In Vitro Metabolism of CRx-119, a Novel Synergistic Combination drug candidate, in Human Liver Microsomes and Recombinant CYP450 Enzymes

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Materials & Methods

Results: (Kinetics and Chemical Inhibitions)

Results: Effect of CRx-119 on Selected CYP450s Probe Substrates

Abstract & Introduction

The inhibition constant (K<sub>I</sub>) values were calculated from IC<sub>50</sub> values, ensuring competitive inhibition, according to the following formula: K<sub>I</sub> = IC<sub>50</sub> / (1 + [S]/K<sub>m</sub>), where S is substrate concentration, IC<sub>50</sub> is the substrate concentration that produces 50% inhibition of CRx-119 or 90% inhibition of 8-OH-AMOX, 50% inhibition of 7-OH-AMOX was strongly inhibited by the CYP2D6 inhibitor, nefazodone (26.5% inhibition by nefazodone: 26.5% inhibition by CRx-119), and 50% inhibition of 7-OH-AMOX was strongly inhibited by the CYP2D6 inhibitor, nefazodone (26.5% inhibition by nefazodone: 26.5% inhibition by CRx-119).

CRx-119: Amoxapine and Prednisolone

In vitro doses studied. Interactions (DDIs) between AMOX and Prednisolone are unlikely during CRx-119 therapy. Clinical pharmacokinetic studies revealed mean K<sub>m</sub> values for inhibition of CYP450-mediated reactions. Experiments were performed using human liver microsomes (HLMs) and 4 = 1, (Y)n (m - H)O HCPXC OX H3 e = 3214 M.

Figure 4: Effect of Chemical Inhibitors and Prednisolone on AMOX (10 μM) Metabolism

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Discussion

Table 1: AMOX and Pred ARH128595 Metabolism in Recombinant Human CYP450 (CYP450)

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The summary of the in vitro data was performed using a gelatinase assay system or an enzymatic method. Inhibitory activity of 50% was observed in HLMs of AMOX (IC<sub>50</sub> = 73.6 μM; K<sub>m</sub> = 37 μM) at concentrations 100 times the expected AMOX therapeutic concentrations. Prednisolone had no effect on CRx-119 metabolism. 5 µM AMOX was not inhibited by Prednisolone and vice versa at clinically relevant concentrations. 8-OH-AMOX was strongly inhibited by the CYP1A2 inhibitor, ketoconazole (26.5% inhibition by ketoconazole: 26.5% inhibition by CRx-119). 7-OH-AMOX was strongly inhibited by the CYP2D6 inhibitor, nefazodone (26.5% inhibition by nefazodone: 26.5% inhibition by CRx-119).

AMOX and Prednisolone are metabolized by multiple enzymes in HLM and recombinant CYP450 enzymes. Three metabolites of AMOX with masses of m/z=320 were detected in HLM and dC8NA-expressed enzyme. 8-OH-AMOX and the unknown Peak 3–5 (D8-AMOX) were observed in HLM and expressed CYP45A4-Prednisolone (m/z=353) and hydroxydiphenidol (m/z=377).

Figure 4: Representative Michaelis-Menten Plots for AMOX Metabolism

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Conclusions

Figure 2: AMOX and Prednisolone Metabolism in Pooled HLM Formed by VP-450 (LC/MS)

Conclusions

Summary

Study

Conclusions

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