

Telomeres

Major finding: A specialized replisome mediates break-induced telomere synthesis to promote telomere maintenance.

Concept: Alternative lengthening of telomeres requires a break-induced replisome lacking canonical components.

Impact: Alternative lengthening of telomeres may be targetable in cancer by blocking the specialized replisome.

ALTERNATIVE LENGTHENING OF TELOMERES REQUIRES A SPECIALIZED REPLISOME

In approximately 10% to 15% of cancers, telomeres are maintained by homologous recombination-dependent alternative lengthening of telomeres (ALT) instead of telomerase upregulation. However, the mechanism by which DNA damage triggers telomere elongation in mammalian cells is not well understood. Dilley and colleagues developed methods to isolate and quantify nascent telomeres after the generation of telomere double strand breaks (DSB). These approaches revealed that telomere specific DSBs induced unidirectional synthesis of long tracts of telomeric DNA in a process they termed break-induced telomere synthesis. Break-induced telomere synthesis occurred by an alternative DNA replication complex, which lacked many of the canonical replisome components. Break-induced telomere synthesis was also independent of DNA damage responsive kinases ATR and ATM, as well as the homologous recombination protein RAD51. Break-induced telomere synthesis required the Pol δ replicative DNA polymerase for synthesis of both the C- and G-rich strands and the Pol δ accessory subunit POLD3. ALT telomere synthesis was independent of several



other polymerases including Pol ϵ , Pol α , Pol η , and Pol ζ , and the canonical replisome associated helicase complex MCM2-7. Altogether, these findings indicate that a minimal Pol δ containing replisome is responsible for break-induced telomere synthesis. Mechanistically, proliferating cell nuclear antigen (PCNA) was loaded by replication factor C (RFC) at damaged telomeres and interacted with POLD3 to recruit the Pol δ complex to ALT telomeres, indicating that RFC-PCNA functions as a telomere damage sensor. Consistent with these findings, POLD3 was essential for Pol δ complex stabilization, and depletion of POLD3 reduced ALT telomere synthesis. Collectively, these data indicate that a noncanonical replisome is involved in break-induced telomere synthesis at ALT telomeres, thereby differentiating ALT from S-phase replication and suggesting the potential for targeting ALT in cancer. ■

Dilley RL, Verma P, Cho NW, Winters HD, Wondisford AR, Greenberg RA. Break-induced telomere synthesis underlies alternative telomere maintenance. Nature 2016;539:54–8.

Breast Cancer

Major finding: Targeting PIM1 induces apoptosis, reduces MYC activity, and up-regulates p27 to suppress TNBC growth.

Clinical relevance: High levels of PIM1 are associated with a poor prognosis in patients with TNBC.

Impact: PIM kinase inhibitors warrant further investigation for the treatment of TNBCs with high levels of MYC.

PIM1 MAY BE A THERAPEUTIC TARGET IN TRIPLE-NEGATIVE BREAST CANCER

No targeted therapies are available to treat triple-negative breast cancer (TNBC). These tumors often display genomic amplification of *MYC*, *MCL1*, and the 6p21-25 genomic region that includes *PIM1*. Brasó-Maristany and colleagues analyzed multiple published TNBC datasets and found that *PIM1* copy-number gains resulted in *PIM1* overexpression in TNBCs compared with non-TNBCs. The majority of TNBC cell lines were dependent on PIM1 expression for survival and proliferation, and depletion of PIM1 induced cell death through the mitochondrial apoptotic pathway by regulating BCL2 expression. However, ectopic overexpression of the antiapoptotic BCL2 only partially rescued cell growth suggesting that PIM1 also promotes TNBC through additional mechanisms. Indeed, PIM1 depletion reduced c-MYC phosphorylation and expression of MYC target genes including *MCL1*, demonstrating that PIM1 activates MYC signaling. Inhibiting PIM1 with the pan-PIM kinase inhibitor AZD1208 reduced the survival of TNBC cells and suppressed tumor growth *in vivo*. Moreover, AZD1208 enhanced the efficacy of chemotherapy in TNBC xenografts and patient-derived xenografts (PDX) expressing high levels of PIM1. Similarly, Horiuchi and colleagues identified PIM1 as a potential druggable target in MYC-overexpressing TNBC cells via an shRNA screen. PIM1 expression was associated

with expression of a MYC-dependent transcriptional signature and poor prognosis in patients with TNBC. TNBC PDXs were sensitive to the pan-PIM kinase inhibitors SGI-1776 and NVP-LGB321, which reduced tumor growth and cellular proliferation and enhanced apoptosis. However, xenografts expressing low levels of MYC were insensitive to PIM inhibition, indicating that MYC-driven TNBCs are specifically dependent on PIM kinase activity. Mechanistically, PIM1 inhibitors suppressed TNBC growth by reducing MYC transcriptional activity and increasing expression of the cyclin-dependent kinase inhibitor p27. Taken together, these studies indicate that MYC-overexpressing TNBCs are dependent on PIM1 expression and provide a rationale for clinical investigation of PIM inhibitors for the treatment of patients with TNBC. ■

Brasó-Maristany F, Filosto S, Catchpole S, Marlow R, Quist J, Francesch-Domenech E, et al. PIM1 kinase regulates cell death, tumor growth and chemotherapy response in triple-negative breast cancer. Nat Med 2016;22:1303–13.

Horiuchi D, Camarda R, Zhou AY, Yau C, Momcilovic O, Balakrishnan S, et al. PIM1 kinase inhibition as a targeted therapy against triple-negative breast tumors with elevated MYC expression. Nat Med 2016;22:1321–9.

CANCER DISCOVERY

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