Toward the Discovery of Small Molecule Inhibitors of APOBEC3 DNA Cytosine Deaminase Enzymes

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Apolipoprotein B mRNA editing enzyme, catalytic polypeptide-like 3B (A3B) catalyzes C to U deamination in ssDNA as a function of innate immune defense against foreign DNA. A3s cause hypermutation in pathogenic DNA causing genomic instability and clearance by the host immune system. However, A3B is overexpressed in various cancer types and at sublethal levels leads to evolution and progression of various cancer types. A3B is shown to be an enzymatic source of mutation in breast cancer, lead to poor clinical outcomes in cancer patients, and promote tamoxifen resistance in ER+ breast cancer. Also, A3B is shown to be nonessential in humans making it a viable target for cancer therapy.

Efforts in the Harki and Harris labs to discover inhibitors of A3 enzymes have yielded several small molecule scaffolds, many of which were shown to inhibit through covalent mechanisms. Recent proteomics studies in our lab have shown covalent adducts between known pan A3 inhibitor MN23 with Cys217 and Cys239 of A3B. A3B constructs with cysteine to alanine mutations at C217 and C239 have been expressed and are being evaluated for their deaminase activity inhibition propensity by MN23 to determine the essential cysteine for enzyme inhibition. This information may be used to rationally design a selective small molecule inhibitor of A3B.

Due to the closed conformation of the active site, a fragment-based discovery approach may also be appropriate for A3B. A new approach for fragment screening called Protein Observed Fluorine (PrOF) NMR utilizing 19F NMR has been developed where the protein itself is fluorinated and perturbations in the resonance shift of the NMR spectrum are monitored to determine ligand binding. This change in shift may be used to both quantify and determine location of binding making this a powerful tool for inhibitor discovery. This poster will highlight recent efforts to develop new A3B inhibitors and unveil the mechanism of action by known covalent inhibitors.

(1) Burns, M. B.; Temiz, N. A.; Harris, R. S. Nature Genet. 2013, 45, 977-983.