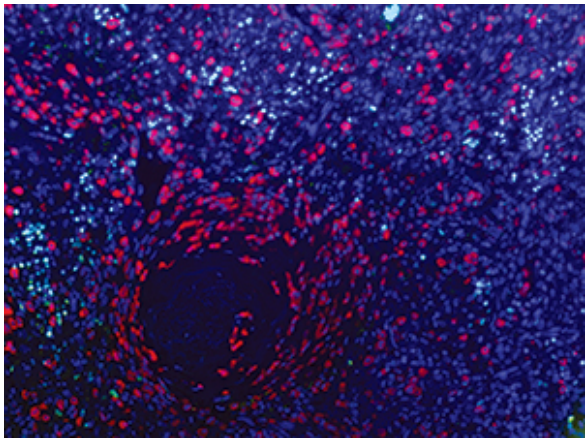
Quizzed in Ansbach, then key to a drug trial for mast cell disease

William Check, PhD

September 2016—In December 2007, American hematopathologist Tracy I. George, MD, spent a weekend in the small town of Ansbach in central Bavaria in the laboratory of Hans-Peter Horny, MD, whom she calls “the father of mast cell pathology.” Dr. Horny was at that time a privately practicing hematopathologist after having spent most of his career in academia. Plans for an international clinical trial were underway to evaluate the investigational drug midostaurin in advanced systemic mastocytosis, a rare group of diseases for which there was no effective therapy, and Dr. Horny would be the study pathologist. Dr. George, who had been diagnosing mast cell diseases for several years, wanted to take part as well.

“There was some concern since I had not published all that much. It was very early in my career,” says Dr. George, who is currently a professor of pathology, division chief of hematopathology, and vice chair of clinical affairs at the University of New Mexico. She put her expertise on the line and traveled to Ansbach. “I had nothing to lose and everything to gain,” she says.

“[Dr. Horny] quizzed me on mast cell disease diagnosis,” Dr. George recalls. “He was incredibly nice, but it was clearly a test. I guess I passed.” Dr. George became the study pathologist for the diagnosis and evaluation of U.S. and Canada patients, while Dr. Horny, who is now a professor in the Institute of Pathology at Ludwig Maximilians University of Munich, handled those from Europe and Australia. “The easy call for study pathologist was professor Horny alone,” Dr. George says, “but I muscled my way in.”



Mast cell leukemia involving spleen. Mast cells (anti-tryptase, red), proliferating cells (anti-Ki-67, green), and nuclei (DAPI, purple); strong colocalization of Ki-67 and DAPI appears white. Image acquired using Nuance spectral camera. Courtesy of Tracy George, MD, and Diane Lidke, PhD, University of New Mexico Department of Pathology.

Results from that trial, reported June 30 in the *New England Journal of Medicine*, were “very dramatic,” she says.

Hematologist Jason Gotlib, MD, MS, Dr. George’s colleague from her time at Stanford and the initiator and principal investigator of the midostaurin trial, tells CAP TODAY that the primary endpoint of the trial was improvement or normalization of organ damage. “Sixty percent of study subjects achieved this criterion,” says Dr. Gotlib, who is an associate professor of medicine at Stanford University School of Medicine and Stanford Cancer Institute.

“Of those patients who reached that mark, 45 percent achieved a major response,” defined as resolution of damage in one or more organs. In addition, the majority of patients had a greater than 50 percent reduction in the percentage of bone marrow mast cells and tryptase levels (Gotlib J, et al. *N Engl J Med.* 2016;374:2530–2541).
Dr. Horny says the trial could not have been successful without the pathologists’ contribution. “The input of the pathologist is highly crucial,” he says. “In particular, it is not possible to establish a diagnosis of systemic mastocytosis and its correct subtyping clinically.”

Dr. Gotlib agrees: “Mastocytosis is a challenging disease to get a handle on.”

Dr. George first became interested in mast cell disease shortly after taking a position as associate director of the hematology laboratory at Stanford following her hematopathology fellowship there. “I’m interested in things that are difficult because I want to learn to do them better,” Dr. George says. “That’s one reason I got into hematopathology—diagnosing bone marrows and lymph nodes is challenging.” For the same reason, she focused on myeloproliferative neoplasms, working with Dr. Gotlib on many cases. “There have been incredible advances in the pathology and treatment of myeloproliferative diseases in the past decade,” Dr. George says.

That interest led her to mast cell disease, which, until recently, had been considered a subtype of myeloproliferative neoplasms. “In 2016 it got its own classification,” she says. Dr. George worked with Dr. Gotlib in 2003 on a patient who had mast cell leukemia. “It is very uncommon,” she says. “That was my first one. Many pathologists have never seen one.”

