# The Centrality of RNA, Then and Now

## *RNA. Life's Indispensable Molecule*

Author: James Darnell
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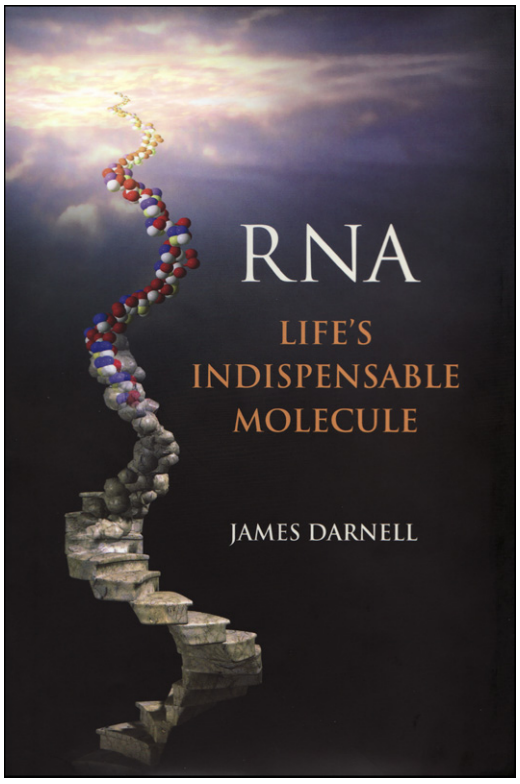
416 pp. $39.

Today, the critical roles of RNA in information transfer are taken for granted. Most graduate students can recite the basic rubrics of molecular biology; DNA is transcribed to make pre-mRNA, which is then extensively processed in the nucleus to yield mRNA. The mRNA is then recruited to ribosomes, where with the obligatory aid of decoding tRNA, it is translated 5′ to 3′ to make proteins, amino to carboxyl. Remarkably, peptide bond formation is catalyzed by ribosomal RNA itself.

Although these "facts" are well known, it is only the very rare student or even faculty member who can describe how we came to learn these fundamentals. Ask how mRNA was discovered, how the genetic code was deciphered, or how the directionality of protein synthesis was established, and one is more than likely to encounter blank stares. Fortunately, there is now a straightforward and enjoyable way to transform those stares into cogent answers by reading James Darnell's book on the history of RNA research.

Darnell tells the story of the early days of the molecular biology of RNA through the eyes of a prolific scientist who lived through those exciting times while making seminal contributions himself. The narrative begins in an era when the concept of macromolecules of any kind was met with stubborn and today unthinkable resistance. Nevertheless, these misguided ideas were dispelled in a series of breakthroughs such as the crystallization of urease and pepsin, the sequencing of insulin, and the elucidation of secondary structural elements in proteins such as the alpha helix. The early history of genetics in peas, the fly, and bacteria is also described, as are the famous experiments that showed quite convincingly that DNA was the genetic material.

After the elucidation of the double-helical structure of DNA, researchers turned toward trying to understand how the information in DNA was interpreted to make protein. Darnell takes the reader down the paths that lead to the discovery of mRNA. Looking back, it is remarkable how much was leaned from "simple" approaches such as base composition analysis of labeled RNA (with [$^{32}$P] orthophosphate) and gradient ultracentrifugation, techniques that provided the first hints of mRNA in bacteriophage-infected bacteria. These experiments presaged an avalanche of studies that showed among other things that ribosomes are the sites of protein synthesis and demonstrated the existence of transfer RNAs (then called soluble RNAs). However, the most important work of the time was the analysis by Jacob and Monod of lactose metabolism in bacteria. Their discovery of the *lac* operon and elucidation of its regulatory circuitry were profoundly influential at the time and still drive our perceptions of gene control today. Darnell describes their work in some detail, but unfortunately, some overzealous (or underzealous) proofreader changed the famous PaJaMo (Pardee, Jacob, Monod) experiment to the PaJaMa experiment, perhaps to bestow a catchier moniker.




The most striking aspect of this period of intense discovery is that the field was driven by the work of a remarkably small group of investigators dominated by the genius of Jacob, Monod, Crick, and Brenner. Moreover, in the days before e-mail and FedEx, the level of communication between the groups involved appears to be amazingly efficient. The confluence of shared ideas and productive collaborations certainly drove the field forward. It makes one long for those "simpler" days when there were not thousands of journals to source and hundreds of meetings to attend where only published work is presented.

