S3. Energy transformation mechanisms. Bioenergetics. Molecular motors

S3.212. Artificial photosynthesis - a promising energy solution

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Fossil fuels are non-renewable, inefficient, and environmentally unfriendly. The use of alternative renewable energy sources is increasing. Natural photosynthesis is a proven for thousands of years example of alternative energy efficiency, producing organic compounds and O2, and under certain conditions H2, from water and CO2 at the expense of solar energy. Burning H2 provides the maximum energy among other fuels and water, an environmentally friendly product. Production of H2 by artificial photosynthesis systems is a promising and high priority. Solar energy can be converted into electricity (in solar cells) or used in systems generating H2 from water. We are investigating the creation and operation of solar energy converters based on phototroph components to produce environmentally friendly energy. We created an original setup to analyze the operation of solar cells based on photosynthetic systems in a wide range of temperatures and light intensities. We have obtained original data on the "work" of solar cells capable of generating photocurrents, based on different components of the photosynthetic apparatus, including thylakoids and photosystem II membranes immobilized on the surface of titanium dioxide under different conditions. Particular attention is paid to the search for efficient catalysts for water oxidation, since such catalysts are key components of solar cells producing molecular hydrogen from water in the light. We found that the most effective catalyst for water oxidation under artificial photosynthesis conditions is a manganese-containing complex. To produce photohydrogen, we modified photosystem I (PSI) in which the secondary electron acceptor, vitamin K, was replaced with platinized naphthoquinone (PtNP), which increases the efficiency of electron transfer to the electrode. With this modification of PSI, it was possible to create an artificial system capable of efficiently generating molecular hydrogen at the expense of light energy.

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S3.213. Changes in mitochondrial membrane potential levels in respiratory chain dysfunction caused by mtDNA mutations

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Introduction

Mutations of mitochondrial DNA can have various effects on cell bioenergetics, in particular on the mitochondrial membrane potential (ΔΨm), decrease or increase of which negatively affects cell viability and can cause pathology development [1]. A decrease in ΔΨm leads to matrix condensation and cytochrome c entry into the intermembrane space, triggering apoptosis [2]. An increase in ΔΨm leads to the hyperproduction of reactive oxygen species, which cause damage not only in mitochondria but in the cell as a whole [3].

The aim of the present study was to determine the effect of mitochondrial DNA mutations on the change and mechanism of ΔΨm maintenance.

Materials and methods

The objects of the study were cytoplasmic hybrid cell lines based on THP-1 cells, each with 5 to 10 mtDNA mutations.

The ΔΨm value as well as the state of mitochondrial FAD were studied using a ZEISS LSM 900 confocal microscope. To assess ΔΨm levels, cells were incubated in 25 nM TMRM solution in Hanks' medium for 45 min at 37 °C without washing. TMRM fluorescence intensity over time was recorded by recording the baseline signal followed by addition of CCCP (2 μM). The value of ΔΨm was estimated from the change in fluorescence intensity. Total mitochondrial content, FAD reduction rate, and FADH2/FAD ratio were estimated from autofluorescence using an excitation wavelength of 488 nm. CCCP (2 μM) and sodium azide (10 mM) were used to convert the coenzyme to a fully oxidized or reduced form, respectively. The state of NADN was investigated using a wide-field fluorescence microscope with a fluorite x20 immersion objective using excitation radiation from a xenon arc lamp. Autofluorescence was recorded in the wavelength range of 430-480 nm using excitation radiation at 340 nm. CCCP (10 μM) to maximize respiration and rotenone solution (10 mM) to block respiratory chain complex I were used to estimate mitochondrial content, NAD reduction rate, and NADH/NAD ratio. Numerical data are presented as (median [Q1;Q3], number of cells analyzed). The % values given are normalized to the median THP-1.

Results

The decrease in ΔΨm relative to THP-1 (100% [86%; 119%], N=45) in mutant cells of HSM1 (76% [55%; 108%], N=48) and MAM3 (72% [56%; 91%], N=60) lines may be the result of increased heteroplasmy mutation in the tRNALeu gene (44 and 32% respectively). The decrease in ΔΨm is explained by the fact that mutations in the tRNALeu gene disrupt tRNA conformation and stability and the efficiency of the aminoacylation reaction. Mutations of tRNAs can lead to disruptions in the translation mechanism and, consequently, to changes in mitochondrial protein synthesis, which leads to a decrease in the activity of respiratory chain complexes I and V [4-6]. However, MAM1 (120% [95%; 156%], N=44) and MAM2 (117% [76%; 257%], N=54) lines showed increased ΔΨm levels with a heteroplasmy tRNALeu gene mutation degree of 23 and 25%, respectively. This can be explained by the activation of compensatory mechanisms of ΔΨm maintenance, which may differ depending on the set of mutations and the level of heteroplasmy. In the case of the MAM2 line such a mechanism for maintaining ΔΨm is represented by a more active ETC complex II, as evidenced by a reduced FADH2/FAD ratio (0.66 [0.44; 1.99], N=48) and a high FADH2 formation rate (253% [121%; 372%], N=99). The cybrid line MAM1 showed a high FADH2/FAD ratio (2.65 [1.28; 3.91], N=36), reduced mitochondrial FAD content (70% [58%; 91%], N=59), and a low FADH2 formation rate (110% [56%; 156%], N=49), indicating reduced ETC complex II function. In this lineage, the mechanism of ΔΨm maintenance may be associated with a large contribution of ETC complex I (NADH/NAD (0.28 [0.18; 0.38], N=102)), which may be explained by a high heteroplasmy (68%) mutation in complex I subunit 5, which, according to literature, is negatively correlated with the development of atherosclerosis [7, 8].

