# O P I N I O N

by Assoc. Prof. Dr. Ruzha Georgieva Harizanova

University of Chemical Technology and Metallurgy, Sofia

for a dissertation thesis in a procedure for acquiring the educational and scientific degree

"**PhD**"

in Scientific area of higher education: 4. Natural sciences, mathematics and informatics

Professional field: 4.1. Physical sciences

Doctoral program: Electrical, magnetic and optical properties of condensed matter

**Author**: Aleksandar Vasilev Grigorov

**Title: „Modified multilayer structures for immobilization of bioactive molecules"**

**Scientific supervisor**: Assoc. Prof. Dr. Asya Petrova Viraneva, Department of Physics at the FPT, Plovdiv University „Paisii Hilendarski", Plovdiv

## 1. General presentation of the procedure and the PhD student

According to order No РД-22-2563 /16.12.2025 of the Rector of Plovdiv University (PU) "Paisii Hilendarski", I have been appointed as a member of the scientific jury to carry out a procedure for the defense of a dissertation entitled "**Modified multilayer structures for immobilization of bioactive molecules**" for awarding the educational and scientific degree "PhD" in the area of higher education 4. Natural Sciences, mathematics and informatics, professional field 4.1. Physical sciences, doctoral program "Electrical, magnetic and optical properties of condensed matter". The author of the dissertation is Aleksandar Vasilev Grigorov – a full-time PhD student at the Department of Physics at PU with scientific supervisor Assoc. Prof. Asya Viraneva, PhD from the Plovdiv University "Paisii Hilendarski".

The materials presented for the procedure by the PhD student include all the documents required: Application to the Rector of PU for the opening of the dissertation defence procedure; CV in European format; opinion of the Departmental council regarding the reporting of the readiness for the opening of the procedure and the preliminary discussion of the dissertation; opinion from the scientific supervisors; dissertation; abstract in Bulgarian and English language; list of the scientific publications on the topic of the dissertation; copies of the scientific publications; declaration of the authenticity and reliability of the attached documents; statement of compliance with the minimum national requirements. The PhD student has submitted copies of 4 publications on the topic of his doctoral thesis (2 are in IF journals and 2 – in magazines with SJR, accoding to the Scopus database). The submitted documents are in compliance with the Regulations for Development of the Academic Staff of the University of Plovdiv and the Law for the Development of the Academic Staff in the Republic of Bulgaria (LDASRB).

Mr. Aleksandar Grigorov has acquired the educational and qualifying degree „BSc“ from the Aberdeen University, Aburdeen, UK in the Professional field 4.1 Physical sciences in 2015, and the MSc degree is awarded by the PU “Paisii Hilendarski” in 2018 in the same professional field, 4.1. From 2020 to 2024 Aleksandar Grigorov works as a researcher at PU and from 2021 till present is an Assist. Prof. at the Department of Physics at the Faculty of Physics and Technology of the PU. Currently he has 9 publications in total on the topic of his dissertation and has taken part into 2 national and 5 international conferences with oral and poster presentations. He has participated into 6 scientific projects.

**2. Relevance of the topic**

Recetly, the development of new generation of multi-layered systems for controlled release of various biologically active substances which anables the efficient targeted delivery of the bioactive substance to the desired places and at the same time decreases the total amount of the substance needed has become of great importance.

With regard to the necessity of efficiency improvement for the already existing and the development of new multi-layered structures for the immobilization and controlled release of bioactive substances, the presented dissertation explores the opportunities for the synthesis by various methods of multi-layered thin films (based on chitosan and xantan), deposited onto modified subtrates from poly-lactic acid (PLA) and poly epsilon caprolactone (PEC); investigates and optimizes their composition and structure. Furthermore, their potential for application as systems for the targeted delivery of several biologically active substances (beta-galactosidase, benzydamine hydrochloride and tolfenamic acid) has been studied.

**3. Knowledge of the problem**

The PhD student, Aleksandar Grigorov became acquainted to and has performed an analytical survey of a large volume of scientific literature on the topic of his dissertation. The number of the cited references is 220 – more than 50 % from them originate from the last 15 years and are published in sources indexed by the scientific databases Scopus and/or Web of Science. This allows concluding that the PhD student is well-acquainted to the current state of the art and the tendencies for the development of the problems solved in his dissertation thesis.

**4. Research methodology**

In the submitted by Mr. Al. Grigorov dissertation, the subject of investigation are thin polymer films appropriately selected as composition and biodegradation ability which are optimized as a combination and their thermophysical and biochemical properties are characterized. The thin films are prepared by utilizing the method of solution casting. Several types of thin films have been prepared – non-porous (smooth), porous with and without liophilization and with the addition of poly-ethylene glycol. Some of the films have been subjected to additional surface modification by corona

discharge aiming the improvement of their hydrophobicity and inducing of unattached side gpoups. The obtained thin layers are based on PLA and PEC as well as on a combination with different ratios of them. Onto the obtained substrates multi-layered polyelectrolyte structures based on chitosan and xantan (or casein) have been deposited. Subsequently, in the obtained multi-layered structures various biologically active substances have been immobilized (beta-galactosidase, benzydamine hydrochloride and tolfenamic acid).

