

Original Article

Patch clamp apply in cardiomyocytes derived from patient's iPS cells for individual anticancer therapy

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Received August 26, 2014; Accepted October 30, 2014; Epub November 15, 2014; Published November 30, 2014

Abstract: Cardiac side-effects of chemotherapy are old dogs for cancer patient at the beginning of cancer treatment. The deleterious side-effects of chemotherapy on the cardiovascular system have been deemed as a serious clinical issue for a long term, especially sudden cardiac death (SCD) due to QT prolongation. Since induced pluripotent stem cells has been firstly reported by Takahashi and Yamanaka in 2006, iPS has become a valuable research tool. Especially, cardiomyocytes have been successfully derived from human iPS cells which carry on corresponding genetic information of disease, and therefore show a great promise in drug screen on the diseasing model. In this study, we hypothesized that iPS cells created from patient with cancer and carried corresponding genetic alteration will provide a genetic background for sensitivity screening of anticancer drug, as well as side-effects of cardiovascular system.

Keywords: Patch clamp, iPS cells, individual anticancer therapy

The cardiotoxicity of anticancer chemotherapy

Cancer is a major public health problem in the United States and many other parts of the world, and now 1 in 4 deaths in the United States is due to cancer [1]. The management of cancer still is a challenge for clinic practice, although the developments of anticancer treatment have dramatically improved the prognosis of cancer patients over the past few years, especially molecular-target drugs chemotherapy. However, the deleterious side-effects of chemotherapy on the cardiovascular system have still been identified with the developments of chemotherapy [2-6].

The cancer chemotherapy drugs can be subclass as anthracyclines, alkylating agents, antimetabolites, microtubule inhibitors, unclassified, antibody-based tyrosine kinase (TK) inhibitors and small molecule TK inhibitors [4]. The common cardiovascular complications related to anticancer chemotherapy include heart failure, myocardial ischemia, hypertension, thromboembolism, QT prolongation, and bradycardia [6]. The incidence of cardiovascular complications ranges from lower to higher [6], and in different patient the incidence of cardiovascular

complications are not the same. QT prolongation related to chemotherapy is one of the most common complications. It has been shown that suppression of rapid component of delay rectifier potassium current, I_{Kr} , represents the principal pharmacodynamic mechanisms, in which lead to heterogeneous prolongation of ventricular action potential and prolongation of the QT interval clinically [7].

The history of iPS cells

In 2006 Yamanaka and co-workers made a breakthrough discovery to the stem cell field. They firstly reported the generation of induced pluripotent stem cells (iPSCs) from murine fibroblasts by the retroviral transduction of Oct4, Sox2, Klf4 and c-Myc transcription factors [8]. Subsequently, in 2007 group from Japan and the other two groups from the U.S.A generated the first human iPS cells from embryonic, neonatal and adult fibroblasts [9-11]. From then on, various other somatic cell types, such as peripheral blood cells, adipose stem cells, mesenchymal cells and melanocytes have been described for reprogramming in creating iPS cells [12].

iPS cells have pluripotent differentiation potential. Various studies have shown iPS cells differentiated into motor neurons (have electrically active) [13], blood cells (have hematopoietic colony activity) [14, 15]. It is inspiring that iPS cells were shown to differentiate into cardiomyocytes that beat rhythmically and respond to cardioactive drugs [16-19].

iPS cell-derived cardiomyocytes

The iPS cell-derived cardiomyocytes can be an in vitro model to test the cardiotoxicity of anticancer drug before apply to cancer patient, which could be a way to create individual medicine for cancer patient. Recently, the paper titled "Patient-specific induced pluripotent stem-cell models for long-QT syndrome", published on New England Journal of Medicine showed the great promise of iPS for disease model [20]. Two studies applied human iPS cell-derived cardiomyocytes together with a multi-electrode assay as a platform to study different agents on the electrophysiological properties of heart cells [21, 22].

Drug screening in human iPS cardiomyocytes

Disease models based on human iPS cardiomyocytes have been established by many groups. This method has been shown a powerful tool to investigate disease mechanisms in vitro and to perform patient-specific drug screening [23-29]. Most of the studies focused on long QT syndrome [24-26, 28]. Matsa E et al established LQT2 (LQTS type 2)-hiPSC cardiomyocytes disease model, and apply multi-electrode array and patch-clamp electrophysiology to investigate the effect of E4031 (an I(Kr) blocker) on LQT2-hiPSC cardiomyocytes. They found that LQT2-hiPSC cardiomyocytes developed EADs when challenged with the clinically used stressor isoprenaline in contrast to control cardiomyocytes. These effects could be reversed by anti-arrhythmia drugs, such as β -blockers, propranolol, and nadolol, etc. Apply experimental potassium channel enhancers, nicorandil and PD118057 in the cardiomyocytes can cause action potential shortening and in some cases could abolish EADs. Furthermore, a combined treatment with isoprenaline (enhancers/isoprenaline) could promote EADs, which could be reversed by nadolol [26]. However, application of cardiomyocytes derived from patient iPS cells has not been reported in anticancer drug screening.

The hypothesis

The functional cardiomyocytes have been successfully derived from human iPS cells, and show great promise in drug screen and disease model [20, 23-29]. Cardiomyocytes derived from iPS, with patch clamp could be a powerful tool for anticancer drug screening in lethal SCDs.

Experimental testing of the hypotheses

Firstly, the patient specific iPS cells will be generated from fibroblasts from patient who need cancer chemotherapy, by expression of a combination of retroviruses encoding the human transcription factors OCT3/4, SOX2, KLF4, and c-MYC [10]. Then, the specific iPCs will be induced to differentiate into cardiomyocytes. [18, 19] which will be verified using reverse-transcriptase-PCR assay on cardiomyocyte phenotype markers. Using the whole cell patch clamp recording, electrophysiological effects of anticancer drugs, such as on kinetics of IKr, will be investigated for side-effect of arrhythmogenesis. Upon successful conduction of this study, a series of specific anticancer drugs without, at least less, side-effect on cardiovascular system will be selected for individual patient for a long-term treatment.

Disclosure of conflict of interest

There are no financial or commercial conflicts of interest in this work.

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