**PRINCIPAL INVESTIGATOR COVER LETTER AND TERMS AGREEMENT**

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Re: **Biofuel Cells Based on Microbial Cells, Enzymes and Nanocomposite Materials for Powering Microelectronic Devices**

I, Principal Investigator (PI) Prof. **Evgeny Katz**, hereby acknowledge that I have submitted a proposal to *2021 U.S.-Ukraine Alternative Energy Research Competition RFP.* I will be collaborating with Prof. **Mykhailo Gonchar** of Institute of Cell Biology, NAS of Ukraine.

If awarded, I undertake this research in good faith and will uphold my portion of the collaborative work as proposed in the submission.

I attest that the information contained in this proposal is truthful and that it has been prepared with the full knowledge and consent of Ms. Shannon Robinson, leadership representative of Clarkson University.

I affirm that I have read and understand CRDF Global's policies and standards outlined within the *2021 U.S.-Ukraine Alternative Energy Research Competition RFP*, including CRDF Global's Plagiarism Policy[1]. I agree to adhere to CRDF Global's Plagiarism Policy, and understand that CRDF Global will not provide funding to an application in which plagiarism exists. If plagiarism is detected, penalties may be imposed up to and including my exclusion from this funding opportunity and barring my participation in future funding opportunities.

Principal Investigator Signature: professor Evgeny Katz — February 02, 2021 *Date*

Institution Leadership Representative Signature: — February 03, 2021 *Date*
Shannon Robinson
Associate Vice President for Research & Technology Transfer
/ Executive Director of Institutional Planning

[1] For more information, please see CRDF Global's Plagiarism Policy

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Re: **Biofuel Cells Based on Microbial Cells, Enzymes and Nanocomposite Materials for Powering Microelectronic Devices**

I, Principal Investigator (PI) Prof. **Mykhailo Gonchar**, hereby acknowledge that I have submitted a proposal to *2021 U.S.-Ukraine Alternative Energy Research Competition RFP.* I will be collaborating with Prof. **Evgeny Katz** from Clarkson University (Potsdam, NY, USA).

If awarded, I undertake this research in good faith and will uphold my portion of the collaborative work as proposed in the submission.

I attest that the information contained in this proposal is truthful and that it has been prepared with the full knowledge and consent of Prof. **Andriy Sibirny**, leadership representative of Institute of Cell Biology, NAS of Ukraine.

I affirm that I have read and understand CRDF Global's policies and standards outlined within the *2021 U.S.-Ukraine Alternative Energy Research Competition RFP*, including CRDF Global's Plagiarism Policy[2]. I agree to adhere to CRDF Global's Plagiarism Policy, and understand that CRDF Global will not provide funding to an application in which plagiarism exists. If plagiarism is detected, penalties may be imposed up to and including my exclusion from this funding opportunity and barring my participation in future funding opportunities.

Head of Department of Analytical Biotechnology,

Prof., Dr., DrSci _____ Mykhailo Gonchar February 01, 2021
Principal Investigator Signature *Date*

Director of Institute of Cell Biology, NAS of Ukraine,

Prof., Dr., DrSci _____ Andriy Sibirny February 01, 2021
Institution Leadership Representative Signature *Date*

[2]For more information, please see CRDF Global Plagarism Policy

## CRDF GLOBAL COVER SHEET

| GENERAL PROJECT INFORMATION | | | | |
|---|---|---|---|---|
| Project Title (*not to exceed 25 words*) | Biofuel Cells Based on Microbial Cells, Enzymes and Nanocomposite Materials for Powering Microelectronic Devices | | | |
| Amount Requested *(excludes cost-shares)* | Total | U.S. Sub-Team | Ukrainian Sub-Team | |
| | $72,000 | $15,000 | $57,000 | |
| Research Categorization[3] | Research Area | Sub-Research Area | Research Focus | |
| | 1. Natural Sciences | 1.6 Biological Sciences; 1.4 Chemical Sciences. | 1.6.4 Biochemistry and Molecular Biology; 1.4.5 Electrochemistry. | |
| Research Involves use of Human/Animal Subjects | None | Project Duration | 12 | |

| UKRANIAN SUB-TEAM | | | | | |
|---|---|---|---|---|---|
| **INSTITUTION INFORMATION** | | | | | |
| Institute Name | Institute of Cell Biology, NAS of Ukraine | | | Institution Type | Research Institute |
| Mailing Address | 14/16 Drahomanov str. | | | | |
| | Lviv | | 79005 | | Ukraine |
| **PRINCIPAL INVESTIGATOR INFORMATION** | | | | | |
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| Highest Degree | DrSci | | Biochemistry | | 2001 |
| Residency Status | Legal Citizen | | | | |
| **INSTITUTION LEADERSHIP REPRESENTATIVE INFORMATION** | | | | | |
| Name | Sibirny | Andriy | Andriyovych | Position/Title | Director, DrSci, Pofessor |
| E-mail | sibirny@cellbiol.lviv.ua | | Telephone # | +380322612108 | |
| Total number of Ukrainian sub-team members, including Ukrainian PI, graduate students, secondary collaborators | | | | | 8 |

[3] Please reference the CRDF Global Research Areas document found here: http://www.crdfglobal.org/sites/default/files/crdf-global-research-areas_updated-june-2015.pdf

| U.S. SUB-TEAM | | | | | |
|---|---|---|---|---|---|
| **INSTITUTION INFORMATION** | | | | | |
| Institute Name | Clarkson University | | | Institution Type | University/Academic |
| Mailing Address | Science Center, Department of Chemistry & Biomolecular Science, 8 Clarkson Ave | | | Institution Congressional District[4] | 21 |
| | Potsdam | NY | 13699 | | |
| **PRINCIPAL INVESTIGATOR INFORMATION** | | | | | |
| Last Name (surname) | Katz | First Name (Given) | Evgeny | Middle | N/A |
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| PI E-mail | ekatz@clarkson.edu | | Alternative E-mail *(optional)* | Email 2 | |
| Highest Degree | PhD | | Electrochemistry | | 1983 |
| U.S. Residency Status | Legal Citizen | | Gender | Male | |
| **INSTITUTION LEADERSHIP REPRESENTATIVE INFORMATION** | | | | | |
| Name | Robinson | Shannon | N/A | Title | Associate VP for Research & Technology Transfer |
| E-mail | srs@clarkson.edu | | Telephone # | 1-315-268-6475 | |
| Have you received a grant under a previous CRDF Global program or been a participant in a CRDF Global-funded workshop? | | | | No | |
| If yes, please list program and grant number or workshop title in the following text box | | | N/A | | |
| Total number of U.S. team members, including PI, students, and secondary collaborators | | | | | 3 |

[4] If you do not know your congressional district, please click on this link and search for your representative using your Institution's zip code.

## PROJECT ABSTRACT

*Should not exceed 350 words*

Biofuel cells (BFCs) are the novel bioelectrochemical devices that use living microorganisms or their enzymes to generate electricity via enzymatic oxidation of organic substrates, including industrial waste products. The aim of the project is a development of new microbial, enzymatic and combined BFSs based on cells of mutant yeast strains or selected mushrooms, as well as purified enzymes immobilized at electroconductive nanocomposite materials. Cheap technological byproducts (alcohols, formaldehyde, phenols) or waste compounds found in environment will be used as the "fuel" for the power production.

The scientific and technical objectives of the project are defined as follows: 1) cultivation of yeast and fungal cells; isolation and purification of alcohol oxidase and laccase; 2) synthesis and characterization of nanocomposite materials; 3) development of laboratory prototypes of microbial BFCs and their functional characterization; 4) construction and characterization of laboratory prototypes of enzymatic BFCs based on purified oxidoreductases and nanocomposites; 5) testing the biofuel cells for the power production for microelectronic chips.

The novelty of the project is based on the idea of combining the oxidoreductase-overproducing yeast and fungal cells and their enzymes with the mediator systems coupled with nanocomposite materials. The obtained nanocomposites will be used for various aims: as electron transfer mediators, matrices for immobilization of biocatalysts, and nanozymes (such as artificial peroxidases) for biocathode construction.

The innovation of the project is based on preparation of a small size (millimeter size) biofuel cell for powering microelectronic chips for various sensors and to provide self-powered operation by extracting power from the environmental sources in remote locations ("install-and-forget"). The sensing will be followed by wireless information transfer, also powered by the installed biofuel cell. The systems will find future applications for environmental monitoring and homeland security. The advantages of a biocatalytic system over lithium batteries will be particularly pronounced upon miniaturization of the power supply.

This project is based on already established collaboration between scientists of the Institute of Cell Biology, NAS of Ukraine and the Department of Chemistry and Biomolecular Science of Clarkson University in Potsdam (NY, USA). The scientist from Ukrainian team is already performing research at Clarkson University, USA.

## PROJECT NARRATIVE

*Should not exceed ten (10) pages. Text should be Arial font size 10 within 1-inch margins*

The search for new renewable energy technologies and approaches for the utilization of technogenic by-products (often toxic) are the main problems of modern science. In this regard, the development of biofuel cells (BFCs) based on microbial cells or enzymes capable of utilizing the by-products as substrates with the simultaneous generation of electricity can help to solve these two actual problems – cheap renewable energetics and environmental protection [Beretta *et al*., 2019; Das *et al*., 2014].

The aim of the project is a development of new microbial, enzymatic and combined BFSs based on cells of mutant strain of the yeast and the selected fungi (overproducers of laccase) and purified enzymes from these sources, immobilized onto electroconductive nanocomposite materials. As substrates of enzymatic reactions and primary energy sources, it is planned to use low-cost and often toxic technogenic by-products for which there is a problem of their utilization, in particular: methanol and formaldehyde – for the mutant producer of alcohol oxidase (AO) and phenol derivates – for the selected fungal producers of laccase. All the proposed mutant strains and classically selected producers of the corresponding oxidoreductases were constructed/selected at the Department of Analytical Biotechnology, Institute of Cell Biology, NAS of Ukraine. The mentioned above enzymes have useful biotechnological characteristics (high thermostability, high affinity for target substrates) but are currently commercially unavailable.

