All this talk about CiPA.....

Comprehensive In Vitro Proarrhythmia Assay
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The Comprehensive In Vitro Proarrhythmia Assay (CiPA) is a proposal by the US Food and Drug Administration (FDA), the Cardiac Safety Research Consortium (CSRC), the Health and Environmental Sciences Institute (HESI) and the Safety Pharmacology Society (SPS) aimed at revising the pre-clinical S7B and removing the clinical E14 guidelines.

CiPA

The S7B and E14 guidelines are based on a predictive link between hERG-block, QT interval prolongation and torsades de points (TdP) but, although QT interval prolongation is a sensitive marker for pro-arrhythmia, it is not very specific. The CiPA proposal aims at moving the evaluation of the pro-arrhythmic risk of a compound away from today’s emphasis on QT interval prolongation to an evaluation based on an electrophysiological understanding of the underlying mechanisms of pro-arrhythmia and TdP using a three component process;

i) candidate drugs are tested on an array of key cardiac ion channels. The proposed ion channels are NaV1.5 (peak and late currents), KV4.3, CaV1.2, hERG, KVLQT1/minK and Kir2.1,

ii) the data generated in these experiments are subsequently used in in silico simulations using a computational cardiomyocyte model to see if the compound yields proarrhythmic markers on a reconstructed human ventricular action potential,

iii) the results of the in silico simulations are verified by comparing them with the electrical activity of human ventricular iPS-derived cardiomyocytes.

One of the consequences of current guidelines is that its lack of specificity means fewer drug candidates in development are progressed further because of early indications of block of the hERG channel. The intention of the CiPA proposal is to increase the efficacy of the drug development process by 1) moving the evaluation of pro-arrhythmic risk to an earlier stage in the drug development process, and 2) enabling compounds with properties today considered as possibly problematic to be further developed and 3) provide a stronger scientific foundation for improved future drug labeling.

The FDA has proposed a deadline of July 15, 2015 for the new guidelines but there are still many questions regarding protocols, validation, translation and more that still awaits answers.

Cardiac Ion Channel Panel for Safety Assessment

Automated Patch Clamp technique allows recordings of single channel ion currents, which makes it suitable for drug development. The assay can be standardized between laboratories and therefore cell lines expressing cardiac ion channels can provide a robust assay for safety assessment.

<table>
<thead>
<tr>
<th>Cardiac Ion Channel Panel</th>
<th>Role</th>
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</thead>
<tbody>
<tr>
<td>NaV1.5 (peak and late currents)</td>
<td>Na+ current</td>
</tr>
<tr>
<td>KV4.3</td>
<td>Transient outward K+ current</td>
</tr>
<tr>
<td>CaV1.2</td>
<td>L-type Ca2+ current</td>
</tr>
<tr>
<td>hERG</td>
<td>Delayed rectifier K+ current</td>
</tr>
<tr>
<td>KVLQT1/minK</td>
<td>Delayed rectifier K+ current</td>
</tr>
<tr>
<td>Kir2.1</td>
<td>Inward rectifier K+ current</td>
</tr>
</tbody>
</table>

Automated Patch Clamp have made it possible to record thousands of data points per day. Critical hardware features enable high fidelity recordings and accurate pharmacological measurements; 1) GΩ seal resistance recordings to provide high quality data, 2) Voltage clamp control to obtain valid data that can be compared between laboratories, 3) Glass-coated measuring plate to avoid issues of sticky compounds, 4) complete solution exchange for accurate presentation of drug concentration.
Cardiac Ion Channel Panel for Safety Assessment

Excellent Pharmacology
To serve as an accurate predictor for a compound’s effect on an ion channel, the results from the automated patch clamp instrument must be unaffected by challenging compound properties such as a lipophilic nature and slow binding kinetics. A number of hERG binding compounds such as astemizole and cisapride provide excellent test cases to demonstrate how the QPatch handles these issues. To minimize loss of compounds through adsorption onto surfaces, users can use glass vials inserted into 96 well compound plates from which QPatch pipetting robot can pipet. Multiple additions and longer compound incubation periods enable the user to be sure that the compound is at steady-state equilibrium binding and compound addition has saturated all non-specific binding reservoirs (ie, other cells).

How QPatch can serve your needs from CiPA

[Fig. 2 a-e]: Currents recorded from cells stably expressing (a) hERG, (b) Kv1.2, (c) NaV1.5, (d) CaV1.2 and (e) Kir2.1. Panels (a) to (d) show current – voltage relationship for the respective channel. Panel (e) shows inhibition of Kir2.1 by increasing concentrations of BaCl₂.
Temperature control
Elevating the temperature of ion channel assays is an option for the QPatch. This enables the execution of assays at physiological temperatures. Since the experimental temperature is being considered as a potentially important parameter by CiPA, QPatch temperature control will enable users to comply with CiPA temperature-related guidelines, if enacted.

[Fig. 3 a-b]: Shows the effect of doing a double addition of each concentration on the measured IC\textsubscript{50} of hERG inhibition by cisapride. For each concentration, a second addition is done in order to achieve true equilibrium binding of cisapride to hERG. The open circles in the graph of the concentration response curve represent inhibition at the end of the first addition and the closed circles is the inhibition at the end of the second addition. At the end of the second addition, the measured IC\textsubscript{50} is 23nM, similar to literature stated values from manual patch clamp.

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Current clamp
Under consideration by CiPA are guidelines on the use of stem-cell derived cardiomyocytes and the comparison between \textit{in silico} calculations of compound effect on the cardiac action potential and direct measurement of action potential in cardiomyocytes. Measurement of the action potential of stem-cell derived cardiomyocytes is made possible by the addition of current clamp on the QPatch.

[Fig. 4]: The right figure shows the left shift in the measured IC\textsubscript{50} of erythromycin in inhibiting the hERG channel. The green curve shows the concentration response curve for a set of experiments executed at 34°C compared to experiments run at 22°C (beige). The IC\textsubscript{50} decreased to 204±35nM at 34°C from 1135±9nM at 22°C.

[Fig. 5]: Spontaneous action potentials in iPS derived cardiomyocytes.

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July 2013  CSRC/HESI Thinktank Meeting: Rechanneling the Current Cardiac Risk Paradigm: Arrhythmia Risk Assessment During Drug Development without the Thorough QT Study & CSRC TREAT IV Meeting
http://www.cardiac-safety.org/php53-3-or111-websitetestlink.com/think-tanks-meetings/2014-meetings/