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Two strikes against CLL

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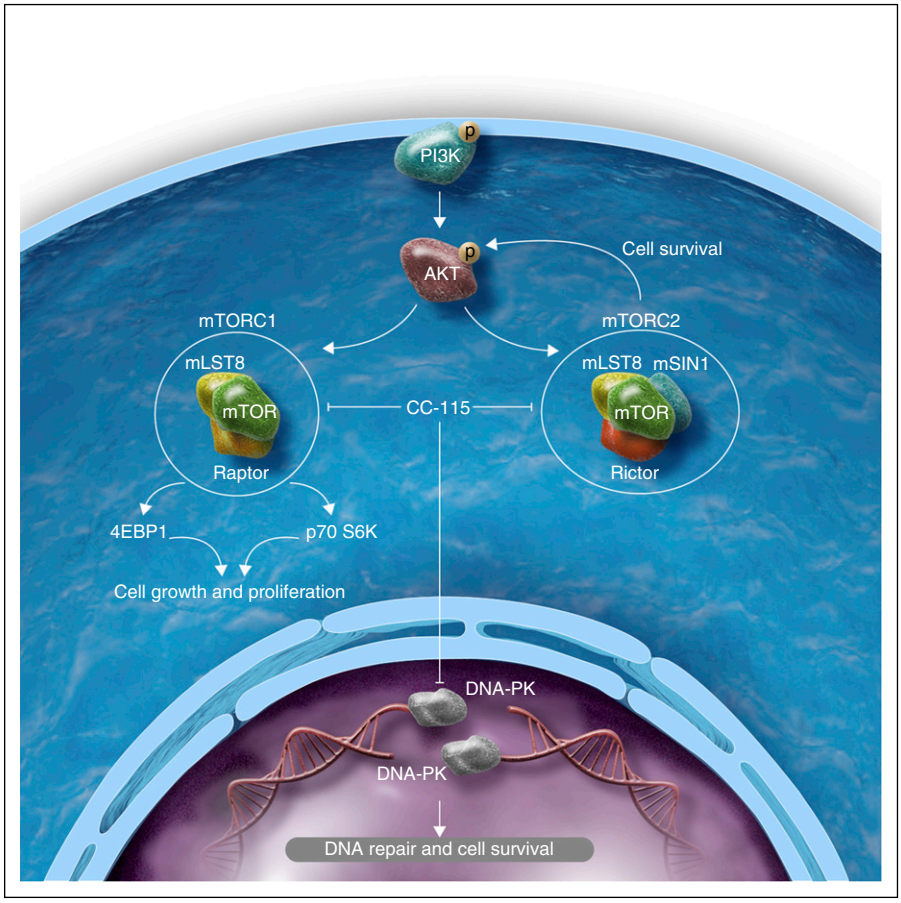
In this issue of *Blood*, Thijssen et al provide evidence that CC-115, a potent, selective, and orally bioavailable inhibitor of DNA-dependent protein kinase (DNA-PK), and the mammalian target of rapamycin kinase (mTORK) may produce the effects outlined below as a single agent.[1]

B-cell receptor (BCR) pathway inhibitors such as ibrutinib and idelalisib show impressive activity in chronic lymphocytic leukemia (CLL)[2] and have revolutionized CLL therapy. However, these drugs have limited cytotoxicity against tumor cells, meaning long-term continuous treatment, lower complete response rates, and lack of disease eradication. Strategies that simultaneously cause microenvironmental egress and direct tumor cell death are more likely to completely eliminate CLL, and this approach is now being tested using a variety of therapeutic combinations.



Model for CC-115 mechanism of action: schematic representing proposed mechanism by which CC-115 induces cytotoxicity in CLL tumor cells. mTOR is a highly conserved serine/threonine kinase downstream of PI3K/AKT that forms multimolecular complexes with key functions in cell growth, proliferation, and survival. The mTOR kinase can complex with Raptor (regulatory-associated protein of mTOR) to form mTORC1, or with Rictor (rapamycin-insensitive companion of mTOR) to form mTORC2. Once phosphorylated by the upstream PI3K/AKT kinases, mTORC1 activates its downstream effectors (eg, 4EBP1, p70S6K1) to stimulate protein synthesis and cell cycle entry. mTORC2 acts upstream of mTORC1 by inducing phosphorylation of AKT on serine 473, which creates a positive feedback loop that contributes to cell survival. The survival signal mediated by mTORC2 is thought to account for the resistance to mTORC1 inhibitors (ie, rapamycin and its analogs). In contrast, CC-115 inhibits both mTORC1 and mTORC2, and leads to growth arrest and apoptosis. Furthermore, CC-115 inhibits NHEJ DNA repair by preventing the recruitment of DNA-PK to DSB. This causes persistence of DNA damage and cell death. Professional illustration by Somersault18:24.

CLL disease progression and chemoresistance result from microenvironment-derived survival signals, as well as acquired aberrations in the pathway being targeted and/or DNA damage repair pathways.[3,4] DNA-PK is a kinase activated upon DNA damage and is involved in repairing DNA double-strand breaks (DSB) through the DNA nonhomologous end-joining pathway (NHEJ),[5] which is thought to be the main pathway for DBS repair in CLL. A direct correlation in CLL patients between DNA-PK activity and clinical resistance to chemo- and radiotherapies has been reported.[6] CLL patients with high-risk cytogenetics (ie, 17p or 11q deletion) are resistant to chemotherapy due to overexpression of DNA-PK.[7] In these cells, pharmacologic inhibition of DNA-PK restores sensitivity to DNA damaging therapies.[7] To date, the promising results obtained in vitro with DNA-PK inhibitors has not been translated to the clinic due to lack of specificity or poor pharmacokinetic properties of available agents.

