Profile of Howard Y. Chang

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Since the 1930s, when RNA was recognized as chemically distinct from DNA, scientists have known of the diversity of RNA molecules. What they could not have fathomed at the time, however, is the variety of roles, both informational and structural, that RNA plays in biology: from protein expression and regulation to chromatin remodeling and epigenetic modification. Over the past two decades, physician-scientist Howard Y. Chang has helped enrich the RNA canon. He has discovered long sequences of RNA that do not code for proteins but instead modulate DNA accessibility. He has also helped develop next-generation sequencing methods that leverage his knowledge of the interactions between DNA, RNA, and proteins. In his Inaugural Article (1), Chang, who was elected to the National Academies of Sciences in 2020 and is a professor of dermatology and of genetics at Stanford University, describes the development of BABEL, a system that uses advanced computational techniques to translate between sets of sequencing data, down to single-cell resolution. The work provides further insights into the mechanisms of DNA regulation and protein expression and may aid the development of therapeutics for diseases such as cancer.

Coming to America

Chang was born in Taipei, Taiwan, but he, his mother, and younger brother moved to southern California when Chang was 12 years old. His father, a physician, remained in Asia. "My parents felt we would have better educational opportunities in the [United States]," Chang says.

America was a major cultural shift for his family, and Chang took English classes to quickly get up to speed. He said that "The social interactions in the [United States], especially with teachers, is very different than in Taiwan." He credits his public high school experience with laying the foundation for a successful career. Debate club helped him gain confidence and develop an understanding of logic and how to construct an argument. In high school, he also performed scientific research; Chang's biology teacher introduced him to a laboratory at the University of California, Irvine that was doing work on immune rejection in transplantation. Chang says that from an early age he was fortunate to have influential mentors.

Howard Y. Chang in his laboratory at Stanford. Image credit: Howard Hughes Medical Institute.

In Love with Research

From Irvine, Chang left for the east coast to attend Harvard University, where he studied biochemistry. There, he joined the laboratory of biochemist Christopher Walsh, now a consulting professor of chemistry at Stanford. Chang recalls his rigorous training there, saying that "Walsh made you start by washing glassware, and only then could you perform experiments." Chang continued research on transplantation, investigating enzymes that were the targets of drugs used in transplantation.

"I fell in love with lab-based research, because I felt that it provided an opportunity to understand something new about disease processes," Chang says. "At the same time, I saw the camaraderie among many scientists; they have peers across the country and around the world. And that was also really appealing to me."

This realization spurred Chang to apply to the joint Harvard/Massachusetts Institute of Technology MD-PhD program, to which he was accepted in 1994. After the first 2 years of medical school at Harvard, he joined molecular biologist David Baltimore's laboratory at the Massachusetts Institute of Technology, where he studied the mechanisms of programmed cell death, apoptosis, and receptor signaling involved in cell death processes.

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In the Baltimore Laboratory, Chang also learned how to approach scientific research. "David ran his lab as a mini-institute with a mindset that we could tackle any scientific question, using any approach, so long as it was effective," he said. "Exposure to this type of thinking helped me believe that if I felt a research direction was worth pursuing, it was, even if I had never worked on that problem before."

Dermatologic Diseases

After completing his education on the east coast, Chang returned to California in 2000 and began a dermatology residency and postdoctoral fellowship at Stanford University School of Medicine's Department of Biochemistry. Chang was drawn to dermatology because it allowed him to combine both aspects of his career. "You can easily visualize the biology of dermatological diseases and have direct access to the tissue. It is a really suitable field in which to make fundamental discoveries."

At Stanford, Chang joined the laboratory of biochemist Patrick Brown, who had recently invented DNA microarray technology. (Brown is now the CEO of Impossible Foods, a private company that makes plant-based meat-product substitutes.) It was in Brown's laboratory that Chang began the investigations that would come to define his research. He started with a project aimed at determining the factors that make skin from different parts of the body different. How did cells know, for example, that they were on the scalp or on the bottom of the foot? Chang examined messenger RNA expression in fibroblasts to discover that these ubiquitous support cells, which were thought to be homogenous, expressed different genes depending on their location (2).

Noncoding RNA

In 2004, when he started his own laboratory at Stanford, Chang continued investigating similarly fundamental questions in cell biology, such as: What made it possible for cells to activate different genes based on position or disease state, even though they all had the same underlying DNA? His studies led him to take a closer look at the noncoding parts of the genome, which account for the vast majority of cellular genetic material.

"Only 2% of the human genome codes for proteins. The rest, 98%, was thought to be junk," said Chang. "We now know that this noncoding DNA plays important regulatory roles."