The novelty and innovative power of the project are based on idea of combining the mutant yeast or fungal cells and their enzymes with the new mediator systems (based on synthetic dyes, ferrocene, cyanoferrate derivatives, etc.), and nanocomposite materials coupled with mediator systems. The obtained nanocomposites will be used for various aims: as electron transfer mediators, as matrices for immobilization of biocatalysts and as nanozymes (such as artificial peroxidases) for biocathode construction. The important novelty is the functional integration of the BFCs producing electric power and microelectronic devices (sensor chips) consuming the produced power.

The aim of the proposed scientific project is a combination of fundamental science with applied biotechnology using advanced interdisciplinary approaches.

The scientific and technical objectives of the project are defined as follows:

1. Cultivation of the yeast and fungal cells; isolation and purification of oxidoreductases (alcohol oxidase and laccase) – promising biocatalysts for the construction of biofuel cells (BFCs);
2. Synthesis and characterization of nanocomposite materials;
3. Development of laboratory prototypes of microbial BFCs and study of their functional characteristics;
4. Construction of laboratory prototypes of enzymatic BFCs based on purified oxidoreductases and nanocomposites, and studying their operational parameters. The hybrid BFCs, which combine microbial cells and enzymes, will be also constructed and tested.
5. Construction of the small ($mm^2$) BFCs compatible with the size of microelectronic chips and activation of the microelectronic sensors with the power produced by the BFCs.

This project is based on collaboration between scientists of the Department of Analytical Biotechnology of the Institute of Cell Biology, NAS of Ukraine (Lviv, UA) and the Department of Chemistry and Biomolecular Science of Clarkson University in Potsdam (NY, USA). Scientific partners (UA and US) are carrying out innovative scientific research in the complementary fields. Both teams are high-level professionals in various fields, including microbiology, enzymology, nanotechnology, biotechnology, analytical chemistry, and bioelectrochemistry. Notably, the teams have a long-term collaboration and Dr. Oleh Smutok (UA collaborator) is presently performing research at Clarkson University (US team) [https://www.clarkson.edu/people/oleh-smutok].

Biofuel cells (BFCs) are the novel bioelectrochemical devices that use living microorganisms or their enzymes to generate electricity through enzymatic oxidation of organic substrates [Gamella *et. al*., 2018]. This innovative technology has a number of potential applications: generation of renewable energy, especially for powering electronic mini- and microdevices [Bollela *et al*., 2020], production of bio-hydrogen, wastewater treatment, water desalination, etc. [Logan *et al*., 2019].

Notably, comparing to the fuel cells based on metal catalysts, the BFCs are capable of converting chemical energy into electricity using biological catalysts. This new technology offers several advantages over traditional batteries, including the use of renewable and clean catalysts, the selectivity of the reaction, fuel flexibility, and the ability to operate at milder temperature [Ganzales–Solino & Di Lorenzo, 2018]. Taken together, all these advantages lead to an economically viable process. While traditional fuel cells designated as “low temperature” operate at approximately 80 °C, biofuel cells can operate in the range of 20 to 40 °C and at physiological pH. These properties make BFCs an interesting alternative especially for devices that need to operate at milder temperatures and conditions [Katz (Ed.) 2014]. Moreover, the variety of reactions that can be catalyzed by enzymes enables the use of various fuel types. BFCs are highly promising due to combination of desirable technological features, such as efficiency, selectivity and biocompatibility of the enzymes and therefore, they could be used to power implantable medical devices [Neifar *et al.,* 2018].

The BFCs operational principle is based on the catalytic oxidation of organic substrates by a biocatalyst, which produces the electrons that are transferred to the anode, generating an electric current. A biocatalytic cathode performs $O_2$ reduction, thus, closing the electric circle. A decisive influence on the BFCs performance is the efficiency of electron transfer from the substrate ("fuel") through biocatalysts to the electrode surface (anode and cathode) [Gunawardena *&* Fernando, 2008; Sayed *&* Abdelkareem, 2017]. There are two main types of BFCs – microbial and enzymatic. Although microbial BFCs are characterized by a low power density (up to 0.56 mW/cm$^2$ - [Ren *et al*., 2016], they exhibit high stability (up to several months); they are cheaper than enzymatic BFCs and capable of simultaneous utilization of different substrates. However, their widespread use is limited by a low current density caused by a slow electron transport through cell membranes to the anode. The enzymatic BFCs typically have a much higher current density, but capable only of partial oxidation of the fuel and have a limited shelf life (7-10 days) due to a low stability of the enzymes [Bullen *et al*., 2006].

**The main aim** of the current project is a construction of new microbial, enzymatic and combined BFCs on the basis of cells of mutant yeast strains and selected fungi – overproducers of oxidoreductases, as well as using purified enzymes from these sources, immobilized onto electroconductive nanocomposites. Low-cost and often toxic technogenic by-products, for which utilization is actually a problem, will be used as substrates of enzymatic reactions (energy sources). Therefore, the project intends to combine two scientific goals – cheap renewable energetics and environmental protection. The designed biofuel cell, after their characterization, will be tested for powering microelecronic sensor devices. For this purpose, we will specifically design small-size (a few mm$^2$) biofuel cells compatible by their size with the electronic chips.

The new BFCs on the basis of mutant cells of thermotolerant methylotrophic yeast *Ogataea polymorpha* – producer of alcohol oxidase (AO) and purified enzyme preparations of AO will be used for generation of electrical energy due to oxidation of methanol or/and formaldehyde – the toxic technogenic pollutants of wood, paint and the other industries. A peculiarity of cells of the mutant yeast strain of *O. polymorpha* C-105 (*gcr1 catX*) is an impairment of glucose catabolite repression and catalase deficiency [Gonchar *et al*., 1998], as well as the thermostability of the enzymes (including AO). AO is a FAD-containing octameric protein with a wide substrate specificity (oxidation of primary alcohols and formaldehyde), which uses oxygen as an electron acceptor. The oxidation of methanol catalyzed by AO and electrochemical current generation looks as follows:

1) $CH_3OH$ + **AO-**FAD → **AO-**$FADH_2$ + $CH_2O$ (enzymatic reaction);

2) **AO-**$FADH_2$ + 2 $mediator_{ox}$ → **AO-**FAD + 2 $mediator_{red}$ + $2H^+$ (mediator reaction);

3) 2 $mediator_{red}$ → 2 $mediator_{ox}$ + 2 $e^-$ (electrochemical reaction).

In general, this enzyme-electrochemical scheme is typical for the other BFCs based on oxidases with a difference in the nature of their cofactor, substrate, enzymatic product or mediator type.

To construct microbial BFCs we will use a novel nanotechnological approach – additional enrichment of the cells of *O. polymorpha* C-105 by the AO immobilized onto gold nanoparticles. The proposed approach for enrichment of yeast cells with the target enzyme significantly increases the local concentration of the enzyme inside the cells and, respectively, improves the efficiency of the biocatalytic energy conversion by increasing the current in the microbial BFCs [Karkovska *et al*., 2015].

For the BFCs based on the cells of selected fungi (laccase producers) as well as for the BFCs based on the purified enzymes, phenol derivatives (a toxic waste product from the chemical and pharmacological industries) will be used as a "fuel" source. Laccase is a copper-containing enzyme with a wide range of substrate specificity and can use oxygen as an electron acceptor releasing water as a by-product [Tominaga *et al*., 2014]. Laccase has a high redox potential (from 400 mV to 800 mV vs. Ag/AgCl reference electrode), making it a promising cathode biocatalyst [Mano *et al*., 2018; Ghosh *et al*., 2019]. The ability of laccase for direct electron transfer to electrode modified with gold nanoparticles, is the basis for the design of mediator-free enzymatic BFCs [Kavetskyy *et al*., 2019].

In the current project, electroconductive nanomaterials based on noble and transition metals or their hybrid forms [Holade *et al.*, 2014], as well as carbon materials (graphene derivatives [Koushanpour *et al*., 2016], carbon nanotubes and carbonated cellulose fibers) will be used for increasing the current density at the electrode. To improve the efficiency of the electron transfer in the BFCs, we plan to use nanocomposites – synthesized nanomaterials, additionally modified by inorganic and organic electron-transfer mediators (synthetic dyes, cyanoferrate derivatives, etc.) [Zhao *et al*., 2017]. For better permeability of the mediator through the cell wall, the permeabilization of the cells will be used. Carbon-based materials, modified with noble and transition metals (electrodeposited Pt, Pt nanoparticles, hybrid metal nanoparticles), including peroxidase-like nanozymes, are planned to be used to improve the BFCs performance [Stasyuk *et al*. 2020]. The main operational parameters (current and power density, maximum operating time, etc.) for the constructed laboratory prototypes of microbial and enzymatic BFCs will be investigated. Thus, the application of nanocomposites with covalently bound enzymes (AO or laccase) will allow us to significantly improve the catalysis rate, both of the anode and cathode reactions, as well as will increase the operating time of the

constructed BFCs up to several months [Gervasio *et al.* 2020]. So, the expected specific power densities of constructed enzymatic BFCs will be in the range from 1.2 to 2.2 $mW \cdot cm^{-2}$.

The original idea of usage of the catalase-deficient mutant cells of the yeast *O. polymorpha* C-105 as a cathode biocatalyst in microbial BFCs will be tested. The hydrogen peroxide secreted outside the cells could be reduced at the cathode. This allows replacing the catalase heat-producing reaction of $H_2O_2$ decomposition by a useful, current-generating process of hydrogen peroxide reduction at the cathode. As a catalyst for the reduction of $H_2O_2$ at the cathode, nanocomposites that exhibited nanozyme properties (artificial peroxidases) will be used. An alternative approach will be based on the use of immobilized microperoxidase (MP-11) as the efficient catalyst for the $H_2O_2$ reduction at the cathode.

