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"THERE'S ALWAYS SOMETHING"

MORE TO COME WITH SICKLE CELL

BY ROBYN K. COGGINS

The shape of the reds was very irregular, but what especially attracted attention was the large number of thin, elongated, sickle-shaped and crescent-shaped forms. These were seen in fresh specimens, no matter in what way the blood was spread on the slide, James B. Herrick wrote in 1910, six years after his intern Ernest Irons showed him a peculiar specimen under a microscope.

The blood came from 20-year-old Grenada native Walter Clement Noel, who was in Chicago to attend dental school. On his voyage from Barbados, Noel had developed an intense

sore on his ankle—similar to more than 20 others that had scarred his legs throughout his childhood in Grenada—which another doctor had treated with iodine. By the time Noel reached Irons and Herrick (an early adopter of microscopic blood exams and best known for having first described myocardial infarction), he had been coughing for five weeks and was feverish, dizzy, and jaundiced.

But the mysterious part of his illness showed in his blood. After Irons, then 27, first examined Noel he performed a smear and noticed "many pear-shaped and elongated forms" on the slide. Thus began a two-and-a-half year search for a diagnosis, following and studying Noel until he returned to Grenada after dental school to open a practice. Herrick published his 1910 paper, and by the 1920s, enough similar cases surfaced that the disease was coined "sickle cell anemia."

The Ewe people in Ghana, Benin, and Togo called the illness "body chewing" and the Adangme, "body biting"—alluding to the extraordinarily painful nature of the affliction.

However, the disease had been known elsewhere for centuries. One history traced the condition through a family in Ghana back to the 1670s. In Nigeria, the Igbo people called sufferers *ogbanje*, or "children who come and go" as evil spirits targeting families. The Ewe in Ghana, Benin, and Togo called the illness "body chewing" and the Adangme, "body biting"—alluding to the extraordinarily painful nature of the affliction.

More than 5 million people worldwide, most of whom have Sub-Saharan African, Middle Eastern, or Mediterranean ancestry, are affected by sickle cell disease (SCD)—a set of recessive, inherited blood disorders. (People of Latin American, Saudi Arabian, and Indian heritage also get SCD.)

Maylen Johnson (not her real name), a Pittsburgh native who's quick to laugh, has the SC type of sickle cell disease, meaning her father and mother each carried a different variant of the SCD trait. Now 40 (but you'd never guess it with her smooth skin and girlish cheekbone freckles), Johnson has been hospitalized for complications related to the disease so many times it's hard to keep track.

She and her twin brother were diagnosed at around 6 months old. Their mother noticed they cried more than usual and had trouble sleeping. "She could tell when we tried to crawl that something was hurting," Johnson says. Her aunt also had SCD and died at age 55; her grandmother, two nephews, and a niece are carriers.

Every red blood cell in our bodies contains hemoglobin, a knotty-looking protein that transports oxygen from the lungs through the rest of the body. Hemoglobin can typically be broken down into alpha and beta subunits. The problem in sickled cells resides in the beta part—it stems from just one nucleotide change, which causes one amino acid change, and results in structurally abnormal hemoglobin.

These irregular hemoglobin molecules clump together, stiffening red blood cells and altering their shape, which prevents them from moving smoothly through the blood vessels, especially capillaries. The result is oxygen loss, anemia, stabbing pain, and sometimes tissue death.

As a child, Johnson was physically active, though it sometimes led to complications. In high school, she ran track and was on the cheerleading team. She wanted to swim, *Who doesn't love to swim?* she asks. But she couldn't handle the cold pool water—even a short dip meant hospitalization for a bad cold or pneumonia. Same thing happened to her brother.

"We didn't understand then why we couldn't do a lot of the things that 'normal' kids could do," she notes.

"I always used to say that the pain felt like a piano dropped on me. When it's at its peak, it's very, very painful."

At least that level of pain isn't constant for Johnson—she experiences it three to five times per year. She mostly deals with chronic lower-level pain in her back, hips, and legs.

