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Supercharging Blood-Forming Stem Cells

Researchers studying the colorfully named zebrafish mutant, *mind bomb*, have discovered a way to replenish blood cells more quickly after exposure to radiation. The studies identify key genetic regulators that boost production of blood-forming stem cells.

The finding could lead to ways to supercharge production of hematopoietic stem cells (HSCs) in cancer patients who have received bone marrow transplants to restore their blood-forming system after chemotherapy or radiation. Supercharging could also enhance effectiveness of such transplantations to treat disorders such as aplastic and sickle-cell anemias, said the researchers.

Leonard I. Zon, a Howard Hughes Medical Institute investigator at Children's Hospital Boston, and his colleagues reported their findings in an article published in the October 2005 issue of the journal *Genes and Development*. Caroline Erter Burns, Elizabeth Mayhall, and Jennifer Shepard, who are members of Zon's laboratory, and David Traver of the University of California, San Diego, were co-authors of the article.

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The researchers chose the zebrafish for their studies because it is an easily manipulable animal model that has a blood-forming system that shares many similarities with that of mammals.

Zon and his colleagues were interested in exploring the possibility that a regulatory gene called *Notch* plays a role in governing HSC production. "The

regulatory program for self-renewal of stem cells is a black box -- very little is known about what controls the ability of a stem cell to replicate," said Zon. Experiments by other researchers hinted that *Notch*, which is known to regulate a wide range of cellular processes, might also play a role in the self-renewal of HSCs. *Notch* produces a transcription factor, a protein that controls the activity of a wide range of genes.

In earlier studies, researchers had permanently activated *Notch* to determine the function of the Notch protein. Researchers could not use the same strategy to determine *Notch's* role in stem cell regulation, however, because permanent activation of the gene has broad effects on cell differentiation.

Zon and his colleagues circumvented that problem by taking an entirely different approach. Beginning with a zebrafish mutant called *mind bomb*, which lacks *Notch* signaling. Among other abnormalities, the mutant fails to produce HSCs when it matures into an adult. But the *mind bomb* mutant was just a starting point. By using some clever genetics, the researchers utilized a new-and-improved technique, in which Notch signaling was activated only when adult fish were exposed to a brief pulse of heat. By employing this technique on adult fish, they avoided the consequences that would result from activating Notch earlier in development. When the researchers gave the fish a pulse of heat, they found that the fish began to produce more HSCs.

The researchers traced the *Notch* regulatory pathway even further, finding evidence that *Notch* controls another gene called *runx1*, which, in turn, regulates HSC production. They then tested whether activating the *Notch-Runx* pathway could restore HSC production in fish whose stem cell production had been damaged by radiation exposure.

"It was, in essence, a clinical trial on the fish, in which we sub-lethally irradiated them, then added heat to activate *Notch*," said Zon. "We found that the blood counts recovered far more quickly in the fish. This suggests that if we had a pharmaceutical compound to activate *Notch* transiently, it could restore the blood system more quickly in patients who are given stem cell transplantation." According to Zon, transient activation of *Notch* would be desirable, because long-term activation may trigger development of lymphoma.

In a new set of studies, Zon and his colleagues are tracing the *Notch* regulatory pathway for HSC formation. They are also using genetic and chemical approaches to explore the machinery of HSC production in the aorta of the fish, and plan to search for compounds that can activate those stem cells directly.