma investigation and about strategies for addressing future challenges. The meeting’s interactive poster sessions and informal networking breaks will provide opportunities to share ideas and establish collaborations. Visit the ASH website for the latest information about the program’s schedule, topics, speakers, and registration requirements and deadlines.

*Meeting attendance is not contingent upon submitting an abstract.

CORRECTION: In the President’s Column in the January/February 2014 issue, we inadvertently stated that ASH Bridge Program applicants must be independent physician scientists. Applicants are not required to be physicians. All eligible independent scientists may apply. Please refer to the ASH Bridge Program website (www.hematology.org/Awards/Bridge-Grants/8669.aspx) for eligibility criteria.
Ask the Hematologist

The Question

When do you recommend light transmission aggregometry as part of the evaluation of a patient with a suspected bleeding disorder?

My Response

Light transmission aggregometry (LTA) remains the reference method for measurement of platelet function in patients with suspected platelet function disorders (PFDs). Remarkably, the core principles on which LTA is based have not changed since it was first described by O’Brien and Born more than 50 years ago. Platelet-rich plasma is stirred in a cuvette that is placed between a light source and a photocell. The plasma is cloudy due to suspension of platelets and allows relatively little light to pass through. Upon the addition of an agonist, platelets aggregate, and the sample becomes clearer, permitting greater light transmission. Transmission of light is detected by the photocell and recorded as a function of time (Figure). An enhancement of modern LTA is the capacity to simultaneously monitor ATP secretion from dense granules using a luciferin-luciferase reagent.

When do I recommend LTA?

Rare PFDs such as Glanzmann thrombasthenia (GT) and Bernard–Soulier syndrome (BSS) are usually diagnosed early in life because of the severity of the bleeding phenotype. In syndromic PFDs, the presence of associated features (e.g., oculocutaneous albinism in Hermansky-Pudlak syndrome) may facilitate diagnosis. Far more common, however, are mild PFDs without associated syndromic features. Even with a detailed bleeding history, these disorders may be difficult to distinguish from normal variation due to the high frequency of mucocutaneous bleeding symptoms in the general population. In one study of healthy adults, epistaxis, easy bruising, and prolonged bleeding after tooth extraction were reported in 25 percent, 18 percent, and 18 percent of subjects, respectively; and 47 percent of menstruant women reported heavy menses.

A bleeding history may be taken using either a conventional approach or a standardized bleeding assessment tool (BAT). Published BATs are effective in discriminating patients with bleeding disorders from healthy controls, but they have not proven effective in predicting the presence of PFDs among patients referred for a suspected bleeding disorder. A recent study investigated the utility of the International Society on Thrombosis and Haemostasis (ISTH)-BAT for this purpose. Twenty-one healthy controls and 79 patients with a suspected bleeding disorder were enrolled. A normal basic bleeding history, these disorders may be difficult to distinguish from normal variation due to the high frequency of mucocutaneous bleeding symptoms in the general population. In one study of healthy adults, epistaxis, easy bruising, and prolonged bleeding after tooth extraction were reported in 25 percent, 18 percent, and 18 percent of subjects, respectively; and 47 percent of menstruant women reported heavy menses.4

Table. Characteristic LTA Patterns of Selected Platelet Function Disorders

<table>
<thead>
<tr>
<th>Disorder</th>
<th>Inherited disorders</th>
<th>Acquired disorders</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Primary ADP</td>
<td>Secondary ADP</td>
</tr>
<tr>
<td>GT</td>
<td>Absent</td>
<td>Absent</td>
</tr>
<tr>
<td>BSS</td>
<td>Normal</td>
<td>Normal</td>
</tr>
<tr>
<td>Dense granule SPD</td>
<td>Decreased or Absent</td>
<td>Variable</td>
</tr>
</tbody>
</table>

In my practice, I employ a conventional approach to the bleeding history. I query patients about spontaneous, mucocutaneous bleeding symptoms (location, frequency, severity, treatment); bleeding with hemostatic challenges (e.g., surgery, trauma, childbirth, menstruation); and family history of bleeding. I also ask about prescription and over-the-counter medications (e.g., anti-platelet agents) and comorbidities (e.g., liver disease, uremia) that could promote bleeding. I use data from the history to estimate the pretest probability of a PFD. I recommend LTA when I judge the pretest probability to be intermediate or high (i.e., ~5-10%). My approach is likely to be highly operator-dependent and result in over-testing (especially about half of the patients I refer for LTA have an identifiable defect). Although BATs hold promise for ameliorating these deficiencies, I have opted not to use BATs in my practice until they are shown to improve diagnostic accuracy compared with conventional approaches to assessment of bleeding.

How is LTA interpreted?

A panel of seven platelet agonists – ADP, epinephrine, collagen, thrombin receptor-activating peptide, the thromboxane A2 mimetic U46619, arachidonic acid, and ristocetin – is recommended by an international consensus panel.1 Aggregation tracings (Figure) are evaluated with respect to a number of parameters including shape change; length of lag phase; slope of aggregation; presence of a secondary wave of aggregation produced by weak agonists, such as epinephrine; maximal percent aggregation; and presence of deaggregation. The pattern of aggregation and/or secretion defects observed with the agonist panel facilitates identification...
of a variety of hereditary and acquired PFDs (Table) and also helps to localize lesions to specific pathways. Classic examples include GT (caused by mutations in integrin αIIbβ3) and BSS (caused by mutations in the platelet glycoprotein Ibα-IX-V complex), both of which have characteristic LTA signatures (absent aggregation to all agonists except ristocetin and absent ristocetin-induced agglutination, respectively).

In practice, LTA abnormalities often do not conform to one of the textbook patterns shown in the Table. Many of these “non-specific” abnormalities are of uncertain clinical significance and molecular pathogenesis. However, critical new insights are emerging from genotype–phenotype correlation analyses such as the United Kingdom Genotyping and Platelet Phenotyping (GaPP) study.1 GAPP is a multicenter study of patients with clinically suspected inherited PFDs. Results from the first 111 patients were recently reported.8 a

The hematologist: Dr. Cuker indicated no relevant conflicts of interest.

Which variables can confound LTA?

Many drugs affect platelet function in vitro. Drugs with reversible effects on platelets (e.g., NSAIDs) should be held for at least three days and drugs with irreversible effects (e.g., aspirin, clopidogrel) for at least 10 days prior to testing. An international consensus panel recommends a short rest period and avoidance of smoking and caffeine prior to blood collection to mitigate the effects of epinephrine release.6 Because chylomicrons in plasma can interfere with LTA, patients should be counseled to avoid fatty meals shortly before testing. Laboratories must adhere to meticulous specifications for sample collection and processing to avoid in vitro platelet activation. The platelet count in platelet-rich plasma should be measured. Results may be inaccurate when the platelet count is < 150 × 10⁹/L. Studies should be completed within four hours after blood collection.6

Conclusion

As they have for decades, a detailed bleeding history and LTA form the backbone of the diagnostic evaluation of patients with suspected PFDs. These approaches are not without shortcomings. The bleeding history has relatively poor specificity because of the frequency of mucocutaneous bleeding symptoms in the general population. LTA is labor-intensive, requires considerable expertise and a fresh blood sample, is not well-standardized, and shows overlap between normal variation and pathology across certain parameters. Continued development of BATs and improved methods for measuring platelet function are needed to overcome these limitations. Genotype–phenotype correlation studies are elucidating novel defects, are providing new insights into platelet function in health and disease, and may ultimately pave the way for more accurate approaches to diagnosis.


Dr. Cuker indicated no relevant conflicts of interest.

Whatever Happened to the Microscope?

Long before the development of the specialty of hematology, the discovery and use of the microscope demonstrated the findings of the blood cells in circulation.1

As every student of medicine knows, and especially anyone who is interested in hematology clearly understands, evaluation of the peripheral blood film by a well-trained clinician is an integral and, indeed, an essential part of the evaluation of most hematologic patients. Thus, the use of the microscope has led the way in diagnosing many hematologic abnormalities.

We take pride in our ability to efficiently diagnose the patient’s problem by evaluating a freshly made and properly stained peripheral blood film. Highlighting the importance of this process, Blood includes a clinically relevant image along with a short case report in a recurring section titled “Blood Works.” The description of the disease process being highlighted in Blood Works is often accompanied by comments such as “This report demonstrates the need for review of the smear in any case of anemia diagnosis” or “A peripheral film review is imperative for correct diagnosis because automated analyzers frequently fail to properly count the giant-sized platelets.”

Yet, two events have conspired in recent years to denigrate the value of the review of the peripheral blood film by the individual physician involved in the care of the patient. First, there has been less emphasis on the teaching of medical students and housestaff about the necessity of microscopically reviewing the peripheral blood film. The microscope has been essentially eliminated from the teaching wards of the hospital and can only be found in the pathology laboratory and is thus usually controlled by the pathologist or the laboratory technician. Second, even though we (hematologists) are recognized by Clinical Laboratory Improvement Amendments (CLIA) as competent to run a clinical laboratory, there is a determined effort to require extraordinary amounts of certification to permit the microscope to be present in a hematologist-run clinical laboratory. This position is also indirectly supported by the Current Procedural Terminology (CPT) reimbursement codes used by the Centers for Medicare and Medicaid Services (CMS) that provide no money for evaluating the peripheral blood film in an outpatient setting and that allow only a one-time payment for evaluation of the peripheral blood film on an inpatient (CPT code 99060). Practically, this code could be used in both the inpatient and outpatient setting, but CMS has chosen to honor it only in the inpatient setting. The single-payment issue persists even though follow-up evaluations of the peripheral blood film are often beneficial in determining response to therapy or in assessing disease status.

