A Cell Based Screening Assay for Identifying Inhibitors of Eosinophil Proliferation

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Eosinophils are white blood cells which contribute to the pathology of asthma. Lidocaine inhibits interleukin 5 (IL-5) mediated survival and activation of human eosinophils and is a useful therapy for chronic asthma; however, lidocaine has many undesired side effects. One of the main goals of this research was to enhance the specificity and overcome the undesired effects of lidocaine. Consequently, a diversity of compounds included commercially available lidocaine analogs and some synthesized compounds designed by modification of lidocaine structure was investigated for inhibitory activity on the proliferation of TF-1 cells, an erythroleukemic cell line model for eosinophil development. TF-1 cell proliferation assays were performed at various concentrations of lidocaine and the investigated compounds to determine their values at which fifty percent growth inhibition was observed (IC-50). Among 75 investigated compounds 25 revealed more potent cell growth inhibitory activity with IC-50 ranging from 1-136 uM compared to lidocaine (IC-50 144 uM). The lowest IC-50 values were: 4-Amino-N-pyridin-3-yl-methyl-benzamide (1 uM), N-[2,4-difluorophenyl]-2-(4-pyrimidin-2-ylperazonyl) ethanamide (5uM), Boc-4-aminochippurinic Acid (19uM), 9,10-difluoro-2,3-dihydro-3-methyl-7-OXO-7H-pyrido[1,2,3-DE]-1,4-benzoxazine-6-carboxylic acid (20uM), 7-Bromo-4-methyl-3,4-dihydro-2H-1,4-benzoxazine (24uM), and N-[2,6-dimethylphenyl]quinoline-4-carboxamide (24uM). Therefore, several investigated compounds were more potent than lidocaine at inhibiting cell growth. The investigated compounds were also tested on cells grown in epoietin-α (EPO). IC-50 values that are higher on cells grown in IL-5 compared to EPO suggests drug specificity for inhibiting IL-5 mediated proliferation. N-[2,6-dimethylphenyl]thiocyanamide, 3,4-ethylenedioxyaniline, 2-methyl-N-[3-pyridinylmethyl]benzamide, and 4-Amino-N-pyridin-3-yl-methyl-benzamide revealed the highest specificity for IL-5 mediated cultures. Consequently, our cell based assay is an effective method for screening chemical compounds and has revealed potential chemicals for the treatment of asthma.