Dr. Gotlib’s Stanford hematology colleague, Caroline Berube, MD, was the first to see the patient. Dr. Berube knew of Dr. Gotlib’s interest in investigational drugs for systemic mastocytosis and asked him, “Do we have anything?” At that time cladribine (2-chlorodeoxyadenosine) and interferon had been tried in small trials in mastocytosis, but they had only partial remitting activity with lack of durable follow-up.

Dr. George

Dr. Gotlib knew of midostaurin (then called PKC412), an investigational agent that was being tried in imatinib-resistant hematologic disorders. He also knew that 90 percent of cases of systemic mastocytosis were intrinsically resistant to imatinib due to the presence of the *KIT* D816V mutation, the genetic variant that activates mast cells and turns on constitutive cell signaling. When he petitioned Novartis to get PKC412 for compassionate use, the company declined. Dr. Gotlib then contacted Gary Gilliland, MD, PhD, a researcher at Harvard who had been working with the drug and had just shown, with colleagues, that it caused significant inhibition of growth in a murine transplant model of a myeloproliferative disease caused by the imatinib-resistant *FIP1L1-PDGFRA* T674I mutation (Cools J, et al. *Cancer Cell.* 2003;3:459–469) as well as potent inhibition of a *KIT* D816V-transformed cell line that was also resistant to imatinib. Based on these findings, Novartis agreed to release PKC412 for compassionate use in the Stanford patient.

“The patient did remarkably well,” Dr. George says. “At the outset she had 40 percent circulating mast cells. Within days of starting treatment they dropped to zero.” Liver function abnormalities also significantly resolved (Gotlib J, et al. *Blood.* 2005;106:2865–2870). “When the patient came in she was at death’s door,” Dr. George says. “After treatment with midostaurin she was able to go home and resume normal life.

“That’s a dramatic story. Unfortunately, she also had an associated myelodysplastic syndrome/myeloproliferative neoplasm.” While the mast cell disease improved, the MDS/MPN evolved to acute myeloid leukemia.

Even so, this result was sufficiently encouraging that Dr. Gotlib and Dr. George initiated a clinical trial of

midostaurin in advanced systemic mastocytosis at Stanford and two other U.S. sites. In their early results they saw findings similar to those of their initial patient. “The main point,” she says, “was that responses were ultimately seen in 18 of 26 patients, an impressive 69 percent.” She and Dr. Gotlib presented these results at the meeting of the European Competence Network on Mastocytosis in November 2008 in Budapest.

Along with the clinical data, Dr. Gotlib presented Dr. George’s images of bone marrow and blood of patients with very high numbers of mast cells being greatly reduced during treatment, and Dr. George gave a presentation on bone marrow evaluation in treated patients. “The data wowed the entire conference,” Dr. George says. “The audience appeared to be stunned by the results Dr. Gotlib presented, showing a highly effective treatment for patients with advanced systemic mastocytosis.” After Dr. Gotlib’s presentation, the audience applauded and asked numerous questions. When the session concluded, physicians and scientists gathered around them “asking for more details about what we had seen in our patients,” Dr. George says. “I can’t even begin to explain how exciting this was.”

Dr. Horny, too, recalls the audience’s response. “The reactions in Budapest in 2008 were enthusiastic!” he told CAP TODAY in an email.

By the time of these presentations, planning had already begun for the international trial and Dr. George had already been “tested” in Ansbach. During that December 2007 weekend, “Professor Horny picked up slide after slide of various types of mast cell disease. He looked at these slides with a microscope that was hooked up to a monitor—video microscope—and I could see the monitor. While he drove the microscope and asked questions over two days, I answered the questions as best I could.”

Dr. Horny

Says Dr. Horny, “We had an open and highly fruitful discussion, finally resulting in the starting point of the trial when we contacted Dr. Gotlib and Dr. [Peter] Valent of the University of Vienna on Saturday evening.”

During planning for the trial, Dr. George says, “What became increasingly clear was that pathologists were incredibly important. I could interpret lab values, I could look at bone marrows and peripheral blood. It wasn’t just mast cells decreasing; it was also eosinophils and monocytes in some patients. I talked with Jason Gotlib about how often to do bone marrows, what samples we should look at. Because we were so thorough, we have a robust understanding of how this drug works for this disease.”

The trial was conducted in patients with advanced systemic mastocytosis, which is an umbrella term for three distinct histologic entities: aggressive systemic mastocytosis (ASM); mast cell leukemia (MCL), defined by a bone marrow aspirate with at least 20 percent mast cells; and systemic mastocytosis with an associated hematologic neoplasm (SM-AHN). All can be fatal, owing to mast cell invasion of bone marrow, spleen, liver, gastrointestinal tract, and lymph nodes.