We make diagnoses in the office setting by reviewing the peripheral blood film while the patient is still present. This efficient approach allows us to order the most appropriate studies (instead of using the uninformative “shotgun” method that results in unnecessary tests), thereby saving both time and money for both the patient and the insurer.

Thus, the patient and society as a whole benefit from review of the peripheral blood film by the hematologist, and, in this day and age of cost containment, review of the peripheral blood film is a well-documented way to provide expert, thoughtfully managed, low-cost medical care. We must encourage use of the microscope by medical students, housestaff, and laboratory fellows, and, of course, we need to enlighten CMS to the advantages of reviewing the peripheral blood film in an office setting.

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1. Van Leeuwenhoek A. Arcana Naturae Detecta, Delphis Batav, 1695.
Exosomes in Hematologic Malignancies

ALDO M. ROCCARO, MD, PHD; SALOMON MANIER, MD; AND IRENE M. GHOBRIAL, MD

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3. Irene Ghobrial, MD, Associate Professor, Dana-Farber Cancer Institute, Harvard Medical School, Boston, MA

Cell-to-cell communication is a fundamental component of both normal physiology and pathophysiology, including tumorigenesis. Among the mechanisms by which cells communicate is the generation and processing of exosomes. Exosomes are nanometer-sized endocytic vesicles that are released into the extracellular compartment by many types of cells. The content of exosomes includes proteins, lipids, mRNA, and microRNA. Examples of exosome-modified processes include cellular immunity, response to infection, and tumorigenesis. For example, antigen-driven, T-cell-derived exosomes containing microRNA transfer unidirectionally to antigen-presenting cells where the transferred microRNA modulates gene expression in recipient cells. Compelling evidence supports a role for exosomes in infectious disease pathophysiology. Exosomes produced by eukaryotic pathogens such as Cryptococcus neoformans or by host cells infected with intracellular pathogens may either facilitate or limit the infection based on the nature of the infectious organism and the cell type that is targeted by the exosome. The function of exosomes in tumor progression can be explained, at least in part, by the capacity of tumor cell-derived exosomes to modulate and mold the host microenvironment and thereby produce conditions that favor proliferation of the malignant cell population. Exosome-mediated processes have been implicated in the pathobiology of both solid tumors and hematologic malignancies. In the case of blood cancers, a pathogenic role for exosomes has been demonstrated for multiple myeloma (MM) and leukemias (Figure).

The capacity of the bone marrow mesenchymal stromal cell (BM-MSC) compartment to generate exosomes has been recently characterized under both normal and pathologic conditions. In the case of plasma cell dyscrasias, MM-BM-MSC-derived exosomes were shown to transfer their microRNA and protein content to the malignant plasma cells, thereby enhancing myeloma tumor growth and increasing dissemination of the malignant cells in vivo (Figure). Characterization of BM-MSC-derived exosomes showed that the microRNA and protein content was different for exosomes derived from MM-BM-MSCs compared with the content of exosomes derived from normal BM-MSCs. Specifically, exosomal microRNA-15a levels were significantly lower in MM compared with the levels from which they were isolated. Thus, analysis of exosome content may provide actionable clinical information. Exosome formation by tumor cells may also affect response to therapy. For example, in patients with B-cell lymphoma, tumor-derived exosomes may limit the efficacy of anti-CD20 immunotherapy. In this case, antigen binding to the B-cell receptor stimulates release of exosomes expressing CD20 into the extracellular space where they bind anti-CD20, shielding tumor cells from antibody attack. Both in vitro and in vivo studies demonstrated that rituximab bound to the CD20-expressing exosomes activated complement, leading to complement consumption that reduced the anti-tumor efficacy of anti-CD20-driven complement-dependent cytotoxicity. In those studies, exosome biogenesis was found to be modulated by the lysosome-related, organelle-associated ATP-binding cassette transporter A3 (ABC3). Pharmacologic blockade of ABC3 using the cyclooxynase type-2 inhibitor celecoxib reduced CD20 expression and thereby reducing the sum effect of CD20-bearing exosomes and enhancing the anti-tumor efficacy of rituximab.

Together, the above studies provide insights into the mechanisms by which exosomes contribute to the pathobiology of hematologic malignancies. In MM, BM-MSC-derived exosomes transfer microRNA and protein to the malignant plasma cells thereby enhancing myeloma tumor growth and increasing dissemination of the malignant cells (Figure). AML-derived exosomes can re-program the bone marrow niche to support tumorigenesis (Figure), and B-cell-derived exosomes uniquely contribute to drug resistance by diverting immunotherapy from its intended target. Continued investigation of the properties of exosomes is warranted both to understand more completely the role of exosomes in the pathobiology of hematologic malignancies and to identify novel approaches to therapy.

Recent studies have shown that primary acute myeloid leukemia (AML) cells release exosomes enriched for both coding and noncoding RNAs. In this case, AML-derived exosomes deliver their content to syntenic cells, including BM-MSCs, consequently modulating proliferative, angiogenic, and migratory properties of co-cultured stromal and hematopoietic progenitor cell lines (Figure). Analysis of exosome content may also have potential as a biomarker of disease, as microRNAs that encode proteins including NPM1, FLT3, CXC4, MMP9, and IGF-IR were detected in exosomes isolated from patient-derived AML blasts and leukemic cell lines. Characterization of these exosomes showed that the microRNA content reflected the FLT3 and NPM1 allelic diversity of the cell population from which they were isolated. Thus, analysis of exosome content may provide actionable clinical information. Exosome formation by tumor cells may also affect response to therapy. For example, in patients with B-cell lymphoma, tumor-derived exosomes may limit the efficacy of anti-CD20 immunotherapy. In this case, antigen binding to the B-cell receptor stimulates release of exosomes expressing CD20 into the extracellular space where they bind anti-CD20, shielding tumor cells from antibody attack. Both in vitro and in vivo studies demonstrated that rituximab bound to the CD20-expressing exosomes activated complement, leading to complement consumption that reduced the anti-tumor efficacy of anti-CD20-driven complement-dependent cytotoxicity. In those studies, exosome biogenesis was found to be modulated by the lysosome-related, organelle-associated ATP-binding cassette transporter A3 (ABC3). Pharmacologic blockade of ABC3 using the cyclooxynase type-2 inhibitor celecoxib reduced CD20 expression and thereby reducing the sum effect of CD20-bearing exosomes and enhancing the anti-tumor efficacy of rituximab.

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Dr. Roccaro, Dr. Manier, and Dr. Ghobrial indicated no relevant conflicts of interest.
Cancer Treatment Parity Act Introduced in Senate

In late December 2013, Senator Al Franken (D-MN) and Senator Mark Kirk (R-IL) introduced the Cancer Treatment Parity Act (S. 1879) in the U.S. Senate. The legislation is similar to the Cancer Drug Coverage Parity Act, H.R. 1801, introduced in the U.S. House of Representatives in April 2013 by Representatives Brian Higgins (D-NY) and Peter King (R-NY). Both bills seek to ensure that patients enrolled in certain federally regulated health plans have access and insurance coverage for all anti-cancer regimens. The bills would require any health plan that provides coverage for cancer chemotherapy treatment to provide coverage for orally administered and self-injectable anticancer medications on a cost schedule no less favorable than that of IV, port administered, or injected anticancer medications.

A number of states have passed oral parity legislation or are currently reviewing related legislation. Federal legislation—such as that introduced in the House and Senate—will ensure a standard across all states so that all cancer patients in the United States can reasonably access both physician- and patient-administered chemotherapy. While legislation will not solve the problem of the high cost of drugs, it does lift much of the financial burden from patients.

ASH strongly supports these legislative efforts, and the Society recently released an ASH Policy Statement in Support of Insurance Coverage Parity for All Cancer Treatments. The Society encourages members to visit the ASH Advocacy Center (www.hematology.org/advocacy) to join the Society’s efforts in urging Congress to support cancer drug parity.

Let ASH Help with Your Questions or Concerns about the Sunshine Act

In 2013, ASH launched a Web page, www.hematology.org/Sunshine, with resources on the Centers of Medicare and Medicaid Services (CMS) Open Payments program, also known as the Physician Payments Sunshine Act. The site was recently updated, and with the following four easy steps, you can prepare for the public disclosure of 2013 Open Payments data:

1. Make sure your disclosures are up-to-date. Financial and conflict-of-interest disclosures required by employers, advisory bodies, and entities funding research should be updated regularly so they are consistent with the data that will eventually be publicly reported under the Sunshine Act.

2. Confirm that your National Provider Identifier (NPI) information is current. The information tied to your NPI, including your specialty, must be accurate to ensure appropriate attribution of payments and other transfers of value that will be listed in CMS’s online database.

3. Request ongoing notification from your industry contacts about the data they report to CMS. Ask your representatives at the manufacturers and group purchasing organizations with which you interact to give you an opportunity to preview and correct, if necessary, information they intend to submit to CMS; 2013 data is due March 31.

4. Track your payments and financial transfers. Download a free smartphone app that will allow you to track reportable transfers. Compatible with Apple® and Android® platforms, “Open Payments Mobile for Physicians” is available through both the Apple Store and the Google Play® Store. A number of security features protect the privacy of the data you capture, which will be stored on one device and cannot be backed up to a cloud or other devices. Also, urge your industry contacts to use the app so you will be able to capture the information you need to ensure accurate reporting.

