Form 21

APPROVED

JINR Vice-Director

“_____” _____20 г.

SCIENTIFIC-TECHNICAL VALIDATION ON THE EXTENSION OF THEME FOR INCLUSION IN THE 2021—2023 JINR TOPICAL PLAN

Theme code:
04-4-1133-2021/2023

Frank Laboratory of Neutron Physics
Sector of Raman Spectroscopy

Field of research: 04 – Condensed Matter Physics; Radiation and Radiobiological Research

Theme title: “Modern Trends and Developments in Raman Microspectroscopy and Photoluminescence for Condensed Matter Studies”

Theme leaders: G.M. Arzumanyan, N. Kučerka

Abstract

Along with neutron and synchrotron research, Raman spectroscopy and microscopy occupy its own niche in the study of properties, structure and diagnostics of condensed matter covering an increasingly wider spectrum of important problems in physics, chemistry, biology and other branches of science and technology. Raman spectroscopy refers to inelastic scattered light from a sample that exhibits a frequency shift reflecting the energy of specific molecular vibrations within the sample of interest. In this manner, it provides a detailed chemical composition of the sample – a chemical fingerprint in essence. The frequency-shifted Raman lines, directly give the most basic information about the possible excitations in the system and provide information about chemical bonds which are the fundamental units in all molecules.

Over the last two decades, there has been tremendous technical improvement in Raman spectroscopy, as overcome by the problems like fluorescence and poor sensitivity. This has led to new trend of applications ranging from ancient archaeology to advanced nanobiotechnology. Among the latter, particular interest has grown in biosensing using fast, non-invasive and highly sensitive analysis with surface-enhanced raman spectroscopy (SERS). The development of biosensing techniques to overcome the problem of reliable detection, identification, and structural study of diverse bioorganic molecules at ultralow concentrations is still an urgent objective of specialists in many spheres including medicine, biology, forensics, ecology, pharmaceutics and so on. This is proven by the statistics on “biosensing” papers, the number of which has increased greatly in the last years. Accuracy and reliability are important requirements of desirable biosensing to reduce risks of false results. Raman spectroscopy has been rightly considered an appropriate approach for biosensing.

The activities carried out under the topic 04-4-1133-2018/2020, on the one hand, demonstrated the possibility of ultra-sensitive registration of organic molecules at the unit level (dyes), or several tens (biomolecules), and, on the other hand, highly-contrast visualization of analyte molecules when mapping by surface-enhanced coherent anti-Stokes light scattering (SECARS). It makes the "CARS" microspectrometer as a competitive tool at the world level in its class of analytical instruments.

The above mentioned items formed the basis for the proposal to extend the theme, which is aimed at applying modern trends of enhanced Raman spectroscopy, achieved over the past three years, in biosensing. Within the framework of the topic, the project “Biophotonics” was developed and formed, which includes fundamental and applied segments. In part of basic research, the work will be aimed at identifying and understanding the mechanisms of the ratio of the intensities of antiStokes / Stokes components in SERS spectrum. This will allow to formulate the conditions for obtaining reproducible SERS spectra during the development of biosensors. Applied tasks are related (i) with spectroscopic studies of netosis: in particular, with the search for Raman markers of this phenomenon, as well as with the identification of mechanisms for triggering the sterile activation of netosis under the influence of UV radiation, and (ii) lipid-protein interaction using a modern membrane mimetic – lipodisc. It should be noted that on the fundamental part of the project, there are quite a lot of publications, however, the issue still remains open and has not been finally resolved. As for the two applied problems, there are currently a very small number of publications around the world.

To realize the tasks of the project, we will employ the multimodal optical platform based on the unique "CARS" microscope, atomic-force microscopy (AFM), dynamic light scattering (DLS), electron microscopy (SEM, TEM), small-angle neutron scattering (SANS) and other instrumentations. Ultralow frequency Raman spectroscopy (~5 cm^{-1}) will become one of the key techniques of the project as well.

Thus, the proposal to extend the theme is in line with modern and innovative trends in the development of enhanced Raman spectroscopy methods, aimed, in particular, at their practical applications in biomedical tasks.

The activities on the theme will be mainly carried out by the staff of the Raman spectroscopy sector of the FLNP, in close cooperation with a number of interested partners from various scientific and educational institutions and organizations of the world, primarily from the JINR member countries.

Project:

“**Raman microspectroscopy in biomedical study**”

Project leaders: G.M. Arzumanyan, N. Kučerka

Project deputy leader: K. Mamatkulov

List of activities

1. Study of the features of the Stokes and anti-Stokes components of the SERS spectra from analyte molecules in order to understand the processes of enhancement in SERS spectroscopy.
2. Testing of SERS active substrates with organic / bio molecules in order to determine the range of pump intensity for recording reproducible aSt / St spectra.

3. Investigation of a possible nonlinear dependence of the SERS spectrum on the intensity and the pump mode used.
4. Stabilization of membrane proteins and studies of their structure using lipodisks by Raman spectroscopy, electron microscopy and SANS.
5. Testing the technique for obtaining Raman spectra of lipid disks with membrane proteins and "empty" lipodisks.
6. Study of the influence of the lipid environment on the structure of the membrane protein.
7. Search for spectral / Raman markers of NETosis.
8. Study of the mechanisms of sterile activation of NETosis under UV radiation.
9. Ultra-Low Frequency Raman spectroscopy ~ (5-10) cm^{-1} – modernization of the optical platform.

Expected main results upon the theme completion:

1. A comparative analysis of the ratio of the intensities of the SERS lines in aSt / St spectral regions depending on the pump radiation power.
2. Determination of the characteristics of the intensity ratio aSt / St depending on the excitation wavelength of analyte molecules.
3. Identification, comparison and characterization of the mechanisms of formation of aSt / St components of SERS spectra in continuous and pulsed modes.
4. A detailed analysis and interpretation of the Raman spectra of lipodiscs with various membrane proteins.
5. Confirmation of the incorporation of membrane protein into lipodisc and determination of the features of its structure.
6. Obtaining new information about the structure of lipodiscs with membrane proteins and "empty" lipodisc.
7. Identification of Raman markers of NETosis in various regions of the Raman spectrum.
8. Determination of the mechanisms of the formation of NETosis under the influence of UV radiation.
9. Gaining experience in ultra-low frequency Raman spectroscopy. ~ (5-10) cm^{-1}.

Collaboration:

Armenia (Yerevan, NNLA), Belarus (Minsk, BSUIR, "SOL Instruments"), Bulgaria (Sofia, ISSP BAS), Egypt (Cairo, CU), Poland (Krakow, JU), Romania (Bucharest, NIMP, Cluj-Napoka, INCDTIM), Russia (Moscow, GPI RAS, MSU, MONIKI), Slovakia (Košice, PJSU), Ukraine (DonNU), Uzbekistan (Jizakh, JSPI).

Terms of the theme implementation: **January 2021 – December 2023**

Estimated cost of the theme implementation according to JINR's Seven-Year Plan:

YEAR	k$
2021	285
2022	280
2023	275

AGREED:

Chief scientific secretary of JINR

_____

"____" _____2020

Director of FLNP

_____

"____" _____2020

Head,
Planning and Finance Department

_____

"____" _____2020

Scientific secretary of FLNP

_____

"____" _____2020

Head,
Science Organization Department

_____

"____" _____2020

Economist of FLNP

_____

"____" _____2020

Team leaders

_____

"____" _____2020

_____

"____" _____2020