

# Z47 - RESEARCH OF THE ELECTROPHYSIOLOGICAL PROPERTIES OF CELL BILAYERS

Physics Laboratory II – academic year 2017/2018

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The main purpose of this exercise I to get acquainted with basic knowledge about electrophysiological studies on living cells performed using Patch Clamp (PC) technique. During classes students will learn about ionic mechanisms taking place in living cell and will register some of them using simple PC protocols. Students should identify the types of channels present in investigated cells and check the impact of selected chemical blockers on cell behavior.

## Preparatory questions

1. Plant and animal cell anatomy
  2. Composition and functioning of the cell membrane
  3. Types, functions and characteristics of ion channels
  4. Basics of Patch Clamp measurements
  5. Significant parameters for electrophysiological measurements
2. Running the data registration and analysis software on the computer
  3. Identifying all important switches, indicators and control windows in the software program (Patch Control)
  4. Carrying out a test measurement, mastering available working modes and software functions
  5. Measurement using living cells
    - I-V curve determination
    - Measurements from hyperpolarization to membrane depolarization (Current Clamp mode)
    - Specific chemical blocker application (external perfusion set)

## Computational assignments

**Exercise 1.** The thickness of lipid bilayer in a neuron is  $8nm$ , the permittivity of fluid filling the inside compartment of bilayer is 3.5. Please calculate the theoretical electrical capacitance of a  $10nm$  by  $10nm$  rectangular section of lipid bilayer in this cell.

**Exercise 2.** Using data from the previous task, please calculate the theoretical capacitance for the entire neuron cell with a diameter of  $30\mu m$ , assuming it is a perfect sphere.

## Apparatus and materials

Experimental setup and materials (with the system picture and element description):

- microcantilever sensor (Fig.1)
- set of microcantilevers
- holder with the sample mounting kit
- Yeast *Saccharomyces cerevisiae*

Student has in her/his disposal an oscilloscope, a PC computer and the copy of Teach Spin manual.

## Data analysis

1. Description of the PC setup together with the diagram
2. Informations about Patch Clamp Technique
  - method description
  - measurement scheme
  - available measurement configurations with detailed descriptions and diagrams
  - information about ion channels (role in organism, construction, action mechanisms)
  - collected I-V curves with interpretation
  - results from all other measurement stages with detailed description

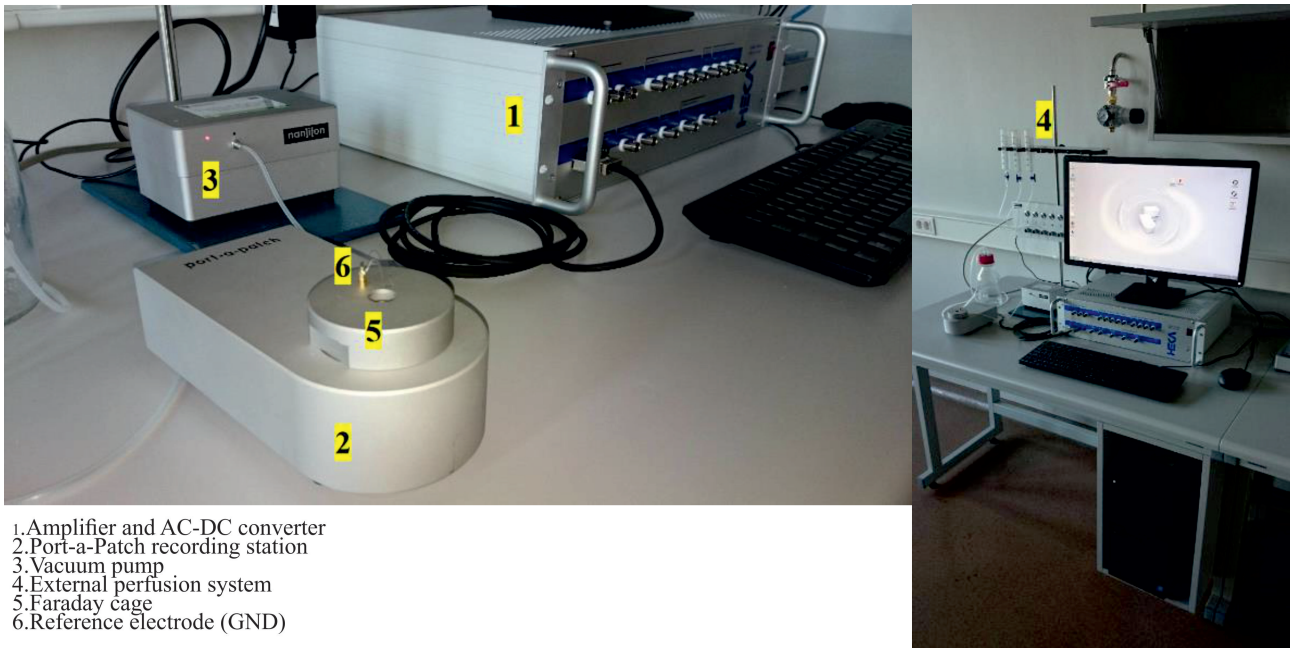
Student in his final report should identify ion channel type present in investigated cells. This fact should be confirmed by analyzing data from I-V curves and results obtained after specific blocker administration.

## Experiment

1. Getting acquainted with all elements of the measuring setup

## Safety rules

**It is advised to wear protective gloves during the work with yeast cells.**



1. Amplifier and AC-DC converter
2. Port-a-Patch recording station
3. Vacuum pump
4. External perfusion system
5. Faraday cage
6. Reference electrode (GND)

Figure 1: Experimental setup

## References

- [1] Bruce Alberts, Alexander Johnson, Julian Lewis, Martin Raff, Keith Roberts, and Peter Walter, , New York: Garland Science; 2002.
- [2] Alan Longstaff, , Garland Science, 2000 – 436
- [3] Online publications related to the subject (using Pubmed or similar scholar browser use phrases like: ion channels, patch clamp, electrophysiology, cell biology, nanion, ion channels blockers)
- [4] <http://www.nanion.de/products/port-a-patch.html>