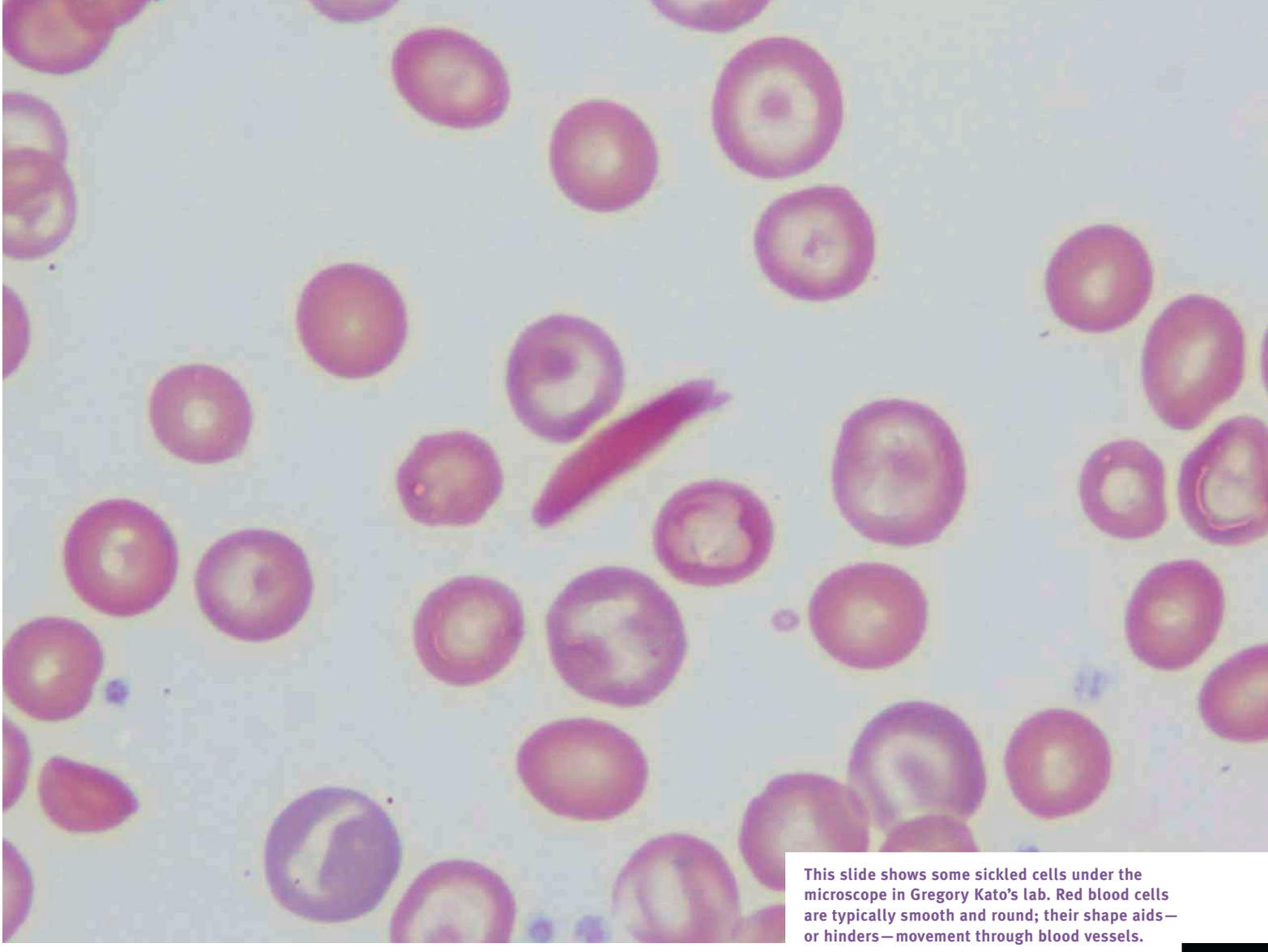
Life-sucking pain and fatigue are probably the most infamous of SCD's manifestations. People with SCD might also experience leg ulcers and jaundice (like Noel), swollen hands and feet, clogged blood vessels, strokes beginning in childhood, multiple organ failure, cognitive deficits, and lung disease (more on these last two in a moment). The extent of symptoms often depends on which of the four main types of sickle cell disease a patient has—HbSS, or sickle cell anemia, is generally most severe. In 1960, the average lifespan of someone with SCD was just 10 years. Today, thanks to improved infection treatment, preventive medicine, and blood transfusions, the average has jumped to about 50.

Often, the best doctors can do are blood transfusions to alleviate bad pain crises and to reduce the risk of strokes.

A stem cell transplant—from bone marrow, umbilical cord blood, or peripheral blood—can cure the disease. But such a transplant, of course, comes with risks of rejection and infection. And finding a donor can be extremely hard. The best match usually comes from a healthy sibling; yet many parents, once they have a child with SCD, opt not to risk having more children with the disease.

