Targeted Epigenetic Therapies: The Next Frontier?

By Rabiya S. Tuma

When researchers look for mutations associated with cancer, they often expect to come up with alterations in signaling molecules or transcription factors. But an increasing number of the mutations found are in genes that regulate the epigenome—a system that alters DNA structure and regulates gene activity without changing the nucleotide sequence itself.

On Sept. 8, investigators published two independent reports online—one in Science and one in the *New England Journal of Medicine*—showing that mutations in an epigenetic regulatory gene, ARID1a, were associated with approximately half of the ovarian clear-cell cancers tested.

The two studies bolster the argument that epigenetics is important in tumor progression and possibly tumor formation, as well as that therapies targeted at specific epigenetic regulators or their effectors may improve patient outcomes. Although the approach’s viability remains unproven, many groups and companies are already working on molecularly targeted compounds designed to restore the epigenetic system.

**Frequent Mutations**

In both studies, the authors initially discovered mutations in ARID1a by sequencing protein-coding genes in ovarian clear-cell tumors. In the *Science* paper, Siân Jones, Ph.D., of the Johns Hopkins Kimmel Cancer Center, and colleagues report that 24 (57%) of 42 ovarian clear-cell cancers carried mutations in ARID1a. In the *New England Journal of Medicine*, Kimberly C. Wiegand, from the British Columbia Cancer Agency in Vancouver, and colleagues report that 55 (46%) of 119 ovarian clear-cell cancers carried ARID1a mutations, as did 10 (30%) of 33 endometrioid cancers. The mutations that both studies detected suggest that ARID1a functions as a tumor suppressor gene and that its role in remodeling chromatin—which can include unwinding DNA to allow transcription machinery access—is required to prevent tumor formation.

“The biology just confirms more and more that these epigenetic regulators are critical,” said Charles Roberts, M.D., Ph.D., assistant professor of pediatric oncology at the Dana–Farber Cancer Institute/Children’s Hospital Cancer Center and Harvard Medical School in Boston. Cancer genome-sequencing projects have uncovered mutations in epigenetic regulators more often than many investigators expected. “I think people who do this for a living are stunned at how frequently these things just keep popping up,” he said.

In addition to the ARID1a discoveries, investigators have found that deletions in SNF5—which, like ARID1a, encodes a chromatin remodeling protein—are associated with pediatric cancers. And mutations in epigenetic effector enzymes, such as the histone methyltransferase EZH2 and histone demethylase UTX, are associated with a variety of adult tumors, including prostate and breast cancers. Moreover, the discoveries keep coming: Researchers from Washington University in St. Louis reported in the Nov. 10 *New England Journal of Medicine* that acute myeloid tumors from 61 (22%) of 281 patients carried mutations in DNMT3A, a DNA methyltransferase.

The best analogy for how important these discoveries are, according to Roberts, is work done in the 1980s showing that the chromosomal translocations associated with leukemias disrupt transcription factors that are master regulators of development. That insight led to “a collective ‘ah-ha’ moment,” Roberts said. “I think epigenetic regulators are the next [regulatory] step up from that. They can regulate multiple transcription factors, not just a single lineage, and in some ways are the ultimate master regulators. I think the ‘ah-ha’ moment has dawned on people who do this for a living, but it hasn’t dawned on the field in general.”

**Getting Specific**

Exactly how to turn these discoveries into therapies that benefit patients, however, remains obscure. The first-generation drugs that target epigenetic modifications are fairly nonspecific. Instead of blocking a particular epigenetic regulator or enzyme, they inhibit a whole family of enzymes, such as histone deacetylases, which remove acetyl groups from histone proteins, or DNA methyltransferases, which add methyl groups to certain nucleotides in the DNA. These pan inhibitors have proven valuable in some cancers. DNA methylation inhibitors, for example, are used to treat myelodysplastic syndrome, a type of leukemia. “These are great but are quite nonspecific targets,” said Jean-Pierre Issa, M.D., a leukemia specialist at the M. D. Anderson Cancer Center in Houston, who has worked extensively with epigenetic drugs.

“We need to start thinking a bit more precisely when we discuss epigenetic therapy,” Issa said. “Given the plethora of distinct abnormalities that are being discovered by mutational screens [of tumor samples], I suspect we will need more tailored therapy approaches than what we have at present.”

Several experts share that view. In pharmaceutical companies and academia, groups are working to develop more targeted approaches to epigenetic therapy, according to Stephen B. Baylin, M.D., a professor of oncology and deputy director of the Sidney Kimmel Comprehensive Cancer Center at Johns Hopkins University in Baltimore, who specializes in epigenetic regulation in cancer.

One company involved is GlaxoSmithKline. “For us, the field has already moved in that
direction, and will only do more so, as more of these mutations are found in epigenetic regulators," said vice president Dash Dhanak, Ph.D.

