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Researchers Distinguish New Type of Leukemia

Researchers who have studied the activity of thousands of genes in a drug-resistant form of childhood leukemia are now proposing that the disease be called mixed-lineage leukemia (MLL) because it is a distinct disease, and not a subtype of the more prevalent acute lymphoblastic leukemia (ALL).

The research team, which was led by Howard Hughes Medical Institute investigator Stanley J. Korsmeyer and colleague Todd Golub, both at the Dana-Farber Cancer Institute at Harvard Medical School, published its findings in the December 3, 2001, issue of *Nature Genetics*.

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— Stanley J. Korsmeyer

According to Korsmeyer, researchers had known that infants with a form of ALL characterized by a specific chromosome break and rearrangement on chromosome 11, called a translocation, suffered severe relapses after chemotherapy. Researchers had discovered that the translocation involved a gene that they called the mixed lineage leukemia gene, or *MLL*.

Korsmeyer, Golub and their colleagues theorized that the MLL translocation might cause aberrations in metabolic pathways that would indicate that the drug-resistant form of leukemia they were studying was genetically different from ALL, and thus a distinct form of leukemia. They decided to use DNA microarrays to test their hypothesis by comparing the expression of genes in the lymphocytes of children with classic ALL versus lymphocytes from children with the chromosome 11 translocation.

DNA microarrays, popularly known as gene chips, are large collections of genes that are arrayed on a postage-stamp-sized chip. To study gene activity in cells, researchers extract collections of RNA from cells and apply those collections to the microarray. By measuring the level of fluorescence of markers attached to the RNA, the researchers are able to determine the level

of gene activity, or expression, of each gene.

In their studies, the scientists compared the gene expression profiles of MLL and ALL cells using a commercial DNA microarray containing more than 12,000 genes. They discovered that about 1,000 genes were under-expressed in MLL compared to ALL, and about 200 genes were expressed at higher levels.

The researchers concluded that the gene expression profiles “show that ALLs possessing a rearranged MLL have a highly uniform and distinct pattern that clearly distinguishes them from conventional ALL or acute myelogenous leukemia and warrant designation as the distinct leukemia, MLL.”

The researchers also found clues about the origin of MLL from the identity of genes that were underexpressed or highly expressed. The underexpressed genes included many important for early development of blood cells. And the overexpressed genes included members of a family known as *HOX* genes, some of which are regulated by the *MLL* gene.

“When we look at these patterns of gene expression and also at the cells of origin of MLL, we see a pattern indicating that they are very early lymphoid progenitor cells,” said Korsmeyer. “This suggests that MLL is caused by arrested maturation of lymphocytes. Once we saw that these cells were nothing like those in ALL, we understood why these children don’t respond well at all to standard chemotherapy for ALL,” he said.

When the scientists compared the genes whose expression is most characteristic of ALL, MLL and AML, they found patterns distinctive enough to be used to distinguish the three leukemias. According to Korsmeyer, this study appears to represent the first time that a whole-genome profile has revealed that a chromosome translocation can switch on a specific gene expression program.

“A central question with respect to these chromosomal translocations is whether they represent simply an oncogenic cancer ‘hit’ that will be followed by additional mutations which dictate whether the cancer becomes a conventional ALL or immature infant leukemia,” he said. “Or, is this translocation really the first dictating event, from which the rest of the leukemic process flows? Our results support the latter mechanism.

“These findings suggest that as we explore more of these cancers, we will discover meaningful prognostic subsets based on gene expression profiles,” said Korsmeyer.

The studies also uncovered promising drug targets that may improve treatment of MLL, said Korsmeyer. Specifically, he cited as an example a gene called *FLT3*, whose increased activity most clearly distinguished MLL from ALL or AML. The *FLT3* gene encodes an enzyme that is a cellular switch called a tyrosine kinase, a type of enzyme that is already targeted by drugs that are in development. The serendipitous discovery of the distinctive expression of *FLT3* activity and of the other MLL-related genes emphasizes

the value of large-scale gene expression profiling, said Korsmeyer.

“The beauty of the gene chip is that, much to our surprise, we could deal from the genomic equivalent of a whole deck of cards and come up with such a distinctive hand,” he said. “We couldn’t have imagined that amidst this vast amount of data, we could not only clearly distinguish MLL, but come up with FLT3 as a testable drug target for treating the disease.”