

Photo-voltage of highly-oriented bacteriorhodopsin in purple membranes: Possibilities for bio solar cells

A. V. VLASOV^a, YU. S. KOVALEV^{a,b}, P. K. UTROBIN^a, YU. L. RYZHYKAU^a, F. V. FROLOV^a, E. V. ZINOVEV^a, A. V. ROGACHEV^{a,b}, A. I. KUKLIN^{a,b,*}, V. I. GORDELIY^{c,d}

^a*Moscow Institute of Physics and Technology, Dolgoprudny, Russia*

^b*Joint Institute for Nuclear Research, Dubna, Russia*

^c*Institut de Biologie Structurale J.-P. Ebel, Université Grenoble Alpes-CEA-CNRS, F-38000, Grenoble, France*

^d*Institute of Complex Systems: Structural Biochemistry (ICS-6), Research Centre Jülich, 52425, Jülich, Germany*

Highly oriented purple-membrane films of bacteriorhodopsin were obtained using external electrical field between two ITO (Indium Tin Oxide) supports. Purple membrane films, highly oriented in one direction showed 300 mV of photo-voltage while being illuminated by cold light lamp. Concentration of bacteriorhodopsin in purple membranes (bR in PM) and salt concentration were screened in order to find optimal conditions for maximizing the photo-voltage. The dependence of electrical properties of the films from light irradiation was investigated. Results obtained in this work might be important for optogenetic tools and for creating bio solar cells based on photosensitive membrane proteins.

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1. Introduction

Nowadays the field of interest of material science and advanced materials moves towards biomaterials [1], [2]. In current work we investigate purple membranes as potential candidates for bio-solar cells.

Purple membranes obtained from *Halobacterium salinarum* are known to consist of 75% (w/w) protein bacteriorhodopsin and actually they are natural membrane protein crystals. Also purple membranes contain several types of different lipids (90% polar and 10% non-ionizable respectively) and have a very complex morphology and architecture as a natural biological membrane [3].

Bacteriorhodopsin is a photosensitive transmembrane proton pump. It absorbs light and pumps protons against their concentration gradient. This creates transmembrane proton potential that drives ATPases and support life in living cells.

In literature oriented purple-membrane films of bacteriorhodopsin were used as a probe for functional studies of the protein. There were trials to investigate bacteriorhodopsin photocycle and, in particular, its' M-state, using external electrical field, where between two electrodes there was a purple-membrane film. The method was proposed in 1986 [4].

Photocycle studying was necessary to get an insight about functional properties of the photosensitive protein. These studies were relevant until the structure of bacteriorhodopsin in M-state was solved by X-ray crystallography in 2000 [5], so the method of bacteriorhodopsin functional studies with external electrical field and electrodes became obsolete.

However, those works gave the idea of investigating the possibility of using bacteriorhodopsin in purple membranes as a basic element for bio solar cells.

In this work we focused on light-induced electrical properties of highly oriented purple membranes and we found 300mV of photo-voltage on photocapacitor ITO – bR in PM – ITO. We argue that this effect can open a possibility for creating solar cells based on photosensitive membrane proteins.

The possibilities of bacteriorhodopsin as a bio solar cell element were investigated recently [6], [7]. However, in literature there are no persuasive results about efficiency of bacteriorhodopsin as a bio solar cell basic element.

Here, we tried to use highly oriented purple membranes of bacteriorhodopsin in order to break through the barrier of low efficiency of one separate membrane and use stack of purple membranes, forming one continuous film. The properties of highly oriented purple membranes are significantly differ from purple membranes, which are badly-oriented or oriented chaotically and drastically changes with concentration of bR in PM and salt concentration.

The results of this work open new possibilities in solar cell industry and broaden horizons of alternative energy sector via using natural converters of solar energy into electricity – photoactivated membrane proteins, which can become efficient, reliable and environmentally-friendly source of energy in the nearest future.

The basic elements of these bio solar cells are photoactive membrane proteins, which transfer ions through the membrane in opposite direction of their concentration gradient.