Johnson takes hydroxyurea (the only FDA-approved SCD drug), pain medication, and occasional blood thinners. She has had the same primary care physician her entire life and has been seeing Enrico Novelli, MD director of UPMC's Adult Sickle Cell Disease Program, for years. For lung checkups a few times a year, she visits Pitt professor of medicine Mark Gladwin.

Since he arrived at Pitt from the National Institutes of Health in 2008, Mark Gladwin—MD division chief of pulmonary, allergy, and critical care medicine and director of the multidisciplinary Heart, Lung, Blood, and Vascular Medicine Institute (VMI)—has summoned a diverse and multitalented sickle cell team that's tackling the disease



COURTESY GREGORY KATO

This slide shows some sickled cells under the microscope in Gregory Kato's lab. Red blood cells are typically smooth and round; their shape aids—or hinders—movement through blood vessels.

with renewed fervor.

"When I came, there [were] modest-sized, strong clinical programs ... but really no research in this space," Gladwin says. At the time, Novelli (Fel '01, Res '02) and his colleagues wanted to do more in the lab to help their 80-some patients, but he and the small staff were overwhelmed simply treating them.

"There's really a critical need for more hematologists doing research," Gladwin says, "but there's a national shortage. So part of this plan of building the VMI was to really enhance benign hematology research. So we're really thrilled now, six years later, that this plan is really coming together."

Gladwin brings a history of National Institutes of Health–backed pulmonary breakthroughs with him, primarily related to nitric oxide's role in blood vessel dilation and constriction. In translational studies, he showed that inhaled nitrite gas can reverse pulmonary hypertension, or high blood pressure in the lungs. Another of his studies showed that hemoglobin in SCD patients scavenges nitric oxide, which narrows blood vessels. Gladwin spent his first three years at Pitt beefing up his own lab and supporting Novelli, assistant professor in hematology/oncology, in developing his.

"[Gladwin] has put sickle cell at the center of the institute," Novelli says. "With him being an expert in sickle cell … he really had the clout to enact change within the institution. And so we've been able to secure funds now even from the Hemophilia Center for Western Pennsylvania and the Institute for Transfusion Medicine, to really boost our services."

In addition to crystallizing VMI's focus, Gladwin bulked up the institute's industry and community partnerships. (VMI collaborates with Ryan Clark's Cure League and has a partnership with Bayer.) UPMC's adult SCD program now has six doctors, two physician's assistants, two outpatient clinical nurses, and a senior clinician/social worker; together, they see more than 160 patients with sickle cell disease. (Children's Hospital of Pittsburgh of UPMC has another team of providers.) The VMI lays claim to a half-dozen labs pursuing vascular research related to sickle cells.

These physicians and scientists hope to create an integrated benign hematology center in Shadyside.

Dominique Stevens-Young (Pitt MSW '89), the program's senior clinician and clinical social worker, specializes in sickle cell disease management. Stevens-Young has been with UPMC since 2000 and says the program has never looked better. She talks a mile a minute about the early Gladwin days: "I used to call Mark and say, *Mark, you promised you were gonna change things when you got here, and things are still the same, and what are you waiting for?"* At a retreat this last September, held just as the new VMI/UPMC team was assembling, Gladwin told her: "I was waiting for now."

This February, Johnson spent a couple of weeks in the hospital for double pneumonia. Nine days in, then less than 24 hours out, then seven more days in.

"That's happened several times before,"

Johnson says of the back-to-back hospitalizations. "There's always something."

A century ago, Noel attributed his lifelong shortness of breath to heavy smoking. Herrick suggested hookworm, syphilis, intestinal parasites, and malaria, never quite landing on the correct interpretation. Nowadays, doctors have the right diagnosis but still don't understand the mechanisms of the disease.

"Most sickle cell patients do have some it feels like a heart attack.") It's not unusual for sickle cell patients to end up with ACS while in the hospital. Ofori-Acquah wants to know precisely how it develops, and has created a mouse model of the condition that any researcher interested in ACS can use. In developing that model, he has identified possible triggers for the syndrome.

Acute hemolysis—a buildup of heme in the blood—may be one. When red blood

"We didn't understand then why we couldn't do a lot of the things that 'normal' kids could do."

baseline lung dysfunction," says Solomon Ofori-Acquah, one of three sickle cell all-stars Gladwin helped bring to Pittsburgh to expand the SCD team here.

