Immunomodulatory drugs (IMiDs) and CelMod-mediated growth suppression of Adult T-Cell Lymphoma/Leukemia (ATL) cells: Functional linkage between Cereblon targets and their down-stream effectors.

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Background：
Adult T-cell lymphoma/leukemia (ATL) is extremely resistant to existing chemotherapy. Recent two clinical studies proved promising curability of Lenalidomide (LEN, a second-generation IMiDs) for ATL patients. IMiDs (immunomodulatory drugs) are known to modulate the substrate specificity of

E3–enzyme cereblon (CRBN) and lead proteasomal degradation of target proteins. We evaluated the machineries of IMiDs-mediated growth suppressive effects on ATL cells.

Methods:
MM cell-lines, NCI-H929 (LEN-sensitive) and RPMI-8226 (LEN-resistant) for control. 13 ATL-related and 3 non-ATL cell-lines for the IMiDs activity assessment. LEN, Pomalidomide (POM, third gen) and Iberdomide (IBE, Cereblon modulator: CelMod) for CRBN modulating activities and growth suppressive effects on ATL cells. Cell Titer assay, Western blot, lentivirus RT-PCR, Gene Knock-Down (KD) were done by standard procedures. Mice xenograft experiments were also conducted.

Results:
Among 13 ATL-related cell lines, HuT102 and TL-Om1 exhibited the best response to LEN doses. CRBN-knockdown (KD) in HuT102 lead accumulation of IKZF1/3 and LEN-resistant growth profiles. IKZF2-KD ED40515 (LEN-resistant) variants turned to LEN sensitive. Oral administration of LEN substantially reduced tumor masses of HuT102-xenograft SCID mice. Finally, we identified IBE-mediated nearly 1000-fold efficient down-regulation of CRBN targets in ATL cells.

Conclusion:
Degradation of LEN/CRBN targets IKZF1/3 and suppression of down-stream effectors IRF4 and cMyc (both have been implied to promote ATL cell malignancy) was confirmed in LEN-responsive HuT102 and TL-Om1. Since they also displayed impaired expression of IKZF2, LEN-mediated growth inhibition was seemingly attributed to functional deprivation of all IKZF1/2/3. Our hypothesis was supported by two gene knockdown experiments (LEN-resistant ED40515 with IKZF2-KD to LEN-sensitive. LEN-sensitive HuT102 with CRBN-KD to LEN-resistant). *In vivo* HuT102-xenographted SCID mice also well responded to LEN oral treatment. Finally, IBE-mediated deeper and wider range of growth suppression to ATL cells implied its potential application as a novel anti-ATL therapeutic agent.

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