ADVANCES IN HEMATOLOGY

Current Developments in the Management of Hematologic Disorders

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Understanding Congenital Platelet Disorders

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H&O What are congenital platelet disorders?

JDP Congenital platelet disorders usually refer to inherited platelet disorders, which are transferred genetically. The strict definition of congenital is that the person is born with the disorder, however, so the term also applies to infants who are born with platelet disorders for other reasons. For example, neonatal thrombocytopenia is often caused by infection before birth. Another example is transient immune thrombocytopenic purpura, which can be transferred to the baby in rare cases if the mother has antibodies against platelets. Congenital platelet disorders that are acquired in utero are transient; the babies usually recover with treatment and do well.

Understanding that congenital platelet disorders can be either inherited or acquired is important when hematologists are called in as consultants for babies who have mucocutaneous bleeding or a low platelet count. Regardless of the initial diagnosis, we as hematologists need to follow up with these patients for at least the first 2 years of life to determine whether the platelet count has normalized.

Bearing this in mind, it is still logical to use the term congenital platelet disorders to refer to those that are inherited.

H&O What are the different types of congenital platelet disorders?

JDP The 2 main categories are disorders in which the platelet count is low—thrombocytopenia—and those in which the platelet count is normal but the function is abnormal.

Among disorders that are characterized by low platelet counts, some patients will have very large platelets (macrothrombocytopenia), others will have average-sized platelets, and others will have very small platelets (microthrombocytopenia). Determining which category the patient fits in is important.

In cases in which the blood count is normal and we suspect abnormal platelet function, the gold standard test is platelet aggregation. If this test reveals aggregation to ristocetin but not to any other agonists, we diagnose Glanzmann thrombasthenia. If the test shows initial aggregation, followed by desegregation in a pattern and a decreased amount of dense granules, we diagnose storage pool defect. If the test shows initial aggregation, followed by desegregation in a pattern and a normal amount of dense granules, we diagnose secretion defect.

Some patients with congenital platelet disorders bleed because they have both low platelet counts and abnormal platelet function, which occurs, for example, in Bernard-Soulier syndrome. This condition is characterized by a combination of thrombocytopenia and giant platelets on the blood film. When we do platelet aggregation tests, we see aggregation to all agonists except ristocetin, which is the opposite of what we see in Glanzmann thrombocytopenia.

H&O What other diagnostic techniques are used in congenital platelet disorders?

JDP The first step in diagnosis is doing a complete blood count, which helps not only to determine the platelet count but also to evaluate other blood cells. The hematologist should also look at a blood film, in order to evaluate platelet morphology under the microscope. Hematologists who are in an academic environment and have access to an electron microscope can examine the

ultra structure of the platelet; this is not essential during the initial diagnostic steps. If the platelet count is greater than 100,000 and a functional defect is also suspected, the next step is to run a platelet aggregation test, which allows us to identify correctly some functional defects.

H&O How common are congenital platelet disorders?

JDP Nobody knows exactly how common most of these disorders are. For example, in the case of hemophilia A, we know that mutations in a single gene—*F8*—are responsible for the disease, and that it affects approximately 1 in 10,000 boys at birth. It is more difficult to estimate the numbers of people with rarer platelet disorders, such as Glanzmann thrombasthenia; we think that this disease affects approximately 1 in 500,000 infants at birth but the number may vary among populations. Congenital platelet disorders that have a less-clear phenotype are difficult to diagnose, but we are picking up more cases as awareness and testing improve. I get a lot of referrals to my practice, and I see approximately 20 to 25 new diagnoses a year.

H&O What recent information have we learned about congenital platelet disorders?

JDP There have been 2 important advances. The first is our improved understanding of the genetics of congenital platelet disorders, despite the fact that the platelets themselves do not have DNA. We have seen an explosion over the last 20 years in the discovery of genes that cause platelet disorders. This advance has allowed us to do testing in animal or cellular models that are helping us to understand better how platelets are formed. For example, we can now knock out a particular gene in an animal or cellular model and see what effect this has on the megakaryocytes, which are the cells that make platelets in the bone marrow. We now know that the genetic makeup of a megakaryocyte will likely determine the kind of platelets a person has. This may determine the way platelets respond to antiplatelet therapy.

The other advance is our improved understanding of the biology of platelets: specifically the role that several receptors play on platelet activation and the way that they transmit signals throughout the platelet (which is known as platelet signaling). Genetic discoveries, coupled with better understanding of platelet biology, have significantly advanced the field over the last 20 years.

