

Friday November 15, 2013

2:00 p.m.

Second Biochemistry Faculty Candidate

Room 156, Science Complex

Novel Mechanisms of Inhibition of DNA Methyltransferase 1

DNA stores genetic information that provides the sequence for protein synthesis. However, more information than just protein sequence is needed – genes must be expressed at the right time and place. Epigenetic mechanisms, heritable changes in genome function that do not alter the underlying DNA sequence, are one way cells can modify gene expression. In humans, a common epigenetic mechanism of gene control is site-specific methylation of cytosine nucleotides in DNA. DNA methylation, which represses gene expression, is essential for normal development and differentiation. A family of proteins known as DNA methyltransferases (DNMTs) are responsible for establishing and maintaining methylation patterns. Changes to the normal methylation pattern are associated with the initiation and progression of many types of cancer, making DNMTs potential cancer drug targets. Understanding the function, regulation and inhibition of these enzymes will aid in our understanding of their biological function as well as their role in carcinogenesis.

Human DNA methyltransferase 1 (DNMT1), the most abundant DNMT, is a very large protein. At one end of DNMT1 is a domain that performs the actual enzyme chemistry of methylating DNA, but several additional domains precede this catalytic domain. Recently I discovered that one of these domains actually binds tightly to the catalytic domain at the place where DNA binds and thus is acting as an autoinhibitory domain. Removing the domain by genetic engineering speeds the methylation of DNA by more than 600-fold. The ability to make an uninhibited DNMT1 opens new ways to study the enzyme, and, importantly, gives us a tool to find new molecules that can inhibit DNMT1. A small molecule that inhibits the enzyme could potentially be developed into a drug that treats cancer. Using semi-automated methods (a so-called high-throughput approach), I tested 2320 compounds and found 9 molecules that inhibit DNMT1. I studied one of these, laccaic acid A (LCA), in detail. LCA binds tightly at the same site used for DNA-binding and seems to have a preference for binding to DNMT1 instead of other methyltransferases. Importantly, LCA treatment increases expression of genes that are normally silenced by methylation in breast cancer cells cultured in the laboratory. Future work will focus on understanding the reaction mechanism of DNMT1 in detail, how the various domains regulate catalysis, and how small-molecule inhibitors work. Collectively, this work will further our understanding of the regulation of DNMT1 enzymatic activity and further define novel small molecule scaffolds for inhibiting DNMT1.