Evaluating post-treatment breast specimens

Karen Titus

January 2023—Laura Esserman, MD, MBA, can still recall her Eureka moment. She had just seen a talk on residual cancer burden by pathologist W. Fraser Symmans, MB.ChB, a pioneer in the field.

“When I saw Fraser present this,” says Dr. Esserman, director, University of California San Francisco Breast Care Center, “I knew immediately that MRI would work and that residual cancer burden would complement it. MRI was basically a snapshot of RCB over time. I realized that we had to institute RCB—we had to standardize our approach.” Until then, she and her colleagues across the I-SPY trial sites relied on individual pathologist assessment for each case. The pathologic complete response rate, or pCR, hovered at about 34 percent.

That insight was soon followed by another. Intrigued by what she heard, Dr. Esserman and her pathologist colleagues from all the I-SPY sites traveled to MD Anderson, where Dr. Symmans helped develop the residual cancer burden system, for training. “We brought them all to Houston on a hot summer day,” she recalls with a laugh. When they returned, they reviewed their cases. The results were startling. Using the new approach, she says, the complete response rate went down to 24 to 25 percent.

The impact on patient care was sobering. “That really taught me a lesson—these standards really matter,” says Dr. Esserman, who is also professor of surgery and radiology, UCSF School of Medicine, and PI for the I-SPY trial network and the WISDOM study. The line between patients’ treatments and a standardized approach to evaluating and reporting post-neoadjuvant therapy breast resections is direct, like a farm-to-table meal.

The urgency around the matter has only grown in recent years.

The seminal paper came out in 2007 (Symmans WF, et al. *J Clin Oncol.* 2007;25[28]:4414–4422). “So this is not new,” says Uma Krishnamurti, MD, PhD, associate professor, Yale School of Medicine, and director, breast pathology service. “It’s just that more and more centers have started giving neoadjuvant therapy.”

With good reason. Neoadjuvant systemic therapy (most typically chemotherapy) has become the standard of care for triple-negative and HER2-positive early-stage breast cancers. The impact can be dramatic. Patients who have a pCR to neoadjuvant chemotherapy have around a 90 percent survival at 10 years, says Veerle Bossuyt, MD, assistant professor, Harvard Medical School, and associate pathologist, Massachusetts General Hospital, “which is incredible for these aggressive tumors.”

Evaluating pathologic response also enables physicians to turn quickly to another regimen if a tumor is responding poorly to treatment. In some cases, tumors will even continue to grow during neoadjuvant treatment, says Sunati Sahoo, MD, professor of pathology and director of surgical pathology, UT Southwestern Medical Center, Dallas.

Dr. Uma Krishnamurti of Yale School of Medicine. She and others highlight the challenges in the pathologic evaluation and reporting of post-neoadjuvant therapy breast resection specimens. [Photo by Karissa Van Tassel]

Deescalating treatment is another reason behind the pCR push. A person who has a complete response to therapy in the breast and lymph nodes may no longer need a mastectomy, for example, and instead choose breast-conserving surgery and avoid axillary dissection. "That's why we, the I-SPY Pathology Working Group, started analyzing core biopsies taken at 12 weeks into therapy to see if it can predict response and help determine the type of surgery or even avoid surgery altogether," says Dr. Sahoo, who is also director of breast pathology services at Clements University and Parkland Memorial hospitals, Dallas.

Moreover, if a patient has a pCR, "they *know* the treatment worked," says Dr. Sahoo, a welcome relief from the uneasy, sometimes years-long wait to see if a treatment was effective.

None of these successes would have been possible without studying pathologic response assessments. "It's unusual that pathology is so central to an advance for treatment," says Dr. Bossuyt. With so many patients now navigating neoadjuvant therapy, she says, the number of pathologists seeing these cases has also risen. "It's not just the big centers anymore," she says.

As Dr. Esserman's own stark experience shows, however, the need for a standardized pathology approach is paramount.

Dr. Sahoo first published on these challenges and inconsistencies in 2009 (Sahoo S, et al. *Arch Pathol Lab Med.* 2009;133[4]:633–642). Given the passage of years, "You'd think this is a dead topic," she says. Clearly, it isn't.