The realization of the current project will allow combining original ideas, the novel technological approaches and experimental skills of the UA-team (specialists in the field of microbiology, enzymology, analytical biotechnology, nanotechnology) and US-group (specialists in the field of bioelectrochemistry, analytical chemistry, nanochemistry). The unique properties of the mutants and selected yeast cells, and fungi as well as of bionanocomposites will allow the construction of new microbial, enzymatic and hybrid BFCs with improved characteristics. The expected results of the project are beyond the current level of technology and therefore are innovative and transformative.

## Milestones

**Task 1. Cultivation of yeast and fungal cells; isolation and purification of oxidoreductases – promising biocatalysts for the construction of biofuel cells (BFCs)**

1.1 Screening of mushroom fungi by their ability to overproduce extracellular laccase in liquid cultures.

Screening of more than 25 strains of the mushrooms belonging to the genera *Trametes* and *Phallus*, available in the collection of the Institute (UA), for the level of extracellular laccase activity in liquid cultures *in vitro* will be performed. The best enzyme's overproducers will be selected for further investigation.

1.2 Cultivation of mutant yeast cells and selected mushrooms, overproducing the target oxidoreductases, alcohol oxidase and laccase.

The optimal conditions for the cultivation of the producers will be determined, ensuring the maximum yield of the target enzymes (composition of the culture medium, intensity of aeration, temperature, time of cultivation, *etc.*). To maintain the highest activity of the enzymes inside the cells (AO) or in extracellular culture (laccase), the biomass (or extracellular culture) of the organisms will be frozen or lyophilized under optimal conditions.

1.3 Isolation, purification, enzymatic and electrochemical characterization of the enzymes: alcohol oxidase (AO) and laccase.

Enzymes, isolated from the mutant yeast cells and the culture medium of the selected fungi will be concentrated and/or purified using ion exchange and/or affinity chromatography. Purified preparations of AO and laccase will be investigated using different approaches: electrophoretic (to test their purity), spectrophotometric (to determine kinetic parameters – $K_M$ and $k_{cat}$) and electrochemical (activity toward electron-transfer mediators, determination of redox potential).

**Task 2. *Synthesis and characterization of nanocomposite materials***

2.1. Synthesis of nanomaterials based on noble and transition metal nanoparticles by chemical reduction.

The nanosized materials of noble and transition metals or their hybrid forms (which will consist of 2-3 different noble or transition metals) will be obtained using chemical reduction from the corresponding ions. Citrate, ascorbate or sodium borohydride will be used as reducing agents in optimal ratios of reactants.

2.2. Carbon-based nanomaterial screening.

The different carbon materials (graphene derivatives, carbon nanotubes, carbonized lignocellulose fibers and their composites) will be characterized by cyclic voltammetry for testing their electrochemical performance in connection to the biocatalytic processes.

2.3. Synthesis of nanocomposite materials with high efficiency of electron transfer from biocatalyst to electrode and characterization of their electron-mediator activity.

To improve communication between the biocatalyst and the electrode, screening and additional modification of the synthesized nanomaterials by inorganic electron transfer mediators (synthetic dyes, electrically conductive films, ferrocene, cyanoferrate derivatives, etc.) will be carried out. The electrochemical characteristics of the obtained nanocomposite materials and their compatibility with BFC biocomponents will be investigated. For enzymatic BFCs, based on laccase, optimization of the methods for direct mediator-less transfer of electrons *via* the nanocomposites will be carried out.

2.4. Screening and synthesis of new nanocomposite materials possessing nanozyme (nanoperoxidase) activity.

In order to create the novel BFC cathodes, the screening and synthesis of new nanomaterials with nanozyme (peroxidase) activity will be performed. The best selected nanozymes will be tested for their ability to reduce hydrogen peroxide at the electrode. The catalytic activity of the nanozymes will be compared to the process catalyzed by the microperoxidase (MP-11). The best catalytic species will be used in the next experimental steps.

**Task 3.** ***Design of laboratory prototypes of microbial BFCs and study of their functional characteristics***

3.1. Design of BFC laboratory models based on mutant yeast and selected fungal cells producing AO and laccase oxidoreductases.

The anode of microbial BFC will be modified with selected nanocomposite materials with the best electron-mediator activity. Carbon electrodes modified with different types of Pt (electrodeposited Pt, Pt nanoparticles, platinum black) are planned to be used as the cathode material. The electrodes with immobilized laccase or nanozymes will also be used as cathodes. Selection of the optimum composition of the anolyte (fuel substrate, buffer, cell concentration, *etc.*) will be performed. To provide more efficient communication between cellular redox systems and the electrode surface, yeast and fungal cells will be used in a permeabilized state. The optimal type of separator membrane will be selected and the possibility of using single chamber BFC will be studied.

3.2. Investigation of the functional characteristics of microbial BFCs.

The functional characteristics of engineered microbial BFCs will be evaluated by the use of inexpensive organic substrates and some by-products, for which there is a problem of remediation: methanol, formaldehyde – for the AO-over-producing mutant yeast strain and phenolic compounds – for the fungal producers of laccase. The enzymatic kits, developed in the laboratory, will be used to study the kinetics and utilization level of the relevant substrates in the BFC system. The possible advantages of the constructed laboratory models of new microbial BFCs based on nanocomposites and enzyme-producing cells will be investigated (high current and power density, long operating time).

When creating microbial BFCs, the original idea of using catalase-free mutant cells of the yeast *O. polymorpha* C-105 as a cathode biomaterial will be tested; in this case, hydrogen peroxide secreted by the yeast cells will be reduced at the cathode. This allows to replacing the catalase-mediated heat-producing reaction of $H_2O_2$ decomposition by a useful, current-generating process of the cathodic reduction of hydrogen peroxide. As a catalyst for the reduction of $H_2O_2$ at the cathode, nanocomposites will be used that exhibit the nanozyme (artificial peroxidase) activity.

**Task 4.** ***Design of laboratory prototypes of enzyme BFCs and hybrid ones on the basis of purified oxidoreductases, microbial cells, enriched by AO and laccase, and nanocomposites and study of their operational parameters***

4.1. Investigation of the possibility of using selected nanomaterials as an immobilization matrix for covalent or physical binding of biocatalysts on the electrode surface.

In order to ensure a high local concentration of the BFC biocatalyst and to increase its stability, screening for covalent and physical immobilization of the purified preparations of AO and laccase on the electrode surface will be carried out. The nanosized carriers will be functionalized by carboxyl and amino groups using appropriate reagents (ω-mercaptohexadecanoic acid, cysteamine – depending on chemical nature of nanomaterial) followed by covalent immobilization of the corresponding enzymes.

4.2. Screening of fixing polymer membranes of different nature.

Screening of various polymeric membranes (electrodeposited, photopolymeric, dialysis) will be carried out for their ability to reliably retain enzymes on the surface of the electrodes, a good electrical conductivity and a high permeability for enzymes' substrates.

4.3. Investigation of the basic operational parameters of enzyme-based BFCs.

The basic operational parameters will be investigated for the designed laboratory prototypes of enzymatic BFCs, based on AO and laccase: current density, power density and maximum operating time. The most effective BFC variants will be selected and compared with existing enzyme-based analogues.

4.4. Development of hybrid cell-enzyme BFCs.

The possibility of creating hybrid cell-enzyme-based BFCs will be investigated. In such novel devices, immobilized alcohol oxidase, hydrogen peroxide-generating yeast cells additionally enriched by the target enzyme will be used as the anode catalytic materials. They will be placed in the biocathode compartment, and nanozyme (artificial peroxidase) will be used as a cathode to catalyze reduction of hydrogen peroxide (Fig.1-2).

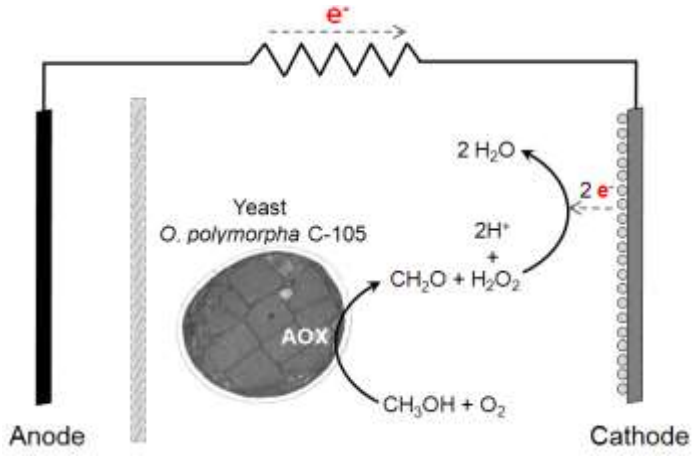


**Fig. 1**. The scheme illustrating the cathode process of reduction of $H_2O_2$, produced by mutant yeast cells.

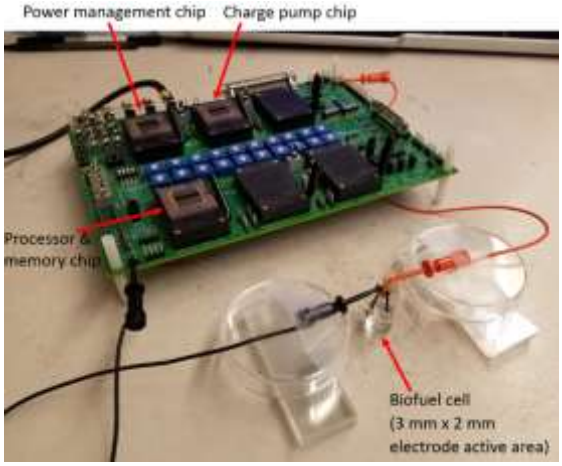


**Fig. 2.** The photo showing an example of the electronic microchip powered by a small size biofuel cell. A similar setup will be used in the proposed project when the new biofuel cell is integrated with a sensor microelectronic chip.