In addition to the clinical improvements already mentioned, Dr. Gotlib shared ad hoc survival data. “Survival is clearly short in patients with mast cell disease,” he tells CAP TODAY. In MCL, for example, survival is typically less than six months. In the trial, eight of 16 patients with MCL responded, and seven of those eight had major responses. “While median survival for the entire 16 MCL patients [including the responders] was 9.4 months, median survival for the eight responders had not been reached at the time of data cutoff,” Dr. Gotlib says. Median survival for those who did not have a response was 7.6 months, significantly less than for responders. Although the trial was not powered to look at survival, he called this finding “encouraging.”

Discussion is ongoing about a possible trial with combination therapy, perhaps midostaurin plus cladribine, Dr. Gotlib says, cautioning: “We must be circumspect about using the two drugs together, since it has never been done.”

In the Netherlands, a trial is already underway of midostaurin in patients with so-called indolent disease, those without frank organ damage. It is being directed by Hanneke C. Kluin-Nelemans, MD, PhD, of the University of Groningen, one of the participants in the advanced systemic mastocytosis trial. Patients with indolent disease have medical symptoms such as flushing, diarrhea, and possibly ulcers. The most serious is anaphylaxis.

Midostaurin is also being submitted to the FDA for treatment of *FLT3*/internal tandem duplication acute myeloid leukemia. A large, collaborative trial was presented at the 2015 meeting of the American Society of Hematology, Dr. Gotlib says (http://bit.ly/midostaurin-ratify).

One reason that pathologists were so crucial to the trial is that mast cell disease is difficult to diagnose. “It is a rare disease,” Dr. Gotlib says, “both from the clinician’s and the pathologist’s standpoint. Not many people have experience with it unless they are at an academic center that tends to see a lot of cases.

“Also, if you don’t think about mastocytosis, you are not going to find it. It has pleiotropic symptoms and pathology, and it often occurs with other myeloid neoplasms,” he says. Like their initial patient, many patients with systemic mastocytosis have an associated MDS or MDS/MPN such as chronic myelomonocytic leukemia (CMML). That makes it hard to decide what disease component is causing symptoms and laboratory abnormalities—the mast cell disease or the associated myeloid neoplasm. Without special stains, such as tryptase or CD25, it is difficult to see the mast cells.

Dr. Gotlib

“I say that mast cell disease lurks in the shadows,” Dr. Gotlib says. He cites a patient with CMML who developed liver disease. Upon biopsy the liver was full of mast cells.
Dr. George has been working with The Mastocytosis Society, a group of patients with possible mast cell disease. (She and Dr. Gotlib are on the group’s medical advisory board.) It has become clear to her that many patients are dealing with clinicians who are not familiar with the disease and not comfortable making the diagnosis, especially in children, who typically have a more benign form of the condition. Often insurance carriers won’t pay for a proper workup. Dr. George has been doing pro bono reading of slides for some patients. “I have seen cases where pathologists have called the bone marrow normal and when I look at it I see abnormal mast cells. That gives me an opportunity to educate pathologists.”

One of the important diagnostic clues, she says: “Neoplastic mast cells don’t look like normal mast cells.” They have a different morphology and can mimic other cells. “I clearly recall a case of a five-year-old girl with primary GI symptoms whose bone marrow was called normal. Yet it was obvious to me on the bone marrow aspirate smear that there were abnormal spindle-shaped mast cells. The referring pathologist had never seen this before. Once you see it you can do ancillary studies to prove they are there.”

Dr. Horny agrees that mastocytosis can mimic other diseases, clinically and morphologically. “Morphologically, aggressive systemic mastocytosis may perfectly mimic bland bone marrow fibrosis,” he says. “And mast cells express various markers or antigens that are usually related to other hematological malignancies, such as CD14, CD25, CD30, CD68, and others. Mast cells, especially in mucosal layers, may even lose expression of tryptase, the antigen most commonly related to mast cells, resulting in an incomplete-aberrant immunophenotype.” This adds

to the difficulty.

Dr. George calls being involved in the study the high point of her academic career. “It has been exciting to be on the cutting edge of diagnosing such a rare disease and being involved in finding effective therapy.”

For Dr. Horny, there has been an added professional benefit that can best be appreciated by other pathologists. “This was the first time for me to be involved as a consultant in an international trial. Never before was it possible for me to see so many cases of the rare category of advanced systemic mastocytosis. Until 2008 I had seen about 40 cases of mast cell leukemia over almost 30 years. Since then,” he says with excitement, “I have seen more than 100.”n

William Check is a writer in Ft. Lauderdale, Fla.