If you have any questions or concerns about how this program will impact hematologists and/or ASH programs, please contact ASH Government Relations and Practice Manager Stephanie Kaplan at skalplan@hematology.org. Your feedback will help the Society assess the impact of the new program on hematology and develop additional resources to help hematologists understand the program.

Become an Advocate for Hematology: Join the ASH Grassroots Network Today

Thanks to ASH’s advocacy efforts, issues important to the future of hematology have been brought to the attention of the U.S. Congress and federal agencies. ASH advocacy has been instrumental in the following legislative processes: securing an increase in NIH funding for the remainder of fiscal year (FY) 2014 that will help lessen the impact of the sequestration cuts that took effect in 2013; preventing scheduled cuts in Medicare physician payment; and calling attention to shortages of drugs and biologics used to treat patients with hematologic diseases. Our work is not done, however, and the Society needs the help of all of its members in continuing to focus attention on these and other issues of importance to hematology.

How You Can Help: To ensure that you get information on the latest advocacy opportunities, please join the ASH Grassroots Network. By doing so, you will receive action alerts on important legislation pending before Congress, and the Network will provide a forum for you to voice your position on issues important to the future of hematology. Members of the Grassroots Network are kept informed both on key developments in issues critical to hematologic research and practice through the monthly “ASH Advocacy Update” e-newsletter and on relevant legislative issues through regular “Action Alerts.” ASH’s online Advocacy Center provides members with a convenient method to contact Congress and the Administration concerning proposed laws and regulations affecting hematologic research, practice, and patients.

To join the Grassroots Network and participate in the Society’s advocacy efforts, visit the ASH Advocacy Center at www.hematology.org/advocacy or contact grassroots@hematology.org.
Unlocking Host Immunity to Lymphoma


Lymphomas should be among the most immunogenic of malignancies because the neoplastic cells express both HLA class I and II antigens at high density. The graft-versus-lymphoma effect seen following allogeneic stem cell transplantation supports this impression. Although attempts to produce a similar effect in the autologous transplant setting have been largely unsuccessful, the development of antibodies with the capacity to inhibit the cellular immune response has generated new interest in immunotherapy for lymphoproliferative disorders. Two recent studies examining pidilizumab give an indication of the potential of this therapeutic approach. Pidilizumab blocks the inhibitory programmed death-1 (PD-1) receptor, which is present on activated T cells, activated B cells, natural killer cells, and myeloid cells (Figure). On T cells in particular, PD-1 is a marker of cellular exhaustion as it conveys signals that inhibit T-cell proliferation and enhance apoptosis, consequently functioning as a negative regulator of the cellular immune response (Figure). Specific ligands (PD-L1 and PD-L2) for PD-1 are expressed by a variety of normal cells and by many tumor cell types, and compelling data indicate that activation of the PD-1 signaling system by PD-L1 and PD-L2 attenuates T-cell activity, thereby allowing tumor cells to escape immune surveillance (Figure). The good news is that the negative effects of the PD-1 signaling system can be limited pharmacologically both by antibodies that block PD-1 and by antibodies that block its activating ligands, PD-L1 and PD-L2 (Figure).

To investigate the clinical efficacy of anti-PD-1 therapy, an international group of investigators led by Leo I. Gordon and colleagues from Northwestern University Feinberg School of Medicine in Chicago administered three doses of pidilizumab at six-week intervals to 66 patients with diffuse large B-cell lymphoma (DLBCL) following treatment with high-dose therapy and autologous stem cell rescue. Although this was a single-arm study with no control group, the finding that 72 percent of patients were free of disease progression 16 months after completing the regimen was promising, especially given that outcomes were the same for the 24 high-risk patients who had a positive FDG-PET at the time of enrollment into the study (i.e., prior to treatment with high-dose chemotherapy followed by autologous stem cell rescue.) Among patients with residual disease visible on CT scans at the time antibody treatment was started, a little over half had an objective response. Although an argument could be made that this outcome might be in part attributable to the ongoing effect of the high-dose therapy, the high response rate for this poor-risk subpopulation is nonetheless encouraging.

A second single-arm study that focused on recurrent follicular lymphoma was led by Sattva S. Neelapu from MD Anderson Cancer Center in Houston. The target group was patients with relapsed disease who had previously demonstrated rituximab sensitivity. Up to 12 doses of pidilizumab were given to patients before and after four weeks of rituximab. The overall response rate was 66 percent (19 out of 29 patients), and a median progression-free survival (PFS) of 19 months was projected.

In both studies, the investigators made attempts to determine the mechanisms of action of pidilizumab and to define markers of response. In the diffuse large B-cell transplant study, flow cytometry was used to quantify the number of both circulating helper T cells expressing PD-L1 and circulating peripheral and central memory T cells following pidilizumab therapy. The results of this analysis suggested an on-target effect of the anti-PD-1 reagent. In the follicular lymphoma study, the density of PD-L1 on T cells and monocytes was predictive of response to the antibody combination, and, using gene-expression profiling, the investigators observed that the presence of transcripts reflecting an activated T-cell phenotype prior to therapy correlated with improved PFS.

Concerns have been expressed about the potential toxicity that might result from disinfected cellular immunity, but the findings in these two studies were reassuring. In the DLBCL study, the most common grade 3-4 toxicity was myelosuppression, with one case of fatal disseminated Herpes zoster infection reported, while in the follicular lymphoma study, no grade 3-4 toxicity was reported. This toxicity profile is more benign than that observed in studies using anti-CTLA-4 that unblocks earlier T-cell activation, as treatment with this antibody can be complicated by severe autoimmune disease. It has been suggested that anti-PD-1 acts to release suppression of the T-cell response in peripheral tissues, accounting for the more favorable toxicity profile compared with treatment with anti-CTLA-4 that results in unrestricted T-cell activation.

Modulation of the cellular immune response using checkpoint-blocking antibodies such as anti-PD-1 and anti-CTLA-4 has recently been identified as a potentially effective therapeutic strategy in the treatment of immunogenic tumor types such as melanoma. The results of the above two trials further reinforce the concept that lymphoma is immunogenic to the host’s autologous cellular immune system, building upon previous studies that used therapeutic approaches such as systemic cytokines, vaccines, and toll-like receptor agonists to stimulate the immune response. While promising, this type of immune modulation is in its infancy as a therapeutic modality with many outstanding issues and unanswered questions such as how to predict who will respond, the precise mechanism of action by which the antibodies disinfibut the cellular immune response, and how to avoid autoimmune-mediated toxicity. The results of studies using anti-PD-1 are encouraging, but the findings need to be confirmed in randomized trials. If the data hold up to rigorous investigation, further studies using anti-PD-1 antibodies either singly or in combination with chemotherapy, targeted kinase inhibitors, immunomodulatory drugs, radiation, other antibodies, and perhaps also vaccines will likely ensue. Activation of host anti-tumor immunity represents an exciting new direction for lymphoma therapy that will certainly be the subject of intense basic and clinical investigation.

Figure

T-cell inhibited by PD-L1, PD-L2 expressed on lymphoma

Anti-PD-1 blocks inhibitory signal, permitting T-cell activation

Peter Johnson, MD
Dr. Johnson indicated no relevant conflicts of interest.
The Great Escape


Many tumors exhibit dormancy that can last for years, making the time to clinical recurrence unpredictable. Minimal residual disease (MRD) represents the survival, following therapy, of tumor cells that gives rise to future relapse. A key element of tumor persistence is evasion of immune surveillance. Thus, MRD may be due, at least in part, to acquisition by the tumor of a phenotype that masks it from immune recognition and clearance. Moreover, the phenotype of the recurrent tumor may be radically different from that of the original tumor. The sensitivity of detection of MRD has increased with the advent of methods such as multi-parameter flow cytometry, fluorescence in situ hybridization, and real-time quantitative PCR. MRD is clinically important, as detection augurs a poor prognosis for many types of cancer.

In the current study, Timothy Kottke and colleagues from the Mayo Clinic in Rochester, Minnesota, investigated, using murine models, tumor-antigen expression and cytokine production during progression from MRD to frank relapse. They observed that transition from MRD to proliferation results in diminished innate immune responses; however, the recurrent tumors were found to be insensitive to the immune effectors, a property that enabled them to escape immune clearance. The investigators studied the efficacy of immunotherapy, oncolytic virotherapy, chemotherapy, and T-cell therapy. They observed that proinflammatory cytokines such as IL-6; interferon; oncolytic virotherapy, chemotherapy, and T-cell therapy. They observed that proinflammatory cytokines such as IL-6; interferon; and serum amyloid P component, a marker of an acute-phase protein response to infection, were associated with emerging recurrence, whereas later and larger, actively growing recurrences exhibited neither proinflammatory cytokines nor the antigens that elicited the immune response. Detection of IL-6 and vascular endothelial growth factor (VEGF) correlated with the onset and early progression of tumor growth in mouse models of melanoma and spontaneous breast cancer. A reporter assay using a luciferase plasmid under control of the VEGF promoter showed a positive signal that was followed by clinical recurrence seven to 12 days later only in the mice that would develop tumors. Kottke and colleagues hypothesized that if the transition from MRD to overt recurrence were induced prematurely, the tumor cells may not yet have acquired the critical properties that allow escape from therapy. In support of this hypothesis, systemic administration of VEGF during MRD led to premature recurrence, with development of angiogenesis and rapid tumor formation, and the induced tumors exhibited neither proinflammatory cytokines nor the antigens that elicited the immune response. Therefore, the authors proposed the alternative approach of flushing out occult MRD by provoking overt recurrence by administering VEGF at a time when tumor cells may not be equipped to evade treatment. Particularly for patients with Fanconi anemia, the possibility for allogeneic bone marrow transplantation for a curative effect of the presence of MRD is associated with a much worse outcome.1

Identifying the best method to eradicate MRD, both prior to and after transplant, has become a great interest, and chemotherapy and immunologic treatment regimens are under investigation. Given the success of cellular immunotherapies such as chimeric antigen receptor (CAR)-T cells, WT-1 specific alloreactive T cells, and similar approaches, it may be time to consider deliberately awakening MRD, perhaps with VEGF as has been demonstrated here, before overt relapse, while enhancing immunologic recognition, to eliminate the malignant cells before they acquire resistance mechanisms.


