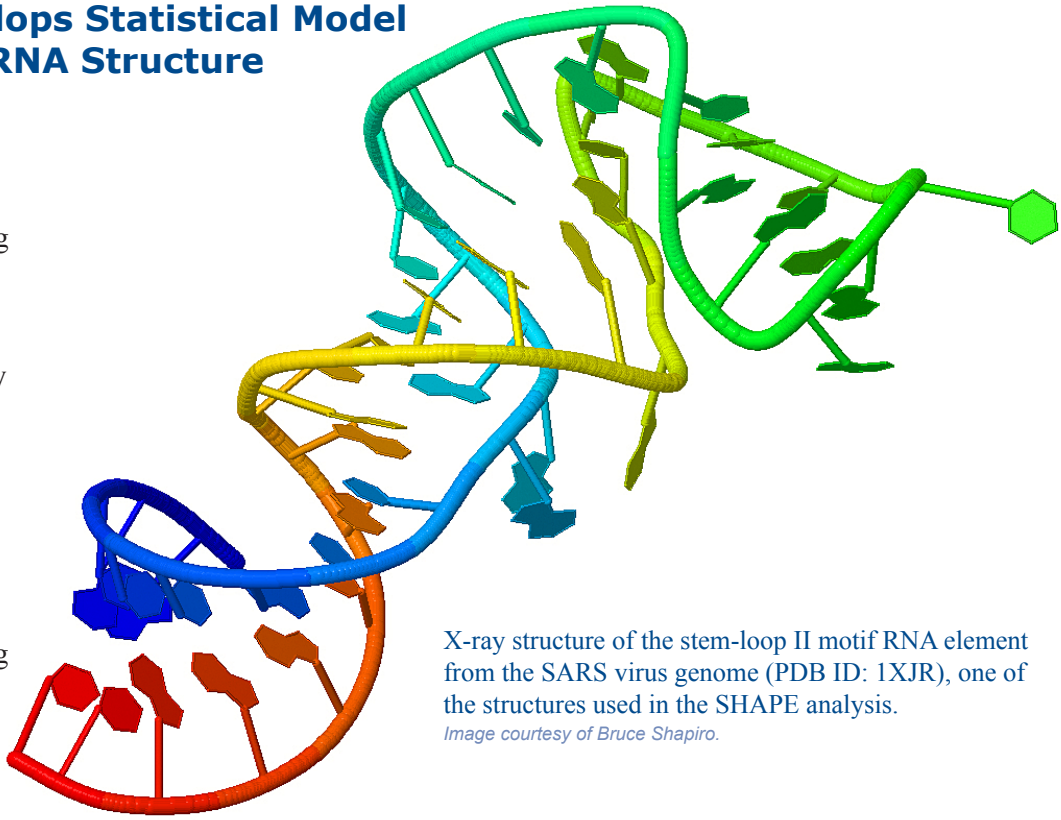
Shapiro's Group Develops Statistical Model for Analyzing RNA Structure

By Nancy Parrish, Staff Writer

RNA is critical to gene regulation and gene expression, or the transformation of DNA into proteins, which are the building blocks of most life forms. It is the RNA that translates the codes from DNA into the codes for proteins, and thus it plays a critical role in the development of healthy organisms, and, conversely, of disease.

However, it has been difficult to determine the structure, or makeup, of RNA, according to Bruce Shapiro, Ph.D., head of the RNA Computational Structure Group, Center for Cancer Research Nanobiology Program (CCRNP). He believes that understanding more about RNA's structure will lead to a better understanding of its function, and, ultimately, to the development of therapeutics to treat diseases like cancer.



X-ray structure of the stem-loop II motif RNA element from the SARS virus genome (PDB ID: 1XJR), one of the structures used in the SHAPE analysis.
Image courtesy of Bruce Shapiro.

Taking Existing Technology a Step Further

In conjunction with Eckart Bindewald, Ph.D., Basic Science Program, SAIC-Frederick, and the CCR Retroviral Replication Laboratory's RT Biochemistry Section, headed by Stuart Le Grice, Ph.D., Shapiro recently used an existing technology to more accurately predict the structure of RNA. The technology, known as selective 2´-hydroxyl acylation analyzed by primer extension, or SHAPE, was developed several years ago to determine RNA structure.

SHAPE methodology identifies only the existence of base pairing, but to date has been unable to provide further structure details, such as the base-pairing partner or the type of base pair, according to Shapiro. His team selected seven RNAs from the Protein Data Bank, whose structure was already known. They prepared RNAs by in vitro transcription and then let the molecules fold into their native structure.

After chemically modifying the RNAs, the researchers subjected the sequences to primer extension. "We then obtained bands on gels that indicated the positions of the stops in the primer extension caused by the chemically modified bases," Shapiro explained. By converting the intensities of the bands into SHAPE values, the group was able to reach several conclusions.

"First, the SHAPE values are impacted to a large extent by the base-pairing state of a residue. This means that the higher the SHAPE value for a given base, the less the likelihood that the base is involved in a base pair," Shapiro said. More importantly, he said, they found "significant correlations with base-pair stacking and a lack of correlations with other base interaction types. By comparing the known structures with the SHAPE data, we developed a method that converts the raw SHAPE values into probabilities of base pairing, potentially enabling RNA structure to be predicted with greater accuracy."

Understanding RNA May Lead to Better Treatments

Shapiro said his group plans to continue working on methods, both computational and experimental, that can help elucidate the structure and function of RNA. Determining the characteristics of these molecules will further the understanding of disease processes and the development of agents (including nanoparticles), some of which may themselves be constructed from RNA, to combat these diseases.

You may read about Shapiro's research in *RNA* (Bindewald et al., Correlating SHAPE signatures with three-dimensional RNA structures, 17:1688–1696, 2011). It was also highlighted in *Genetic Engineering and Biotech News* (31:18, October 15, 2011). In addition, Shapiro's method is available online at http://knetfold.abcc.ncifcrf.gov/rnashape. ■