S T A T E M E N T

by Prof. Dr. Reni Emil Kalfin, PhD - Institute of Neurobiology, BAS

on dissertation for awarding the educational and scientific degree “Doctor”

Field of higher education 4. Natural sciences, mathematics and informatics

Professional area 4.2. Chemical Sciences

Doctoral program "Bioorganic chemistry, chemistry of natural and physiologically active substances"

Author: Assistant Boryana Krasimirova Yakimova, MSc

Form of PhD studies: Independent preparation

Topic: *"Design and synthesis of biologically active peptides as potential inhibitors of angiotensin converting enzyme (ACE I)"*

Scientific adviser:

Prof. Ivanka Stoyneva, PhD, DSc

1. General presentation of the procedure and the PhD student

The author of the dissertation is assistant Boryana Krasimirova Yakimova, a PhD student in the Laboratory "Chemistry and Biophysics of Proteins and Enzymes" at the Institute of Organic Chemistry with Center for Phytochemistry (IOCCP) at BAS with supervisor Prof. Ivanka Stoyneva.

The set of materials presented to me by the PhD student in electronic version and two thesis abstracts in Bulgarian and English on paper is in accordance with the Regulations for the development of the academic staff of IOCCP and fully meets the criteria of IOCCP-BAS for obtaining the scientific and educational degree "Doctor" for PhD students enrolled before January 1, 2019, such as Boryana Yakimova.

In connection with the dissertation, the PhD student has attached to her documents 4 publications of scientific articles and 14 abstracts from her participations in scientific forums in Bulgaria and abroad.

Presentation of the PhD student: Boryana Krasimirova Yakimova was born on October 3rd, 1982 in the town of Veliko Tarnovo. In 2001 she graduated from the Vasil Drumev High School of Natural Sciences and Mathematics in the same town. Boryana obtained a professional qualification "Master in Industrial Biotechnology" in 2007 at the Faculty of Biology of Sofia University with "Excellent" success.

Yakimova was enrolled in the doctoral program of independent training in the scientific specialty "Chemical Sciences" on December 1, 2015, and 4 years later she was expelled with the right to defense.

Assistant Boryana Yakimova is a member of the Bulgarian Peptide Society and the European Peptide Society. She is fluent in English in her work. He has participated in grants financed by the National Science Fund and in a project under the Operational Program "Human Resources Development". She was a scientific consultant to three successfully defended MSc students.

2. Actuality of the topic

Cardiovascular disease continues to be the leading cause of death in the world, with hypertension underlying morbidity and mortality. Hypertension is a chronic disease that increases the risk of developing atherosclerotic complications 2 to 3 times, namely the occurrence of ischemic heart disease, heart failure, stroke, renal failure. Arterial hypertension is of two types: essential or secondary (symptomatic). The incidence of hypertension in the European Union is between 30 and 45% of the population. In Bulgaria, cardiovascular diseases are the number one cause of death and disability. Vasodilators, diuretics, calcium channel blockers, angiotensin II receptor blockers and angiotensin converting enzyme I inhibitors are used in clinical practice to treat hypertension. As the patient's body becomes accustomed to the drugs administered during treatment, the search for new angiotensin-converting enzyme inhibitors as modulators of the renin-angiotensin system is essential for clinical practice.

The PhD thesis is dedicated to the synthesis of biologically active peptides as potential inhibitors of angiotensin converting enzyme, which makes its topic especially relevant in scientific and scientific-applied terms.

3. Knowledge of the problem

The numerous literature references cited in the thesis show that the PhD student Boryana Yakimova is very well acquainted with the state of the researched problem, object of her dissertation. The literature review consists of four sections, illustrated with 7 figures, 15 chemical formulas and 1 scheme.

The doctoral student describes very well the renin-angiotensin system, its function in the human body and some pathophysiological disorders underlying cardiovascular and renal diseases. The second section of the literature review is devoted to hypertension as a socially significant problem, its prevalence in the world by country, mortality from cardiovascular disease in men and women in Bulgaria, ways to prevent and treat hypertension. In the third section, Boryana presents a classification of angiotensin-converting enzyme inhibitors, their chemical formulas and mechanism of action. In the fourth section of the literature review, the PhD student analytically makes a brief historical

overview on the synthesis of the peptide bond, protecting groups, coupling agents and modern methods for peptide synthesis.