Fanconi Anemia Bone Marrow Stem Cells: Innately Complex


Fanconi anemia (FA) is an uncommon, autosomal recessive, multisystem disorder characterized by near uniform loss of bone marrow function by late adolescence. Much has been learned about the activity of the 16 Fanconi genes whose coordinate function guides DNA double-strand break repair that prevents accrual of DNA damage, thereby protecting cells from malignant transformation and clonal evolution. Met with initial skepticism when first proposed, the FA pathway has gained credence (and expanded to a state of dizzying complexity) over the past decade and now serves as a roadmap for understanding the molecular and cellular basis of the disease. However, the discovery of the overlap of some genes of the FA pathway with some genes of the BRCA pathway (i.e., the pathway that is defective in patients with inherited forms of breast and ovarian cancers and now called the FA/BRCA pathway) skewed FA research in the direction of identifying and characterizing the mechanisms that underlie neoplastic transformation and away from investigation both of the basis of the vulnerability that erodes the stem cell pool and of the physiologic function of FA proteins in stem cell self renewal. But now a study from the laboratory of Elizabeth Eklund at the Feinberg School of Medicine and the Robert H. Lurie Comprehensive Cancer Center, Northwestern University, Chicago, provides a new perspective on the basis of bone marrow failure in FA.

The studies of Liping Hu and colleagues focused on a connection between emergency granulopoiesis, a component of the innate immune response, and hematopoietic failure in a murine model of FA. Emergency granulopoiesis is induced in response to an infectious or inflammatory challenge and is characterized by rapid expansion and differentiation of granulocyte/monocyte progenitor populations, a process that is due in part to a shortened S-phase of the cell cycle. Based on experiments that showed that transcription of the FA gene FANCc is augmented by interleukin-1β (IL-1β), an essential emergency granulopoiesis cytokine, the authors hypothesized that the FA/BRCA pathway contributes to genomic stability during emergency granulopoiesis. The authors challenged their hypothesis by inducing emergency granulopoiesis in wild-type and Fancc-deficient mice using the vaccine adjuvant alum. Those experiments showed failed emergency granulopoiesis in Fancc-deficient mice, and with repeated successive rounds of challenge with the inflammatory stimulant, the knockout animals experienced progressive anemia and neutropenia. The peripheral blood cytopenias were found to coincide with apoptotic loss of hematopoietic stem cells and myeloid progenitors in the bone marrow. The effect was mediated by G-CSF and by IL-1β augmentation of the interferon regulatory transcription factor IRF8, which initiates transcription of Fancc, and ameliorated by blockade of the interleukin receptor-1 (IL-1R). The results elucidated the mechanistic upregulations of prior work that showed that FA stem cells were abnormally susceptible to interferon-γ and may explain the relatively poor response to G-CSF mobilization observed in Fancc mice and in some FA patients. The experimental findings also appear to validate the long-held notion that “protective” animal husbandry in an infection-free environment may inadvertently account for the lack of spontaneous post-natal hematopoietic failure that plagues many murine FA models.

The novel findings of Hu and colleagues bring to mind Einstein’s view that “it is the theory which decides what we can observe,” as these and other studies suggest multiple functions for individual FA proteins, with activity outside the canonical repair of interstrand crosslinks or cytokine defense. In the current study, IL-1β and G-CSF transcriptionally upregulated Fancc and FANCb, but did not consistently alter Fancc2D levels. The DNA pathway function of FA proteins may continue to be instructive for understanding alkylating agent detoxification, but not necessarily for understanding stem cell self-renewal. Involvement in aldehyde detoxification, recently shown by others, and innate immune defense, shown here, serve to reinvestigate, and reorient, our approach to understanding hematopoietic failure and the physiologic role of FA proteins.

Other research models of acquired bone marrow failure support the role of innate immunity in regulating the hematopoietic stem cell pool.1 The article by Hu and colleagues now extends those observations in an unexpected way to FA. The findings need to be validated, but they suggest that stem cells from individuals with FA (and perhaps FA carriers) are vulnerable to recurrent infectious stimuli. The work raises additional questions including the following: Does innate immunity trigger DNA damage? How does emergency granulopoiesis relate to stem cell function at a mechanistic level? Notably, the paper suggests a novel therapeutic strategy that may yet preempt the loss of bone marrow stem cells in children with FA.

F or decades, vitamin K antagonists (VKAs) such as warfarin have been the standard oral anticoagulant used to prevent or treat thromboembolic disease. Although the VKAs are effective, their narrow therapeutic window, combined with the significant inter-individual variability in dose response, makes this class of medications challenging to use. Once the decision has been made to initiate treatment with a VKA, the therapeutic focus is on maximizing the percentage of time in the therapeutic range (TTR), as a higher percentage of TTR correlates with better clinical outcomes.1

In the last 10 years, scientists have identified common genetic polymorphisms that impact the pharmacodynamic effect of VKAs. Specifically, testing for single-nucleotide polymorphisms in the genes VKORC1 (Vitamin K epoxide reductase complex subunit 1) and/or CYP2C9 (cytochrome P450 2C9) improves the accuracy of warfarin dose prediction, raising the hope that genotype-guided dosing might improve the safety and efficacy of coumarin therapy. Until now, however, sufficiently powered clinical studies designed to test whether genotype-guided dosing improves TTR or clinical outcomes have been lacking.

Recently, Stephen E. Kimmel et al. of the COAG group and Talitha I. Verhoef et al. of the EU-PACT group compared dosing algorithms based on both clinical and genotypic variables with algorithms based solely on clinical variables. Patients in the COAG trial had INRs checked twice a week for two weeks, then weekly for four weeks. Patients in the EU-PACT trial had a similar evaluation timeline with the addition of INR checks at eight and 12 weeks. Neither study found a significant difference between genotype-guided and control dosing groups for the primary endpoint of TTR during warfarin initiation (Table). Murin Pirmohamed et al. used a slightly different trial design, comparing a dosing algorithm based on genotype and clinical variables with a three-day standardized loading dose regimen in which all control-group patients received an identical dose for the first three days (except those older than 75 years who received a smaller dose on day 1). In this trial, the genotype-guided group had a modestly greater TTR (67.4% vs. 60.3%, P<0.001). The experimental group also experienced fewer supratherapeutic INRs and, on average, reached a first therapeutic INR more quickly (21 vs. 29 days, P<0.001).

Two of these important clinical trials suggest that genotype-guided dosing of VKAs will not improve upon traditional VKA management. The statistically significant TTR difference seen in the Pirmohamed study is intriguing, but difficult to interpret, because the initial dose for patients in the experimental group was based on both their genotype and clinical characteristics, whereas dosing for the control group was pre-defined and independent not only of pharmacogenetic but also of most clinical information.

Together, these three randomized, controlled trials support the use of clinically based dose calculators (e.g., www.warfarindosing.org) to optimize TTR during VKA initiation; however, they also indicate that, in practice, adding pharmacogenetic testing to clinical dose estimation plus frequent INR monitoring will not affect a clinical outcome.


**Table**

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<th>Randomized Controlled Trials of Pharmacogenetic Testing</th>
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<td><strong>Number of Patients</strong></td>
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PG = group of patients whose dose was estimated with knowledge of their VKORC1 and CYP2C9 genotypes. TTR = time in therapeutic range, the primary endpoint for all three studies.

*All patients were given 10 mg on day 1 and 5 mg on days 2 and 3 except for patients >75 years old who received 5 mg daily on days 1-3.

EBV-associated lymphomas are characterized by expression of a limited number of viral proteins, the pattern of which is determined by the particular latency program that is active (there are three latency programs: type I, type II, and type III). Lymphomas that arise in immunocompromised hosts (e.g., PKU) express the latency III program that is defined by expression of EBV nuclear antigens (EBNAs) 1, 2, 3A, 3B, 3C, 6/LP, and latent membrane proteins (LMPs) 1 and 2. Immunosuppressed patients are at risk for developing EBV-driven PTLD after donor stem cell or solid organ transplantation because immunosuppressive therapy renders them unable to mount an adequate T-cell response against the latency III panel of EBV proteins that is otherwise highly immunogenic. Patients with PTLDs that develop after stem cell transplantation have been successfully treated with ex vivo-generated, donor-derived CTLs that recognize latency III-expressing, EBV-infected B cells. On the other hand, lymphomas such as EBV-associated classical HL, DLBCL, and NK/T-cell lymphoma express the less immunogenic latency II program that consists only of EBNAs 1, LMP1, and LMP2. Consequently, treatment of patients with latency II-type lymphomas has been marginally effective.

To generate CTLs that are effective against latency II-expressing lymphomas, the investigators constructed an adenoviral vector-encoding LMP2 and later an LMP2 combined with a truncated version of LMP1. The vector was used to transduce both bone-marrow cells and EBV-transformed B-lymphoblastoid cell lines that were then used to stimulate antigen-specific CTLs from patient-derived peripheral blood mononuclear cells.