Ukrainian and USA partners (UA and US) have all the necessary specialized equipment to carry out the planned research under the current project.

**UA** has the material and hardware support for the cultivation of the yeast and fungal cells (culture media, sterile boxes, autoclaves, thermostats, thermostatic shakers, disintegrators, centrifuges "Sorvall", "Eppendorf", lyophilizer "Christ alpha 1-2 LDplus"; the equipment for isolation, purification of enzymes and study of their physicochemical and kinetic characteristics (chromatographic columns, chromatographic carriers, apparatus for vertical electrophoresis of proteins VE-2M Helicon, peristaltic pumps, fraction collectors, refrigerators, spectrophotometers "SHIMADZU UV-1650", fluorometer "Quantech filter"); equipment for synthesis and characterization of micro- and nanoparticles (atomic force microscope "Solver P47-PRO (NT-MDT)", scanning electron microscope "SEM-microanalyser REMMA-102-02", transmission electron microscope "REM-100", fluorescence microscope "Axio Lab. A1", optical microscopes); apparatus for conducting electrochemical studies (commercial electrodes of different configuration and different surface area values, electrochemical cells, potentiostats "CHI 1200A" and "PGstat16"; potentiostat/galvanostat MTech SPG-500L, enzymatic kits for methanol, formaldehyde and phenols assay to evaluate the fuel substrate consumption in BFCs.

**US** has a high-tech modern equipment for the design and operation of BFCs: Electrochemical equipment: Potentiostat/Galvanostat (Autolab, ECO Chemie) PGSTAT12 (two sets) – with additional modules (FRA2 impedance measurements module; BIPOT bipotentiostat module; ADC750 fast sampling module for fast chronoamperometric measurements; SCAN-GEN fast potential scanning module for ultra-fast cyclic voltammetry; ECD module for low current measurements on nanoelectrodes; FI20 filter and integrator module for chronocoulometric measurements; Rotating disk-ring electrode setup); Surface plasmon resonance instrument (Autolab, ECO Chemie) SPRINGLE connected with the potentiostat/galvanostat for in situ electrochemical-SPR measurements; Quartz crystal microbalance (QCM) (Autolab, ECO Chemie) connected with the potentiostat/galvanostat for in situ electrochemical-QCM measurements; Optical Contact Angle and Surface Tension Meter (KSV Instrument Ltd) Modular CAM 200 connected with the potentiostat/galvanostat for in situ electrochemical-contact angle measurements; Orion 3-Star pH Meter; Keithley 236 Source-Measure Unit; ELISA instrument; SpectraMax i3x Microplate Reader (Molecular Devices, LLC., CA, USA). Optical/biochemical equipment: Shimadzu Spectrophotometer UV-2401PC; Barnstead Nanopure Diamond Lab Water System; Refrigerated Universal Centrifuge (Labnet) Z300K; Clinical and Educational Incubator (Barnstead); Excellence XS Analytical Balance (Mettler). Available shared equipment (University facilities): AFM, STM, SAXS and WAXS techniques, HPC, SLS, DLS, particle size analyzers, high resolution SEM, TEM, NMR, Perkin Elmer LS 50B Fluorescence spectrometer, and mass spectrometer for proteomic analysis, Leica TCS SP5 II Laser Scanning Confocal Microscope, dynamic light scattering instrument (Malvern Zetasizer Nano), etc. US team has a great experience in developing small BFCs for powering electronic mini- and microdevises (Fig. 2).

**The list of project authors/participants:**

**US-Team:**

Prof. Evgeny Katz;
Dr. Oleh Smutok;
MS Daniel Massana (PhD student)

**UA-Team:**

Prof. Mykhailo Gonchar;
PhD Nataliya Stasyuk;
PhD Galina Gayda;
PhD Olha Demkiv;
PhD Andriy Zakalskiy;
PhD Tetyana Prokopiv;
MS Oksana Zakalska;
MS Bohdan Pshoniuk.

## REFERENCES CITED

*This section must only include bibliographic citations and not be used to provide parenthetical information outside of the Project Narrative*


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**STUDENT/POST-DOCTORAL RESEARCHER MENTORING PLAN**
*This should not be used to circumvent the Project Narrative page limit*

**Ukrainian Sub-Team Participant**

A young researcher Bohdan Pshoniuk graduated with a Master Degree from the Stepan Gzhytskyi National University of Veterinary Medicine and Biotechnologies in 2020. We plan to expand his level of knowledge and experience in related fields, namely, Molecular Biology and Analytical Biotechnology. We encouraged Bohdan to attend lectures at the Ivan Franko National University of Lviv and the Institute of Cell Biology, NAS of Ukraine for additional training courses: Genetic Engineering, Bioinformatics, Molecular Biology, and a special training course on Microbial Protein Production. Attending by the young researcher new disciplines will ensure his personal and professional growth. He will receive a thorough knowledge in modern protein bioengineering: selection of protein overproducers, immobilization of the enzymes to create bioreactors, acquaintance with modern fundamentals of protein engineering, approaches to obtaining artificial, semi-synthetic proteins.

Publications and presentations are expected to result from the work supported by the grant. These will be prepared under the direction of Prof. Gonchar M. and in collaboration with researchers from his team in the Institute of Cell Biology. Bohdan Pshoniuk will receive guidance and training in the preparation of manuscripts for scientific journals and presentations at conferences. Bohdan Pshoniuk will be encouraged to travel to attend scientific meetings and conferences, and to present his research in the form of seminars or poster presentations. It will promote his social activity and formation of high efficiency and a responsible attitude to the educational and scientific process. Bohdan will receive appropriate social and psychological counseling and support from the Department staff if necessary.

Technology Transfer activities will include regular contact with researchers in the Institute of Cell Biology. Young researcher Bohdan Pshoniuk will be given an opportunity to become familiar with the university-industry relationship including applicable confidentiality requirements and preparation of invention disclosure applications.

All this will help Bohdan in participation in research projects of the Department of Analytical biotechnology, Institute of Cell Biology, and will be a prerequisite for continuing his studies as a PhD student for his future career.

**STUDENT/POST-DOCTORAL RESEARCHER MENTORING PLAN**

*This should not be used to circumvent the Project Narrative page limit*

**US Sub-Team Participants**

As a group, we have been very successful in the mentorship of junior researchers, as indicated by their successful placement. Dr. Marcos Pita, the first postdoc in Dr. Katz's group at Clarkson University, has received a tenure-track position in Spain, after being rated No.1 on the national competition list, later he received tenure and promotion to the next academic level. Another postdoc, Dr. Vera Bocharova, received the prestigious Wigner scholarship in Oak Ridge National Laboratory; she is presently a head of a research group. Dr. Jan Halámek received a tenure position of the Director of the Forensic Science Institute at the Texas Tech University. Dr. Roberto Luz returned back to Brazil where he received a tenure-track professor position. The most recent postdoc, Dr. Maria Gamella, returned back to her home country (Spain) where she received an academic position. A few more postdocs found jobs in industry in the USA. Dr. Nataliia Guz is in the process of getting an academic position after 2-year maternity leave. In the last five years, seven Ph.D. graduates from Dr. Katz's group received postdoc positions in universities and government labs, or industry jobs.

Our mentorship of junior researchers has included research guidance, and emphasizing independent creativity balanced in the framework of a group effort. In addition, all our students and postdocs receive support and advisement in preparing their CVs, web sites, job searches, career stages, job market environment and networking. This is accomplished in group and individual discussions. Presentations are scheduled in seminar/interview formats and then critiqued. We even discuss interview behavior and dress code.

Junior researchers are also exposed to our network of colleagues and seminar visitors, as well as attend and present at conferences. The former research networking has included joint work and co-authorship with leading researchers and their groups locally and at other institutions: most recently, with Prof. Joseph Wang (UCSD; http://joewang.ucsd.edu/), Prof. Sergiy Minko (University of Georgia; http://nsmlab.com/), Prof. M.J. Schöning (Aachen University of Applied Sciences, Germany; https://www.fh-aachen.de/menschen/schoening/), Prof. D.M. Kolpashchikov (University of Central Florida; https://sciences.ucf.edu/chemistry/people/kolpashchikov-dmitry/), and many others (https://webspace.clarkson.edu/~ekatz/Collaborators.html). Generally, our publication record is in top journals, and high citation of articles (https://webspace.clarkson.edu/~ekatz/publications_most_cited.htm) has facilitated career development of graduate students and postdocs. Particularly, numerous highlights on our research on implantable bioelectronics and biosensors, including highlights in *Nature*, *Science*, *Scientific American*, *New Scientist*, *New York Times*, as well as in various magazines, radio and TV programs have encouraged students and postdoctoral scholars.

Dr. Nataliia Guz (from the Katz group) received an award for the best poster presented at the conference: "Soft Magnetic Materials and Systems",

Postdoctoral researchers and graduate students of Dr. Katz have received numerous awards (including those from ACS, ECS, NYS and others) for research and presentation quality.

Specifically, with postdoctoral researchers, we have also encouraged development of communication and management skills by mentoring graduate and undergraduate students, and sharing leadership roles in the team, as well as preparing presentations, publications, and occasionally helping write grant proposals.

