Available online at www.sciencedirect.com

Integrative Medicine Research

journal homepage: www.imr-journal.com

Review Article

Electrophysiological properties and calcium handling of embryonic stem cell-derived cardiomyocytes

*Jae Boum Youm**

National Research Laboratory for Mitochondrial Signaling Laboratory, Department of Physiology, College of Medicine, Cardiovascular and Metabolic Disease Center, Inje University, Busan, Korea

ARTICLE INFO

Article history:
Received 5 December 2015
Received in revised form
28 December 2015
Accepted 30 December 2015
Available online 7 January 2016

Keywords:
calcium handling
cardiomyocytes
electrophysiology
embryonic stem cell
ion channels

ABSTRACT

Embryonic stem cell-derived cardiomyocytes (ESC-CMs) hold great interest in many fields of research including clinical applications such as stem cell and gene therapy for cardiac repair or regeneration. ESC-CMs are also used as a platform tool for pharmacological tests or for investigations of cardiac remodeling. ESC-CMs have many different aspects of morphology, electrophysiology, calcium handling, and bioenergetics compared with adult cardiomyocytes. They are immature in morphology, similar to sinus nodal-like in the electrophysiology, higher contribution of trans-sarcolemmal Ca^{2+} influx to Ca^{2+} handling, and higher dependence on anaerobic glycolysis. Here, I review a detailed electrophysiology and Ca^{2+} handling features of ESC-CMs during differentiation into adult cardiomyocytes to gain insights into how all the developmental changes are related to each other to display cardinal features of developing cardiomyocytes.

1. Introduction

Embryonic stem cell-derived cardiomyocytes (ESC-CMs) are greatly promising for stem cell therapy against cardiovascular diseases such as myocardial infarction and potentially life-threatening arrhythmias because of their ability to differentiate into sinus–nodal, atrial, or ventricular-type of cardiomyocytes.[1,2] ESC-CMs also have a potential to be a novel pharmacological tool in, for example, high-throughput screening tests of newly developed drug for cardiotoxicity.[3] In addition, ESC-CMs could be a powerful new model system to study mechanisms of inherited cardiomyopathies.[4]

ESC-CMs are immature in both morphological and functional aspects. They have underdeveloped contractile machinery and lack transverse-tubular (T-tubular) system.[5,6] They display diastolic potentials similar to those of adult sinus–nodal cells and slower upstroke velocity.[1,2] They have fewer mitochondria[5] and are predominantly glycolytic.[7] Ca^{2+} handling is also significantly different from that of their adult counterpart displaying slower kinetics and smaller amplitude.[8] This review will primarily focus on the

* *Corresponding author.* National Research Laboratory for Mitochondrial Signaling, Department of physiology and Biophysics, College of Medicine, Cardiovascular and Metabolic Disease Center, Inje University, 75, Bokji-ro, Busanjin-Gu, Busan, 614-735, Korea.
E-mail addresses: youmjb@inje.ac.kr, jaeboum@gmail.com
http://dx.doi.org/10.1016/j.imr.2015.12.007

Table 1 – Model parameter values in mathematical model*

Model parameters	Nodal type (AP)	Atrial type (AP)	Ventricular type (AP)
E_{CaL}, mV	47	47	47
g_{CaL}, nS	11.0140	12.5874	12.5874
k_1	0.9	1.1	1.8
k_2	1.0	1.3	2.2
$g_{HCN,Na}$, nS	0.0529	0.0529	0.0529
$g_{HCN,K}$, nS	0.0851	0.0851	0.0851
$g_{to,UF}$, nS	0.3346	1.3382	0.6691
$g_{to,F}$, nS	0.1338	0.5352	0.2676
$g_{to,S}$, nS	0.2091	0.8364	0.4182
g_{Ks}, nS	0.0411	0.2057	0.4114
$g_{b,Ca}$, nS	0	0	0
$g_{b,Na}$, nS	0.170	0.122	0.122
$g_{b,K}$, nS	0.035	0.025	0.025
g_{CaT}, nS	0	0	0
g_{Kl}, nS	0	0.6075	2.2275
g_{Kr}, nS	1.5319	0.6383	0.6383
$g_{Na,1.5}$, nS	0.0059	0.0237	0.0296
$g_{Na,1.1}$, nS	0.0059	0.0237	0.0296
g_{st}, nS	0	0	0
g_{sus}, nS	0	0	0

* See glossary of Kharche et al's[9] model for detailed explanation of each channel.
Note. Adapted from "Dual modulation of the mitochondrial permeability transition pore and redox signaling synergistically promotes cardiomyocyte differentiation from pluripotent stem cells," by S.W. Cho, J.S. Park, H.J. Heo, S.W. Park, S. Song, I. Kim et al., 2014, *J Am Heart Assoc*, 3, e000693. Copyright 2014 The Authors. Reprinted with permission.
AP; action potential.