She revisited this topic with a recent paper (Sahoo S, et al. *Arch Pathol Lab Med.* Published online Aug. 17, 2022.

doi:10.5858/arpa.2022-0021-EP), in which the I-SPY Pathology Working Group offered its recommendations for handling these specimens. One hope, she says, is that the group's work will help pathologists as they await updated guidance from groups such as the CAP and the AJCC.

(The CAP Cancer and Pathology Electronic Reporting committees are evaluating emerging standards in pathology reporting for breast cancer following neoadjuvant therapy. Liaisons are involved in the AJCC Breast Working Group to ensure alignment with AJCC recommendations for the standards.)

These specimens can be taxing. Pathologists' experience so far has been mostly derived from standards developed for cases seen in clinical trials, says Dr. Krishnamurti. This includes recommendations from the Breast International Group–North American Breast Cancer Group collaboration (Bossuyt V, et al. *Ann Oncol.* 2015;26[7]:1280–1291).

Among other things, the BIG-NABCG recommendations addressed the various systems for identifying response to neoadjuvant therapy. "Instead of being subjective and saying 'good,' 'excellent,' or 'poor,' how do you have a more objective method of assessment?" says Dr. Krishnamurti. Though there are several options, "The MD Anderson residual cancer burden is an important way of assessing response," in large part because it uses residual tumor size and residual tumor cellularity in the breast as well as responses in the lymph nodes.

The online RCB calculator (https://bit.ly/RCB-calc) provides both score and class and "is an excellent method" for predicting event-free five- and 10-year survival, Dr. Krishnamurti says. RCB-0 indicates a complete pathologic response; RCB-I is minimal residual disease; RCB-II, moderate residual disease; and RCB-III, extensive residual disease. "Within each of these groups, as well as across groups, and for each receptor subtype, the RCB is a continuous parameter for prognosis," Dr. Krishnamurti says. The RCB has held up well in multiple reproducibility studies; more recently, Dr. Esserman says, a pooled analysis of more than 5,100 patients from 12 sites and trials showed RCB score and class were independently prognostic in all subtypes of breast cancer (Yau C, et al. *Lancet Oncol.* 2022;23[1]:149–160). "It's a very consistent biomarker."

The RCB website has links to graphical illustrations for estimating the percentage of cancer cellularity and to the pathology protocol for macroscopic and microscopic assessment of RCB, and thus walks pathologists through the intricacies of handling these specimens, Dr. Krishnamurti says. Microscopically, residual invasive tumor can extend beyond what is grossly seen; the RCB calculator uses the primary tumor bed area. "It explains what to do when your residual invasive tumor is present only in a small portion of the grossly seen tumor bed," she says.

Pathologists already do much of the heavy lifting on these samples—histologic type, grading, tumor size, single tumor versus multifocal, lymphovascular invasion. Plugging numbers into the RCB calculator, Dr. Krishnamurti says, "is an extra few minutes. Or not even that. You have the information already; it's certainly not tedious."

Pathologists do have to calculate overall tumor and invasive tumor cellularity; again, says Dr. Krishnamurti, it's a relatively easy task, one that can be done in a matter of minutes if the specimen was handled properly.

It's worth the effort, says Dr. Esserman, who agrees taking those extra steps up front isn't particularly burdensome. "It's a huge burden, however, if you don't do it from the get-go. It matters how much disease you have," she says. The difference between RCB-0/I and RCB-II/III "is a meaningful split" and will determine whether patients will need additional therapy.

Given the relative ease of using RCB, Dr. Bossuyt says the question she gets most often from her clinical colleagues is: Why isn't everyone doing it?

Pathologists are not required by current protocols or guidelines to include RCB values in the synoptic report or in the final diagnosis report, says Dr. Krishnamurti; instead, many are reported using phrases like "probable or definite response" and "no definite response."

Dr. Bossuyt suggests there remains a lack of familiarity with it. "Maybe there's an aura of it being an ivory tower thing," she adds. But once pathologists start using it, these specimens become less overwhelming. "The sign-outs become much shorter, and you end up submitting fewer sections."

