

# Establishment of a multi-ion channel assay in accordance with ICH E14/S7B Q&As best practices

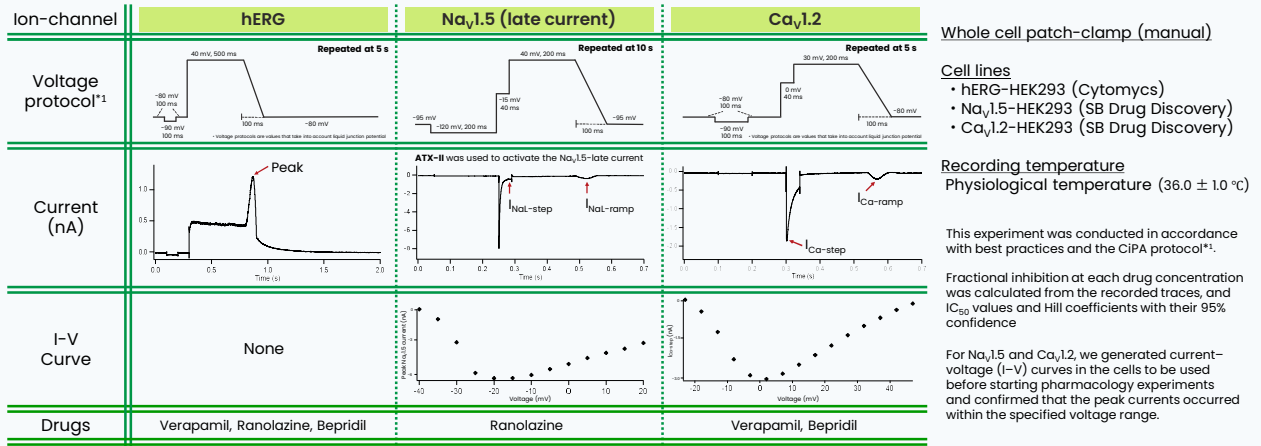
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## Background and Objective

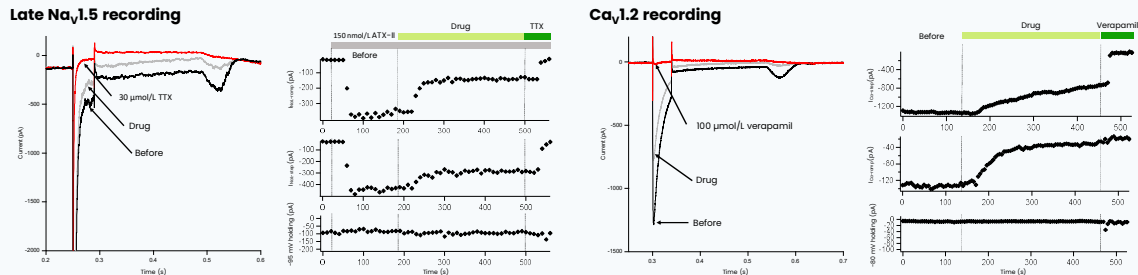
While the implementation of the hERG channel assay based on ICH-S7B has suppressed the occurrence of **torsade de pointes (TdP)** caused by new drugs in clinical settings, this approach may have unduly constrained the development of potentially promising new compounds. Accordingly, as a solution, CiPA (Comprehensive In Vitro Proarrhythmia Assay) proposes a risk assessment method that uses multiple cardiac ion channels, including **hERG, Na<sub>v</sub>1.5 (late-current), and Ca<sub>v</sub>1.2**, which affect the action potential duration of cardiomyocytes. However, test systems that collect Na<sub>v</sub>1.5 and Ca<sub>v</sub>1.2 channel data in accordance with the **CiPA protocol and the ICH-S7B E14/S7B Q&As** have yet to be widely adopted. Therefore, this study attempted to establish a test system using a multi-ion channel assay.