For the characterization of the substrates and the multi-layered structures, containing the bioactive substances, deposited onto them several appropriately selected methods have been used: differential scanning calorimetry – served to determine the ratio amorphous/ crystalline and the compatibility of the obtained materials as well as the impact of these factors on their potential applicability; scanning electron microscopy is used for the study of the morphology of the obtained films. The hydrophobicity or hydrophilicity of the obtained surfaces have been evaluated by the method of the static drop which is a standard method for the characterization of that type of biomaterials. For the evaluation of the enzymatic activity of the prepared lolyelectrolyte films the enzymological method has been utilized – taking into account the type of the enzyme used. The enzyme release kinetics characteristics have been studied by using UV-Vis spectrophotometry. Aiming to probe the potential applicability of the prepared structures as a drug-delivery system, pilot experiments have been performed in media simulating human saliva. In order to elucidate the mechanism of drug release from the polymer matrix, various mathematical models have been considered (first order model, Korsmeyer-Peppas model, Weibull model) depending on the specificity of the modeled system.

**5. Characteristics and evaluation of the thesis and the scientific contributions**

The submitted dissertation is written on 165 standard pages, contains 65 figures and 12 tables. 220 refernces have been cited and more than half of them are from the last 20 years. The dissertation is structures as follows: introduction; theoretical background of the importance of the investigated problem, materials and the choice of the research methods for their characterization and the achievement of the goals and objectives of the work. The main results and their discussion are presented in a separate chapter. At the end of the dissertation the main conclusions of the work are summarized and the scientific contributions of the presented study are given. A list of the references used is attached as well as the required declarations for authorship and the lists of the authored publications and conference and contract participations are also appended.

The PhD student has performed a thorough survey on the topic of the subject of the dissertation and has carried out a remarkable as volume experimental work as well as he has made an analytical discussion and interpretation of the obtained results. Based on the obtained results and their interpretation, the 6 main scientific contributions of his dissertation are formulated which are both of fundamental and applicable type. In the dissertation of Mr. Al. Grigorov a thorough study of the possibilities for the utilization of electrical field in order to influence the kinetics of release of biologically active

substances from multilayered structures onto modified composite liophilized and chemically modified substrates from PLA and PEC is peformed for the first time. In his work pilot investugations concerning the possibilities for the obtaining of multilayered structures on corona-charged porous composite substrates based on PLA and PEC are contained and establishing of the the role of the porous morphology on the immobilization and subsequent controlled release of the various biologically actibe substances is carried out.

**6. Evaluation of the publications and the personal contribution of the PhD student**

In the procedure for the defence of his dissertation, the PhD student has submitted 4 publications on the topic of his thesis which are published in indexed and referred in the Scopus/Web of Science databases journals: 2 works are in a magazine with Q1 and IF=2.8, and the rest are 1 – in a Q3 and 1 – in a Q4 journals. All publications are co-authored and in 1 of them the PhD student is the first author which undoubtedly shows his involvement and personal contribution in the preparation of the manuscripts. For one of the publications 1 independent citation has already been noticed.

Aleksandar Grigorov has participated into 7 scientific forums - 2 national and 5 international conferences with oral and poster presentations. He took part as a member of the team of 6 research projects on topics close to his dissertation topic or dedicated to the qualification improvement of PhD students and young researchers which resulted in broadening of his knowledge and gaining experience in the respective scientific area. These facts as well as the acquaintance with his dissertation allow me to conclude that Mr. Aleksandar Grigorov has a significant and well-defined personal contribution in obtaining and the interpretation of the results included in his dissertation and publications.

The 4 publications submitted for the procedure correspond to 77 points which exceeds the 30 points required according to the Regulations. The number and the quality of the publications fully corresponds to and even exceeds the minimum criteria and corresponds to the Regulations of PU and of the Faculty of Physics and Technology.

**7. Abstract**

The abstract is submitted in Bulgarian and English langauges and is written on 32 pages including the contributions and lists of the publications and participation in conferences and research contracts. It reflects in a compact and exact form the contents and the main results as well as contributions of the dissertation.

**8. Recommendations for future use of the dissertation contributions and results**

Having in mind the relevance and the importance of the investigayed topic, I would like to recommend to the PhD student to continue this research and to pay attention to the mechanical properties and their potential correlation to the physico-chemical properties of the obtained materials.

**CONCLUSION**

After reading the dissertation, abstract and publications on the topic of the dissertation of Mr. Aleksandar Grigorov, I am convinced in his leading role in obtaining the presented results and in their interpretation. In the materials submitted for the procedure, the PhD student has demonstrated a deep and thorough knowledge of the investigated topic and shows both theoretical knowledge and practical skills for performing of research work as well as for solving of scientific problems.

The dissertation thesis submitted by Mr. Aleksandar Grigorov contains both scientific and fundamental with elements of application results which represent an original contribution to science and fully comply with the requirements of the LDASRB, the Regulation for its Application and the Regulation for the conditions and procedure for the acquisition of scientific degrees at the University of Plovdiv.

Based on the above stated, I give my **positive assesment** for the research carried out and presented in the dissertation and the abstract and would like to recommend with conviction to the esteemed scientific jury to award the scientific and educational degree "PhD" to Aleksandar Vasilev Grigorov in Scientific area of higher education 4. Natural sciences, mathematics and informatics, Professional field 4.1. Physical sciences, Doctoral program "Electrical, magnetic and optical properties of condensed matter".

11.02.2026 The opinion is prepared by:

Assoc. Prof. Dr. Ruzha Harizanova