Dual inhibition of AKT and KIT is synergistic in Gastrointestinal Stromal Tumor

Abstract

Purpose: The majority of gastrointestinal stromal tumors (GIST) harbor oncogenic mutations in the receptor tyrosine kinase KIT or in the platelet-derived growth factor receptor alpha (PDGFRα). Small molecule kinase inhibitors such as imatinib mesylate (IM) have significantly improved the clinical management of GIST by targeting these mutant receptors. However, despite strong overall response rates to IM and other second-line targeted therapies, disease progression generally does occur in time. It is clear that certain mutations in KIT and PDGFRα pathways provide a resistance mechanism to IM therapy. Therefore, inhibiting targets other than, or perhaps in addition to, traditional tyrosine kinases may provide additional therapeutic benefit in GIST. Both KIT and PDGFRα activate AKT and recent studies associate PI3-kinase/AKT pathway activity with the survival of IM-resistant GIST cell lines and tumors. Experimental Design: Here, we performed experiments to assess the potential benefit of combining IM with an ArQule AKT inhibitor, either ARQ 092 or ARQ 751, in a panel of IM-sensitive (GIST-T1, GIST882) and resistant GIST cell lines (GIST-T1/829, GIST430). To evaluate in vitro drug sensitivity, cells were subjected to drug treatment for 72 hours before measuring viability with the Cell Titer Blue sensitivity, cells were subjected to drug treatment for 72 hours before measuring viability with the Cell Titer Blue Viability Assay. Synergy between IM and each AKT inhibitor was quantified using the Chou-Talalay algorithm to calculate Combination Index (CI) values. CI values <1 are considered synergistic. Results: The 3:1 molar ratio of ARQ 092:IM demonstrated synergistic CI values in all four GIST lines. Immunoblot assays confirmed that drugs hit their intended targets (phospho-KIT, phospho-AKT) in each cell line following six-hour drug treatment. Interestingly, a significant decrease in the activation of a downstream signaling protein, p-S6, was observed in the combination-treated cells compared to cells treated with single agents. Conclusion: These data provide strong rationale for further testing of these combinations in GIST xenograft models.

ARQ 092 and IM have synergistic effects on in vitro GIST cell growth

ARQ 751 and IM have synergistic effects on in vitro GIST cell growth

Combination of IM and AKT inhibitor displays synergism in in vitro GIST cell lines

Biochemical potency and selectivity of ARQ 092 and ARQ 751

Response markers of IM, ARQ 092 and ARQ 751

Conclusions and future directions

- ARQ 092 and ARQ 751 are potent and selective allosteric AKT inhibitors.
- In vitro studies demonstrated drug synergy between ARQ 092/ARQ 751 and IM in 3:1 molar ratio in a panel of IM-sensitive and -resistant GIST cell lines.
- We provide evidence for the initiation of preclinical studies evaluating the use of IM in combination with AKT inhibitors in in vivo models of GIST.


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