Dr. Sahoo empathizes with those who are starting to see more neoadjuvant specimens, recalling her early encounters with these cases. In 2004, “When I started as a faculty, I was struggling, like everyone else, with how to report these treated carcinomas,” she says, given the lack of widespread experience among pathologists. “And then slowly it seemed like every day one was coming into the lab.”

Adding to their difficulties, she and her colleagues didn’t have reliable access to information through an electronic health record system. “We didn’t know half the time—most of the time, actually—that the person had been treated with chemotherapy” prior to surgery. What they did know was that some of these tumors “looked weird,” as Dr. Sahoo puts it. “So we learned to look for that information in the patient’s chart when we suspected it.”

She also taught residents and fellows to look closely at the dates of core biopsy. If a core biopsy of a tumor was done several months earlier, she’d tell them, they needed to think about the possibility of neoadjuvant therapy. “The person wasn’t sitting at home doing nothing about it. A breast cancer freaks everyone out,” Dr. Sahoo says.

Things improved with the arrival of a more robust EHR as well as adopting the RCB system by the mid-2000s. For Dr. Sahoo, the latter has been a lifeline. “Currently, the majority of our breast cancer patients who are eligible for adjuvant chemotherapy receive it neoadjuvantly,” she says.

Still, challenges remain. The grossing template at UTSW contains a field for neoadjuvant therapy: yes/no. The first step is to track down that bit of history. “I always check myself; so do my colleagues,” Dr. Sahoo says. “Even during frozen sections of sentinel lymph nodes, I cannot rely on the surgeons to tell me if the patient had prior treatment, so we proactively look for that history.” Knowing the type of cancer (luminal versus triple negative versus HER2 positive) the patient had and the result of the prior biopsied node is extremely important, she says. “All this information helps me when I examine these treated nodes intraoperatively to help my surgeon colleague determine the next step in the axillary management.”

Depending on the type of tumor, some might be completely gone from the breast and lymph nodes when the pathologist sees the post-treatment resection specimen. The only remaining evidence might be the tumor bed.

Those are the easy cases, Dr. Sahoo says, and the pathologist can confidently report there is no residual tumor.

When there is residual tumor, pathologists face more forks in the road. What’s the best way to measure that residual tumor?

Even as experience with these cases grows, standardization has lagged. Says Dr. Bossuyt: “There’s been a real ask from pathologists to figure this out.”

The recent I-SPY working group recommendations, Dr. Sahoo says, might nudge the conversation along. “It was time to revisit the topic when the group acknowledged that things are not standardized.”

She and her coauthors started there, but discussions soon led to more in-depth discussion about controversies in the field. Then came a leap of faith, she says. “At the beginning, I have to tell you, most of my colleagues wanted to just highlight the problems, just like everybody else.”

To be clear, even absent standardization, she says, most of the work is done “very thoughtfully. But it takes several cycles of work, and you need feedback,” especially as each new unusual situation arises. “We need to be very clear why we want this paper to read as a recommendation. If we just keep going back and forth on the issues, they’re not going to be addressed.”

She adds: “It’s pretty clear based on the published surveys that in academic centers in the United States, we are not very consistent in the way we report treated tumors. And some pathologists are not aware there are certain important elements to include in the report.”

Dr. Sahoo

Even within her own group, she adds, disagreements can arise. What is the best way, for example, to measure tumor size post-therapy? “It’s not always easy,” Dr. Sahoo says. “Tumor cells are often scattered haphazardly over the tumor bed. Where do you put the ruler? Do you go with the number of slices that have tumor, combine them, or do you take one slide and measure the largest focus?”

Surgeons and medical oncologists also have a stake in these conversations, Dr. Esserman notes. “It’s not like we’re just doing mastectomies and it doesn’t matter what you find. It *does* matter. And so RCB is a way of standardizing evaluation of specimens. We don’t let people use whatever MRI protocol they want. They have to follow a certain protocol because it’s like a biomarker. RCB is a biomarker.”

The technical challenges can seem even more fraught for centers that are just starting to see these specimens, Dr. Bossuyt says. Many neoadjuvantly treated tumors today are easier for pathologists to assess, she says, than the advanced-stage, inoperable tumors that all centers were seeing before.