**KEY PARTICIPANT INFORMATION FORM**
*Complete ONE for each participant involved*

| **TEAM MEMBER INFORMATION** | | | | Ukrainian Sub-Team Participant | |
|---|---|---|---|---|---|
| Last Name (surname) | Gonchar | First Name (Given) | Mykhailo | Middle | Vasylyovych |
| Current Position | Head of Department of Analytical Biotechnology | | Classification on Project | | Researcher/Engineer |
| Institute Name | Institute of Cell Biology, NAS of Ukraine | | | | |
| Complete Mailing Address | 14/16 Drahomanov str. | | Lviv | 79005 | Ukraine |
| E-mail Address | mykhailo1952@gmail.com | | Telephone # | | +380322612144 |
| Highest Degree/ Year Awarded | DrSc | | Biochemistry | | 2001 |
| Gender | Male | | | | |

**Description of project role** (responsibilities, expertise, level of effort on project):

Principal investigator, supervision of the UA-team research, preparation of scientific reports and international publications. Representation of the results on the international conferences and forums.

| **TEAM MEMBER INFORMATION** | | | | Ukrainian Sub-Team Participant | |
|---|---|---|---|---|---|
| Last Name (surname) | Gayda | First Name (Given) | Galina | Middle | Zufarivna |
| Current Position | Senior Researcher | | Classification on Project | | Researcher/Engineer |
| Institute Name | Institute of Cell Biology, NAS of Ukraine | | | | |
| Complete Mailing Address | 14/16 Drahomanov str. | | Lviv | 79005 | Ukraine |
| E-mail Address | galina.gayda@gmail.com | | Telephone # | | +380322612144 |
| Highest Degree/ Year Awarded | Dr | | Bioorganic chemistry | | 1983 |
| Gender | Female | | | | |

**Description of project role** (responsibilities, expertise, level of effort on project):

Coordination and realization of experiments focused on microbiology (screening the fungal producers of laccase) and enzymology (isolation, purification and enzymatic characterization of alcohol oxidase, laccase). Biofunctionalyzation of the nanocarriers by purified enzymes for the construction of biocatalysts for biofuel cells. Screening of new nanocomposite materials possessing nanozyme (nanoperoxidase) activity. Participation in preparing scientific reports and international publications.

| TEAM MEMBER INFORMATION | | | | Ukrainian Sub-Team Participant | |
|---|---|---|---|---|---|
| Last Name (surname) | Zakalskiy | First Name (Given) | Andriy | Middle | Yevstakhovych |
| Current Position | Researcher | | Classification on Project | | Researcher/Engineer |
| Institute Name | Institute of Cell Biology, NAS of Ukraine | | | | |
| Complete Mailing Address | 14/16 Drahomanov str. | | Lviv | 79005 | Ukraine |
| E-mail Address | andriy.zakalskiy@yahoo.com | | Telephone # | | +380322612144 |
| Highest Degree/ Year Awarded | Dr | | Biochemistry | | 1985 |
| Gender | Male | | | | |

**Description of project role** (responsibilities, expertise, level of effort on project):

Cultivation of mutant yeast cells and selected fungi, overproducing the target oxidoreductases. Participation in construction of laboratory prototypes of microbial biofuel cells and studying their functional characteristics.

| TEAM MEMBER INFORMATION | | | | Ukrainian Sub-Team Participant | |
|---|---|---|---|---|---|
| Last Name (surname) | Prokopiv | First Name (Given) | Tetyana | Middle | Markyanivna |
| Current Position | Researcher | | Classification on Project | | Researcher/Engineer |
| Institute Name | Institute of Cell Biology, NAS of Ukraine | | | | |
| Complete Mailing Address | 14/16, Drahomanov str. | | Lviv | 79005 | Ukraine |
| E-mail Address | tetyanaprokopiv@gmail.com | | Telephone # | | +380322612144 |
| Highest Degree/ Year Awarded | Dr | | Microbiology | | 2013 |
| Gender | Female | | | | |

**Description of project role** (responsibilities, expertise, level of effort on project):

Screening and modification of the synthesized nanomaterials by electron transfer mediators (synthetic dyes, electroconductive films, ferrocene, cyanoferrate derivatives, etc.). Screening of various polymeric membranes (electrodeposited, photopolymeric, dialysis) with an ability to reliably retain enzymes on the surface of the electrodes, possessing a good electrical conductivity and a high permeability for enzymes' substrates. Participation in the design of new enzymatic biofuel cells and studying their operational parameters.

| TEAM MEMBER INFORMATION | | | | | Ukrainian Sub-Team Participant |
|---|---|---|---|---|---|
| Last Name (surname) | Stasyuk | First Name (Given) | Nataliya | Middle | Yevgenivna |
| Current Position | Researcher | | Classification on Project | | Researcher/Engineer |
| Institute Name | Institute of Cell Biology, NAS of Ukraine | | | | |
| Complete Mailing Address | 14/16 Drahomanov str. | | Lviv | 79005 | Ukraine |
| E-mail Address | stasuk_natalia@ukr.net | | Telephone # | | +380322612144 |
| Highest Degree/ Year Awarded | Dr | | Physical Chemistry | | 2013 |
| Gender | Female | | | | |

**Description of project role** (responsibilities, expertise, level of effort on project):

Synthesis of nanocomposite materials with a high efficiency of electron transfer from biocatalyst to electrode and characterization of their electron mediator activity. Structural and morphological characterization of the obtained nanomaterials. Testing the possibility of using selected nanomaterials as an immobilization matrix for covalent or physical binding of biocatalysts on the electrode surface. Participation in the design of laboratory prototypes of enzymatic and hybrid biofuel cells and studying their operational parameters.

| TEAM MEMBER INFORMATION | | | | | Ukrainian Sub-Team Participant |
|---|---|---|---|---|---|
| Last Name (surname) | Demkiv | First Name (Given) | Olha | Middle | Mykhailyvna |
| Current Position | Researcher | | Classification on Project | | Researcher/Engineer |
| Institute Name | Institute of Cell Biology, NAS of Ukraine | | | | |
| Complete Mailing Address | 14/16 Drahomanov str. | | Lviv | 79005 | Ukraine |
| E-mail Address | demkiv@yahoo.com | | Telephone # | | +380322612144 |
| Highest Degree/ Year Awarded | Dr | | Microbiology | | 2013 |
| Gender | Female | | | | |

**Description of project role** (responsibilities, expertise, level of effort on project):

Screening of strains of the mushrooms for their synthesis of extracellular laccase in liquid cultures. Isolation, purification, enzymatic and electrochemical characterization of the laccase from the best enzyme producers. Synthesis of nanocomposite materials with a high efficiency of electron transfer from biocatalyst to electrode and characterization of their electron mediator activity. Participation in construction of laboratory prototypes of microbial biofuel cells and studying their functional characteristics.

| TEAM MEMBER INFORMATION | | | | Ukrainian Sub-Team Participant | |
|---|---|---|---|---|---|
| Last Name (surname) | Zakalska | First Name (Given) | Oksana | Middle | Myroslavivna |
| Current Position | Engineer | | Classification on Project | | Technical/Scientific Support |
| Institute Name | Institute of Cell Biology, NAS of Ukraine | | | | |
| Complete Mailing Address | 14/16 Drahomanov str. | | Lviv | 79005 | Ukraine |
| E-mail Address | zakalska@yahoo.com | | Telephone # | | +380322612144 |
| Highest Degree/ Year Awarded | MS | | Microbiology | | 1986 |
| Gender | Male | | | | |

**Description of project role** (responsibilities, expertise, level of effort on project):

Technical support in screening, cultivation, and stabilization of mutant and selected fungi - overproducers of laccas. Participation in the synthesis of nanocomposite materials for application in the construction of biofuel cells.

| TEAM MEMBER INFORMATION | | | | Ukrainian Sub-Team Participant | |
|---|---|---|---|---|---|
| Last Name (surname) | Pshoniuk | First Name (Given) | Bohdan | Middle | Vasylyovych |
| Current Position | Engineer | | Classification on Project | | Student |
| Institute Name | Institute of Cell Biology, NAS of Ukraine | | | | |
| Complete Mailing Address | 14/16 Drahomanov str. | | Lviv | 79005 | Ukraine |
| E-mail Address | psoniukbohdan@ukr.net | | Telephone # | | +380322612144 |
| Highest Degree/ Year Awarded | MS | | Biotechnology | | 2020 |
| Gender | Male | | | | |

**Description of project role** (responsibilities, expertise, level of effort on project):

Technical support in screening, cultivation, and stabilization of mutant and selected fungi - overproducers of laccas. Participation in the synthesis of nanocomposite materials for application in the construction of biofuel cells.

| TEAM MEMBER INFORMATION | | | | US Sub-Team Participant | |
|---|---|---|---|---|---|
| Last Name (surname) | Katz | First Name (Given) | Evgeny | Middle | N/A |
| Current Position | Full Professor | | Classification on Project | | Researcher/Engineer |
| Institute Name | Clarkson University | | | | |
| Complete Mailing Address | Science Center, Department of Chemistry & Biomolecular Science, 8 Clarkson Ave | | Potsdam, NY | 13699 | USA |
| E-mail Address | ekatz@clarkson.edu | | Telephone # | | 1-315-268-4421 |
| Highest Degree/ Year Awarded | PhD | | Electrochemistry | | 1983 |
| Gender | Male | | | | |

**Description of project role** (responsibilities, expertise, level of effort on project):

Project management, supervision of the US-team research, preparation of scientific reports and international publications. Representation of the results on the international conferences and forums.

| TEAM MEMBER INFORMATION | | | | US Sub-Team Participant | |
|---|---|---|---|---|---|
| Last Name (surname) | Smutok | First Name (Given) | Oleh | Middle | N/A |
| Current Position | Research Associate | | Classification on Project | | Researcher/Engineer |
| Institute Name | Clarkson University | | | | |
| Complete Mailing Address | Science Center, Department of Chemistry & Biomolecular Science, 8 Clarkson Ave | | Potsdam, NY | 13699 | USA |
| E-mail Address | osmutok@clarkson.edu | | Telephone # | | 1-315-268-2389 |
| Highest Degree/ Year Awarded | DrSc | | Microbiology | | 2019 |
| Gender | Male | | | | |

**Description of project role** (responsibilities, expertise, level of effort on project):

Planning, coordination, and realization of experiments related with electrochemistry (construction and characterization of microbial, enzymatic biofuel cells on the base of microbial cells, oxidoreductases, and nanocomposite materials). Preparation of scientific reports and international publications.

| TEAM MEMBER INFORMATION | | | | US Sub-Team Participant | |
|---|---|---|---|---|---|
| Last Name (surname) | Daniel | First Name (Given) | Massana | Middle | N/A |
| Current Position | PhD student | | Classification on Project | | Student |
| Institute Name | Clarkson University | | | | |
| Complete Mailing Address | Science Center, Department of Chemistry & Biomolecular Science, 8 Clarkson Ave. | | Potsdam, NY | 13699 | USA |
| E-mail Address | masim@clarkson.edu | | Telephone # | | 1-347-524-3426 |
| Highest Degree/ Year Awarded | BA | | Chemistry | | 2017 |
| Gender | Male | | | | |
| **Description of project role** (responsibilities, expertise, level of effort on project): | | | | | |
| Performing electrochemical experiments on modified electrodes. | | | | | |

**CRDF GLOBAL BUDGET NARRATIVE FORM**

*Describe and justify the expenses included in each budget line item. If a category doesn't apply to your budget, please insert N/A for "not applicable" in the space provided.*

| **Institution Name:** | **Institute of Cell Biology, NAS of Ukraine** | **Applicant type:** | **Primary Institution** |
|---|---|---|---|

**Individual Financial Support (IFS)**
Describe the level of effort projected for the PI and other team participants. Provide justification for pay rate and any fringe benefits included.