Following the discovery of mRNA, interest turned toward deciphering the genetic code. Darnell lucidly takes the reader through the thinking at the time and describes the famous "n = 3 experiment," a clever genetic analysis proving that nonoverlapping triplets of nucleotides were the code words. He then details how Ochoa and Nirenberg broke the code, each using different experimental approaches. With the solving of this fundamental problem the book shifts gears and departs the prokaryotic world to enter the maze of eukaryotic RNA metabolism. It is worth noting that the story up to this point closely parallels the content of another engaging book, *The Eighth Day of Creation* by Horace Freeland Judson, an eminently readable and highly detailed account of the early days of molecular biology told from the perspective of an historian of science but not a scientist himself.

As Darnell delves into eukaryotic RNA metabolism, the tone of the book changes and becomes more personal, undoubtedly because Darnell himself was intimately involved in this area of research. He recounts the development and importance of tissue culture cells and animal and plant viral propagation without which the work would have been impossible. The reader is led through the discovery of ribosomal RNA processing and tRNA processing and learns that it was Darnell himself that coined the term ''RNA processing.'' Nevertheless, the most engaging part of this portion of the book is the description of the mystery of heterogenous (in length) nuclear RNA, hnRNA. Pulse labeling of HeLa cells revealed the presence of hnRNA, and base composition analysis showed that it was distinct from ribosomal RNA. Subsequent to the discovery of the 5′ methylated cap and serendipitous discovery of 3′ polyadenylate tails (interesting stories in their own right), it was shown that hnRNA possessed both. However, much of the label in hnRNA never left the nucleus. Although labeling strategies suggested a precursor product relationship between hnRNA and mRNA, it remained unclear how hnRNA was reduced in size. A number of clever and technically difficult approaches such as UV transcription mapping successively narrowed down the possibilities, and Darnell describes how tantalizingly close his group was to discovering splicing. However, this was not to be as the Roberts and Sharp groups proved using electron microscopy that removal of intervening sequences (introns) occurred in viral pre-mRNAs. Soon after, it became clear that splicing caused the reduction in size of hnRNA to mRNA. Following the discovery of splicing, the (at the time) astounding discovery of catalytic RNA is briefly described, and the attention is turned toward transcription and its regulation in animal cells.

To RNA biologists, the detailed account of the discovery of eukaryotic RNA polymerases and the subsequent description of transcription factors and their roles in controlling gene expression seem like a distraction because, perhaps unfortunately, transcription and RNA processing have evolved into quite distinct fields. The overwhelming complexity of each precludes an in-depth discussion of them both. Nevertheless, the large portion of the book devoted to transcription as well as chromatin and epigenetic histone marks serves as an excellent introduction to these topics for students and faculty alike. One minor criticism is that transcription and its control could be seen to be discussed at the expense of other interesting topics. For example, eukaryotic mRNA turnover is presented in a little over a page, and mRNA translational control is not even mentioned. This aside, to his credit Darnell does cover nearly every aspect of modern RNA biology, and the referencing is remarkably current. Although one may have preferred more detailed treatments of small noncoding RNAs such as microRNA, piwi-interacting RNAs, and small-interfering RNAs, the references provide ready access to anyone who wants to explore these topics in more depth.

The final chapter of the book provides a nice summary of the RNA World hypothesis. In a nutshell, a number of lines of evidence suggest that RNA was ''the'' molecule (because of its catalytic and informational capacities) essential for the origin of life. Perhaps this explains the title of the book, which is to say that RNA was indispensible for life, as we know it, to have begun. In this view, RNase P, the splicesome, the ribosome, and telomerase, among others, are relics of an ancestral ''life form'' that preceded proteins and DNA. It is clear that the RNA World hypothesis will continue to stimulate new ideas and experimental analyses.

In sum, Darnell has succeeded in writing an appealing and cogent account of the rise of RNA molecular biology and its continued centrality in research today. This is an excellent book that should be required reading for graduate students and more senior investigators alike. The emphasis on hypotheses-driven experimental analysis and inclusion of informative figures, many showing primary data, are a significant plus as is the up-to-date referencing. Although quite technical in parts, the concepts are explained in clear enough terms that any reader of *Cell* should be able to understand and appreciate them.

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