2. Experimental

Bacteriorhodopsin in purple membranes (bR in PM) was obtained from *Halobacterium salinarum*. It was expressed and purified according to protocols developed by Gordeliy V.I. [8].

Dialysis was done in order to change buffer on H₂O (mQ). bR in PM was concentrated on Eppendorf centrifuge 5417R (14000 rcf, 30 min). Final concentration of bR in PM in mQ was approximately 20 mg/ml.

Two transparent polymeric electrodes – Indium Tin Oxide (ITO) were used on glass basement and a drop of bR in PM ($V = 100$ ul) was placed between two electrodes using a syringe. In order to make highly-oriented bR in PM films we used current source and applied 4V potential between two ITO electrodes for approximately 30 seconds in order to create an external electrical field in a drop of bR in PM. Purple membrane actually has a dipole momentum because of different bR aminoacids on the different sides of a membrane. The scheme of sample preparation is shown on [Fig. 1].

When purple membranes precipitated on the cathode the rest liquid that became transparent was removed with the help of syringe.

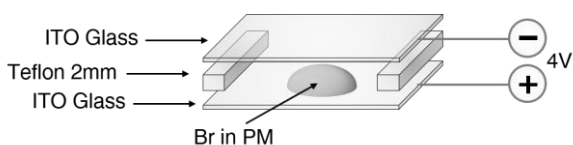


Fig. 1. The scheme of sample preparation bR in PM film on ITO glass. The scheme is actually a capacitor connected to DC voltage source. The potential between two ITO electrodes - 4 V

Purple membranes were dried at 50% humidity 24h. Thin scotch tape was used to separate two electrodes. ITO glass was placed over dry bR in PM and formed a photocapacitor ITO – bR in PM – ITO [Fig. 2].

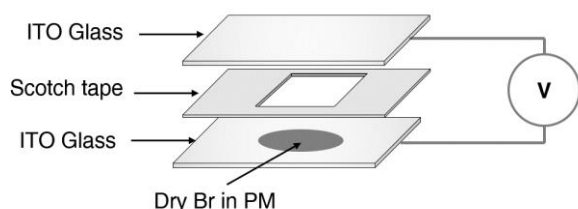


Fig. 2. Photocapacitor ITO – bR in PM – ITO. Oscilloscope acts as a voltmeter

The photocapacitor was exposed to light illumination of KL2500 LCD lamp. The photograph of experimental installation is shown on [Fig. 3].

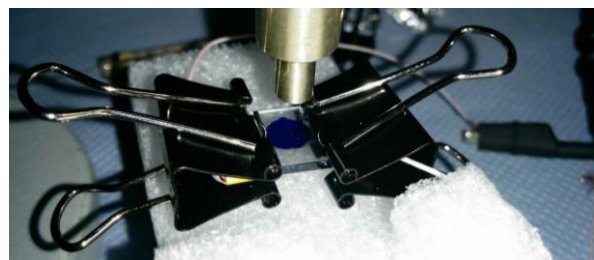


Fig. 3. Experimental installation for photopotential measurements of ITO – bR in PM – ITO photocapacitor. On the photograph there is a tip of light guide of KL2500 LCD lamp

3. Results and discussion

When illuminated by KL2500 LCD lamp the photocapacitor ITO – bR in PM – ITO gave a photopotential of 300 mV [Fig. 4].

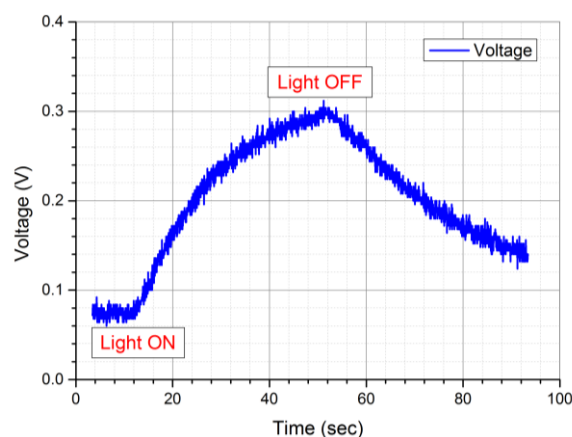


Fig. 4. Photovoltage obtained from the photocapacitor ITO – bR in PM – ITO versus time at two modes: «Light ON» and «Light OFF» of KL2500 LCD lamp. The peak photovoltage value is 300 mV

The initial part of the photovoltage curve (see Fig. 4) shows the signal increases up to 0.3 V, and after «Light OFF» there is an exponential decay of the signal.