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S3.214. Direct participation of hydrogen ions in ATP synthase F1-factor functioning

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An analysis of the experimental data closely related to the work of our research team shows that transmembrane-transferred hydrogen ions are involved in the work of the F1 factor of mitochondrial and chloroplast ATP synthases. After transmembrane transfer through the Fo protons are transferred to the F1-factor via the gamma subunit, which also transmits torque to the catalytic centers of ATP synthase. The existence of kinetic barriers for proton transfer on both sides of the inner mitochondrial membrane is also shown. Mechanical mixing of the interface during rotation of the ATP synthase rotor ensures lowering of the kinetic barrier for proton transfer from the membrane surface to F1-factor. A model has been proposed and discussed, according to which, during the operation of F1-Fo ATP synthase the mechanical movement of the gamma subunit in parallel with the conformational rearrangement of the active center of the enzyme (Boyer's scheme) provides the replacement of magnesium ion by hydrogen ions ensuring the effective displacement of the ATP molecule.

S3.215. Electrogenesis in the root environment of various lettuce varieties

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The occurrence of a potential difference in living systems is due to a complex of physicochemical processes that ensure the maintenance of an uneven distribution of ions at the cellular, tissue and organism levels. In the process of plant development, an electrical potential gradient arises along the entire organism, due to ion diffusion, concentration effects, and differences in the intensities of biochemical processes.
Bioelectrochemical systems based on electroactive processes in the root environment of plants and associated microorganisms - plant-microbial fuel cells - are a new promising environmentally friendly source of renewable energy. Although the possibility of practical use of bioenergetic resources has already been shown in many studies, the nature of electrogenesis, including its dependence on the genetically determined physiological characteristics of plants and their state during development, has yet to be revealed.
The purpose of this research was to study the dynamics of the potential difference formation in the root environment of various lettuce varieties.
Measurement of electrical characteristics was carried out by placing biocompatible corrosion-resistant electrode systems in the root environment, which provided surface electrical contact with the root and root zone. Potential difference changes were monitored using the Arduino hardware platform every 15 minutes during the entire growing season (28-32 days). The experiments were carried out under controlled conditions of the agrobiopolygon.
To reveal the role of plants in the formation of electrogenic reactions in the root environment, changes in the potential difference in a bioelectrochemical system containing a nutrient solution without plants (control) and with plants were measured using the lettuce variety Typhoon as an example. At the initial stage of the experiment, a potential difference of the order of 70–100 mV was observed in the nutrient solution, apparently due to differences in the concentrations of the nutrient solution components at the upper and lower electrodes. We can say that the nutrient solution acts as an analogue of the electrolyte in a galvanic cell. Over time, the voltage in the control cell decreased, most likely due to the leveling of concentrations. When growing plants, the potential difference, on the contrary, increased to ~200 mV and was stable throughout the entire growing season for lettuce. Probably, the increase in voltage in the bioelectrochemical system when plant objects are placed in it is associated with the development of the root system, the vital activity of rhizosphere microorganisms, the transport of mineral substances and, as a result, the intensification of diffusion processes.
To select the most promising plants in terms of obtaining electricity during the cultivation of plant products, a study was made of the electrophysiological properties of the following lettuce varieties, which differ in the efficiency of the photosynthetic apparatus: Solos F1, Chinese curly, Chinese red-green, Mercury, Dubrava, Ballet, Robin, Cockade. The plants were grown in peat soil (Agrobalt C, Russia) in the vegetative and irradiation installations developed by us under controlled conditions of the ARI agrobiopolygon.
The dynamics of the potential difference for the studied varieties was similar: an increase in values from ~200 mV from the beginning of the vegetation cycle to more than 300 mV by the 15th day and then stabilization was observed. The average value of the potential difference in the root environment-plant system was 281±32 mV for Solos F1, 221±42 mV for Chinese curly, 206±47 mV for Chinese red-green, 306±32 mV for Mercury, 291±35 mV for Dubrava, 289±27 mV for Ballet, 286±31 mV for Robin, 272±37 mV for Cockade.
The highest value of the potential difference in the root environment-plant system was typical for lettuce of the Mercury variety – it reached 430 mV. At the same time, the electrical characteristics of plants did not directly correlate with biomass indicators. The mass of the aerial parts of plants in one cell was 47.8±8.6 g for Solos F1, 48.1±13.5 g for Chinese curly, 58±15.3 g for Chinese red-green, 71±9.3 g for Mercury, 42.6±13.3 g for Dubrava, 113.1±25.8 g for Ballet, 54.6±11.6 g for Robin, 82±26.4 g for Cockade.
On the basis of the data obtained, ideas about the electrical characteristics in the root environment of various lettuce varieties were formed and the design of a phytofuel cell, a bioelectrochemical system based on electrogenic processes in the root environment-plants system, was proposed.
Thus, the possibility of using electrogenesis in the root environment-plant system as a new green source of electricity was shown. The potential for using the bioelectrochemical systems described above includes the provision of power supply to environmental sensors, light sources, wireless sensor networks, the Internet of things, phytomonitoring systems in natural conditions and protected ground, remote areas, partial power supply of plant life support devices in artificial agroecosystems.