PRODUCT NOTE

vCor.4U™

VENTRICULAR ENRICHED CARDIOMYOCYTES

Pharmacological Characterization Utilizing Manual Patch Clamp

Independent analysis of the **spontaneous and pacing stability** of vCor.4U™ and their **predictive response to selective pharmacological agents**

OVERVIEW

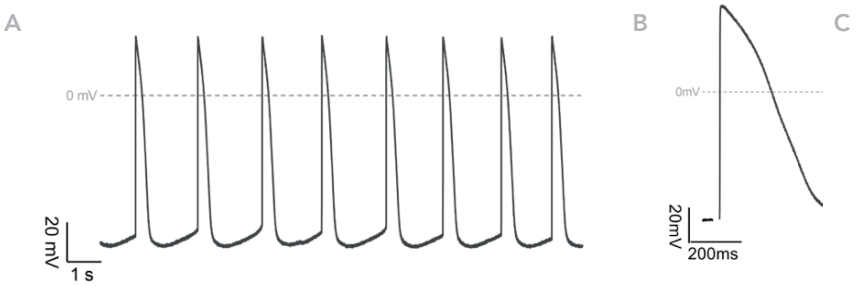
Extensive cardiac ion channel profiling at Metrion using selective pharmacological tools and manual patch clamp has enabled us to define the electrophysiological characteristics of vCor.4U™ human iPSC-derived cardiomyocytes (iPSC-CM).

vCor.4U™ elicit spontaneous action potentials (AP) and can be paced at a range of frequencies (0.2 - 1 Hz; Figure 1). The electrophysiological characteristics of evoked AP resemble primary human cardiomyocytes, with the cells having an average resting membrane potential of -73 mV and action potential duration at 90 % of repolarization (APD90) of 426 ms when paced at 1 Hz (Figure 1C).

vCor.4U™ AP are stable over time in both spontaneous and evoked recording conditions confirming the suitability of vCor.4U™ for compound screening experiments (Figure 2). Additionally, we confirm that vCor.4U™ express a predominantly ventricular phenotype and exhibit the appropriate pharmacology in response to core cardiac channel modulators, including early after depolarizations (EADs) following I_{Kr} inhibition (Figure 3).

In the light of the incoming CiPA guidelines, Axiogenesis vCor.4U™ iPSC-CM represent a suitable model for *in vitro* pre-clinical cardiac safety evaluation of compounds to identify potential human proarrhythmic liabilities.

vCOR.4U™ HAVE ELECTROPHYSIOLOGICAL CHARACTERISTICS THAT RESEMBLE PRIMARY HUMAN CARDIOMYOCYTES



AP parameter	Spontaneous	Evoked
RMP (mV)	-72.0 ± 0.3	-73.3 ± 0.6
Vmax (V/s)	56.8 ± 3.7	50.1 ± 4.8
APA (mV)	106.5 ± 0.8	119.9 ± 1.7
APD20 (ms)	133.1 ± 3.8	99.3 ± 3.0
APD50 (ms)	249.4 ± 4.9	211.6 ± 3.8
APD90 (ms)	502 ± 16.1	426.8 ± 10.9
Frequency (Hz)	0.43 ± 0.01	1

Figure 1: Characteristics of spontaneous and evoked vCor.4U™ action potentials
Representative traces of spontaneous **(A)** and evoked **(B)** AP (1 Hz) recorded from vCor.4U™ iPSC-CM under control conditions. **(C)** Average parameters for spontaneous (n = 89) and evoked (n = 75) AP in control conditions. RMP, resting membrane potential; V_{max}, maximum upstroke velocity; APA, action potential amplitude; APD20-90, action potential duration at 20, 50 or 90 % repolarization.