In the clinical arm of the study, 50 patients with latency II- or latency III-expressing EBV-positive lymphomas were treated, including 25 patients with HL, 11 with NK/T-cell lymphoma, seven with DLBCL, two with PTLD, and one with peripheral T-cell lymphoma. Twelve patients had high-risk disease in first remission. Seventeen patients had relapsed disease in remission after salvage therapy. Of these 29 patients with high risk of disease recurrence, none relapsed, and the event-free survival (EFS) was 82 percent at two years. Nine patients died of complications related to prior therapy. Twenty-one patients on the study had active disease after standard therapy. Of these 21, 13 responded (11 complete responses and two partial responses). The EFS was 50 percent. The therapy was well tolerated without significant infusion-related toxicity.

The technique developed by Bollard and colleagues to generate EBV antigen-specific CTLs that can be used to treat both latency II- and latency III-expressing lymphomas represents a major advance in the field of immunotherapy. Based on an understanding of the characteristics of EBV-driven lymphoproliferative disorders, the investigators developed highly specific T cells capable of eradicating a broad array of lymphomas, even in the refractory-disease setting. In the future, their approach may have broader application to include treatment of other virally induced cancers.

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**Ann Lacasce, MD**

Dr. LaCasse indicated no relevant conflicts of interest.
Inhibiting Platelets to Treat a Disorder of Hemoglobin


Sickle cell disease (SCD) is, fundamentally, a disorder of hemoglobin and red blood cells, but complex and dynamic interactions between erythrocytes, platelets, leukocytes, and endothelial cells underlie the pathophysiology of the vaso-occlusive ischemia and tissue infarction that account for much of morbidity and mortality of SCD. Inflammation also contributes to these processes, and there is compelling laboratory evidence that SCD is a hypercoagulable state. In particular, aberrant platelet function appears to play a role in the pathophysiology of SCD. Thrombocytosis occurs commonly in patients with SCD and is often exacerbated during vaso-occlusive events, and there is evidence of platelet activation at baseline (i.e., under steady-state conditions), with markers of activation increasing during acute vaso-occlusive events. The plasma concentration of ADP is abnormally high in SCD, and ADP may promote thrombosis and inflammation by directly activating platelets and by inducing release from platelets of CD40 ligand, respectively. Together, these observations provide a rationale for studying anti-platelet agents in the treatment of SCD.

Anti-platelet therapy for SCD is not a new idea, and several trials of aspirin or ticlopidine, a first-generation ADP-receptor antagonist, were conducted about 30 years ago.1,2 In addition, studies showed a response based on assessment of biomarkers, or both, but the impact of those studies on the field was muted because of limitations in experimental design (e.g., small sample size, short duration of therapy). A study of anti-platelet therapy in the management of SCD has remained uncertain. However, with the advent of newer anti-platelet agents, this question is being readdressed. Prasugrel is an oral, third-generation platelet P2Y12 ADP-receptor antagonist. It irreversibly inhibits ADP-mediated platelet activation and aggregation, and it is currently approved for the treatment of patients with acute coronary syndrome who are managed with percutaneous coronary intervention. In patients with SCD, prasugrel has been tested in one phase I and two phase II studies. The phase I study demonstrated that prasugrel had comparable pharmacokinetics and produced a similar degree of platelet inhibition in patients with SCD compared with healthy volunteers.3 One of the phase II studies (conducted in adults) reported that treatment with prasugrel was safe (there were no hemorrhagic events requiring medical intervention), decreased platelet activation based on biomarker analysis, and possibly lessened painful crises. A phase IIb study has been completed in children, but the results are not yet published. A phase III study is currently recruiting subjects.

The manuscript by Joseph A. Jakubowski and colleagues provides further analysis of data from the phase I study. Cellular and soluble biomarkers of platelet activation and coagulation were compared between adult SCD patients and healthy volunteers at baseline and following 12 days of once daily treatment with prasugrel. The investigators confirmed that patients with SCD who had increased platelet activation at baseline compared with healthy volunteers, as measured by percentages of monocyte-platelet aggregates, neutrophil-platelet aggregates, and platelets expressing CD40 ligand. Soluble prothrombin fragment 1.2 (F1.2) and thrombomodulin B2 levels were also increased in adults with SCD compared with the volunteer group. A significant reduction in platelet-monocyte aggregates was observed for the patients with SCD treated with prasugrel but not in their control counterparts. Post-treatment, SCD patients also had lower levels of both platelet-erythrocyte aggregates and soluble tissue factor, but maintained high levels of monocyte-platelet aggregates and soluble F1.2 compared with volunteers.

These results support earlier findings of chronic platelet activation in patients with SCD, even during baseline, steady-state conditions, and the investigators have now shown that this activation can be partly attenuated by prasugrel. Because prasugrel is a platelet P2Y12 ADP-receptor antagonist, these findings suggest that ADP contributes to platelet activation in SCD. The source of excess ADP in SCD is speculative but may be erythrocyte-derived and released by hemolysis. Alternatively, or additionally, it could be platelet-derived and be released as a consequence of the stimulation of platelets by thrombin. Whether these observations, in conjunction with ongoing studies, will ultimately support the use of prasugrel for the treatment of SCD is unknown, and we anxiously await the completion of the ongoing phase III study.

A Stealthy Gene Therapy Virus


Figure

A denoviruses are the gene therapy vectors used most frequently in humans, and adenosine type 5 (Ad5) is the most commonly used of the adenoviral vectors. Many viral vectors are recognized by natural antibodies (i.e., antibodies that develop in the absence of prior antigen exposure). Such natural IgM antibodies limit vector utility because IgM is a pentameric immunoglobulin that efficiently activates the classical pathway of complement, thereby neutralizing the vector as a consequence of C3b opsonization (Figure). Notably, however, Ad5 is not neutralized by this natural antibody/complement mechanism, which makes it a more effective vector.

Zhli Xu and colleagues, in the laboratory of Andrew Byrnes at the Center for Biologics Evaluation and Research at the U.S. Food and Drug Administration, now find that factor X (FX) enhances liver transduction by Ad5 vectors by protecting Ad5 from attack by the classical complement pathway (Figure). This finding is congruent with reports that adenosinurids bind coagulation factors. In particular, high-affinity binding of FX to Ad5 is required for efficient transduction of hepatocytes.4 In support of this observation, the authors found that the Ad5 mutant vectors, Ad5BAP and AdHVR7, which weakly bind FX, inefficiently transduce mouse livers. Xu et al. also observed that transduction of liver in vivo by a wild-type Ad5 vector was inhibited in normal mice treated either with warfarin (which reduces the activity of vitamin K-dependent proteins, including FX) or with the FX-binding protein, ksp. However, liver transduction was normal in warfarin- or ksp-treated Ad5-transgenic mice, which lack mature T and B lymphocytes and therefore cannot generate antibodies. Transplantation of normal mouse bone marrow into Rag1−/− mice restored the requirement of FX for efficient transduction. This result set the stage for experiments designed to identify the cell that limits transduction efficiency in the absence of FX. Warfarin treatment had no effect on liver transduction in SCID mice, which lack B and T cells, or in IgM−/− or IgD−/− mice, both of which lack B cells. In contrast, warfarin treatment inhibited liver transduction in nude mice, which lack T cells, but not B cells. Thus, B cells are necessary for the immunologic neutralization of Ad5 in the absence of FX. Further, warfarin did not inhibit transduction in membrane immunoglobulin-transgenic (mIgTg) mice, which have B cells that express membrane-bound IgM but do not secrete IgM. However, warfarin inhibited liver transduction by Ad5 of JHD mice treated with purified IgM. Transduction was not inhibited by warfarin in C1q-deficient and C4-deficient mice (i.e., mice that lack the capacity to generate C3b opsonization through classical pathway activation). Collectively, these results support the hypothesis that activation of the classical pathway by natural anti-IgM antibodies is the major mechanism involved in suppressing liver transduction by Ad5 in FX-deficient mice.

In vitro experiments were performed to further characterize the interaction of natural antibodies, complement, and FX during Ad5 vector transduction. Normal mouse serum, which was used as a source of FX, efficiently inhibited liver transduction in nude mice, but not in IgM−/− or IgD−/− mice, both of which lack B cells.

Western blotting experiments showed that, in the absence of functional FX, C3b bound covalently to Ad5 virions (Figure). In these experiments, purified normal Ad5 or AdHVR7 virions were incubated with serum-containing complement and isolated by density centrifugation. Following SDS-PAGE, which dissociates non-covalent complexes, covalently bound C3b was detected bound to AdHVR7 proteins. However, covalent binding of C3b to normal Ad5 proteins was not observed. Additionally, incubation of AdHVR7 with mouse plasma resulted in the conversion of C3 into C3a and C3b (a finding indicative of complement activation), but C3 conversion was not observed in samples containing the normal Ad5 vector unless X-ksp was added to inhibit FX binding (Figure). Together, these results demonstrate that FX protects virions from C3b opsonization mediated by IgM antibody-induced activation of the classical pathway of complement (Figure).

The authors have identified a defense mechanism in which FX enhances virus survival by inhibiting attack by natural antibodies and complement. This observation suggests that FX might not inhibit the infectivity of a normal Ad5 vector. However, it markedly neutralized the non-FX-binding mutant AdHVR7. Purified IgM added to IgM-deficient serum, but not IgM-deficient serum alone, neutralized AdHVR7. Furthermore, the normal Ad5 vector was neutralized by mouse serum when FX binding was blocked by X-ksp.