**Gonchar Mykhailo Vasylyovych** will work as a Principal Investigator/Project Director for this project and will commit 12 calendar months and dedicate 48 % of his working time during this period;
**Gayda Galina Zufarivna** will work as a Researcher for this project and will commit 12 calendar months and dedicate 45 % of her working time during this period;
**Zakalskiy Andriy Yevstakhovych** will work as a Researcher for this project and will commit 12 calendar months and dedicate 30 % of his working time during this period;
**Prokopiv Tetyana Markyanivna** will work as a Researcher for this project and will commit 12 calendar months and dedicate 30 % of her working time during this period;
**Stasyuk Nataliya Yevgenivna** will work as a Researcher for this project and will commit 12 calendar months and dedicate 48.5% of her working time during this period;
**Demkiv Olha Mykhailyvna** will work as a Researcher for this project and will commit 12 calendar months and dedicate 48.5% of her working time during this period;
**Zakalska Oksana Myroslavivna** will work as a technical/scientific assistant for this project and will commit 12 calendar months and dedicate 20 % of her working time during this period
**Pshoniuk Bohdan Vasylyovych** will work as a graduate research assistant for this project and will commit 12 calendar months and dedicate 17.2 % of his working time during this period.

**Equipment, Supplies and Services (ESS)**
Justify the purpose and cost rationale of each ESS line item included in the budget. General or non-descript line items such as "supplies" or "services" are not acceptable. Please itemize.

We request an amount of $2,276 for the General Supplies and Expendable Materials.
Planar "DropSens" electrodes (4 mm gold - $ 250.0, and 4 mm platinum - $ 250.0), glass carbon electrode ($ 231.0), gold screen printed electrode ($ 425.0), platinum screen printed electrode ($ 450.0), copper working electrode ($ 200.0), glass electrochemical cells ($ 250.0) and Amicon centrifugal filter unit ($ 220.0) will be used for construction of model biofuel cells. The price of each position of supplies and materials is calculated as the producer's catalog price including the costs for transportation to Institute.

**Travel**
Explain the need for travel - how the travel will benefit the project's aims - and your calculations of travel costs for domestic and foreign travel. Break down by airfare, hotel, per diem, etc.

We request $ 17494.0 for foreign travel from Lviv to the Clarkson University (Potsdam, NY, USA). DrSc Gonchar M., Dr. Stasyuk N., and Dr. Demkiv O. are suggested to attend U.S. PI laboratory for collaborative work. The financial calculation is based on $850 Amount airfare per person, $151 Amount International lodging/Per Diem rates per person for 31 nights, $160 Amount visa fee per person and $60 Amount medical insurance per person based on the U.S. Government allowances in effect at the time of travel.
We request $ 259.0 for domestic travel as Conference Registration Fees $ 200 and Other Travel Expenses $59.0 for attendance at the International Scientific Conference (Ukraine).

**Indirect Costs (IDCs)**
Justify indirect costs % of the total sub-team direct expenses requested. Indicate if a NICRA or other institutional IDC certification is applicable.

Indirect costs (10 %) will be used for the cover of communal payments, communication items (internet, telephone and mail), office and accounting expenses.

**Cost Share (optional)**
If applicable, describe the cost share that is being undertaken. Describe what items the cost-share will be applied to.

Not applicable

**CRDF GLOBAL BUDGET NARRATIVE FORM**

*Describe and justify the expenses included in each budget line item. If a category doesn't apply to your budget, please insert N/A for "not applicable" in the space provided.*

| Institution Name: | Clarkson University | Applicant type: | Primary Institution |
|---|---|---|---|

**Individual Financial Support (IFS)**
Describe the level of effort projected for the PI and other team participants. Provide justification for pay rate and any fringe benefits included.

Funding is requested for one Postdoctoral Research Associate to devote approximately 10% effort toward this project.

Funding is also requested for one part-time graduate student who will also devote approximately 20% effort toward this project.

Fringe benefits are charged on the Research Associate's salary at the anticipated rate of 29.1%.

**Equipment, Supplies and Services (ESS)**
Justify the purpose and cost rationale of each ESS line item included in the budget. General or non-descript line items such as "supplies" or "services" are not acceptable. Please itemize.

$1,050 is requested for the purchase of the lab consumables required for the completion of the proposed project.

**Travel**
Not applicable

Not applicable

**Indirect Costs (IDCs)**
Justify indirect costs % of the total sub-team direct expenses requested. Indicate if a NICRA or other institutional IDC certification is applicable.

Indirect Costs are calculated in accordance with Clarkson University's Indirect Cost Rate Agreement with the Department of Health and Human Services. Currently, Clarkson is using a rate of 53% of modified total direct costs (MTDC), per DHHS Agreement dated 03/11/20. If any rate other than the appropriate negotiated rate is applied to a project due to published program restrictions or sponsor policy, all unrecovered indirect costs are designated as cost sharing by Clarkson University. Clarkson's threshold for equipment is $5,000 as allowed by the Uniform Administrative Requirements, Cost Principles, and Audit Requirements for Federal Awards (UG 2 CFR § 200).

**Cost Share (optional)**
If applicable, describe the cost share that is being undertaken. Describe what items the cost-share will be applied to.

Not applicable

**CRDF Global PROJECT MILESTONE PLAN (TEMPLATE/ SAMPLE)**

*Copy template to complete. Information should match the proposal Project Narrative and Project Budget*

| **Reporting Period (Complete for each quarterly segment applicable top project duration.)** | | | **Responsible Team** | |
|---|---|---|---|---|
| ***First Quarterly Reporting Period*** | | | ***Mark all that apply*** | |
| Milestone: | Description: | Associated Deliverable(s): | U.S. Sub-Team | Ukrainian Sub-Team |
| Cultivation of yeast and fungal cells; isolation and purification of oxidoreductases – promising biocatalysts for the construction of biofuel cells (BFCs) | Over 25 strains of fungi will be screened for their ability to produce laccase, and prospective producers of this enzyme will be selected. For cells of mutant strains of the yeast and selected fungi (overproducers of laccase), the cultivation conditions will be optimized. Enzymes, isolated from the mutant yeast cells and the culture medium of the selected fungi will be concentrated and/or purified using ion exchange and/or affinity chromatography. Purified preparations of AO and laccase will be investigated using different approaches: electrophoretic spectrophotometric and electrochemical. | Scientific report | $3.750 | $8.500 |
| Total Amount Requested for this Reporting Period: | | $12,250 | $3.750 | $8.500 |
| ***Second Quarterly Reporting Period*** | | | ***Mark all that apply*** | |
| Milestone: | Description: | Associated Deliverable(s) | U.S. Sub-Team | Ukrainian Sub-Team |
| Synthesis and characterization of nanocomposite materials | The methods of synthesis and functionalization of metal nanoparticles will be optimized and more than 10 variants of mono- and bimetallic nanoparticles hybrids (based on noble and/or transition metals) will be obtained. Among newly synthesized nanomaterials, artificial peroxidases (nanoperoxidases) will be selected. Conductive carbon materials – derivatives of graphene, carbon nanotubes and carbonated cellulose fibers will be tested and characterized. For the obtained nanomaterials, their physicochemical, catalytic and electrochemical characteristics will be investigated. Screening and additional modification of the synthesized nanomaterials by inorganic electron transfer mediators will be carried out and the electrochemical characteristics of the obtained nanocomposite materials and their compatibility with the BFCs | Scientific report | $3.750 | $12.300 |

| | | | | |
|---|---|---|---|---|
| | biocomponents will be investigated. | | | |
| Total Amount Requested for this Reporting Period: | | $ 16,050 | $ 3.750 | $ 12.300 |
| ***Third Quarterly Reporting Period*** | | | ***Mark all that apply*** | |
| Milestone: | Description: | Associated Deliverable(s) | U.S. Sub-Team | Ukrainian Sub-Team |
| Design of laboratory prototypes of microbial BFCs and study of their functional characteristics | New laboratory prototypes of microbial BFCs will be constructed on the basis of mutant yeast and selected fungi cells, producing oxidoreductases, and their functional and operational characteristics will be investigated. | | $3.750 | $4.226 |
| Analysis of Experimental Data | Both teams will conduct analysis on data collected by two institutions to combine them for the next researching stages | Scientific report | - | $1.500 |
| Total Amount Requested for this Reporting Period: | | $ 13,450 | $ 3.750 | $ 5.726 |
| ***Fourth Quarterly Reporting Period*** | | | ***Mark all that apply*** | |
| Milestone: | Description: | Associated Deliverable(s) | U.S. Sub-Team | Ukrainian Sub-Team |
| Design of laboratory prototypes of enzyme BFCs and hybrid ones on the basis of purified oxidoreductases, microbial cells, enriched by AO and laccase, and nanocomposites and study of their operational parameters | New laboratory prototypes of enzymatic BFCs based on purified preparations of AO and laccase as well as nanoperoxidases and electron-mediated nanocomposites will be obtained and characterized. For BFCs, the possibility of creating the third-generation mediator-free enzyme bioelectrodes based on laccase in combination with nanomaterials will be investigated. | Scientific report | $3.750 | $4.225 |
| Travel to US Institution | Members of the Ukrainian team will visit their partner institution in the USA. The main objective is face-to-face interaction in providing joint experiments and future publications | Trip Report<br>Travel documents (copies of boarding passes, visa, other receipts)<br>Photos | - | $17.494 |
| Completion of manuscript to be submitted for publication and final CRDF-report | The teams will work to prepare a final manuscript to be submitted for publication in the peer-reviewed journal and Final CRDF-report | Copy of the manuscript<br>Final CRDF-report | - | $1.755 |
| Total Amount Requested for this Reporting Period: | | $ 30,250 | $ 3.750 | $ 23.474 |