A portion of neoadjuvant cases pathologists encounter now are small lumpectomies, with relatively few sections. Submitting even one cross-section will allow them to give a quantitative assessment of residual tumor. “So then the treating physician can have a conversation with the patient about the risks and benefits of additional treatment, based on a precise estimate of their individual risk.

“A lot of these specimens now are incredibly straightforward,” Dr. Bossuyt continues. “If you get a small lumpectomy and sample it appropriately, and there’s no more tumor left, you get to say, ‘No residual carcinoma, and the patient has a great prognosis.’ What could be better than that?”

That’s one hand. What about the other?

“These specimens are technically challenging, and they’re very disorienting for pathologists,” Dr. Bossuyt says, “because we’re all used to all the important prognostic factors in breast cancer, and when you give the chemotherapy first, they’re all altered.”

That includes difficulty finding the tumor—there’s no good correlation between imaging findings and pathology findings. Imaging may be negative with a lot of residual disease on pathologic evaluation, or imaging may still see a lesion but there is no residual viable carcinoma microscopically.

Once the correct area is identified and sampled, “the second step is identifying tumor,” Dr. Bossuyt says. That’s easy enough if a lot of tumor remains. But in most cases, breast cancer is not a ball of tumor that shrinks. Instead, the cancer will often percolate, so to speak, through normal tissue, she says. Areas with relatively normal-looking breast tissue can alternate with areas containing tumor. If there are few tumor cells left, it can be hard to identify them.

And often there are multiple areas of concern and the specimens are extremely complex. “So these are not easy specimens for the pathologist to handle,” Dr. Bossuyt says.

“But it’s also an opportunity where we add a lot of value.”

Hence the need for two Big C’s: clips and communication.

The first may be more straightforward. When neoadjuvant treatment leads to pCR—which happens in most HER2-positive breast cancers, says Dr. Krishnamurti—it can be difficult to find the tumor bed. A biopsy clip can help

direct pathologists to the right spot. At Yale, she says, the radiologists put a clip in the breast biopsy site, even in cases not involving neoadjuvant therapy.

Placing a clip in the biopsied lymph node is equally important. Following neoadjuvant therapy, the tumor can completely resolve, but the lymphatic channels may become fibrotic from the treatment, preventing the sentinel node dye or radioisotope from reaching its mark.

The clip ensures that surgeons have the right node. “When we give the frozen section report, in addition to telling them whether there is tumor in the lymph node, we also tell them that a clip was found,” says Dr. Krishnamurti. That can extend to complicated cases involving multiple tumors, she adds, a not infrequent topic of discussion at tumor boards.

When UT Southwestern began doing more neoadjuvant chemotherapy, Dr. Sahoo recalls, the subject of lymph nodes began to dominate tumor board discussions. One case still stands out for her, involving a patient who had a positive lymph node before therapy; during the surgery, the surgeon removed sentinel lymph nodes. The pathology revealed residual tumor in the breast, while the sentinel lymph nodes that were removed were all negative.

“So my question was, ‘Where is that lymph node that was positive? How do we know that the biopsied node was removed?’” The surgeon’s response: Anything highlighted by the blue dye or hot (radioisotopes) was removed. But as Dr. Sahoo pointed out, she wasn’t certain which node was biopsied without seeing changes of a clip site. “So that’s when we started putting a biopsy clip marker in the lymph nodes if we know somebody’s going to receive neoadjuvant chemotherapy.”

“In order to do those things you have to work as a team,” Dr. Sahoo says. She can tell her surgeon colleagues that even though she’s reporting that all the nodes are negative, it doesn’t mean the positive node is accounted for. At the same time, she says, they realize, *I can’t rely on blue dye—I have to specifically remove that positive lymph node.* “Unless you do that, you can’t tell if the patient has complete pathologic response.” Her surgeon colleagues got ahead of the game, she says, and started to localize the biopsied node prior to surgery with radioactive seed to ensure removal of the node at surgery, instead of relying on tracers (blue dye or radioisotope).

Enter that other C, communication.

While wider adoption of EHRs has made things easier for pathologists, Dr. Sahoo says, that’s not the end of the discussion. Surgeons and oncologists might ask why the pathology report lacks the “y” for treatment in the staging, for example, or ask pathologists to repeat a marker. Surgeons and oncologists are clearly “keeping us on our toes,” she says. “We are constantly making sure everything is addressed in the pathology report.”