Repeatability of photovoltage occurrence when flat capacitor ITO – bR in PM – ITO was illuminated by lamp KL2500 LCD was confirmed [Fig. 5].

The long-term time dependence of photopotential on ITO – bR in PM – ITO photocapacitor was investigated at «Light ON» and «Light OFF» modes of KL2500 LCD lamp.

The system was found to come to saturation (capacitor charging by light), and produced exponential decay of the signal (capacitor discharging after switching off the light) [Fig. 6].

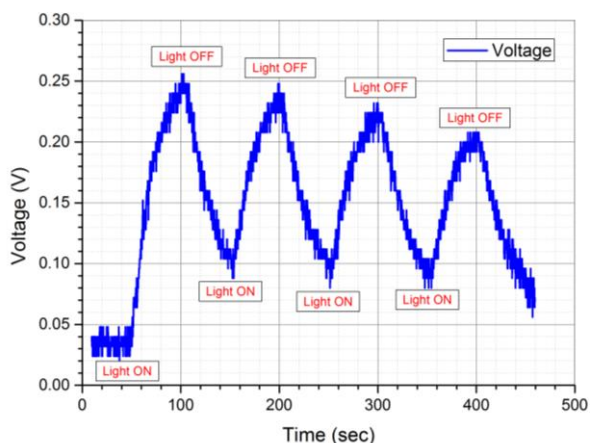


Fig. 5. Photovoltage obtained from the photocapacitor ITO – bR in PM – ITO versus time at periodical changing modes «Light ON» and «Light OFF» of KL2500 LCD. Repeatability of photovoltage occurrence is shown

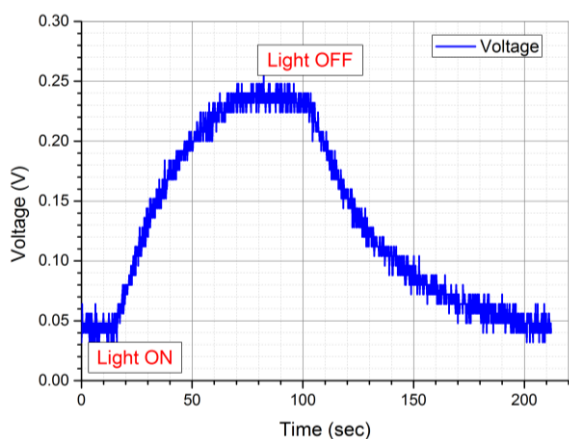


Fig. 6. Photovoltage obtained from the photocapacitor ITO – bR in PM – ITO versus time at long time periods (100 sec) at modes «Light ON» and «Light OFF» of KL2500 LCD lamp. There is a capacitor charging at «Light ON» mode, and discharging at «Light OFF» mode

4. Conclusion

In this work we built photocapacitor based on highly-oriented bR in PM films. The photopotential of 300 mV was obtained for ITO – bR in PM – ITO flat photocapacitor when illuminated by KL2500 LCD lamp. The effect of photopotential occurrence has proved its repeatability. At long-term period (~100 sec) the system came to saturation and capacitor charging by light and when the light was off the capacitor was discharging with exponential decay of the signal.

The biological system (bR in PM film) might be degrading if the light illumination is too strong. It can be detected at periodically changing light [Fig. 6].

The results of this work will be useful for further optimization of different parameters for increasing photopotential from systems based on membrane proteins.

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*Corresponding author: kuklin@nf.jinr.ru