The authors have described a mechanism by which FX enhances virus survival by inhibiting the attack by natural antibodies and complement. This observation suggests that FX might not inhibit the infectivity of a normal Ad5 vector. However, it markedly neutralized the non-FX-binding mutant AdHVR7. Purified IgM added to IgM-deficient serum, but not IgM-deficient serum alone, neutralized AdHVR7. Furthermore, the normal Ad5 vector was neutralized by mouse serum when FX binding was blocked by X-ksp.


Conversion of Renal Erythropoietin-Producing Cells to Myofibroblasts: A Single Mechanism for Both Anemia and Fibrosis in Kidney Disease


Chronic kidney disease (CKD) is characterized by erythropoietin (EPO)-deficiency anemia, renal fibrosis, and scarring. A large majority of circulating EPO is produced by a population of interstitial fibroblast-like cells in the renal cortex and outer medulla. In healthy individuals, the number of these renal EPO-producing cells (REPs) is low, but the number increases when decreased oxygen delivery due to hypoxia triggers hypoxia-inducible factor-mediated EPO production. In CKD, anemia develops as the number of REPs and the resultant concentration of circulating EPO are decreased despite the tissue hypoxia caused by anemia. Thus, EPO deficiency is the major cause of anemia in renal failure.1 This EPO deficit in CKD may be due to loss of REPs, locally increased oxygen levels in the microenvironment of the REPs, or an altered relationship between local oxygenation and EPO production by REPs.2

In CKD, renal fibrosis and scarring are mediated by myofibroblasts, a subset of mesenchymal cells that produce collagen and can be identified by their expression of desmin and α-smooth muscle actin (α-SMA). In mice with renal inflammation that is induced by unilateral ureteral obstruction (UUO), the majority of kidney myofibroblasts proliferate in response to inflammatory cytokines and are derived from cells that reside in the kidney, but are neither epithelial nor vascular pericytes.2 A minority of myofibroblasts in this model are derived from differentiation of marrow-bred progenitors that do not proliferate in renal tissue. The origin of the intrarenal cells that divide and differentiate into myofibroblasts is unknown, but REPs are likely candidates, because, with inflammatory stimuli, they show decreased expression of EPO while desmin and α-SMA expressions are induced.

Using mice in which REPs can be both identified and their state of EPO production determined, Tomokazu Souma et al. demonstrate that renal inflammation from UUO transforms REPs into myofibroblasts that do not produce EPO, but rather mediate kidney fibrosis. Similar results were found in these mice using unilateral ischemia reperfusion injury and protein overload nephropathy models. This transformation was associated with both cell proliferation and morphologic transformation. Angular EPO-producing REPs that bridged capillaries and proximal tubules decreased, while an increase was observed in stellate REPs that no longer produced EPO and had less extension and more rounded nuclei characteristic of myofibroblasts. Although UUO induced the majority of REPs to assume the characteristics of myofibroblasts, expressing α-SMA and collagen, when the UUO was relieved after two days, the morphologic and biochemical changes were completely reversed over the ensuing 12 days. The recovery of EPO production and loss of collagen expression was hastened when the mice that had been subjected to short-term UUO were treated with dexamethasone. The transformation of EPO-producing REPs to myofibroblasts by UUO appeared to be induced by inflammatory cytokines produced in the REPs themselves, as expression of TGFβ1 and NFκB increased, and their respective signaling factors, Smad 2/3 and phosphorylated p65, showed nuclear accumulation. The recapitulation of reversible transformation of REPs to myofibroblasts, including the suppression of EPO expression following lipopolysaccharide (LPS) injection and the attenuation of this effect by dexamethasone, suggested that the NFκB pathway had an important role in suppression of EPO production during transformation of the REPs. The inability of TGFβ1 injections to suppress EPO expression in REPs suggested that the major effect of this inflammatory cytokine on EPO expression in myofibroblasts.

This study by Souma and colleagues demonstrates that, in the short term, the fibroblast-like interstitial cells that produce the majority of EPO in the body are not destroyed as a consequence of renal inflammation, but rather they are transformed into myofibroblasts that cause fibrosis and scarring in CKD. The duration of reversibility and corticosteroid responsiveness of the transformed REPs is unknown. Indeed, these features may vary depending upon the cause of renal disease, but the characteristics of reversibility and steroid responsiveness suggest an opportunity for therapeutic intervention in the early stages of renal injury. Cytokine suppression of renal EPO production is not only confirmed by this study, but the experimental evidence supports the hypothesis that REPs are a source of the cytokines. Interruption of this cytokine-mediated transformation process may not only ameliorate EPO deficiency in patients with CKD, but also inhibit the progression to end-stage renal disease.


Register for the Highlights of ASH® Meetings in Singapore and Brazil

The 2014 International Highlights of ASH meetings will take place this spring in Singapore and Florianópolis, Brazil. A select group of ASH members will review handpicked abstracts that were presented at the 2013 annual meeting in New Orleans. The abstract reviews are organized programmatically based on hematologic subspecialties with speakers who are experts in a particular programmatic area (e.g., non-Hodgkin lymphoma, bone marrow failure). Abstracts for review are chosen because of their predicted impact on the field. Both basic and clinical research are represented throughout the program. The meeting format encourages interaction between speakers and attendees, and attendees are encouraged to discuss, with the speakers, challenging cases from their own experience. The collegial atmosphere of these meetings supports national and regional networking opportunities for those in attendance.

Both meetings will be co-chaired by Dr. Marc J. Kahn and Dr. Wendy Stock. The international co-chairs for each meeting are Dr. Wee Joo Chng and Dr. Sin Tiong Ong (Asia) and Dr. Carlos Sérgio Chiattone and Dr. Roberto Passetto Falcão (Latin America).

Highlights of ASH in Asia will take place March 29-30 at the Suntec Singapore International Convention & Exhibition Centre in Singapore. For this meeting, ASH will partner with the Cancer Science Institute of the National University of Singapore. Advance registration is open until March 3, and on-site registration is available on the first day of the meeting. For more information on the Highlights of ASH in Asia meeting, please visit www.hematology.org/highlightsasia14.

In partnership with the Associação Brasileira de Hematologia, Hemoterapia e Terapia Celular, Highlights of ASH in Latin America has been scheduled for April 26-27 at Costão do Santinho Resort in Florianópolis, Brazil. Simultaneous translation from English to Spanish and Portuguese will be available at this meeting. Early-bird registration is open until March 10, and advance registration is open from March 10 through April 7. For more information on the Highlights of ASH in Latin America meeting, please visit www.hematology.org/highlightslatinamerica14.
Ankle Ulcers in Sickle Cell Disease: A Topical Solution to a Vexing Problem?

STUDY TITLE: A Phase I Study of the Use of Topical Sodium Nitrite for Chronic Leg Ulcers in Adult Patients with Hemolytic Disorders

CLINICALTRIALS.GOV IDENTIFIER: NCT01316796

SPONSOR: National Heart, Lung, and Blood Institute

LOCATION: National Institutes of Health Clinical Center

ACCRUAL GOAL: 44

STUDY DESIGN: This study was developed originally using a non-randomized, dose-escalation, phase I strategy to examine topical sodium nitrite cream as a treatment for chronic leg ulcers in patients with sickle cell disease (SCD) or other red blood cell disorders. The protocol was subsequently amended, converting the study into a double-blind, placebo-controlled trial designed to determine the maximum tolerated dose of the sodium nitrite compound. Secondary endpoints focus on the extent and duration of response to treatment. Adverse effects, including hypotension and methemoglobinemia, that could occur as a consequence of sodium nitrite absorption from the ulcerated skin will be documented. To be eligible, patients must be at least 18 years of age, have SCD or another red cell disorder, and have had a leg ulcer > 2.5 cm² but < 100 cm² for more than four weeks. Exclusion criteria include recent use of nitroglycerin, nitroprusside, nitric oxide, arginine, sildenafil, or methemoglobin-inducing drugs or a history of methemoglobinemia or infection.

RATIONAL: Chronic leg ulcers are painful, debilitating complications of SCD and other hemolytic anemias that contribute to early mortality, poor quality of life, and increased disability of affected patients. In SCD, the incidence of leg ulcers varies from 8 to 75 percent depending on variables such as geographic location of the patient, genotype of hemoglobin (HbSS more than HbSC), age (most affected patients are between the ages of 18-25), contribution of trauma to lesion, concentration of hemoglobin (lower worse than higher), an accompanying hypercoagulable phenotype, or the presence of the lupus anticoagulant. (Bazuyu GN et al. J Med Sci. 2010;8:190-194; Minniti CP et al. Am J Hematol. 2010;85:831-833; Delaney KM et al. Hemoglobin. 2013;37:325-32). Decreased tissue blood flow appears to be the major contributor to both ulcer formation and delayed healing. The pathophysiologic process that underlies abnormal tissue perfusion is multifactorial with vaso-occlusion by microthrombi, upregulated integrin expression that promotes platelet and granulocyte aggregation, increased venous pressure, bacterial infections, abnormal autonomic nervous system control with excessive vasoconstriction when in the dependent position, and degree of anemia having been proposed as contributing factors. Recent studies by Caterina Minniti’s group that examined histopathology and assessment of the microcirculation by laser imaging and thermography showed evidence of venostasis, inflammation, and vasculopathy (Minniti CP et al. Am J Hematol. 2014;89:4-6). Blood vessels were increased and activated endothelium with evidence of microthrombi and fibrin deposition along with thrombosis/recanalization were observed. Remarkably similar findings were noted in chronic venous ulcers in individuals without SCD, suggesting that leg ulcers may be a common end-organ complication of endothelial dysfunction.

