TKI Combination Therapy: Strategy to Enhance Dasatinib Uptake by Inhibiting Pgp- and BCRP-Mediated Efflux

R. R. D'Cunha, S. Bae, G. An
University of Iowa

Purpose
The overexpression of efflux transporters, especially P-glycoprotein (Pgp, ABCB1) and Breast Cancer Resistance Protein (BCRP, ABCG2), represents an important mechanism of multidrug resistance (MDR). Tyrosine kinase inhibitors (TKIs), a novel group of target-specific anticancer drugs, have recently been found to interact with Pgp and BCRP and serve as both substrates and inhibitors. Considering this dual role of TKIs, we anticipate that combination TKI therapy may represent a promising strategy to reverse the efflux transporter mediated TKI resistance. Presently, the investigations on these interactions are very limited. To fill in the literature gap, in the current study we used dasatinib as the model drug and evaluated the effect of various other TKIs on Pgp- and BCRP-mediated dasatinib efflux.

Methods
Cell uptake studies were performed using LLC-PK1 cells and its subclone transfected with human Pgp, and MDCK-II cells and its subclone transfected with human BCRP. For the concentration-dependent studies, the study protocol was the same as the cell uptake study except that the concentrations of the tested TKIs varied from 0.5 μM- 200 μM. BCA assays were carried out to normalize dasatinib intracellular concentrations. An LCMS/MS method was developed to analyze dasatinib concentrations in all cell lysates. The mean values and their difference from the negative control were analyzed for significance using a one-way ANOVA or Student’s t-test, followed by the Holm-Sidak post-hoc test for multiple comparisons.

Results
Among the 14 TKIs screened, afatinib, axitinib, bosutinib, erlotinib, imatinib, lapatinib, nilotinib, pazopanib and sorafenib greatly inhibited Pgp-mediated dasatinib efflux at 50 μM. Further concentration dependent studies showed that imatinib, nilotinib and pazopanib were potent Pgp inhibitors with IC50 values of 2.42, 6.11 and 8.06 μM, respectively. Additionally, 50 μM of axitinib, erlotinib, imatinib, nilotinib and pazopanib greatly increased the intracellular concentration of dasatinib through BCRP inhibition. Concentration dependent studies revealed that imatinib, erlotinib, nilotinib, axitinib and pazopanib were potent BCRP inhibitors with IC50 values of 0.94, 2.23, 2.50, 6.89 and 10.4 μM, respectively.

Conclusion
Our findings point to potential combinations of TKIs that could help enhance intracellular concentrations of the targeted TKI, overcome MDR and improve TKI efficacy. Further in vivo studies are warranted to confirm the efflux transporter-mediated TKI-TKI interaction and chemosensitizing potential of the potent TKI inhibitors.