## Materials and Methods

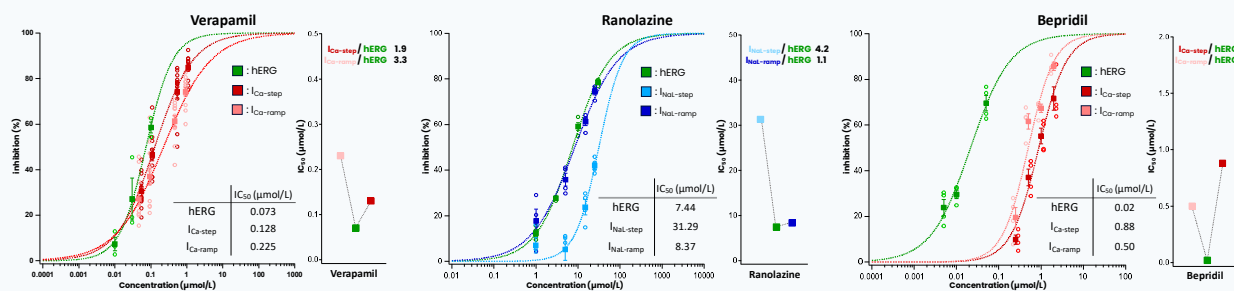


## Results

### Recording quality of manual patch-clamp



### IC<sub>50</sub> values of the 3 compounds for each ion channel



### Data comparison (MDF data set vs Reference data set)

	Verapamil				Ranolazine				Bepridil					
	TdP risk : LOW <sup>+2</sup>				TdP risk : LOW <sup>+2</sup>				TdP risk : HIGH <sup>+2</sup>					
	MDF data		Manual <sup>3)</sup>		MDF data		Manual <sup>3)</sup>		MDF data		Manual <sup>3)</sup>		HTS <sup>3)</sup>	
	IC <sub>50</sub> (μmol/L)	nH	IC <sub>50</sub> (μmol/L)	nH	IC <sub>50</sub> (μmol/L)	nH	IC <sub>50</sub> (μmol/L)	nH	IC <sub>50</sub> (μmol/L)	nH	IC <sub>50</sub> (μmol/L)	nH	IC <sub>50</sub> (μmol/L)	nH
hERG	0.07 (0.05 - 0.11)	1.2 (0.6 - 1.7)	0.5	1.1	7.44 (6.90 - 8.02)	1.0 (0.9 - 1.1)	6.5	0.8	3.4	1.1	0.02 (0.02 - 0.03)	0.9 (0.7 - 1.1)	0.2	0.9
I <sub>Ca-step</sub>	0.13 (0.11 - 0.15)	0.8 (0.7 - 0.9)	0.2 <sup>A</sup>	1.1							0.88 (0.75 - 1.05)	1.3 (1.0 - 1.7)	2.8 <sup>A</sup>	0.7
I <sub>Ca-ramp</sub>	0.23 (0.19 - 0.27)	0.7 (0.6 - 0.8)									0.50 (0.41 - 0.60)	1.4 (1.0 - 1.9)		638 <sup>B</sup>
I <sub>NaL-step</sub>					31.3 (23.83 - 41.08)	1.5 (0.8 - 2.3)								4.6
I <sub>NaL-ramp</sub>					8.37 (6.83 - 10.27)	0.8 (0.7 - 1.0)			7.9 <sup>C</sup>	1				
	Verapamil similar protocol <sup>4)</sup>				Ranolazine similar protocol <sup>5)</sup>									
	IC <sub>50</sub> (μmol/L)	nH			IC <sub>50</sub> (μmol/L)	nH								
I <sub>Ca-step</sub>	0.9	0.4			28.3	1.2								
I <sub>Ca-ramp</sub>	0.4	0.5			24.5	1.2								

A: Action potential protocol, Ba<sup>2+</sup> as charge carrier, physiological temperature  
 B: ICWG protocol 1, Ca<sup>2+</sup> as charge carrier, Ambient  
 C: ICWG protocol 2, veratridine as I<sub>NaL</sub> enhancer, physiological temperature  
 D: ICWG protocol 2, ATX-II as I<sub>NaL</sub> enhancer, Ambient

## Conclusion

The values obtained in this study were generally consistent with those reported in previous studies using the same or similar protocols.<sup>\*3 \*4 \*5</sup>  
**This demonstrates that the methodology adopted in this study can serve as a Best Practice Multi-Ion Channel Assay.**



Acknowledgement  
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## References

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- \*5: Wu M, et al. J Pharmacol Toxicol Methods. 2019 Nov-Dec;100:106608.