The literature review is generally written with a competent use of scientific terminology. An analysis of the problem of hypertension, cardiovascular diseases and their treatment is made, peptide synthesis is presented, literature data are creatively evaluated, both established dependencies and unresolved issues are identified, which determines the goals and objectives of the dissertation.

4. Research methodology

The selected experimental methods by the PhD student and her scientific supervisor are adequate and allow obtaining an answer to the tasks in the dissertation, namely the synthesis of biologically active peptides that inhibit angiotensin converting enzyme.

5. Characteristics and evaluation of the PhD thesis and contributions

The dissertation is written on 124 pages according to the standard scheme, keeping the ratio between the individual parts. The work is very well written - intelligently and with lot of knowledge. It is richly illustrated with 39 figures, 21 diagrams and 6 tables. The bibliography consists of 124 literature sources, all in Latin.

The set tasks are 5 in number and correspond to the aim of the PhD thesis to synthesize angiotensin-converting enzyme inhibitors with potential application in biomedicine.

Own results are presented on 33 pages, illustrated with 28 figures and 4 tables. All this contributes to the authenticity of the experimental material on which the contributions of the dissertation are built. Own results are explained with the help of data from the literature.

THE CONTRIBUTIONS summarize the original experimental data obtained by the PhD student. The achievements of the dissertation are scientific and scientific-applied. Data demonstrating that the synthesized proline peptides have a pronounced antihypertensive effect and could find potential application for prevention and therapy in medicine and as food supplements are of a contributing nature. The developed procedure for solid-phase synthesis of new short-chain peptide sequences, not described so far in the literature and proven as inhibitors of angiotensin-converting enzyme, is original.

In conclusion, this is a serious, labor-intensive, using many experimental methods dissertation that deserves respect.

6. Assessment of the PhD student's publications and personal contribution

Assistant Boryana Yakimova personally participated in the dissertation research. The contributions of the dissertation, noted by the PhD student, are her own work, obtained by the support of the scientific supervisor Professor Ivanka Stoyneva, PhD, DSc. The results of Yakimova's research in connection with the dissertation are summarized in 4 scientific articles in co-authorship, in two of which the PhD student is the first author. The fact that all articles are published in journals with an impact factor is excellent and makes the results obtained "visible" to the international scientific community. The PhD student presented the results of the thesis at 14 scientific forums in Bulgaria and abroad.

7. Thesis abstract

The dissertation abstract is written on 47 standard pages and fully corresponds to the content of the PhD thesis. From this abstract one can get an idea of the conducted scientific research. The abstract is very well shaped and richly illustrated with 38 figures and 6 tables. All main results of the dissertation**,** the conclusions and the contributions of the PhD thesis are included.

CONCLUSION

The dissertation presented for defense on the topic: "Design and synthesis of biologically active peptides as potential inhibitors of angiotensin converting enzyme (ACE I)" is sufficient in volume, the methods are adequately selected and the work deserves a positive assessment. The dissertation contains original scientific and scientific-applied results, which are popularized through well-formed scientific articles. The obtained results are also of interest for clinical practice.

The dissertation shows that the PhD student Boryana Krasimirova Yakimova has in-depth theoretical knowledge and professional skills in the scientific specialty "Bioorganic Chemistry, Chemistry of Natural and Physiologically Active Substances" by demonstrating qualities and skills for independent research. The presented results and scientific publications in connection with the dissertation fully comply with the specific requirements reflected in the Regulation for obtaining the educational and scientific degree "Doctor", an integral part of the Regulations on the terms and conditions for obtaining scientific degrees and holding academic positions in the Institute of Organic Chemistry with the Center for Phytochemistry, BAS and meet the requirements of the Law for the Development of the Academic Staff in the Republic of Bulgaria and the Regulations for its application.

Having in mind all stated above, I confidently give my positive assessment on the study presented by the peer-reviewed dissertation, abstract, results and contributions, and propose to the esteemed Scientific Jury to award the educational and scientific degree “Doctor” to Assistant Boryana Krasimirova Yakimova in the field of higher education 4. Natural sciences, mathematics and informatics, Professional area 4.2. "Chemical Sciences", doctoral program "Bioorganic Chemistry, Chemistry of Natural and Physiologically Active Substances".

Prepared the statement:

Prof. Reni Kalfin, PhD