Current treatment options for leg ulcerations, including antibiotics, compression bandages, dressing changes, Unna boots, silver and zinc oxide gauze, skin grafts, and maggot therapy, rely mostly on bacterial containment, stimulation of tissue granulation, and reduction of venostasis to ameliorate disease severity. A recent Cochrane Database review, however, identified only six treatment-focused clinical trials over the past 20 years, three involving systemic interventions (L-carnitine, arginine butyrate, isosuximide) and three based on topical interventions (Solcosery® cream, RGD peptide dressing, topical antibiotics) (Marti-Carvalho AJ et al. Cochrane Database Syst Rev. 2012;11:CD008394). Studies using three interventions (arginine butyrate, RGD peptide, and L-carnitine) reported on change in ulcer size. Of these, only the study that used an RGD peptide matrix (to block integrin-receptor binding) showed a significant reduction in ulcer size compared with the control group. Three trials – those using isoxsuprine, arginine butyrate, and RGD peptide matrix – reported on the incidence of complete closure of the ulcers. None found a significant effect. No trial reported on the time to complete ulcer healing, ulcer-free survival for patients with SCD, quality-of-life measures, or incidence of amputation. There was no reported information on the safety of these interventions.

Nitrre oxide (NO) bioavailability is diminished in SCD as a consequence of associated processes including hemolysis, oxidative stress, and arginine depletion. NO, a soluble gas with a half-life of a few seconds, is continuously synthesized in endothelial cells from the amino acid L-arginine by NO synthase. In addition to its antimicrobial activity, NO mediates a number of essential biologic processes, including vasodilation, wound healing, and angiogenesis. Moreover, NO has anti-platelet effects and influences availability of several growth factors involved in endothelial homeostasis. Based on these properties, topical delivery of NO is a logical approach for treating leg ulcers in SCD. Sodium nitrite is thought to act as a reservoir for NO. The nitrite anion acts as a vasodilator in vivo by generating NO in tissues with low oxygen tension and pH. The mechanism involves a novel physiologic function of hemoglobin and myoglobin as oxygen- and pH-dependent nitrite reductases. Therefore, nitrite provides the ideal substrate for NO generation along the hypoxic gradient likely to be present in chronic wounds.

COMMENT: This topical solution to healing skin ulcers in hemolytic disease appears to have merit. As described above, many factors are involved in the pathogenesis of the ulcers related to hemolytic disease, including inflammation, oxidative stress, venous insufficiency, hypercoagulability, hypoxia, and infection. The lack of efficacy of therapeutic approaches with a limited spectrum of activity underscores the complexity of this pathogenesis. In chronic venous-stasis ulcers not related to hemolysis, improving blood flow is paramount. Under hypoxic conditions, nitrite cream releases NO to promote vasodilation and thereby improve blood flow (Umbrello M et al. J Physiol. 2014. Ebaph ahead of print). The hypoxic environment of the ulcer may further induce additional NO generation as a consequence of NO release from deoxygenated hemoglobin. Nitrite also has significant antibacterial activity (a property that has been appreciated by butchers for years) that can inhibit bacterial growth in the ulcer. Production of methemoglobin by the oxidative effects of nitrite may have a beneficial effect by releasing heme that can interact with l-arginine receptor 4 expressed on the vasculature to promote an inflammatory response that could contribute to the healing process (Belcher JD et al. Blood. 2014;123:377-90). Potentially, nitrite may also affect pain pathways via local mast cell activation as recently described (Vincent LE et al. Blood. 2013;122:1853-62). The main toxicity concern is whether the dose of nitrite delivered by the treatment compound will cause systemic hypotension, as absorption from an open wound can be significant. As noted above, the present study will assess hypotension and methemoglobinemia as adverse events.

The devastating toll that leg ulcers exact from patients with SCD and other hemolytic anemias is under-appreciated. This well-designed study will generate substantive new information on the strategy of topical delivery of NO as treatment for this challenging complication of SCD vasculopathy.

-- Gregory M. Vercellotti, MD
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The Hematologist: ASH NEWS AND REPORTS

A Snapshot of the Media Coverage at the 2013 ASH Annual Meeting

We know that hematologists who attend the annual meeting do so for various reasons, whether it’s to hear the breaking research being presented, to learn a new treatment approach, or to network with experts in the field. We also have reporters from around the world who attend the meeting to cover the research and science being presented.

The 2013 meeting had a record number of reporters covering the meeting. Two hundred and sixty-two reporters (79 domestic and 183 international) from more than 150 outlets in 26 countries traveled to New Orleans to cover the meeting site. An additional 16 reporters covered the annual meeting remotely, conducting interviews via phone, participating in teleconferences, and accessing the online press kit. The following is a sampling of the coverage that ASH received during and after the annual meeting:

The Associated Press via USA TODAY (online, 12/7/13; print, 12/8/13): “Gene Therapy Scores Big Wins Against Blood Cancers”
CNN/CNN.com (online/broadcast, 12/7/13): “Killing Cancer Like the Common Cold”
National Public Radio (online, 12/7/13): “Gene Therapy Keeps ‘Bubble Boy’ Disease at Bay in 8 Children”
Time.com (online, 12/9/13): “Gene Therapy for Cancer Shows Continued Progress”
Bloomberg News (online, 12/9/13): “New Leukemia Medicines Bring Chemotherapy-Free Potential”
Good Morning America (online/broadcast, 12/12/13): “‘Bubble Boy’ Disease, Nearly Always Fatal, Could Have Cure”

ASH member Dr. Joseph Mikhael speaks with reporters at the conclusion of a press conference during the annual meeting.

January 23, 2014


In this study, Jabbour and colleagues present three-year follow-up data from a phase III trial comparing dasatinib and imatinib for the initial therapy of chronic myeloid leukemia. Data demonstrate that patients who received dasatinib experienced more rapid and deeper molecular responses compared with those who received imatinib. Perhaps more importantly, achievement of an early, deep molecular response was predictive of better progression-free and overall survival for patients in both arms of the study, suggesting that early assessment of molecular response to tyrosine kinase inhibitors is clinically relevant.

January 16, 2014


Mahlangu and colleagues report the results of a pivotal phase III study in which they evaluated the safety, efficacy, and pharmacokinetics of rFVIIIc, a novel recombinant FVIII Fc fusion protein with a prolonged half-life that was developed to allow lengthening of the interval between prophylactic injections for patients with hemophilia A. The efficacy of rFVIIIc was evaluated for prevention of bleeding in the prophylactic setting, for treatment of acute bleeding, and for perioperative hemostatic control in 165 previously treated males aged 12 and older with severe hemophilia A. Analysis showed that the recombinant protein was well tolerated and lowered bleeding rates when dosed once to two times per week.

January 2, 2014


Hematopoietic stem cells maintain stemness through various protective mechanisms. Among these defenses, heat shock proteins preserve cell homeostasis during stress responses, safeguarding stem cells against potentially traumatic metabolic insults. In this Blood article, Tai-Nagara and colleagues report that the mitochondrial heat shock protein, mortalin, plays an essential role in maintaining stem cell properties by modulating response to oxidative stress. While inhibition of mortalin function results in abnormally high levels of reactive oxygen species in stem cells and reduces stem cell numbers, over-expression of mortalin induces the reverse effect, indicating that mortalin is a necessary component of hematopoietic stem cell homeostasis.


Clinical multiple myeloma (CCM) is preceded by an asymptomatic monoclonal gammapathy (AMG), classified as either mononclonal gammapathy of undetermined significance (MGUS) or asymptomatic multiple myeloma (AMM). Dhodapkar and colleagues detail results of their data analysis from a prospective observational clinical trial of AMG patients (n=311). Baseline data from clinical variables, gene-expression profiles of purified tumor cells, and skeletal magnetic resonance imaging were correlated with the risk of progression to CMM. The authors write that a high-risk score based on serum-free light chain concentration, M-protein concentration, and a 70-gene expression profile identified a subset of AMM patients with a high probability of progression to clinical myeloma. On the other hand, a low-risk score based on these factors predicted a probability of progression similar to that observed in patients with MGUS. Detection of multiple (-1) focal lesions by MRI also conferred an increased risk of progression. The results of this prospective study suggest that gene-expression analysis can contribute to the management of AMG by aiding in the identification of patients at high risk for progression to CMM.

December 19, 2013


Although superficial vein thrombosis (SVT) is largely regarded as a benign process, it is associated with an increased risk of deep-vein thrombosis (DVT). However, there are almost no data to guide identification of SVT patients who are at risk for subsequent development of DVT. Roach and colleagues analyze results from a case-controlled study of nearly 5,000 patients and more than 5,700 controls to identify risk factors associated with development of DVT in patients with a previous SVT. Consistent with previous data, patients with previous SVT had a nearly six-fold greater risk of developing DVT than the control group, and the relative risk increased in patients with concomitant acquired thrombocytic risk factors. There was a 9.3-fold increase in SVT patients with a mild thrombocytic risk factor such as smoking or obesity, whereas the risk was 31.4-fold greater for SVT patients with a strong risk factor such as surgery, hospitalization, oral contraception use, plaster cast immobilization, or malignancy. This study identified subsets of patients with prior SVT who may benefit from risk-based management ranging from life-style modification to thromboprophylaxis.