But EHRs don’t automatically dispense information, either. As Dr. Bossuyt notes, neoadjuvant specimens aren’t always labeled as such. For all breast specimens, “It’s really important to figure out why we have these specimens.”

At tumor board meetings, Dr. Krishnamurti says conversations often revolve around multiple tumors. Another challenge involves receptor profiles.

At Yale, “We routinely repeat the ER, PR, and HER2 on all neoadjuvant-treated specimens,” she says, “but by the international consensus you’re not required to repeat predictive markers unless the patient is in a trial or the oncologist requests it.”

Sometimes the tumor profile changes after treatment, however. Most often, receptors may be lost or decrease, she says. Sometimes a new receptor, which was not expressed before, now is, possibly due to tumor heterogeneity.

Dr. Esserman

Dr. Esserman has her own spin on the importance of communication, and she doesn't absolve her surgeon colleagues of responsibility.

"We use pathology tracking sheets," she says. "We've been doing this for years to make sure the pathologist knows: Here's where the tumor is located; what treatment the patient had ahead of time; were they on I-SPY; treatment specifics; making sure the pathologist knows to look for the clip in the tumor bed and where.

"It's essential to communicate that to pathologists," she says. "They can't do their job unless you do that." She developed the worksheet after talking with pathologists about what they needed. That also led to more standardization among her surgeon colleagues as far as marking specimens. "The more we can standardize what we do, the easier it is for them."

Likewise, the surgeons asked the pathologists to have a standard way of grossing specimens. "That lets me look down and say, *OK, they went from medial to lateral, and I know where the margins are,"* says Dr. Esserman. "You're trying to figure out, if you have to go back, *where* you have to go back."

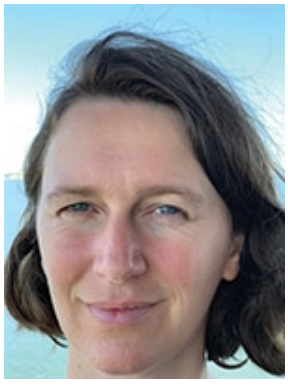
Now that pathologists no longer come to the OR regularly, she says, "I ink my own specimens. I do it in six colors because I want to make sure I know the orientation. There's no way for a pathologist to know that unless they come to the OR. And if you're not set up at your institution to do that, the surgeons can be taught to ink the specimens."

For the most complicated cases, she continues, "I've actually asked the pathologist to make a map of the specimen—where it's positive, where it's not. On these complex cases, if you sit down and talk to someone about it, and you sort it out, you can figure out who really needs to go back to the OR and who can avoid an unnecessary procedure."

Handling these specimens is "a team sport," Dr. Esserman says. "First of all, it's fun to work with your colleagues if you know them and everybody knows what their jobs are. There's no substitute for talking to each other. And having some camaraderie and saying, *How can I make your job easier so you can make my job easier?"*

Talking to clinical colleagues "actually makes it more pleasant and more rewarding for the pathologist," says Dr. Bossuyt. "Because you're working closely with your colleagues, and you know what's happening to the patient."

If it's not clear by now, the complexities of these specimens can make pathology reports more byzantine as well.



Dr. Bossuyt

Say, for example, a pathologist gets a request for Ki-67 analysis to see if a patient would benefit from abemaciclib. The interpretation of the Ki-67 result is dependent on whether the specimen has been pretreated, which may not be immediately clear from the report, Dr. Bossuyt says. “You can look at the ypT stage, but that’s all the way at the end of the report.” And while elements in current reporting try to address the neoadjuvant setting, things can still be confusing, she says.

Among her concerns: If there is no residual tumor because it was removed in the core biopsy, “then information from the original biopsy is added in the synoptic report. In the neoadjuvant setting, that’s not appropriate because the report for that surgical specimen needs to have the information at that point in time.” Dr. Bossuyt would like more clarity: “This is the information post-treatment.”

She’d also like to make it easier for clinicians to identify in the report whether there’s been a pCR. They might read, for example, that there’s no residual invasive carcinoma, then encounter a note referring to, say, lymphovascular invasion. “You’ll stage it as ypT0, and that’s prominent in the report,” Dr. Bossuyt says, “but buried somewhere else is that it’s not a pCR. It can be very confusing.”

