Bio-informational Pharmacology

1. Staffs and Students (April, 2010)

Professor	Tetsushi Furukawa, MD, PhD
Associate professor	Junko Kurokawa, PhD
Assistant professor	Yusuke Ebana, MD, PhD
Adjunctive Assistant professor	Tetsuro Sasano, MD, PhD

2. Purpose of Education

This laboratory focuses on understanding fundamental pathophysiological roles of ion channels and transporters in cardiovascular system. We employ multidisciplinary approach (patch-clamp, cell biology, optical recording, genetic analysis, and computational analysis) in order to seek novel regulatory mechanisms and modulatory molecules/compounds of ion channels and transporters in cardiac myocytes, vascular smooth muscle and endothelial cells, and circulating cells in vessels (monocytes and macrophages). Our ultimate goal is to discover novel diagnostic and therapeutic strategy for intractable and common cardiovascular diseases.

3. Research Subjects

1. Gender-specific medicine (GSM) for cardiovascular diseases

Susceptibility of several diseases and responsibility to various drugs and therapy exhibit gender-difference, and cardiovascular system show unique gender difference. We have previously reported that non-genomic actions of sex hormones play an important role creating gender-difference in cardiac arrhythmia. As a next step, we focused on the role of XY chromosome in gender difference. Using a novel ink-jet method to visualize mouse coronary artery, we found gender differences in coronary artery circulation not related to sex hormone differences.
(collaboration with Prof. Y. Kurihara in the University of Tokyo Graduate School of Medicine)

2. Pathogenesis of atrial fibrillation

Atrial fibrillation is the most frequent persistent arrhythmias, reaching more than 3.5 million patients in Japan. Associated cerebral infarction due to cardiogenic thrombosis (250,000 patients /year in Japan) causes reduced QOL and is one of the main causes of bedridden old people. We have taken following 2 approaches to establish protection and treatment of atrial fibrillation;

a. GWAS (genome-wide association study) in atrial fibrillation

We carry out most extensive GWAS (genome-wide association study) in Japan to determine gene polymorphisms associated with atrial fibrillation. Our data show 2 genetic risks highly related to atrial fibrillation which have not ethnic difference, and 2 intermediately significant risks specific to Japanese and 2 intermediately significant risks specific to Caucasian. Based on these genetic risks, we could provide framework for risk stratification of atrial fibrillation in Japan.

(collaboration with Prof.. Nakamura Y in The Institute of Medical Science, The University of Tokyo, Dr. Tanaka T. in RIKEN, Dr. Sawabe M. in Tokyo Metropolitan Geriatric Hospital, and Department of Cardiology in this University)

b. Inflammatory and immunological mechanisms in atrial fibrillation

Pathogenesis of many metabolic syndrome-related diseases, including atrial fibrillation, involves chronic, mild inflammation. We examine environmental and genetic factors to cause recruitment of inflammatory cells (e.g. T lymphocytes, macrophages, mast cells) to atrial tissues. Our data show that physical stretch of atrial myocytes secrete some danger signal, which plays a critical role as an autocrine/paracrine factor to recruit inflammatory cells, both in vitro and in vivo.

3. Pathogenesis of ventricular tachyarrhythmias and sudden cardiac death

Despite extensive effort by many researchers for years, ventricular tachycardia and fibrillation remain the main cause of sudden death, and the biggest challenge in arrhythmia research. Our laboratory approaches this issue using 2 genetically engineered mice;

a. Analysis of *NOS1AP* (NOS1 associated protein) KO mice

In recent GWAS performed in Western countries, *NOS1AP* is surprisingly the most closely related gene to sudden cardiac death. We analyzed *NOS1AP* KO mice to clarify the mechanism of association between *NOS1AP* and sudden cardiac death. Our data showed that *NOS1AP* KO mice are highly susceptible to sudden death after transverse aorta

constriction (TAC) procedure.
(collaboration with Dr. N. Kato in National Center for Global Health and Medicine)

b. Analysis of KO mice for a transcription regulator specific to Purkinje fiber
Recent clinical data implicate the importance of Purkinje fiber in development of ventricular tachycardiac/fibrillation, and cardiac sudden death. We created mice deficient of a transcription regulator specifically expressed in Purkinje fibers. Mice are highly susceptible to lethal arrhythmias caused by exercise (swimming) or sympathetic stimulation.
(collaboration with Prof. N. Miura Hamamatsu University School of Medicine)

4. Use of iPS cells for arrhythmia research

Traditional arrhythmia researches have been performed in cardiomyocytes of species other than human, or in cultured cells, in which human ion channel genes have been heterologously expressed. The milieu different from human cardiac myocytes (especially the lack of excitation-contraction coupling machinery) is the huge limitation for arrhythmia research. Cardiomyocytes differentiated from human iPS cells could overcome this critical limitation, and would bring marked advance in arrhythmias researches. We take following 2 approaches;

a. Establishment of human iPS-derived cardimyocytes (hiPS-CM) from LQT patients
We try to establish and characterize iPS cell-derived cardiomyocytes from human fibroblasts obtained from congenital LQT patients. We have able to established 1 line for type 1 LQT and 2 lines for each of type 2 and 3 LQT. Our data showed that hiPS-CM from LQT patients maintain electrophysiological phenotype found in LQT patients' hearts.
(collaboration with Prof. K. Fukuda in Keio University School of Medicine)

b. Drug screening system using human iPS cells-derived cardiomyocytes
New drugs are developed in a research consisted of ①discovery of a lead compound→②pre-clinical study in animals→③clinical study. Because of cardiac toxicity in human, many candidates compounds advanced to clinical trials fail to pass, which is because of the lack of assay system in human tissues in pre-clinical study step. Cardiomyocytes derived from human iPS cells could provide an assay system in human tissues in pre-clinical study step, and facilitate new drug discovery and prevent unexpected drug side effects.
(collaboration with Dr. Y. Kanda in National Institute of Health Sciences)

4. Publications List

Original Article

1. Yamashiro K, Sasano T, Tojo K, Namekata I, Kurokawa J, Sawada N, Suganami T, Kamei Y, Tanaka H, Tajima N, Utsunomiya K, Ogawa Y, Furukawa T. (2010). Role of transient receptor potebntial vanilloid 2 in LPS-induced cytokine production in macrophages. ***Biochem. Biophys. Res. Commun***. 398, 284-289.
2. Yang PC, Kurokawa J, Furukawa T, Clancy CE. (2010). Acute effects of sex steroid hormones on susceptibility to cardiac arrhythmias: a simulation study. ***PLoS Comput. Biol***. 6, e1000658.
3. Uejima T, Koike A, Sawada H, Aizawa T, Ohtsuki S, Tanaka M, Furukawa T, Frase AG. (2010). A new echocardiographic method for identifying vortex flow in the left ventricle: Numerical validation. ***Ultrasound Med. Biol***.
4. Furukawa T. (2010). Transmural dispersion of repolarization and drug-induced Torsade de Pointes – A 3-D simulation study. ***Circ. J***. 75, 49-50.