Malignant peripheral nerve sheath tumors (MPNST) are incurable tumors of the Schwann cell lineage that occur sporadically or in the context of the disease Neurofibromatosis type 1 (NF1). In NF1 patients, MPNSTs arise from malignant transformation of existing benign plexiform neurofibromas (PNF); however, predicting which PNFs will undergo transformation has been very difficult, and MPNSTs have a poor prognosis by the time they are identified. Because NF1 patients carry a mutation in the NF1 gene encoding neurofibromin that inhibits RAS signaling, most of the clinical trials aimed at treating MPNSTs have focused on inhibitors of the receptor tyrosine kinase (RTK)/RAS/MEK/AKT/mTOR signaling pathway to reduce RAS signaling at different levels in the pathway. Unfortunately, none of these trials have yet shown strong effects against MPNSTs (1, 2), suggesting that new targets or combination approaches are desperately needed for MPNST.

In this issue, Watson and colleagues (3) take an unbiased approach to identifying pathways critical for MPNST tumorigenesis. Using their Sleeping Beauty (SB) model, they have driven insertional mutagenesis events in the Schwann cell lineage using the promoter for CNP, a gene involved in myelination. The SB system uses a mutagenic transposable element to tag insertions/disruptions that promote cancer in a particular cell type, allowing for rapid identification of cancer genes. Pathway analysis of the identified gene insertions showed a significant enrichment in WNT signaling pathway genes. The use of pathway analysis is critical in the identification of important targets, as multiple mutations can feed into the same pathway and may be targeted by common inhibitors. In addition, targeting single points in a pathway can lead to unexpected results through interfering with negative feedback loops, as has been seen in the case of mTOR inhibitors (4). Therefore, understanding all of the steps in the pathways that can contribute to MPNST may help to design combination therapies that reduce resistance mechanisms.

The canonical WNT signaling pathway is regulated at many levels (5), and there is increasing evidence from other systems for crosstalk between WNT signaling and other pathways important in MPNST tumorigenesis (6–8), converging on β-catenin (Fig. 1). β-catenin is a multifunctional protein with distinct molecular roles in cell adhesion at the plasma membrane and in transcription within the nucleus. Its function within the cell is regulated by its localization and post-translational modifications. In the absence of a WNT signal, β-catenin’s activity is blocked by localization in the cytoplasm in the destruction complex with AXIN, APC, CK1, and GSK3β, where it can be phosphorylated and subsequently degraded. Tankyrase (TNKS) can destabilize the destruction complex through effects on AXIN. The small-molecule inhibitors IWR-1 and XAV-939 stabilize AXIN through inhibition of TNKS and promote β-catenin cytoplasmic localization and degradation. Upon WNT signaling to FZD at the membrane, LRP and FZD associate and AXIN/GSK3β/APC/CK1 are localized to the LRP/FZD complex. β-catenin is released and localizes to the nucleus, where it interacts with LEF1 and CBP to activate progrowth and survival transcriptional programs that include MYC, CCND1, and BIRC5. R-spondin 2 (RSPO2) promotes WNT signaling, possibly through interactions with LRP and FZD.

Watson and colleagues (3) identify Ctnnb1 (encoding β-catenin), Tnks, Crebbp (encoding CBP), Gsk3β, and Axin1 as common insertions found in the SB model of MPNST. In addition, they present additional data from a gene expression microarray analysis of human MPNSTs that members of the destruction complex, APC, CK1 subunits, and GSK3β are underepressed in MPNST cells and primary tumor samples, whereas RSPO2/3 and FZD1/7/10 are overexpressed. This expression data correlates with canonical WNT signaling playing an oncogenic role in MPNSTs, with the destruction complex playing a tumor suppressive role. Watson and colleagues confirm that β-catenin is localized to the nucleus with increasing grades of Schwann cell tumors and
that downstream transcriptional targets become activated in human tumor samples. Through a variety of gene manipulations at different points in the WNT signaling pathway, they show that the pathway is necessary for full transformation of MPNST cells and robust growth in vivo, but it may not be sufficient for transformation of normal Schwann cells. As the authors point out, cooperation with other mutations, such as in the \( NF1/RAS \) pathway, are likely to be important for MPNST transformation.

Even if activation of \( \beta\text{-catenin} \) by the canonical WNT pathway is not sufficient to drive transformation, it can still provide a very valuable marker for MPNST progression, particularly from benign PNFs. In a comparison of benign neurofibromas, PNFs, and MPNSTs, the authors show that higher-grade tumors are more likely to show strong positivity for \( \beta\text{-catenin} \) with more nuclear localization. Nuclear localization of \( \beta\text{-catenin} \) has been suggested as a score for aggressiveness or grade in other tumor types (9, 10), and further studies are needed to determine whether \( \beta\text{-catenin} \) staining will have any clinical value in managing patients with MPNST. Because a subset of PNFs show nuclear \( \beta\text{-catenin} \), it would be particularly important to determine whether these PNFs are more likely to transform to MPNSTs. This could potentially be used in a wide range of cancers for which WNT signaling plays a role.

It becoming evident that treating MPNSTs with molecularly targeted monotherapies is unlikely to be successful. EGFR was one of the first molecular targets identified for treatment of MPNSTs (6), resulting in a clinical trial for MPNSTs with the EGFR inhibitor erlotinib (clinicaltrials.gov; study NCT00068367). To date, this trial has not led to improved treatment for patients with MPNST. Cross-talk between the WNT signaling pathway and other signaling pathways such as EGFR is now well established (7, 8), and recent data suggests that inhibition of both EGFR and \( \beta\text{-catenin} \) may show some synergy for inhibition of brain cancer (11). In addition, \( \beta\text{-catenin} \) can be stabilized by inhibition of GSK3\(\beta\) by AKT, downstream of CXCR4 (8). Activation of AKT downstream of RTKs such as EGFR may similarly inhibit GSK3\(\beta\) and stabilize \( \beta\text{-catenin} \) (Fig. 1). ERK signaling downstream of EGFR/RAS may also affect activity of \( \beta\text{-catenin} \), and this signaling pathway could be potentiated by loss of neurofibromin. The complicated crosstalk between many distinct signals at the plasma membrane and oncogenic effects on transcriptional programs in the nucleus are still difficult to predict. Unraveling these
relationships will be critical for designing rational combination therapies; however, β-catenin seems to be a key node in many of these pathways. Interestingly, Watson and colleagues (3) found specific synergy between the TNKS inhibitors IWR-1 and XAV-939 and the mTORC1 inhibitor RAD-001 (Fig. 1), suggesting that the β-catenin and mTORC1 pathways may be able to at least partially compensate for each other in the growth of MPNST cells. The work of Watson and colleagues (3), as well as the recent study by Mo and colleagues (8), lays the groundwork for future investigation of combination therapies for MPNST treatment. In both of these studies, novel targets for inhibition of canonical WNT signaling and β-catenin activation are shown.

Because MPNSTs are ultimately rare tumors, the possible combinations of therapeutic approaches far exceed the ability to test combinations in the patient population. One of the greatest challenges facing scientists and clinicians in the development of MPNST clinical trials is in establishing criteria for rational combination therapies. Unbiased screens, such as described by Watson and colleagues (3), pinpoint causal pathways in MPNST tumorigenesis. A detailed understanding of how these signaling networks interact will lead to more rational therapy design.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

REFERENCES

Extending the Convergence of Canonical WNT Signaling and Classic Cancer Pathways for Treatment of Malignant Peripheral Nerve Sheath Tumors
Karlyne M. Reilly

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