CODE BREAKERS

By Holly Korschun

If human beings were assembled like automobiles, with each molecule, enzyme, protein, and cell laid out on a conveyor belt, the array would span a series of football field–long factories. The delicate piecing together of all those parts into a functioning whole of blood, tissues, and organs surrounded by skin and hair would require engineering genius and unimaginably marvelous machinery.

Among those collaborating to break the histone code are biochemist Xiaodong Cheng, microbiologist Jeremy Boss, biologist Bill Kelly, mathematician Eva Lee, and cancer researcher Paula Vertino.

Scientists face the opposite task. They are unraveling this elaborate human packaging from the outside in, like biochemical mechanics probing under high-performance human hoods.

Sequencing the human genome promised medical breakthroughs by targeting thousands of variations within human genes, acquired through inherited mutations or through changes resulting from environmental influences or disease.

Now scientists are uncovering more layers of variable influences on gene functioning that turn out to be just as important as the genome itself. This ever-expanding field of discovery, called epigenetics, refers to the biochemical neighborhood inhabited by each DNA molecule. After the genetic code is set, these neighborhoods decide whether genes are turned on or turned off. If turned on, they will undergo the process of transcription and manufacture proteins that carry out bodily processes. If turned off, they will wait silently in the wings while genes appropriate to a necessary function are expressed.

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Epigenetic influences include proteins and enzymes powered by biochemical reactions that place tags on individual genes, which in turn alert other enzymes and proteins to interact with them or not.

A multidisciplinary group of Emory scientists is studying an aspect of epigenetics called methylation—a chemical reaction that marks genes or their associated molecules. These marks dictate whether or not a gene will be expressed.

For the past several decades, scientists have known about DNA methylation, which marks genes during embryonic development and when cells divide. In recent years, scientists have discovered another kind of methylation that plays an integral role in gene expression. Biochemist Xiaodong Cheng, a Georgia Research Alliance Eminent Scholar, and his colleagues are pioneers in this new area of discovery called histone methylation. Histones are proteins that are part of the complex packaging of DNA within the nucleus of cells.

Strands of DNA wrap around groups of histones, like ribbon around a package, creating a bundle called a nucleosome. Several more compact layers tightly group into a chromosome, a complex arrangement the most accomplished LL Bean packer could be proud of.

Until a few decades ago scientists believed that histones were the most boring of molecules, serving only as a scaffold onto which DNA was wound and with little relationship to gene function. Now scientists know that many processes involving histones influence whether genes are turned on or off. Like DNA, histones can be methylated or marked and communicate with their neighboring histones or DNA. In 2000, the first histone methyltransferase—one of the enzymes that tags histones—was identified. This was the first time scientists had demonstrated the biochemical process of histone methylation. Since then, more than 75 histone methyltransferases have been identified. Cheng's group in the past two years provided the first crystal structure of a histone methyltransferase and illustrated how it interacts with the histone.

Now scientists are trying to crack the entire "histone code" by identifying all the modified histones (including methylation) and their enzymes, such as methyltransferases performing the chemical reactions and how they interrelate with the other epigenetic processes. It turns out that when certain histones are methylated, corresponding gene regions are not and vice versa. Histones can be methylated in different locations and tagged up to three times (with three methyl groups). The numerous permutations and combinations result in a mind-boggling variety of influences on gene expression.

Researchers are now discovering that problems with DNA methylation and histone methylation are linked to cancer and other diseases. If just one part of the histone code malfunctions, the wrong gene could be silenced. In the case of a growth-regulating gene, the result could be tumor formation. Winship Cancer Institute investigator Paula Vertino discovered several years ago that a tumor-suppressor gene called TMS-1 plays a role in breast cancer development when it is silenced abnormally. Others now have linked the same problem to glioblastomas, lung cancer, and melanoma. And scientists have discovered that histone methyltransferases can themselves be mutated in some cancers, including leukemia and lymphoma.

"There is a lot of interest now in how the different epigenetic signals are interrelated," says Vertino. "We haven't decided yet who's driving and who's following. Do DNA methyltransferases read histone alterations, or is it the other way around?

"The histone code is another layer of information superimposed on the genome," she explains. "This gives us another level of detail about what a gene looks like when it's on and when it's off. All of these marks are part of the normal way in which the genome is segregated into active and inactive genes. When genes are aberrantly methylated in some cancers, it's the result of a normal set of codes being applied in an inappropriate time and place."

Vertino is working with mathematician Eva Lee to identify patterns in DNA that dictate normal and abnormal methylation on a genome-wide basis. Lee uses mathematical algorithms to predict regions of the genome that are more likely to be aberrantly methylated, based only on the sequence of their DNA. Thus far, Lee and Vertino have applied the algorithm to chromosomes 21 and 22 and are testing their predictions on tumor samples.

"This is the first time anyone has approached aberrant DNA methylation from a mathematical standpoint," Vertino says. "Differences in DNA structure or binding proteins may offer us clues as to why some genes are aberrantly methylated."

Cheng and Vertino, along with scientists in a variety of Emory departments, are studying methylation using different models. Biochemist Cheng and his team are investigating the histone code from a broad perspective, using chrystallography, while Vertino concentrates on cancer.

Pathologist Paul Wade, who studies frog eggs, was one of the first scientists to uncover interactions between DNA methylation and histone modification. Biochemist Danny Reines uses yeast to study how methylation silences the FMR gene and leads to the development of fragile X syndrome, the most common form of inherited mental retardation. Microbiologist and GRA scholar Sam Speck studies methylation's role in allowing the Epstein Barr virus to hide from the immune system. Biologist Bill Kelly is looking at how methylation functions during the development of germ cells in the worm *C. elegans*, and biologist John Lucchesi is examining methylation in the sex chromosomes of fruit flies. Microbiologist Jeremy Boss researches methylation related to gene regulation in the immune system.

"Emory has a unique strength in epigenetics because our work crosses so many different disciplines and model systems," Vertino says. Emory scientists' exchange of research findings and ideas is a perfect example of how collaboration can advance the leading edge of scientific discovery. ■

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Talking in code

Just when you thought you were getting all that **genetics gab** down pat, here are some new additions to your vocabulary **sure to impress your friends.**

Epigenetics: changes in gene function that occur without changes in DNA sequence

Transcription: process in which DNA, through its genetic coding, manufactures proteins to carry out bodily processes

Methylation: a chemical reaction that places a recognizable marker on molecules such as DNA and proteins

DNA methylation: marking of certain genes during embryonic development and when cells divide

Methyltransferase: an enzyme that performs the chemical reaction that marks DNA or histones

Histones: family of five basic proteins that associate tightly with DNA in chromosomes

Histone methylation: a chemical modification that marks histones

Histone octomer: a set of eight histones

Nucleosome: structural unit for packaging chromatin; consists of a DNA strand wound around a histone octomer

Chromatin: a package of nucleosomes

Chromosome: single large DNA molecule and its associated proteins, containing many genes; stores and transmits genetic information

Histone code: combination of all the biochemical modifications that can occur in histones and associated DNA