Inhibition of cytokine release from HT-29 cells and ulcerative colitis biopsies is potentiated by combination of selective kinase inhibitors and such effects are mimicked by TOP1210, a Narrow spectrum kinase inhibitor (NSKI).

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Introduction

• Intracellular kinase activation plays a key role in inflammation and kinase inhibitors have been proposed as potential therapies in chronic inflammatory disorders such as ulcerative colitis.

• Selective kinase inhibitors, however, have proved disappointing, particularly in the treatment of rheumatoid arthritis and inflammatory bowel disease (IBD).

• Multi-kinase inhibition has been investigated as a strategy to improve efficacy.

• In this study the inhibitory effects of multi-kinase inhibition by either selective kinase inhibitors in combination or TOP1210, an NSKI inhibiting p38, Src and Syk kinases, have been tested and compared to those of selective kinase inhibitors alone.

Methods

• BIRB-796, dasatinib and BAY-61-3606 were used as selective inhibitors of p38, Src and Syk respectively and TOP1210 as an exemplar NSKI.

• Inhibitory effects on recombinant p38, Src and Syk kinases was assessed in an ATP dependent ZYLETM based assay.

• Cellular assays were performed after 2hr preincubation with compound or vehicle. HT29 cells, an epithelial cell line, were stimulated with IL-1β for 24hrs prior to collection of supernatants for measurement of IL-8 concentration.

• Biopsies from inflamed ulcerative colitis patients were incubated with compound (24hr) and spontaneous cytokine release measured.

• Inflammatory cytokines in supernatants were measured by ELISA. Results are expressed as means ± s.e.m of at least 3 determinations.

Results

• Generally, a combination of selective kinase inhibitors resulted in significantly potentiated potency and more than additive inhibitory effects.

• TOP1210 potently inhibits IL-8 release from HT-29 cells with greater efficacy than the selective kinase inhibitors alone.

• TOP1210 is superior to selective kinase inhibitors alone in down-regulating pro-inflammatory cytokine release from Ulcerative Colitis biopsies. TOP1210 activity is comparable to a combination of all three selective inhibitors.

Conclusions

• Selective kinase inhibitors individually demonstrate limited efficacy and potency in HT29 and biopsy assays. In contrast, TOP1210, demonstrates potent and efficacious inhibitory activity in both HT-29 cells and UC tissues. These studies suggest that the inhibition of multiple kinases, either by a combination of selective kinase inhibitors or by the NSKI TOP1210, results in more than additive effects with increased potency and efficacy as inhibitors of inflammatory mediators. Hence, TOP1210 offers significant advantages over existing selective kinase approaches and potentially offers a much improved therapeutic benefit in IBD.