CHO-Kv1.3 Cell Line

The Kv1.3 potassium channel has shown promise as a pharmaceutical target for such diseases as multiple sclerosis, obesity, and more recently, diabetes\(^1\). In order to screen large compound libraries against Kv1.3, a reliable expression system is needed.

Our CHO-Kv1.3 cell line was specifically designed to express high levels of Kv1.3 channels, and to show uniform expression over time.

Our CHO-Kv1.3 cell line:

- Stably expresses Kv1.3 potassium channels
- Validated using electrophysiology and efflux assay
- Suitable for high-throughput screening (HTS)

Electrophysiology

Electrophysiology experiments were conducted using standard patch clamp techniques. The bath solution contained (in mM) 0.90 CaCl\(_2\), 2.67 KCl, 1.47 KHPO\(_4\), 0.50 MgCl\(_2\), 138 NaCl, and 8.10 Na\(_2\)HPO\(_4\). The pipette solution contained (in mM) 140 KCl, 1 MgCl\(_2\), 1 EGTA, and 20 HEPES.

For I-V plots, cells were held at -80mV, and then stepped to a depolarizing voltage for 1s to record the peak current.

For IC\(_{50}\) experiments, a test pulse was applied until the peak current stabilized (Fig. 2).

Fig. 1. I-V response of the CHO-Kv1.3 cell line.

Fig 2. IC\(_{50}\) curve of Agitoxin using the CHO-Kv1.3 cell line as determined by patch clamp.
Rubidium Efflux Assay

Using Aurora Biomed’s Rubidium Efflux Assay protocol, basal efflux was measured at 8.5%. After a 6 minute activation time using 63mM KCl, the maximal activation-induced efflux was 49.3% resulting in about six fold window of detection (Fig. 3).

The concentration-response curves of two commonly known Kv1.3 blockers, Agitoxin and Margatoxin were determined by Rubidium Efflux Assay using the CHO-Kv1.3 cell line (Fig. 4). Table 1 is included to summarize some results and information on our Kv1.3 cell line.

Table 1. Summary of data for CHO-Kv1.3 cell line.

<table>
<thead>
<tr>
<th>Cell Line</th>
<th>Activation KCl (mM)</th>
<th>Activation period (min)</th>
<th>Published EP IC$_{50}$ for Agitoxin (nM).$^1$</th>
<th>Aurora EP IC$_{50}$ for Agitoxin (nM).</th>
<th>Aurora Flux IC$_{50}$ for Agitoxin (nM).</th>
</tr>
</thead>
<tbody>
<tr>
<td>CHO Kv1.3</td>
<td>63</td>
<td>6</td>
<td>0.2</td>
<td>0.3</td>
<td>0.655</td>
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References