The Hedgehog's spine

By Lev Osherovich, Senior Writer

The primary cilium, a lone spiky protrusion on the cell surface, once was thought to be a functionless vestige of humankind's unicellular past. However, genetic defects in the primary cilium turned out to be linked to polycystic kidney disease and certain birth defects. Now, new studies from two independent teams at the **University of California, San Francisco**, implicate the primary cilium in a well-known cancer pathway—Hedgehog signaling.^{1,2}

In addition to providing a rationale for targeting the primary cilium in cancer, the findings could help companies identify patients who may respond well to Hedgehog (Hh) signaling inhibitors.

The Hh pathway normally shapes the growth of tissue during embryonic development. Later on, however, inappropriate activation of the pathway can cause basal cell carcinoma (BCC) and medulloblastoma, a childhood brain cancer. These tumors, as well as others, often result from inactivating mutations in Patched 1 (PTCH1), an inhibitory cell-surface receptor in the pathway, or through activating mutations in Smoothened homolog (SMO), a receptor downstream of PTCH1.³

Several companies, including **Roche's Genentech Inc.** unit, **Infinity Pharmaceuticals Inc.** and **Bristol-Myers Squibb Co.**, have SMO inhibitors in development for various cancer indications. **Fate Therapeutics Inc.** is pursuing an undisclosed target in the Hh pathway in a preclinical program.

Because PTCH1 and SMO reside in the primary cilium as part of normal embryonic development, the UCSF teams asked whether the primary cilium played a role in cancer.

One team was led by Jeremy Reiter, assistant professor of biochemistry. The other team was led by Arturo Alvarez-Buylla, professor of neurological surgery.

Reiter's team tested the primary cilium's role in tumors by knocking out kinesin family member 3A (KIF3A), a cytoskeletal motor protein needed for cilium development, in mice with a tumor-promoting mutation in SMO. Indeed, the absence of a cilium disrupted SMO signaling and Kif3a knockouts had lower tumor growth rates than controls with intact cilia.

Meanwhile, Alvarez-Buylla's team also used Kif3a knockouts to test the role of cilia in medulloblastomas and obtained results similar to those of Reiter's team.

Alvarez-Buylla's team then used gene expression profiling to show that ciliated tumors had higher levels of Hh pathway activity than non-ciliated tumors.

Both groups also looked at the distribution of cilia in human pathology samples. Reiter's team found that 5 of 8 BCC tumors had cilia, whereas Alvarez-Buylla's team found that 9 of 24 medulloblastomas had cilia.

Have cilium, will travel

Altogether, the findings suggest that tumors driven by mutations in SMO or PTCH1 require an intact cilium for growth. Conversely, the studies imply that tumors lacking cilia grow independently of SMO and PTCH1.

Thus, even though tumors can grow in either case, the data suggest that tumors may have different growth mechanisms depending on whether or not they have cilia. As a result, treatment strategies should be different in the presence or absence of cilia.

Reiter thinks the results suggest that tumors lacking cilia, which appear to be a minority of BCCs but a majority of medulloblastomas, may be highly aggressive and nonresponsive to SMO inhibitors.

"The presence or absence of cilia might be an important biomarker for diagnosis" of tumors, he said. "Having this biomarker might direct what kinds of therapies to use for these tumors."

Alvarez-Buylla agreed. "We would predict that in patients without primary cilia, inhibiting

Smoothened shouldn't be very effective," he told *SciBX*.

Frederic de Sauvage, VP of molecular biology at Genentech, said the findings are consistent with earlier studies of the primary cilium in embryonic development, in which the structure coordinates Hh-dependent growth cues.

"In the absence of the primary cilium, you would predict that Hedgehog tumors would not develop," said de Sauvage.

Genentech and partner **Curis Inc.** have the most advanced SMO inhibitor, GDC-0449, in Phase II trials for solid tumors. Other SMO inhibitors in Phase I for solid tumors include IPI-926 from Infinity, and BMS-833923 (XL139) from BMS and **Exelixis Inc.**

"In order for the pathway to be active, these tumors need to have primary cilia and therefore you could use this as a diagnostic tool," said de Sauvage.

However, de Sauvage did note that Hh involvement in tumor growth can already be assessed using gene expression profiling.

Reiter countered that genetic tests are relatively expensive but pathologists routinely diagnose and grade tumors by cellular characteristics. Thus, adding a visual test for cilia to the battery of standard lab analyses would be relatively simple.

Getting cilia

From a therapeutic standpoint, the studies suggest that disrupting the primary cilium could be beneficial in SMO- or PTCH1-driven tumors. According to Reiter, KIF3A is an enzyme that is "presumably targetable," but better targets may become apparent through screens for

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genes involved in cilium development.

His next step is to test whether the presence or absence of primary cilia plays a role in the progression of other types of tumors. Alvarez-Buylla plans to examine the involvement of cilia in the Wnt pathway, another tumor-promoting signaling pathway.

de Sauvage said the therapeutic prospects for blocking cilia are limited by the current lack of pharmacological inhibitors of ciliogenesis and potential concerns about off-target effects.

Reiter and Alvarez-Buylla did not patent their discoveries. Alvarez-Buylla said he explored patenting the use of cilia as a diagnostic but that UCSF's technology transfer office "wanted us to submit over 1,000 samples before they could decide about whether to patent this."

Finding that many patients with medulloblastoma is a formidable challenge, Alvarez-Buylla noted.

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COMPANIES AND INSTITUTIONS MENTIONED

Bristol-Myers Squibb Co. (NYSE:BMJ), New York, N.Y.
Exelixis Inc. (NASDAQ:EXEL), South San Francisco, Calif.
Curis Inc. (NASDAQ:CRIS), Cambridge, Mass.
Fate Therapeutics Inc., La Jolla, Calif.
Genentech Inc., South San Francisco, Calif.
Infinity Pharmaceuticals Inc. (NASDAQ:INF), Cambridge, Mass.
Roche (SIX:ROG; OTCQX:RHHBY), Basel, Switzerland
University of California, San Francisco, Calif.