Chang developed a platform to study the activity of noncoding DNA and discovered a large amount of unexpected transcription. His group found that the RNAs from this DNA have distinct patterns of expression and can modulate gene activity. In a 2007 article, they identified HOTAIR, a 2.2-kb noncoding RNA that represses transcription at one of the *HOX* genetic loci (3). The *HOX* loci encode a highly conserved family of transcription factors that play crucial roles in embryonic development.

In 2010 Chang's laboratory also uncovered a link between breast cancer and the dysregulation of long-noncoding RNAs (lncRNAs) at *HOX* loci. The expression level of HOTAIR, in particular, emerged as a predictor of metastasis and poor clinical outcome (4). The work drew attention to lncRNA sequences as well as the techniques used to isolate and study their functions.

According to Chang, "In the last two decades, the RNA field has undergone a revolution. In the early 2000s, people were very excited about small RNAs: siRNAs and microRNAs, which obviously have tremendous regulatory roles. In parallel, my lab and others started to highlight the pervasive transcription of lncRNAs in the human genome. We know now that there are three-times the number of lncRNAs as there are protein-coding genes."

Chang emphasizes that technological advancements were crucial to both discovering lncRNAs and identifying their biological roles. "The technology and the biology go hand-in-hand," he said, "What we think of as the transcriptome depends entirely on how you detect it."

Evolving Dogma

Chang's group further characterized lncRNAs to discover that they were modular: Much like different subunits on proteins, these RNAs have different domains, some that can recruit enzymes and others that interact directly with chromatin. They found that HOTAIR, for example, serves as a molecular scaffold for histone modification complexes and can regulate the availability of DNA to transcriptional machinery (5). "We can understand a lncRNA by breaking down the different components and looking at what each piece does," he says.

Chang points out that this work has helped evolve the central dogma of biology, which is that information encoded in DNA flows downhill: first to RNA and then into protein. "The central dogma that we all grew up learning is more complicated," he said. "RNA has a way to talk back to the DNA, not by changing it directly, but by changing its accessibility via the chromatin structure."

In 2013, Chang, with his Stanford colleague William Greenleaf, applied this knowledge to develop a method to rapidly sequence accessible DNA in cells (6). The assay for transposase-accessible chromatin using sequencing, or ATAC-seq, relies on the prokaryotic enzyme transposase, which can copy-and-paste parts of the genome, but only at certain sites and only if they are available for transcription. ATAC-seq revealed the interplay between open chromatin, epigenetic modifications, and DNA regulatory complexes.

Another benefit of ATAC-seq, says Chang, was that it required many fewer cells than traditional DNA-sequencing methods. He said that "We went from needing nearly 10 million cells to at most a few tens of thousands of cells." In 2015, the group extended ATAC-seq to single-cell resolution (7).

"It's great when investigators tell me how they've used ATAC-seq," Chang said. "I've heard from plant and evolutionary biologists, among many others. I'm delighted that it has had such a profound impact."

His group has also used the technique to look at genome-wide chromatin accessibility in 23 of the most common human cancers as cataloged by The Cancer Genome Atlas (8). The study identified DNA regulatory elements, noncoding regions, tied to genetic loci associated with cancer.

For his wide-ranging contributions, in 2018 Chang received the National Academy of Sciences Award in Molecular Biology, joining his mentors Patrick Brown (2000) and David Baltimore (1974) in the honor.

Writing the Genome

In his Inaugural Article (1), Chang's group pushed sequencing technologies further, taking advantage of the last decade's computational revolution. BABEL is a deep-learning method that allows researchers with one set of genetic information, single-cell ATAC-seq data, to translate it into another, single-cell RNA-sequencing data, and vice versa.

"You can translate across chromatin, RNA, and protein," said Chang. "This allows you to build multiomic profiles, very rich datasets, from a single cell." With just the single-cell ATAC-seq information, Chang's group was able to build a single-cell RNA profile from a patient-derived basal cell carcinoma sample, despite the program having never seen basal cell carcinoma data.

Chang says artificial intelligence and machine learning are already advancing genomic research, and that he expects that in silico methods will help narrow down the range of physical experiments scientists must perform. He believes this will allow a transition to occur, in which scientists will not only be reading genetic information but regularly writing that information into cells as well.

"This idea—changing genomic sequences for therapeutic benefits—is already moving into clinics. It's an exciting reality, but there are also a lot of challenges to be met. How do you actually write something meaningful? What edits and changes to the genome do you want to make?" Change says. He envisions a future in which RNA regulation can be used to tackle some of the most challenging diseases, such as cancer.

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