Ofori-Acquah has a PhD in molecular genetics; he investigates the bases of SCD complications, specifically in the lung.

"Even when [patients] are clinically well, they may have slightly [lower oxygen levels in the blood]," he says.

Less oxygen means less energy—Johnson's major symptom. Plus, hypoxemia often coincides with low levels of nitric oxide in SCD patients (one of Gladwin's areas of study). The oxygen and nitric oxide molecules, only partially absorbed by hemoglobin and therefore red blood cells, bind together and stick to vessel walls, causing deficiency of these important gases.

As patients age, the compounding effects of lung complications and damage become permanent and debilitating. Pneumonia after a stint of traveling and a bath ultimately killed Noel at age 32. Johnson has pulmonary hypertension and asthma; she has had two blood clots in her lungs and sleeps with oxygen at night.

One common and especially devastating lung condition in SCD is acute chest syndrome (ACS). The number two reason for hospital admissions and the leading cause of death among adults with SCD, ACS ravages the lungs, resulting in inflammation and fluid buildup. No one knows exactly what leads to it. (Johnson doesn't have ACS, but says lots of people in her support group do: "They say

cells break down, heme (and probably some iron) is released into the bloodstream. Since sickled cells die more quickly than healthy cells, a heme overload results. What signals this heme? What then causes ACS to progress so rapidly in mice and in humans? That's one set of mysteries Ofori-Acquah's NIH-funded lab hopes to crack.

His lab is also investigating the development of chronic lung injury and what might slow down its damage. When cells are stressed, they release enzymes to sweep up excess heme in the blood and halt damaging buildup. Ofori-Acquah thinks NrF-2, a transcription factor that induces those cytoprotective enzymes, might be a good target for future drug treatments. He wonders, *Is this the right pathway? Could drugs provoke the body's protective response?*

"It's almost like the cancer paradigm, where you are looking at a prognosis, and you take chemo to give you another 10 or 15 years," Ofori-Acquah says of potential therapeutics that might come out of his lab. It wouldn't be a cure, he says. But such a treatment could help patients "live a better quality of life with lungs that still function and that can still do a six-minute walk or jog."

Throughout our conversation about her life and illness, Johnson struggled to remember exactly when her hospitalizations were. Perhaps that's not surprising, as there've been quite a few; still, she says that her memory is "terrible."

"There [are] people from school I don't

new area of investigation for Novelli, as well as for SCD researchers generally. (Since he was a junior faculty member at Pitt, Novelli has focused much of his investigations on pulmonary hypertension and tissue pathways.) Physicians have long known that blockages in blood vessels affect organs and limbs, but not much, outside of anecdotes, is known about the effects on the brain.

Novelli says that, as a clinician, he sees how cognitive impairment makes it difficult for patients to adhere to plans for their care: "Sometimes we try and explain to patients, 'You have to do this; you have to take this medication; this is what happens.' And we think they understand. But then we realize that, indeed, their level of understanding and their memory are also very limited."

These deficits, in conjunction with the need for high doses of pain medication, can set up patients for derision. Sometimes callous providers accuse sickle cell patients of looking for a high with prescription meds or not taking responsibility for their own care.

"You want to get along with your doctor," Johnson says. "Dr. Novelli is very good at what he does. He's very nice." But she has encountered doctors elsewhere who didn't treat her so kindly and accused her of exaggerating her pain.

And many with SCD have trouble getting to appointments in the first place, because of mobility problems or a lack of access to transportation. Stevens-Young says the burnout rate for her colleagues who work closely with SCD patients is about five years. (Regarding her own experiences, she says, "I've got a lot of stories,

but most of them are sad stories.")

Though both Johnson and Stevens-Young sing the praises of those who've given long-standing care here, they've noticed the recent efforts to bolster the clinical team. "Never in the history of the program have I ever seen this much care and devotion towards our patients," Stevens-Young says. "I've been telling the patients, just hold on, things are gonna get better. And now it's here."

Pitt's Sruti Shiva, PhD assistant professor in pharmacology and chemical biology, and Novelli may have made the most intriguing SCD breakthrough at the VMI to date. With help from colleagues at Children's Hospital of Pittsburgh of UPMC, they revealed that SCD patients exhibit mitochondrial dysfunction.

