ACADEMIA ROMÂNĂ
SCOSAAR

Anexa nr.6

ABSTRACT OF THE HABILITATION THESIS

TITLE: “From Molecules to Applications: Exploring the Supramolecular Assemblies of Nucleic Acids, Guanosine Quartets, and Cyclodextrin Inclusion Complexes in Chemistry, Biomaterials, and Cellular Imaging”.

Habilitation domain: **Chimistry**

Author: **Dr. Rotaru Alexandru**

Abstract

The Habilitation thesis under consideration investigates various aspects of Supramolecular Chemistry, including DNA sequence assembly, the assembly of guanine nucleoside networks, and the formation of cyclodextrin "host-guest" inclusion complexes, with the aim of developing functional entities for a wide range of applications. The thesis provides a summary of the most important results obtained by Dr. Alexandru Rotaru during the period ranging from 2006 to 2024, following the completion of his PhD thesis. The academic research and experimental studies described within this thesis falls within the field of Chemistry.

The thesis starts with the results obtained in the field of synthetic nucleotides, investigating simple DNA assemblies and complex DNA nanostructures, and exploring their design, preparation, and remarkable properties for chemical and biomedical research. The term "Caged" nucleic acids is introduced which refers to DNA molecules that can be triggered into action using light of a specific chosen wavelength. One such wavelength is red light, which is notably less harmful than UV light and has the advantage of being able to penetrate deep into the tissues. This chapter also introduces the concept of DNA Nanotechnology, specifically the DNA origami method, and its potential for conducting and visualizing chemical reactions at a single molecule level on a DNA origami nanostructure using atomic force microscopy (AFM). The results demonstrated remarkable efficiency and selectivity in successive cleavage and bond-forming reactions, allowing for post-assembly chemical modification of DNA nanostructures. Moreover,

this chapter explored the combination of light, AFM, and DNA nanostructures to investigate the generation of singlet oxygen from a single photosensitizer molecule attached to a specific DNA origami staple strand. The results illustrated that the oxidation of organic components at precise locations on the DNA origami is influenced by the distance from the central photosensitizer in each origami nanostructure. Furthermore, the chapter discusses the precise arrangement of streptavidin patterns on DNA origami and their subsequent efficient transfer onto a functionalized surface.

The next chapter of the thesis explores the design and characterization of guanosine quartet-based supramolecular hydrogels employing natural guanosine, diboronic acid, and a variety of templating and bridging cations, along with additional components such as cyclodextrin, carbon nanotubes, or dextran. The obtained hydrogels display a wide range of physical and functional properties that are influenced by the selection of primary constituents in their structure. Some of these hydrogel systems exhibit potential for applications in cell growth, as evidenced by the significant cell proliferation observed in the initial *in vitro* experiments or demonstrate notable antimicrobial properties. This innovative approach combines dynamic polymeric and guanosine quartet supramolecular chemistry, providing a versatile means of creating adjustable biocompatible hydrogels with thixotropic characteristics. Such materials hold considerable promise for various applications, including 3D bioprinting and regenerative medicine.

In continuation, this thesis investigates the effectiveness of supramolecular "host-guest" cyclodextrin inclusion complexes as versatile tools for the preparation of non-toxic fluorescent probes for fluorescent imaging of cells and cellular components. Since cyclodextrins are unique cyclic oligosaccharides capable of forming "host-guest" inclusion complexes with various guest molecules, including fluorescent dyes, this chapter examines the design and synthesis of inclusion complexes between β-cyclodextrin and a series of fluorescent indolizinyl-pyridinium salts. The formation of these complexes was thoroughly investigated using various experimental techniques, as well as theoretical investigations with molecular docking studies, confirming the formation of both 1:1 and 2:1 host-guest species. The key features of the investigated inclusion complexes distinguish them as promising candidates for labeling acidic organelles such as lysosomes or mitochondria. These include their noncytotoxic nature, cellular permeability, long-lasting intracellular fluorescence, and selective accumulation within acidic organelles. Given the success of cyclodextrin formulations in improving the properties of the investigated fluorescent

dyes described in this chapter, we believe that this methodology may considerably expand the range of fluorescent molecules considered for cell labeling and staining applications.

The final chapter of the thesis presents future research directions, including the development of ultra-sensitive diagnostic tools using DNA nanotechnology to improve early disease detection. The integration of hybrid DNA nanostructures with metal nanoparticles is expected to reveal novel applications in nanoelectronics and biomedicine. Further advancements in metallic nanoparticle functionalization are expected to enhance their catalytic, antimicrobial, and theranostic properties. Additionally, guanosine quartet-based supramolecular systems will be explored for their potential in targeted drug delivery and responsive material design. These directions aim to advance interdisciplinary research while enabling transformative innovations through molecular precision and multifunctional materials.