

MAY 18, 2007

Kidney Cancer Shuts Down Protein Destruction Complex

New evidence shows that Wilms tumor - a rare kidney cancer that affects children - promotes its own growth agenda by taking over a genetic program used by normal cells during development. The studies demonstrate that Wilms tumor exploits the same signaling pathway that is targeted by colorectal cancer cells, but it goes about hijacking that pathway in an entirely different way. The finding suggests that drugs targeting this pathway may be effective against a variety of cancers.

Misguided Wnt signaling is now understood to be the culprit in a variety of cancers and other diseases. Normally, Wnt works closely with an intracellular foot soldier called β -catenin, to trigger changes within cells. In the absence of Wnt, β -catenin is actively degraded by a multi-protein destruction complex. When a cell detects Wnt that has been secreted from a nearby neighbor, however, that destruction complex is deactivated, and β -catenin is allowed to accumulate. As its level rises, it gets to work activating the expression of Wnt-target genes.

"One can pursue drugs and therapies blindly, and hope that something works, or one can use the insights into biology from the past 100 years to make educated guesses about how biology and diseases work, and thus perhaps make more effective drugs, and do so more quickly."

— **Randall T. Moon**

Howard Hughes Medical Institute investigators have now shown that efficient function of the β -catenin destruction complex depends on a factor that is often mutated in kidney cancer cells. Using multiple experimental systems, Howard Hughes Medical Institute investigator Randall Moon and colleagues report in the May 18, 2007, issue of *Science* that a protein called WTX is a necessary component of the army of destruction proteins that target β -catenin when Wnt signaling is off.

The study demonstrates how a perfectly normal form of cell-to-cell communication can be misappropriated. There are a relatively small number of molecules that allow cells to talk to one another, said Moon, who is at the University of Washington. Wnts are among these molecules, and the pathways they trigger are also at the heart of many cancers, retinal diseases, osteoporosis, and likely some neurodegenerative diseases.

WTX protein was previously shown to be mutated in a significant number of Wilms tumor samples. However, the connection between WTX and Wnt signaling in those tumors was not recognized until lead author Ben Major, a postdoctoral fellow in the Moon lab, began his investigations.

Our studies began over three years ago and preceded identification of WTX mutations in Wilms cancer, said Major. We had no idea at the outset that this protein was so intimately associated with cancer.

Using proteomic methods, which can determine whether proteins in a cell touch each other, Major first found that WTX physically interacts not only with β -catenin but with another factor in the protein-destruction complex — the one that marks β -catenin for destruction by tagging it with a small protein called ubiquitin in a process known as ubiquitination. Most ubiquitinated proteins are automatically targeted for degradation in a cell.

The investigators then investigated WTX activity both in the test tube and in living organisms. For the first set of experiments they showed biochemically that that WTX partners with other destruction-complex proteins to promote β -catenin ubiquitination and degradation. For the second they used a classical manipulation of frog embryos to test the effects of Wnts in the presence and absence of WTX.

Because Wnts regulate development of anterior structures in vertebrates, frog embryos injected with the *Wnt* gene make too much Wnt protein and develop two heads. However, when investigators spiked the *Wnt* injection with the gene for WTX, the resulting tadpoles developed milder head anomalies, confirming that WTX antagonizes Wnt signaling. The researchers saw similar results with experiments using zebrafish embryos.

The investigators are now focusing on learning how WTX does this biochemically. We are performing experiments designed to elucidate the mechanism by which WTX promotes β -catenin ubiquitination, said Major, adding, We are also looking for WTX mutations in cancers outside of the kidney, such as in lung and skin.

Noting the increasing association of aberrant Wnt activity with cancer, Moon said, This study shows that mutations in the Wnt signaling pathway can be directly linked to yet another cancer. This suggests that as drug companies work on drugs to block this pathway, such drugs will be useful in more types of cancer than originally thought — drugs under development for treating colon cancer, which involves activation of β -catenin signaling, may be equally effective in treating Wilms tumors and other cancers.

Moon credits the success to an outstanding postdoc with an advisor smart enough to stay out of his way and to the community of developmental biologists who have spent years defining how Wnt signaling works in organisms from sponges to humans. One can pursue drugs and therapies blindly, and hope that something works, or one can use the insights into biology from the past 100 years to make educated guesses about how biology and diseases work, and thus perhaps make more effective drugs, and do so more quickly, he said.