Their results, published in *Blood* this May, are noteworthy for a couple of reasons. First, no one had ever studied mitochondrial function in this patient population before. And second, the dysfunction they discovered is associated with increased platelet activation in the blood, which is, in turn, associated with increased red blood cell death, hemoglobin abnormalities, and pulmonary hypertension. The researchers think that this "bioenergetic aberrancy" may be caused by complex V—an important link in the chain of mitochondrial energy transfer that's dysfunctional in SCD patients. All of this makes the mitochondrial issue a potential target for treatment.

And there's more to look forward to. Aes-103—a drug in phase 2 trials that binds to hemoglobin and may block sickling altogether—has been shown in some patients to increase oxygen absorption and stabilize red blood cells, stopping the cells from dying. Pitt/UPMC's new MD hire, Gregory Kato (pronounced kah-toe), led development of this prophylactic treatment as former director of the Sickle Cell Vascular Disease Section at the NIH's National Heart, Lung, and Blood Institute.

Also on Kato's short list: a 2010 cover story in *Blood* about the role of placental growth factor in the development of pulmonary hypertension in SCD and an upcoming *Blood* article showing that excess iron from repeated blood transfusions stimulates abnormal production of placental growth factor. He's also building a sickle cell registry at UPMC.

Kato joins other 2013 all-star hires Ofori-Acquah and Laura De Castro, an MD and national leader in clinical sickle cell research from Duke University. De Castro runs the adult sickle cell clinical programs with Novelli and is director of clinical translational research for the UPMC Sickle Cell Disease Center of Excellence, as well as a clinical faculty member in Pitt's Division of Hematology/Oncology.

De Castro focuses on mental health issues and end-organ damage in SCD, as well as finding novel treatments. She has been the principal investigator or coprincipal investigator on 20-plus NIH- and industry-sponsored clinical studies for hemoglobinopathies. Preliminary data from one recent study showed that 28 percent of SCD participants studied had indices of depression; further, De Castro found a "statistically significant association between the presence of depression and low scores for neurocognitive function domains," she reports.

De Castro's, Kato's, and Novelli's patient relationships should help inform the academic interests of the VMI labs and spur development of more effective treatments for SCD. Such collaborations are a bit like a relay race, with each party handing off a baton of knowledge to the next; with each revolution, the race distance gets shorter.

De Castro is quick to point out that Herrick's study of SCD was published more than 100 years ago—it's time to make strides toward better treatments, she says.

Ofori-Acquah will look toward Africa for insight. In his native Ghana, two of every 100 newborns have SCD, compared to one in every 5,000 in the United States generally and one of every 500 African Americans. This May, Pitt and the Kwame Nkrumah University of Science and Technology in Ghana signed a memorandum of understanding to help grow an international collaboration.

"There's a very small number of eligible patients for clinical trials" in the United States, Ofori-Acquah says. "What we're trying to do is develop collaborations in Ghana and elsewhere where there is a large patient population of sickle cell disease so that if we find a potential therapeutic, the clinical trials will not be such a headache."

It would mean the world to Johnson to have some of her energy back. Her passion is hair—twisting, curling, dyeing, cutting. Johnson trained as a stylist but doesn't have the stamina needed for that work now. She'd like to open her own salon—maybe someday. She has to pace herself. For a friend's wedding this June, Johnson did her aunt's hair. "But it took everything out of me," she says.

Johnson wants to build awareness about sickle cell disease. "There needs to be commercials, billboards, radio shows," she says. "It's not a nice disease. It needs to be more known."

"We're understanding the disease better; we're figuring out how to study it," Gladwin says. "The drugs are getting better, and companies are starting to focus on these diseases more and more. So this has become a kind of perfect storm here in Pittsburgh."

Johnson has learned to understand her disease better, too. She tires easily, but she tries to be active—getting out to her sickle cell support group, visiting with family and friends. She listens to her body, takes her medications, aspires to eat right.

And after a childhood spent avoiding pools, Johnson gave swimming another shot in her mid-20s. "I was definitely nervous, because it affected us when we were little. But you take chances, you know? I just took a chance."

At first, she just dipped her feet in. Then, she inched the rest of her body into the water, making sure it wasn't too cold.

She didn't get sick. "I can go swimming," she says with a grin. "I don't know why, but I can.

"This illness will give you some curveballs. A lot of them. You cannot predict it at all. When you think, *Oh I got this in the bag. I know everything about it*—no! No, you don't. I believe you can learn something until the day you die, because there's always gonna be something new.

"There's so much more to come about this disease." ■