**PI OTHER SOURCES OF SUPPORT FORM**

| **PI Name** | **Mykhailo Gonchar** | | |
|---|---|---|---|
| **If no other sources of support, check "None." Otherwise, complete table below for each source (duplicate as needed).** | | | ☐ **"None"** |
| | | | |
| **Project/Proposal Title** | Development of biosensors and biofuel cells based on redox enzymes | **Location of Research** | Ukraine/Lithuania |
| **Support** | √ Current ☐ Pending Submission Planned in Near Future | | |
| **Source of Support** | Ministry of Education and Science of Ukraine | **Level of Effort (%)** | 10 % |
| **Award Amount** | $ 8,000 | **Period Covered** | 03/20 – 12/22 |
| | | | |

**PI OTHER SOURCES OF SUPPORT FORM**

| **PI Name** | **Evgeny Katz** | | |
|---|---|---|---|
| **If no other sources of support, check "None." Otherwise, complete table below for each source (duplicate as needed).** | | | ☐ **"None"** |
| | | | |
| **Project/Proposal Title** | HFSP Collaborative Research Grant | **Location of Research** | Clarkson University |
| **Support** | √ Current ☐ Pending Submission Planned in Near Future | | |
| **Source of Support** | Human Frontier Science Program | **Level of Effort (%)** | 0% |
| **Award Amount** | $420,000 | **Period Covered** | 10/18 – 09/21 |
| | | | |
| **Project/Proposal Title** | EAGER: NSF-BSF: Quorum Biosensing using Magnetic Field-Activated Molecular Machines | **Location of Research** | Clarkson University |
| **Support** | √ Current ☐ Pending Submission Planned in Near Future | | |
| **Source of Support** | National Science Foundation | **Level of Effort (%)** | 0% |
| **Award Amount** | $150,000 | **Period Covered** | 08/19 – 01/21 |
| | | | |
| **Project/Proposal Title** | Wearable-implantable Sensors for Continuous Monitoring of Biomarkers in Animals and Humans | **Location of Research** | Clarkson University |
| **Support** | ☐ Current √ Pending ☐ Pending Submission Planned in Near Future | | |
| **Source of Support** | Queensland University of Technology (DOD-prime) | **Level of Effort (%)** | 0% |
| **Award Amount** | $200,000 | **Period Covered** | 09/20 – 02/22 |
| | | | |
| | | | |

**PREVIOUS CRDF GLOBAL AWARD FORM**

*Individual forms should not exceed one page.*

| **CRDF Global Award Number**: | UKB2-9044-LV-10 | | |
|---|---|---|---|
| Title of Previous Project: | Production of trial series and preparation of technical documentation for bioanalytical kit for enzymatic analysis of L-lactate in clinical diagnostics and beverage industries | | |
| Start Date | 05/2011 | End Date | 05/2012 |

Please briefly describe the previous research project. Be sure to provide specific information regarding results and objectives. Were all objectives of the research plan achieved? If not, what prevented you from doing so? Please list scientific publications and conference reports that were published as a result of CRDF Global award.

As a result of this CRDF-project, the optimization of a process of L-lactate cytochrome *c*-oxidoreductase (flavocytochrome $b_2$, FC $b_2$) production and its isolation have been performed. It was obtained preparation with 70 units of enzyme activity that allows produce 3 000 “Lactatest” kits. Optimization of composition parameters of the “Lactatest” (concentration of enzyme, chromogen, and buffer), storage form of the kit’s components and optimization of reaction conditions were done.

The manual (instruction) for using the proposed kit and description of laboratory technology to produce “Lactatest” were prepared according to the standards of Ukraine.

The trial series of the kit “Lactatest” was prepared and its testing on the real samples of food products (dairy products, juices, ketchups etc.) as well as human liquids was done. The analysis of L-lactate by the use of “Lactatest” in sweat samples obtained from the trained athletes was also performed. The correlation level for obtained results with the reference methods was done.

Several international publications with a high impact factor were published and acknowledgment of CRDF has been indicated.

The list of publications with affiliated CRDF-grant # UKB2-9044-LV-10:

1. Smutok O., Broda D., Smutok H., Dmytruk K., Gonchar M. (2011) Chromate reducing activity of the *Hansenula polymorpha* recombinant cells overproducing flavocytochrome $b_2$. ***Chemosphere***, *83*(4), 449-454 (IF - **3.613**). https://doi.org/10.1016/j.chemosphere.2010.12.078;

2. Smutok O., Gayda G., Dmytruk K., Klepach H., Nisnevich M., Sibirny A., Puchalski C., Gonchar M., Sibirny V. Amperometric Biosensors for Lactate, Alcohols, and Glycerol Assays in Clinical Diagnostics / in epy “**Biosensors - Emerging Materials and Applications**”, (2011), ISBN 978-953-307-328-6, pp. 401-446.

How will the work accomplished during this project contribute to the proposed research? Please address specific project results (data, models, methods) that the proposed project will further develop and/or build upon.

Due to the realization of the previous CRDF-project, a more close relationship between the scientific group of the Institute of Cell Biology, NAS of Ukraine and the company “Engineering Laboratory Ltd.” was established. On the base of acquired experience, it will be easier to introduce our innovations into the practice, especially, in the fields of amperometric biosensors’ and biofuel cell construction.

**PREVIOUS CRDF GLOBAL AWARD FORM**
*Individual forms should not exceed <u>one page.</u>*

| | | | |
|---|---|---|---|
| **CRDF Global Award Number**: | UKB1-9048-KV-10 | | |
| Title of Previous Project: | Development of technology for production of recombinant human arginase as anticancer enzyme and element of biosensor for arginine | | |
| Start Date | 05/2011 | End Date | 05/2012 |

Please briefly describe the previous research project. Be sure to provide specific information regarding results and objectives. Were all objectives of the research plan achieved? If not, what prevented you from doing so? Please list scientific publications and conference reports that were published as a result of CRDF Global award.

The objective of the previous CRDF-project was to develop an innovative technology for expression of recombinant human arginase in yeast host and its purification to homogenous state and to make this product commercially available. This enzyme is currently not commercially available but is of great interest for anticancer enzymotherapy (medicine) and as an element of novel biosensor for arginine (analytical biotechnology).

As a result of the project, we optimized the purification protocol for recombinant human arginase expressed in two species of yeasts, bakers' yeast *Saccharomyces cerevisiae* and methylotrophic yest *Hansenula polymorpha* to make it cost-efficient. We achieved production of the purified preparations of the recombinant arginase sufficient for the use in bioanalytical devices and for further manipulations to be used in animal studies and clinical trials (in PEGylated form).

Obtained in the frame of previous CRDF-project enzyme was used also for the other scientific investigations. As a result, some international publications with a high impact factor were published and acknowledgment of CRDF has been indicated.

<u>The list of publications with affiliated CRDF-grant # UKB1-9048-KV-10:</u>

1. Stasyuk N., Smutok O., Gayda G., Gonchar M., Koval'chuk. Y. (2011) A new bi-enzyme potentiometric sensor for arginine analysis based on recombinant human arginase i and commercial urease. ***Journal of Materials Science and Engineering: A***, *1*, 819-827;

2. Stasyuk N., Smutok O., Gayda G., Koval'chuk. Y., Vus B., Gonchar M. (2012) Bi-enzyme L-arginine-selective amperometric biosensor based on ammonium-sensing polyaniline-modified electrode. ***Biosens. & Bioelectron.***, *37*(1), 46-52 (IF - **5.63**) https://doi.org/10.1016/j.bios.2012.04.031

Zakalskiy A,, Zakalska O., Rzhepetskyy Y., Potocka N., Stasyk O., Horak D., Gonchar M. (2012) Overexpression of (His)(6)-tagged human arginase I in *Saccharomyces cerevisiae* and enzyme purification using metal affinity chromatography. ***Protein Expr Purif***., *81*, 63-68. (**IF – 1.64)** DOI:10.1016/j.pep.2011.09.001

How will the work accomplished during this project contribute to the proposed research? Please address specific project results (data, models, methods) that the proposed project will further develop and/or build upon.

The methods developed during this project will be valuable for the proposed project, namely, for isolation of the enzymes from cell extracts and their purification using ion exchange and affinity chromatography. There will be also used methods for stabilization of the enzymes and their immobilization on the electrodes, proposed in the earlier project.