Where we still need to make major advances is in testing platelet function. Our most useful test is platelet aggregation, which has changed little since it was introduced 40 or 50 years ago.

H&O Has treatment improved in recent years?

JDP The availability of thrombopoietin in the 1990s was a major advance, and in 2008 was saw the approval of 2 thrombopoietin mimetic drugs—romiplostim (Nplate, Amgen) and eltrombopag (Promacta, GlaxoSmithKline)—that increased the platelet count. These advances have allowed us to treat more patients with acquired and hereditary thrombocytopenia successfully.

But for most patients with congenital platelet disorders, we have not seen much improvement—we still use platelet transfusions or administer ancillary agents, such as antifibrinolytics. Also, the use of appropriate therapy may complicate the disease even further. For example, one problem in Glanzmann thrombasthenia is that some patients lack the fibrinogen receptor, and may develop antibodies against that receptor after receiving several platelet transfusions.

H&O How are refractory cases handled?

JDP We have reports on the successful use of human recombinant coagulation factor VIIa (NovoSeven RT, Novo Nordisk) in patients with Glanzmann thrombasthenia that has become refractory to treatment. Although at this point the US Food and Drug Administration has not approved it for this use, we do not have many other options for these patients when platelet transfusions do not work. I have used desmopressin in some patients with mild platelet disorders, but this does not usually work well in patients with Glanzmann thrombasthenia, so the best treatment for these patients remains platelet transfusions. Some patients have undergone bone marrow transplants because, for example, they have become refractory to platelet transfusions or have a very poor lifestyle.

In 2011, Dr David Wilcox of the BloodCenter of Wisconsin reported on the use of gene therapy for Glanzmann thrombasthenia in dog models (the first author is Fang). That gives me some hope that someday we will be able to treat these patients by correcting their genetic defects using some type of genome-editing technology.

H&O Which congenital platelet disorders do you focus on in your laboratory?

JDP We focus on inherited platelet disorders, especially gray platelet syndrome, which is characterized by the deficiency of alpha granules in platelets. A couple of years ago, our laboratory—working in collaboration with the laboratories of Dr Walter Kahr at the University of Toronto and Dr Andrew Weyrich at the University of Utah—found the gene responsible for gray platelet syndrome. That research appeared in *Nature Genetics* in 2011

(two other groups from the US and Europe also reported the gene at the same time). We recently also helped Dr Kahr to characterize the gene deficiency in a mouse model and it appears to mimic the human condition. Now that we have a good model of disease, we are currently working on *NBEAL2*, trying to understand what it does and why it causes thrombocytopenia.

My laboratory also focuses on understanding how platelets behave in the circulatory system. One technique we have developed is microfluidic technology (in collaboration with Dr Keith Neeves at the Colorado School of Mines), which allows us to study platelets under flow in real time. We hope that characterizing platelet function under flow could someday be an alternative to the current platelet assays for diagnosis.

We also are working to understand the common genetic variants that affect platelet function. Some people have platelets that are more reactive, and we still do not understand why. That could be important in cardiovascular diseases, for example—people with platelet hyperreactivity might be at increased risk for cardiovascular disease or from complications of cardiovascular disease.

H&O What direction should future research efforts take?

JDP First, we need to continue to work on understanding the genetic origin of rare disorders, such as gray platelet syndrome and Glanzmann thrombasthenia. I think that sequencing the genes of entire families with rare platelet disorders is going to give us a lot of information about platelet formation and function. We also need to learn more about the complex network of RNA that makes platelets behave differently from individual to individual.

There is a very elegant study that was published recently by Dr Paul Bray's group from Thomas Jefferson University in *Nature Medicine*—the first author is Edelstein—that shows that the platelet RNA repertoire in blacks is different from whites. This difference in platelet RNA might put some blacks at increased risk for cardiovascular disease because of their platelets' increased "reactivity." I expect to see more of this type of research that differentiates among individuals.

H&O Is there anything else that you would like to add?

JDP The production of platelets, or megakaryopoiesis, is a highly complicated process. Stem cells become differentiated and evolve into megakaryocytes, which produce platelets. This is a unique biological event—no other cell does what the megakaryocyte does, which is to build up a nucleus with extraordinary amounts of DNA and then make cellular fragments that will play different roles. As we learn more about megakaryopoiesis and tissue engineering in the coming decades, we should be able to make platelets in vitro from the patient's own megakaryocytes. Eventually, platelet transfusion from donors will be a thing of the past.

Suggested Readings

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