December 12, 2013


Ruxolitinib is a potent Janus kinase 1/2 (JAK1/JAK2) inhibitor that has demonstrated reductions in splenomegaly and improvement in both disease-related symptoms and quality of life in patients with myelofibrosis (MF). The analysis published in Blood reports the three-year follow-up (median 151 weeks) of the efficacy and safety of COMFORT-II, a clinical trial comparing ruxolitinib with best available therapy in 219 patients with intermediate-2 and high-risk MF. In the ruxolitinib arm, 97 percent of patients (132 of 135) with post-baseline assessments experienced some degree of reduction in spleen volume at any time during the study. Of the 51 percent of patients on the ruxolitinib arm whose spleen size diminished by a 35 percent, the probability of sustaining this response through at least 144 weeks on the study was 50 percent (95% CI, 36%-63%). At the time of this analysis, 45 percent of the patients randomized to ruxolitinib remained on treatment. There was a 52 percent reduction in the risk of death in the ruxolitinib arm compared with the best available therapy arm. Thus, long-term analysis shows that ruxolitinib treatment is well tolerated and results in durable reduction of splenomegaly and in improvement in survival.
It’s a Small World: Hematology Research Opportunities Around the Globe

The past 20 years have seen an unprecedented interest in global health among faculty and students in North American universities. The impact of basic and translational research combined with resources provided through the President’s Emergency Plan for AIDS Relief (PEPFAR) on the global epidemic of HIV has been transformational. Recognizing that cardiovascular and pulmonary diseases, diabetes, and cancer are now the leading causes of death worldwide, the United Nations General Assembly held a high-level meeting on non-communicable diseases in September 2012, only the second such meeting on a health topic.

In 2008, the National Heart, Lung, and Blood Institute (NHLBI) established an Office of Global Health to guide its activities in the global health arena. The NHLBI has been a leader in addressing non-communicable diseases at the National Institutes of Health (NIH), co-hosting the trans-NIH working group on global health and the Global Alliance for Chronic Diseases. Essential to effective engagement is the development of a diverse, multilocalized workforce of U.S. and international investigators.

The NHLBI has been an active participant in the Global Health Program for Fellows and Scholars, which provides early-stage investigators from the United States and low- and middle-income countries opportunities to enhance their global health research expertise and their careers. The program is run by the Fogarty International Center at NIH, with funds provided by categorized Institutes, including NHLBI, for work within our mission areas. The program has enabled U.S. subspecialty trainees to gain international experience with mentors from their home institutions, and from international centers. Two cardiology trainees supported under this program have published about their experience, emphasizing both the importance of overseas training and home institutional support to successful research collaborations and the personal challenges of the experience.

Many potentially productive research opportunities in hematology require engagement in collaborative global investigation. Basic and clinical research on Burkitt lymphoma in Africa has informed cancer biology and treatment. Scientific discoveries can be made by studying diseases that are rare in the United States, but are common in other areas, such as hemoglobinopathies. Basic and translational research, identification of new targets for therapy, and health systems research opportunities abound. Developing new approaches to bringing the benefits of existing interventions and creating new approaches to health-care delivery may come from collaborations with investigators in diverse settings. In addition, developing new approaches to improving the availability of blood resources would improve health in areas in which trauma, complications of pregnancy and delivery, cancer, and surgical interventions lead to preventable death and disability. As cancers are defined by their molecular signatures, international partnerships will be essential to ensure adequate study participation to assess new therapies. Iron deficiency remains the world’s most common hematologic disorder, adding significantly to the global burden of disease and actually worsening over the past two decades in children under age five. Common and rare disorders of thrombosis and hemostasis require studies at a population level.

The NHLBI is eager for hematologists to join the cardiologists and pulmonologists now taking advantage of opportunities to train and do research in international settings. Strong support from home institutions, collaborations with international partners who are full participants in design and conduct of research, and creation of sustainable career pathways will enable the hematology community to exploit opportunities and improve health with broad impact and new frontiers. You can learn about the program at www.fic.nih.gov/Programs/Pages/scholars-fellows-global-health.aspx and by contacting Dr. Myat Htoo Razak, Division of International Training and Research in the Fogarty International Center at NIH (myathtoo.razak@nih.gov).


Hematology Research Experience Available for Minority Medical Students

Minority medical students who are interested in hematology and are in their early years of medical school are invited to apply for a summer research experience and career-development program through the ASH Minority Medical Student Award Program (MMSAP). Participants will receive a stipend to support an eight- to 12-week research experience in the lab of an ASH member. After completion of the project, MMSAP participants will present their findings at the Promoting Minorities in Hematology event at the ASH annual meeting in December in San Francisco. Students will remain involved with ASH and will be assigned a career-development mentor for the remainder of their time in medical school and through residency.

This program is open to minority medical students from the United States or Canada who are enrolled in a DO, MD, or MD/PhD program. Visit www.hematology.org/Awards/MMSAP/2624.aspx for specific eligibility requirements.

If you are eligible or know someone who may be interested in this opportunity, apply today or encourage others to submit an application. Completed applications are due by March 10.
### ASH Guides Mobile App

ASG Guides is a mobile app that features the Society’s collection of clinical quick-reference guides. The app now includes guides on immune thrombocytopenia (ITP), von Willebrand disease, and a recently updated guide on the evaluation and management of heparin-induced thrombocytopenia (HIT). In 2014, ASH will introduce additional mobile versions of the Society’s Clinical Quick-Reference Guide collection, including recommendations for use of epoetin and darbepoetin, management recommendations for thrombocytopenia in pregnancy, anticoagulant dosing recommendations and management of anticoagulant-associated bleeding complications, and recommendations for red blood cell transfusion.

To install the app (iOS and Android compatible), simply search for “ASH Guides” in your device’s app store. For more information about the guides and app, go to www.hematology.org/Practice/Guidelines/2934.aspx.

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### WHAT’S ON THE WEB

As technology and the Web have evolved, so too have ASH’s online offerings. Now you can download ASH apps for your smartphone or tablet, follow ASH on Twitter (www.twitter.com/ASH_hematology), find ASH videos on YouTube (www.youtube.com/user/ASHWebmaster), and visit ASH on Facebook at www.facebook.com/AmericanSocietyofHematology.

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### MARK YOUR CALENDAR

#### March

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<th>Date</th>
<th>Event Description</th>
<th>Location</th>
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<tbody>
<tr>
<td>3</td>
<td>Abstract submission site opens for ASH Meeting on Lymphoma Biology</td>
<td>Colorado Springs, CO</td>
<td><a href="http://www.hematology.org/meetings">www.hematology.org/meetings</a></td>
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<tr>
<td>10</td>
<td>Application deadline for the ASH Minority Medical Student Award Program (MMSAP)*</td>
<td>Washington, DC</td>
<td><a href="http://www.hematology.org/awards">www.hematology.org/awards</a></td>
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<tr>
<td>18</td>
<td>Online application deadline for the ASH-Harold Amos Medical Faculty Development Program (AMFDP) Award</td>
<td>Washington, DC</td>
<td><a href="http://www.hematology.org/awards">www.hematology.org/awards</a></td>
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<td>19</td>
<td>Deadline for postmark of all supporting documents and hard copies for ASH-Harold Amos Medical Faculty Development Program (AMFDP) Award Washington, DC</td>
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<td>28</td>
<td>Application deadline for ASH Clinical Research Training Institute</td>
<td>Washington, DC</td>
<td><a href="http://www.hematology.org/awards">www.hematology.org/awards</a></td>
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<tr>
<td>29-30</td>
<td>Highlights of ASH in Asia</td>
<td>Singapore</td>
<td><a href="http://www.hematology.org/meetings">www.hematology.org/meetings</a></td>
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<td>4</td>
<td>Deadline to submit nomination package for ASH Mentor Award</td>
<td>Washington, DC</td>
<td><a href="http://www.hematology.org/awards">www.hematology.org/awards</a></td>
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<td>5-9</td>
<td>American Association for Cancer Research Annual Meeting</td>
<td>San Diego, CA</td>
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<td>10-12</td>
<td>Thrombosis &amp; Hemostasis Summit of North America</td>
<td>Chicago, IL</td>
<td><a href="http://www.ithana.org">www.ithana.org</a></td>
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<td>11</td>
<td>Deadline to claim CME credits and print a CME certificate for the 55th ASH Annual Meeting</td>
<td>Washington, DC</td>
<td><a href="http://www.hematology.org/meetings">www.hematology.org/meetings</a></td>
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<tr>
<td>23-24</td>
<td>ASH Clinical Research Training Institute in Latin America***</td>
<td>Florianópolis, Brazil</td>
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<td>25-26</td>
<td>Highlights of ASH in Latin America</td>
<td>Florianópolis, Brazil</td>
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<td>29</td>
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<td>Colorado Springs, CO</td>
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<td>2</td>
<td>Application deadline for the ASH Visitor Training Program</td>
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<td>14-17</td>
<td>American Society of Pediatric Hematology/Oncology Annual Meeting</td>
<td>Chicago, IL</td>
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<td>1</td>
<td>Application deadline for ASH Bridge Grant</td>
<td>Washington, DC</td>
<td><a href="http://www.hematology.org/awards">www.hematology.org/awards</a></td>
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*To submit an application for the MMSAP, you must have requested a mentor by January 6, 2014.

**To submit an application for the Clinical Research Training Institute, you must have submitted an eligibility review by January 8, 2014.

***To attend the Clinical Research Training Institute in Latin America, you must have applied by January 22, 2014, and have been notified of acceptance by February 2014.

For additional meeting dates and award deadlines, go to www.hematology.org/Calendar.