Signaling via the BCR stimulates growth and survival of CLL cells, and inhibits apoptosis by activating PI3 kinase/protein kinase B (AKT), mTOR, extracellular signal-regulated kinase (ERK), and other pathways. The mTOR complexes mTORC1 and mTORC2 are the main downstream kinases of the phosphatidylinositol 3 kinase (PI3K)/AKT pathway, and contribute to proliferation and antiapoptotic signaling in lymphoid malignancies including CLL. In particular, the Raptor-containing complex mTORC1 mediates cell growth and proliferation by activating p70 S6 kinase, and the eukaryotic initiation factor 4E binding protein 1 (4EBP1), thereby regulating translation of proteins critical for progression from G_1 into S phase (eg, cyclin D1 and c-myc). The Rictor-containing complex mTORC2 phosphorylates AKT on Ser473, resulting in AKT/ERK activation and prosurvival through a positive feedback loop.[8] Evidence indicates that mTORC2 overexpression is required in CLL cells for AKT phosphorylation, activation, and downstream signaling.[9]

Rapamycin analogs, which are allosteric mTORC1 inhibitors, have shown clinical activity in CLL and other lymphoid malignancies, but responses are usually partial

and transient due to the mTORC2-induced compensatory feedback loop affecting AKT.[8]

On the other hand, specific inhibition of mTORC2 is more active in vitro in inhibiting the downstream phosphorylation of AKT Ser473 and inducing apoptosis of CLL cells.[9]

This suggests that simultaneous inhibition of mTORC1/2 could effectively block CLL cell proliferation and survival. Indeed, there has been substantial interest in improving the anticancer activity of mTOR-targeting agents. Toward this end, several non-rapamycin-based mTOR inhibitors targeting both mTORC1 and mTORC2 have been developed (eg, OSI-027) that show enhanced preclinical antilymphoma activity.[8] A recent first-in-human trial evaluating tolerability, pharmacokinetics, and pharmacodynamics of OSI-027 in patients with advanced solid malignancies shows that this agent inhibits mTORC1/2 in a dose-dependent manner; however, doses above the tolerable levels are required for a continuous target engagement in tumor cells.[10] Further clinical development of this compound as a single agent has been discontinued, emphasizing the continued need for mTORC1/2 inhibitors with improved tolerability.[10]

Here, Thijssen et al take the concept of dual mTORC1/2 inhibition one step further by assessing the effect of simultaneously targeting mTORC1/2 and DNA-PK in CLL tumor cells with CC-115. They hypothesize that concurrent inhibition of mTORC1/2 will provide a substantial break in cell proliferation and survival, whereas inhibition of DNA-PK will overcome microenvironment-induced protection and chemotherapy resistance. The authors tested the potency of CC-115 in primary CLL samples of different prognostic subgroups with respect to induction of cytotoxicity, as well as inhibition of CD40-mediated chemoresistance and proliferation. Their data clearly show that CC-115 inhibits mTORC1/2 and also affects the DNA damage repair pathway (as evidenced by inhibition of radiation-induced γH2AX), in both ataxia telangiectasia mutated (ATM) and wild-type CLL cells, with simultaneous sensitization to chemotherapeutic agents such as chlorambucil and bendamustine. As expected, in contrast to the mTORC1-only inhibitor CC-214 that blocks proliferation with minimal cytotoxicity, CC-115 induces caspase-dependent cell death irrespective of p53 and ATM status while sparing normal B and T cells. Furthermore, CD40-mediated chemoresistance is completely reversed by CC-115 through upregulation of Bim and downregulation of antiapoptotic proteins (ie, BclXl, Mcl-1, and Bfl-1). Interestingly, the CC-115 induced cytotoxicity, and reduction of the downstream BCR and CD40L signaling was also observed in CLL cells derived from idelalisib-resistant patients, suggesting the possibility of using this inhibitor in CLL patients with developed resistance to PI3K targeting agents. More importantly, preliminary clinical testing of CC-115 in CLL patients with ATM alterations shows a promising decrease in lymphoadenopathy in 8 out of 9 patients studied, with one partial response and three partial responses with lymphocytosis. This is particularly interesting, as in contrast to their in vitro data showing an ATM- and p53-independent mechanism of action, clinical results suggest enhanced dependency on DNA-PK for DNA damage repair in cells with biallelic alteration of ATM. This small study included only patients with mutated ATM, however, and the clinical relevance of this class of inhibitors remains to be investigated in a broader patient subset. Also, as more patients emerge with resistance to Bruton tyrosine kinase–targeting agents (eg, ibrutinib), it will be interesting to determine if CC-115 and related drugs can bypass such acquired resistance.

Overall, CC-115 is a novel and exciting molecule that acts through the inhibition of several critical pathways for CLL survival and chemoresistance (see figure). This study represents a significant step forward in the development of novel strategies for CLL that can overcome microenvironment-derived survival stimuli while directly killing tumor cells. This work warrants future in vivo studies, especially in combination with DNA damaging agents, toward the ultimate goal of disease eradication.

Conflict-of-interest disclosure: The author declares no competing financial interests. ■

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