vCOR.4U™ ADVANTAGES

- Predictive and physiological cell model; applicable for drug development and preclinical research (e.g., ventricular hypertrophy)
- Established assays for cardiac contractile force measurements, impedance measurements, calcium transients analysis and metabolic liability
- Ideal for bioengineering including disease and tissue modeling

vCor.4U™

Human Induced Pluripotent Stem Cell-Derived Ventricular Cardiomyocytes

AP STABILITY ENABLES vCOR.4U™ FOR PHARMACOLOGICAL STUDIES

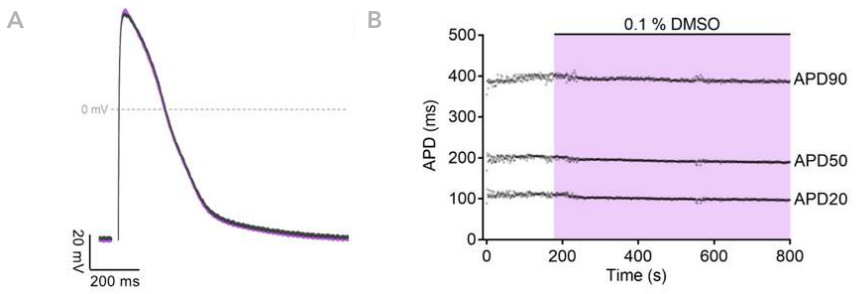


Figure 2: vCor.4U™ elicit stable action potentials
(A) Representative trace showing evoked AP stability following 0.1 % DMSO application (10 min). **(B)** Action potential duration stability of evoked AP over time (APD vs. time) during the control period and following application of 0.1 % DMSO.

vCOR.4U™ ARE SUITABLE TO ASSESS COMPOUNDS' PRO-ARRHYTHMIC RISK

A

Compound	Target	Expected human effect	Response
Lidocaine	I_{Na}	↓ Vmax, ↓ BR	✓
Nifedipine	$I_{Ca,L}$	↓ APDs	✓
Dofetilide	I_{Kr}	↑ APD90, ↓ BR	✓ EADs
Carbachol	I_{KACh}	↓ RMP, ↓ APDs, ↓ BR	X

B

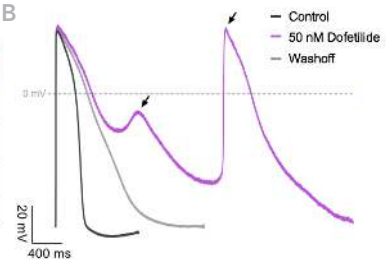


Figure 3: Pharmacological sensitivity of vCor.4U™ to cardiac channel modulators
(A) Table summarizing the response of vCor.4U™ to key cardiac channel modulators. Carbachol was used to determine if the atrial current I_{KACh} could be detected. No response was observed indicating a ventricular phenotype. **(B)** Application of dofetilide, an I_{Kr} inhibitor with a high Torsade de Point (TdP) risk, elicited EADs (arrows) from spontaneous AP, suggesting that vCor.4U™ are a suitable model to assess the pro-arrhythmic risk of compounds.

PRODUCT SPECIFICATIONS

Cell type	iPSC-derived ventricular cardiomyocytes
Source	iPSC of 26 y/o Caucasian female
Species	Human
Purity	Approx. 76% ventricular; fibroblast-free
Assay window	Stable beating after 72h. Refer to our protocols for assay-specific recommendations

EACH LOT IS TESTED QUALITY CONTROL PHARMACOLOGICALLY

Cooperation partner
metrion biosciences

AXIOGENESIS OVERVIEW

DIFFERENTIATED HUMAN CELLS

Axiogenesis is a leading expert in providing commercial-grade *in vitro* differentiated cell types derived from human induced pluripotent stem cells (iPSCs).

Core products include Cor.4U® cardiac myocytes and fibroblasts, as well as Peri.4U™, Dopa.4U™, CNS.4U™ and Astro.4U™ neural cells.

VALIDATED ASSAYS & PROTOCOLS

Axiogenesis enables customer efficiency by providing ready to use cells along with validated protocols.
Assays for each cell type have been developed for advanced drug discovery, safety pharmacology, *in vitro* toxicology applications, and disease and tissue modeling.
Based on its in-house assay capabilities, Axiogenesis is able to provide expert scientific support in order to facilitate selection and quick implementation of validated assays and technologies.

iPSC-derived neurons

iPSC-derived cardiomyocytes

vCor.4U™

Metrion Ion Channel Screen Service

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