2002 PROGRAM

8:30 a.m. **Registration and Continental Breakfast Atrium (Room 1-65), William T. Young Library**

9:00 a.m. **Welcome by Dr. Lee T. Todd, Jr., President, University of Kentucky - Auditorium (Room 1-62), William T. Young Library**

9:05 a.m. **Introductory Remarks - Dr. Stephen M. Testa, University of Kentucky**

9:10 a.m. **Dr. Alan M. Lambowitz, University of Texas at Austin**
"Group II Intron Mobility by Reverse Splicing into DNA and its Potential Applications in Targeted Gene Disruption and DNA Insertion"

Group II introns are both catalytic RNAs ("ribozymes") and mobile genetic elements. Mobility occurs by a novel and highly efficient mechanism in which the intron RNA uses its ribozyme activity to insert directly into one strand of a double-stranded DNA target site by reverse splicing. The inserted intron RNA is then reverse transcribed by an intron-encoded reverse transcriptase, and the resulting cDNA is integrated into genomic DNA by cellular recombination or repair mechanisms. Because the DNA target site is recognized in part by base pairing of the intron RNA, it is possible to retarget group II introns to insert efficiently into virtually any desired target DNA. The retargeted group II introns can be used for highly efficient chromosomal gene disruption in *E. coli* and other bacteria at frequencies ranging from 0.1-45% without selection. In addition, the introns can be used to introduce targeted chromosomal breaks, which can be repaired by transformation with an homologous DNA fragment, enabling the introduction of point mutations. We are attempting to develop similar approaches for targeted gene disruption and site-specific DNA insertion in eukaryotic organisms.

10:30 a.m. **Poster Session, Room 137, Chemistry-Physics Building**

11:50 p.m. **Buffet Lunch, Faculty Club [Please return registration card by April 17, 2002 for reservations.**

2:00 p.m. **Dr. Ronald R. Breaker, Yale University**
"Exploring Ancient and Modern Biochemistry using RNA Engineering"

Recent findings indicate that RNA and DNA have a far greater potential for catalytic function than what is manifest in extant organisms. Numerous new ribozymes have been created using a combinatorial selection strategy termed "*in vitro* evolution". This approach exploits the Darwinian process of molecular "survival-of-the-fittest" wherein new functional molecules are isolated from a population of trillions of different sequence variants. For example, new ribozymes have been created that catalyze many of the chemical transformations that are essential for life, such as RNA polymerization, nucleotide synthesis, aminoacylation, and peptide bond formation. These findings provide compelling support for the "RNA world" theory, which holds that life passed through an evolutionary stage wherein all metabolic functions were guided by RNA enzymes. If true, then we can expect that an incredible array of RNA enzymes have been lost during the unforgiving process of evolution. Our laboratory has embarked on broad-based program to study the functional properties of nucleic acids. We have recently identified a new class of "self-cleaving" ribozymes (the "x motif") that has a catalytic potential for RNA destruction that is predicted to equal the power of natural protein enzymes. We have used similar ribozymes to create a prototype "RNA biochip" that can be used as a universal biosensor platform for the rapid examination of genomic, proteomics and metabolomics parameters. Finally, our laboratory has created new enzymes that are made of DNA, including a series of deoxyribozyme that catalyze the reactions of DNA cloning. The most significant advances made by our laboratory in these areas will be summarized in an effort to address the depth of this untapped potential for nucleic acid function.

3:10 p.m. **Break**

3:30 p.m. **Dr. Stephen J. Lippard, Massachusetts Institute of Technology**
"Cisplatin: From DNA Damage to Curing Cancer"

cis-Diamminedichloroplatinum(II), or cisplatin, is in widespread use for the treatment of genitourinary, head and neck, and a variety of other cancers. Cisplatin is activated in cells by aquation to form cationic species that bind to DNA. The major adducts are 1,2-intrastrand cross-links, also formed by carboplatin, which has fewer dose-limiting side effects. The structural distortion in DNA that accompanies platination affects replication and transcription. Details of the structure have been revealed by X-ray crystallographic investigations. HMG-domain and TATA-binding proteins bind with specificity to the major cisplatin-DNA adducts, forming a stable platinum-DNA-protein ternary complex. The structure of one such complex has also been determined crystallographically. Binding of HMG-domain proteins to the major cisplatin-DNA adducts shields them from nucleotide excision repair. The adducts block RNA polymerase, stimulating ubiquitination of the large subunit. HMG1 levels in human cancer cells bearing the estrogen or progesterone receptor (ER or PR) are elevated following hormone treatment, increasing sensitivity towards cisplatin. These findings form the basis for a clinical trial that is currently in progress.

4:40 p.m. **Meet with Speakers, Auditorium (Room 1-62), William T. Young Library**

(http://www.chem.uky.edu/seminars/naff/welcome.html)

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Twenty-Eighth Annual Symposium on

Chemistry & Molecular Biology

established in the memory of
Anna S. Naff

Nucleic Acids

SPEAKERS

Ronald R. Breaker
Alan M. Lambowitz
Stephen J. Lippard

Friday, April 26, 2002

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