Bioeng 6460
Electrophysiology and Bioelectricity

Microstructural Basis of Conduction II
Introduction to Experimental Studies

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Overview

- Microstructural Basis of Conduction
  - Fibroblasts
    - Arrangement
    - Differentiation
    - Role in Electrophysiology
- Introduction to Experimental Studies
  - Whole Heart
  - Tissue Preparations
  - Cell Culture
- Summary

Group work
Fibroblasts

- most numerous cells in myocardium
- primary producer of extracellular matrix proteins
- change of phenotype in response to pathological conditions, eg infarction, inflammation and injury
- chronic activation can lead to fibrosis, hypertrophy and heart failure
- spatial organization in tissue still topic of research
- role in electrical conduction is still not completely understood

Fibroblast Organization in Rat Neonatal Myocardium

Discoidin domain receptor (DDR) - Fibroblasts

Actin - Myocytes

Cx43 - Gap Junctions

Arrows indicate gap junctions of fibroblasts

(E. C. Goldsmith et al, Develop Dyn 2004)
Fibroblasts in Mouse Ventricular Myocardium

Fluorescent microsphere in blood vessels

DAPI – Nuclei

DDR - Fibroblasts

(Sounders et al, Circ Res, 2009)
Fibroblasts in Normal Rat Ventricular Tissue

(M. Arp et al, Biomed Tech, 2011)

50 µm

WGA – Extracellular space
DAPI – Nuclei
Cx43
Vimentin - Fibroblasts

(M. Arp et al, Biomed Tech, 2011)
Fibroblasts in Rat Ventricular Tissue (Zoom)

WGA – Extracellular space
DAPI – Nuclei
Cx43
Vimentin - Fibroblasts

(M. Arp et al, Biomed Tech, 2011)
Fibroblast Differentiation

(Tomasek et al, Nat Rev, 2002)
Role of Fibroblasts in Electrophysiology

- electrically inexcitable

- passive role
  - septa due to fibrosis
  - reduced volume fraction of myocytes
  - reduced lateral coupling

- active role
  - electrical myocyte-fibroblast coupling via gap junction channels
  - electrical bridging of myocytes in culture: over distances up to 300 µm (G. Gaudesius et al, Circ Res 2003)
  - additional sink or source for activation of myocytes

- role dependent on phenotype of fibroblast

(Jong et al, J Cardiovasc Pharm, 2011)

(Rook et al, Am J Physiol, 1992)
Discuss effects of fibrosis on extracellular electrograms.

Speculate on effect in the near (interstitial space) and far field (ECG).
Electrical Signaling in the Heart

(from Malmivuo and Plonsey)
Experimental Studies of Cardiac Electrical Conduction

**Measurement methods**
- Electrode arrays: Extracellular voltages (similar ECG measurements on body surface)
  - Sampling rate up to several kHz
  - Channels up to 2000
- Optical: Transmembrane voltages
  - CCD-camera
  - Photodiode array

**Preparations**
- Cell strands - Purkinje fibers
- Small muscles - papillary muscle, trabeculae
- Sections - wedge preparations from ventricles
- Atria/ventricle
- Whole heart

in vivo/in vitro

Color-coded visualization of extracellular voltages measured on surface of canine ventricles
Epicardial Electrical Mapping System for Mouse Heart

Sohn et al, IEEE TBME, 2011
Electrical Mapping of Canine Ventricles

http://www.cvrti.utah.edu/?q=node/36
Optical Mapping of Canine Ventricular Area

http://www.cvrti.utah.edu/?q=node/36
Isotropic/Anisotropic Propagation of Excitation (2D)

- Long axis of myocytes parallel to y-axis
- Stimulus at point (0,0)

**Isotropic x/y - 1/1**
Velocity $v$: 1 / s

**Anisotropic x/y - 1/3**
Velocity $v_x$: 1 / s, $v_y$: 3 / s

Simplifications
- Homogeneous tissue
- Neglect of microstructure
In-/Outflow of Currents during Excitation

10 ms

20 ms

30 ms

40 ms

50 ms

60 ms
In-/Outflow of Currents during Repolarization

110 ms

130 ms

150 ms

170 ms

190 ms

210 ms
Dipole Approximation and Surface ECG
Species: Adult New Zealand White rabbits (1.5-3.0 kg)
1. Anti-coagulated with heparin and anesthetized with pentobarbital
2. Hearts are rapidly excised and moved to dissection tray
3. Retrograde perfusion via aorta with modified Tyrode solution
4. Opening of right ventricle
5. Selection and excision of papillary muscle including onset of chordae tendinae
   **Criteria:** Small diameter, large length, unramified
6. Transfer to horizontal flow-through chamber
7. Fixation of muscle
8. Measurement

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**Diagram:**
- **Stimulus position**
- **EG measurement**
- Oxygenated HEPES solution, 37 °C
Measurement Results: Electrograms

- Stimulus artifact
- Distance to stimulus site
Experimental Studies of Conduction in Cell Culture

Myocyte strand with fibroblast insert

Optical mapping using voltage sensitive dyes

(Gaudesius et al, Circ Res, 2005)
Optical Mapping of Co-Culture of Rat Myocyte/Myofibroblast

DAPI - nuclei
Actin - myocytes
α-smooth muscle actin - myofibroblast

(Zloicher et al, Biophys J, 2008)
Optical Mapping of Reentrant Arrhythmia

(Zloicher et al, Biophys J, 2008)
Identify the major mechanisms of cardiac conduction! What would be an electrical engineering description of those?

Which other systems exhibit similar phenomena? List at least 10.
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