Regional Differences of Acetaminophen (APAP) and Naproxen (NPX) Enterotoxicity in Human Small Intestine
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Introduction

A novel in vitro experimental system of the small intestine, cryopreserved human intestinal mucosa (CHIM) is being applied in our laboratory in the evaluation of drug-induced intestinal injuries. The nonsteroidal anti-inflammatory drugs (NSAIDs) known to cause intestinal mucosal damage, acetaminophen and naproxen, were evaluated using CHIM prepared from different regions of the small intestine.

Materials & Methods

Human intestine. Human small intestines were obtained from the International Institute for the Advancement of Medicine (IMP, Exton, PA) as known interval, but not used, for transplantation.

Intestinal mucosa isolation and cryopreservation. Isolation of mucosa from human intestine was performed on ice-cold ligatures of the intestinal lumens (Li et al. 2009). The intestinal mucosal epithelia released from the intestinal lumens were partially purified by differential centrifugation, resuspended in a proprietary cryopreservation medium and cryopreserved using a single-vial, liquid-nitrogenic, 2-step method. Vials were stored in liquid-nitrogen.

Recovery of CHIM. CHIM vials were removed from liquid-nitrogen storage and thawed on ice for 15 min, both by approximately 1. The content of each cryopreserved sample was recovered and resuspended by gentle agitation. Cryopreserved Enterotoxicity Recovery Medium (CERM)™, in Vitro ADMET Laboratories, Columbia, MD, was prepared in a single vial/liquid-nitrogen method.

Preparation of CHIM from different regions of the small intestine

CHIM were isolated from 10 consecutive 12-centimeter segments, starting right after the pyloric valve.

Results

IC50 values for APAP (blue) and NPX (gold) measured in CHIM segments

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<tr>
<th>DONOR</th>
<th>CONCENTRATION (mg/mL)</th>
<th>RELATIVE VIABILITY (%)</th>
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<tbody>
<tr>
<td>A</td>
<td>B</td>
<td>C</td>
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<tr>
<td>APAP (Donor 1, CHIM6023)</td>
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<td>APAP (Donor 2, CHIM6037)</td>
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<td>APAP (Donor 3, CHIM6038)</td>
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Dose-response enterotoxicity for APAP (upper figures) and NPX (lower figures)

Summary and Conclusions

- Cryopreserved human intestinal mucosa (CHIM) were successfully isolated from ten 12-centimeter segments of the small intestine from three human donors.
- In the CHIM from all segments from the three donors, a dose dependent decrease in viability, quantified by measuring cellular ATP contents, was observed for both APAP and NPX.
- In general, NPX had higher enterotoxicity than APAP, an observation consistent with the clinical observation that NPX is more prone to cause intestinal damage than APAP.
- Apparent regional differences in response were observed that were both drug-specific and donor-specific.

Our results suggest that CHIM represents a useful in vitro experimental system for the measurement of drug-induced enterotoxicity. One important application is the identification and elimination of drug candidates with unacceptable enterotoxicity in humans.

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