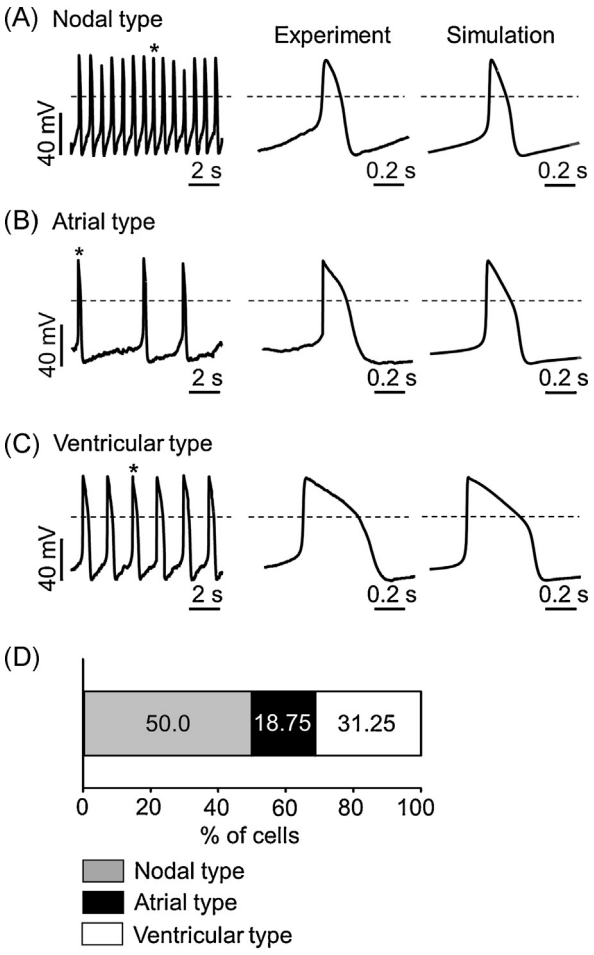

Fig. 1 – Representative action potential (AP) morphologies from patch clamp recordings and mathematical modelling. (A) Nodal type, (B) atrial type, and (C) ventricular type APs recorded from different embryonic stem cell-derived cardiomyocytes in a current–clamp mode. Each AP trace in the middle panel corresponds to an expanded trace of single AP denoted with an asterisk in the left panel. Each AP trace in the right panel corresponds to a simulated AP trace. Dotted lines indicate zero voltage level. (D) Percentile distribution of three different types of APs in embryonic stem cell-derived cardiomyocytes. See Table 1 for model parameters used in the mathematical model.
***Note.* Fig. 1D is from "Dual modulation of the mitochondrial permeability transition pore and redox signaling synergistically promotes cardiomyocyte differentiation from pluripotent stem cells," by S.W. Cho, J.S. Park, H.J. Heo, S.W. Park, S. Song, I. Kim et al., 2014, *J Am Heart Assoc*, 3, e000693. Copyright 2014 The Authors. Reprinted with permission.**

electrophysiological properties and Ca^{2+} handling features of ESC-CMs during differentiation into adult cardiomyocytes.

2. Electrophysiological properties of ESC-CMs

Early studies on the differentiation of pluripotent mouse embryonic stem cells (ESCs) into adult cardiomyocytes revealed that the rhythmic action potentials (APs) of early differentiated cardiomyocytes are very similar to those of sinus–node cells.[1,2] Terminally differentiated cardiomyocytes generated APs similar to those of adult sinus–nodal, atrial, and ventricular myocytes, suggesting that there should be developmental changes in cardiac ion channels and calcium handling properties along with differentiation into adult cardiomyocytes. Representative traces of nodal-like, atrial-like, and ventricular-like APs from patch–clamp recording and mathematical modelling are demonstrated in Fig. 1. The mathematical model was based on the Kharche's model[9] and modified parameters are summarized in Table 1. In the following, functional changes in electrophysiological properties during differentiation are described in detail.

2.1. *Capacitance*

In one early study on mouse ESC-CMs, membrane capacitance was found to steadily increase from 24.5 pF to 50.0 pF during differentiation.[10] As the specific membrane capacitance was calculated to be 0.85–0.86 μF/cm^2, it was concluded that the T-tubular system apparently was not developed during differentiation.[10] T-tubule formation was not observed at the ultrastructural level in human pluripotent stem cell-derived cardiomyocytes, when electron microscopy or membrane staining by Di-8-ANNEPPS were used.[6]