## CRDF INSITUTIONAL DATA FORM (UKRAINE INSTIUTION)

| | | | |
|---|---|---|---|
| Institution Name | Institute of Cell Biology | | |
| Institutional Website | https://www.cellbiol.lviv.ua/ | | |
| Type of Organization | ☐ International Organization ☐ Government ☐ Corporation ☑ University | | |
| Small Business (US Orgs Only) | ☐ Small Business ☐ WOSB ☐ SDB ☐ HUB-Zone ☐ VOSB ☐ SDVOSB ☐ N/A | | |
| DUNS Number | [565754314] | TIN/EIN (US Tax Liability Only) | [TIN/EIN] |

Organizations must have a DUNS number to receive federal funding. For help applying for a DUNS number and more guidance on completing this form, please click here.

| **Non-Profit/Charitable Status (Non-US Organizations Only)** | | | |
|---|---|---|---|
| Is your organization registered as a charitable/non-profit entity? | | ☑ Yes | ☐ No |
| Country of Registration | Ukraine | Registration # | 1713044601024 |

| **Financial Controls & Audits** | | |
|---|---|---|
| Did your organization expend more than US $750,000.00 in U.S. Government Federal Funding (Grants, Contracts, Subgrants, Subcontracts) in the previous fiscal year? If yes, please provide a copy of your single audit report, which is required under 2 CFR 200. | ☐ Yes | ☑ No |
| Have you been audited in the past 3 years? **If yes, please send a copy of the current report.** | ☐ Yes | ☑ No |
| Were there any material or significant findings in the audit report? | ☐ Yes | ☑ No |
| Has your organization ever had a grant or contract terminated for cause? | ☐ Yes | ☑ No |
| Does your organization utilize a financial manual to authorize expenses? | ☑ Yes | ☐ No |
| Does your organization utilize an accounting system to track expenses? | ☑ Yes | ☐ No |
| Does your organization have an ethics policy? | ☐ Yes | ☑ No |
| Does your organization have a timekeeping system for labor such as timesheets? | ☑ Yes | ☐ No |
| Will your organization use an institutional bank account to receive funding? | ☑ Yes | ☐ No |
| Please identify the authorized institutional signatory for agreements:<br>Sibirny A.A., Prof., Dr., DrSci.<br>Director of Institute of Cell Biology, NAS of Ukraine | | |

| Executive/Management Reporting Requirements | |
|---|---|
| CRDF Global may be required to publicly report the names and total compensation of the five most highly compensated individuals at the awardees' institution. If you meet any of the criteria below, you are exempt from this requirement. Please find and check any applicable exemption: | |
| 1 In the previous tax year, institutional gross income from all sources was LESS than $300,000. | ☐ Exempt |
| 2 The institution received LESS than 80 percent of its annual gross revenues in U.S. federal funding (Contracts, Grants, Subgrants, Subcontracts or Loans). | V Exempt |
| 3 The institution received LESS than $25,000,0000 in annual gross revenues from U.S. federal funding sources (Contracts, Grants, Subgrants, Subcontracts or Loans). | V Exempt |
| 4 Executive compensation is publicly reported under section 13(a) or 15(d) of the Security Exchange Act or section 6104 of the Internal Revenue Code. | ☐ Exempt |
| I do not meet any of the exemptions above. I will provide the names and total compensation of the five most highly compensated executives. Click here for more information. | ☐ Not Exempt |

The information requested above must be provided in full and signed by an authorized institutional signatory, certifying that the information is true to the best of their knowledge. CRDF Global cannot proceed with an award to the institute without this information.

Past Performance

Please list any applicable grants or contracts received from outside organizations. Successful completion is defined as zero suspensions or terminations for cause, audit findings or other discrepancies.

| Funding Source | Agreement Number | Total Funding | Successful Completion? | Type of Project |
|---|---|---|---|---|
| NATO | CBP.NUKR.SFP 984173 | 59.500 € | V Yes ☐ No | Research Grant |
| EU-Poland | IPBU.03.01.00-18-452/11-00 | 47.555 $ | V Yes ☐ No | Research Grant |
| CRDF | UKB2-9044-LV-10 | 9000 $ | V Yes ☐ No | Research Grant |
| CRDF | UKB1-9048-KV-10 | 9000 $ | V Yes ☐ No | Research Grant |

The information requested above must be provided in full and signed by an authorized institutional signatory, certifying that the information is true to the best of their knowledge. CRDF Global cannot proceed with an award to the institute without this information.

Signature | Andriy Sibirny, Prof., Dr., DrSci. Name and Title | February 04, 2021 Date

Address Drahomanov Street, 14/16, Lviv 79005, Ukraine
Phone +38 032 261-21-48
E-Mail sibirny@cellbiol.lviv.ua

## CRDF INSITUTIONAL DATA FORM (U.S. INSTIUTION)

| | | | |
|---|---|---|---|
| Institution Name | Clarkson University | | |
| Institutional Website | https://www.clarkson.edu/ | | |
| Type of Organization | ☐ International Organization ☐ Government ☐ Corporation ☑ University | | |
| Small Business (US Orgs Only) | ☐ Small Business ☐ WOSB ☐ SDB ☐ HUB-Zone ☐ VOSB ☐ SDVOSB ☑ N/A | | |
| DUNS Number | 041590993 | TIN/EIN (US Tax Liability Only) | 15-0543659 |

Organizations must have a DUNS number to receive federal funding. For help applying for a DUNS number and more guidance on completing this form, please click here.

| **Non-Profit/Charitable Status (Non-US Organizations Only)** | | | |
|---|---|---|---|
| Is your organization registered as a charitable/non-profit entity? | | ☑ Yes | ☐ No |
| Country of Registration | USA | Registration # | 15-0543659 |

| **Financial Controls & Audits** | | |
|---|---|---|
| Did your organization expend more than US $750,000.00 in U.S. Government Federal Funding (Grants, Contracts, Subgrants, Subcontracts) in the previous fiscal year? If yes, please provide a copy of your single audit report, which is required under 2 CFR 200. | ☑ Yes | ☐ No |
| Have you been audited in the past 3 years? **If yes, please send a copy of the current report. (The report can be downloaded here:** FY2019**)** | ☑ Yes | ☐ No |
| Were there any material or significant findings in the audit report? | ☐ Yes | ☑ No |
| Has your organization ever had a grant or contract terminated for cause? | ☐ Yes | ☑ No |
| Does your organization utilize a financial manual to authorize expenses? | ☑ Yes | ☐ No |
| Does your organization utilize an accounting system to track expenses? | ☑ Yes | ☐ No |
| Does your organization have an ethics policy? | ☑ Yes | ☐ No |
| Does your organization have a timekeeping system for labor such as timesheets? | ☑ Yes | ☐ No |
| Will your organization use an institutional bank account to receive funding? | ☑ Yes | ☐ No |
| Please identify the authorized institutional signatory for agreements:<br>Shannon Robinson, Ed.D.<br>Associate Vice Provost for Research & Technology Transfer<br>Clarkson University \| 315-268-7766 | | |

| **Executive/Management Reporting Requirements** | |
|---|---|
| CRDF Global may be required to publicly report the names and total compensation of the five most highly compensated individuals at the awardees' institution. If you meet any of the criteria below, you are exempt from this requirement. Please find and check any applicable exemption: | |
| 1 In the previous tax year, institutional gross income from all sources was LESS than $300,000. | ☐ Exempt |
| 2 The institution received LESS than 80 percent of its annual gross revenues in U.S. federal funding (Contracts, Grants, Subgrants, Subcontracts or Loans). | ☑ Exempt |
| 3 The institution received LESS than $25,000,0000 in annual gross revenues from U.S. federal funding sources (Contracts, Grants, Subgrants, Subcontracts or Loans). | ☐ Exempt |
| 4 Executive compensation is publicly reported under section 13(a) or 15(d) of the Security Exchange Act or section 6104 of the Internal Revenue Code. | ☐ Exempt |
| I do not meet any of the exemptions above. I will provide the names and total compensation of the five most highly compensated executives. Click here for more information. | ☐ Not Exempt |

The information requested above must be provided in full and signed by an authorized institutional signatory, certifying that the information is true to the best of their knowledge. CRDF Global cannot proceed with an award to the institute without this information.

| **Past Performance** | | | | |
|---|---|---|---|---|
| Please list any applicable grants or contracts received from outside organizations. Successful completion is defined as zero suspensions or terminations for cause, audit findings or other discrepancies. | | | | |
| Funding Source | Agreement Number | Total Funding | Successful Completion? | Type of Project |
| National Science Foundation | 032221 | 200,000 USD | ☑ Yes ☐ No | Research Grant |
| National Science Foundation | 100099 | 330,000 USD | ☑ Yes ☐ No | Research Grant |
| National Science Foundation | 032570 | 450,000 USD | ☑ Yes ☐ No | Research Grant |

The information requested above must be provided in full and signed by an authorized institutional signatory, certifying that the information is true to the best of their knowledge. CRDF Global cannot proceed with an award to the institute without this information.

| Signature | Shannon Robinson, Ed.D.<br>Associate Vice Provost for Research & Technology Transfer | February 03, 2021<br>Date |
|---|---|---|


Address 8 Clarkson Avenue, CU Box 5630, Clarkson University, Potsdam, NY 13699-5630
Phone 315-268-7766
E-Mail srs@clarkson.edu

**SUGGESTED REVIEWERS AND REVIEWERS NOT TO INCLUDE**
*(Optional)*

| Suggested Reviewers | | | | |
|---|---|---|---|---|
| **#** | **Name** | **Affiliation** | **Email** | **Brief Justification** |
| 1. | Sergiy Minko | University of Georgia, USA | sminko@uga.edu | Expert in advanced functional materials |
| 2. | Michael J. Schöning | Aachen University of Applied Sciences, Germany | schoening@fh-aachen.de | Expert in modified electrodes and bioelectronics |
| 3. | Antonio López | Instituto de Catálisis y Petroleoquímica, CSIC, Spain | alopez@icp.csic.es | Expert in bioelectrochemistry, bioelectronics and biofuel cells |
| 4. | Sergey Shleev | Malmö University, Sweden | sergey.shleev@mah.se | Expert in bioelectrochemistry and biofuel cells |
| 5. | | | | |