Just as important is response in the lymph nodes. It’s been an area of longstanding confusion, she says. A report that indicates no residual tumor, but that there is carcinoma in the lymph nodes, is not a pCR—and it portends a worse prognosis. “We need to clearly identify those patients.” Moreover, she says, “In untreated specimens, isolated tumor cells are less important. They’re not going to affect prognosis or treatment in a big way. But in the neoadjuvant setting, *any* disease in the lymph nodes is important.”

Cellularity also plays a key role, says Dr. Esserman. “It can make the difference between more chemotherapy or not.” But sampling and location challenges muddy the picture for everyone.

Dr. Sahoo still encounters questions from her colleagues on this, even after two decades of experience, tumor board discussions, and consistent use of the RCB in addition to AJCC stage in their reports.

She gives one example of how complicated these cases can be. “Let’s say a case was reported out as pT1a by AJCC staging criteria,” indicating a tumor that is greater than 1 mm but less than or equal to 5 mm post-therapy. The pathologist had measured the largest contiguous focus in the tumor bed, which was 4 mm. But what if there are 10 or 20 foci (“Who’s counting?” she asks), some of them with single cells; what does that mean? “The surgeon asks, ‘You say the tumor bed is 20 mm, but then you say it’s pT1a. Which one is it?’”

With RCB in the report, pathologists can explain that even though the tumor bed is 20 mm, the scattered tumor cells only constitute 10 percent of tumor cellularity (compared with 100 percent before treatment). “Versus if I say, ‘It’s a 20-mm area of residual tumor but the cellularity is 80 percent.’” Which, Dr. Sahoo says, indicates the neoadjuvant treatment had minimal impact; on the other hand, reporting a cellularity of one, five, or 10 percent suggests a strong response, with only a few scattered tumor cells remaining.

The current AJCC recommendation for doing T stage is based on the largest contiguous focus, but that can be hard to pin down. “How much stroma do you need in between tumor clusters to call it contiguous or noncontiguous?” Dr. Sahoo asks.

“Nobody counts the number of foci,” she points out. “After five or six sections, each slide could have, say, five or 10 foci.” Totting them up “is not practical,” nor would it be easy to explain their size. “It is difficult for the oncologists to picture in their mind what the residual tumor looks like from reading a report, unless I am able to translate what I see under the microscope in a standardized manner.”

This is true of any organ system where the tumor has been treated neoadjuvantly, she adds. When the goal is to reduce the tumor volume, and ideally make it disappear, pathologists can be left with the equivalent of a locked-room mystery: “When you get the specimen, you are trying to make sense of what little is left—and *how* to tell the surgeon and the oncologist what is left, given the entirety of what you see.” Easier, perhaps, for poets to figure out how to capture the sensation of moonlight on a river.

Dr. Sahoo is sympathetic to the demands placed on each group of specialists. “You have three different people—oncologist, pathologist, surgeon—doing three different things.” Of the three, the pathologist is probably best positioned—like a baseball catcher—to see everything that’s happening with the patient and to talk to everyone involved.

Dr. Sahoo reports that the volume of these samples has increased to the point where, “believe it or not, some weeks our first-year residents will say, ‘I have not grossed or seen a breast cancer that hasn’t been treated with neoadjuvant chemotherapy yet.’” With untreated tumors fast disappearing, so are the old ways of looking at things.

If that’s astounding for pathologists, it’s even more so for patients. But this approach is demanding for them as well, Dr. Bossuyt says.

“Instead of going to the surgeon and having the tumor taken out, we’re asking patients to not do that immediately,” she says. “And they get treatment that is very difficult to tolerate. Patients have to live with this tumor for six months. Then they want to know: ‘Was it helpful? How did the tumor do?’” And when pathologists can then offer a detailed, quantitative response assessment, “Patients can do something with that number.”

It’s true, agrees Dr. Esserman. Ultimately, refining and standardizing the approach will only make things better. As she puts it, “That will let us get the right drugs to the right people at the right time.”

Karen Titus is CAP TODAY *contributing editor and co-managing editor.*