His group already has what he calls a selective and potent small-molecule inhibitor of the histone methyltransferase EZH2. Dhanak said EZH2 is a potential target in oncology because a variety of cancers, including breast and prostate cancer, overexpress it. Also, mutations in UTX, a histone demethylase that works in opposition to EZH2, are associated with the same cancer types. He declined to give specifics but said that the inhibitor appears active in a variety of tissue culture and animal models and that the company plans to take the compound into phase I trials sometime next year.

If the EZH2 inhibitor that Dhanak describes is truly selective, that would be a step forward. “At the moment, getting a specific methyltransferase inhibitor hasn’t been easy,” said Peter A. Jones, Ph.D., director of the University of Southern California Norris Comprehensive Cancer Center in Los Angeles. Part of the challenge is that all the enzymes that methylate proteins use the same donor molecule, S-adenosylmethionine, according to Jones. He expects, however, that, the community will find a way around the problem.

“I think we are where we were with the kinases 30 years ago: Everyone said that because ATP is the universal phosphate donor we were never going to be able to come up with a specific kinase inhibitor,” Jones said. “And of course we now know that is not true.” As with the kinases, Jones predicts that once chemists understand the chemical backbone of small-molecule methylase inhibitors, they will be able to develop more and more selective agents.

Of course, GlaxoSmithKline isn’t the only group working to overcome the hurdle of targeted epigenetic therapies. At least two biotechnology companies, Epizyme Inc. and Constellation Pharmaceuticals, both in Cambridge, Mass., have bet their very existence on the idea, with the sole aim of identifying and developing targeted epigenetic therapies.

For histone methyltransferases, “that has proven to be a very doable thing,” said Robert Copeland, Ph.D., executive vice president and chief scientific officer at Epizyme. Although the team has not published data demonstrating that selectivity, they reported in an online publication in the Proceedings of the National Academy of Sciences Nov. 15 that a mutant form of EZH2, which is associated with B-cell lymphomas, requires the presence of wild-type EZH2 to drive tumorigenesis.

Jean-Pierre Issa M.D.

**Ramifications of Mutations**

Such biological complexity points to another challenge in developing targeted epigenetic therapies. Although an enormous amount of information has emerged in the last decade, the biology is only partially understood. A mutation in a gene that encodes a signaling protein or transcription factor affects a limited number of pathways in the cell, but mutations in epigenetic regulators or effectors can affect whole developmental programs, altering the expression of hundreds of genes all at once. And usually, researchers don’t know yet what effects individual epigenetic regulators or enzymes have at either the DNA or cellular level. “We need to know the ramifications of mutations,” said Johns Hopkins’ Baylin. “We need to know what pathways are altered, what specific epigenetic changes are really correlated with the presence of a mutation.”

With those issues in mind, both Baylin and Issa think that an epigenome cancer project is warranted. “Essentially, what we need to figure out is the epigenome of, say, ovarian cancers with ARID1a mutations different from the epigenome of ovarian cancers without that mutation?” Issa said. “And how is it different precisely? Until we have a very good handle on this [type of information], we are still going to be moving a bit in the dark in terms of epigenetic therapy.”

Roberts agrees that more information about how epigenetics works is needed. “I think we are getting close but are not quite there yet, understanding those mechanisms well enough to target them,” he said.

However, that doesn’t mean one has to wait for all the data to come in before starting the effort, Roberts points out. Working with researchers at Novartis Institutes for Biomedical Research in Cambridge, his group reported in the Nov. 14 Nature Medicine that deletion of SNF5, a chromatin remodeling protein like ARID1a, leads to aberrant activation of the Hedgehog–Gli1 pathway in malignant rhabdoid tumors, which are poorly differentiated tumors affecting the brain and several other tissues. Although the team doesn’t understand all the mechanisms or implications of the SNF5 deletion, they have identified a possible therapeutic target because the tumors appear dependent on Gli1 activation. Missing from this approach, he admits, is the knowledge of what other pathways might also be disrupted by the loss of SNF5.

“I would say we are going at it two ways, understanding individual pathways that are important [and deregulated] and the regulators themselves,” Roberts continued. “In my lab we are already thinking about targeting the epigenetic regulators. As we gain more and more understanding of how these epigenetic regulators work, we will be able to think about targeting them more specifically.” Until then, blocking one affected pathway at a time is worth trying.

But not everyone is convinced. “The question in my mind is whether the Hedgehog–Gli1 pathway is the only thing that is really wrong in these tumors,” Issa said. “And if it is not the only thing, will that kind of targeted therapy be enough to induce remission in patients? It is worth testing, but I’m skeptical that it will work as a single agent.”

With complete information or not, the researchers interviewed for this article agreed that targeted genetic therapies are the next step forward. “Certainly the existing epigenetic therapies have already shown success in the clinic,” Jones said, “and I think there is every reason to believe that being more specific would be more helpful. But it is very early days.”