EVALUATION REPORT

by Prof. Dr. Valerij Christov Christov, PhD, DSc,
Pensioner, Konstantntin Preslavsky University of Shumen
in regard to the thesis submitted for awarding the educational and scientific degree **Doctor**
field of higher education **4. Natural Sciences, Mathematics and Informatics**, professional field
4.2. Chemical Sciences, Organic Chemistry Doctoral Programme

Author: Zhanina Stoyanova Petkova
Dissertation: „Synthetic Approaches Towards Chiral and Biologically Active Compounds“
Scientific Advisors: Prof. Dr. Vladimir Dimitrov, PhD, DSc and Assis.-Prof. Dr. Malinka Stoyanova, PhD †

Further to the Protocol of the meeting of the Scientific Jury from 05. 10. 2020, I was appointed by Order № РД-09-05-291/28. 09. 2020 signed by the Director of the Institute of Organic Chemistry with Centre of Phytochemistry (IOCCP) to be a member of the scientific jury (reviewer) for awarding the educational and scientific degree Doctor on thesis named ” Synthetic Approaches Towards Chiral and Biologically Active Compounds”, field of higher education **4. Natural Sciences, Mathematics and Informatics**, professional field **4.2. Chemical Sciences**, **Organic Chemistry** Doctoral Programme. It is Zhanina Stoyanova Petkova`s thesis. Zhanina Petkova is a PhD student at Laboratory of Organic Synthesis and Stereochemistry, IOCCP. His scientific advisors are Prof. Vladimir Dimitrov, PhD, DSc and Assis.-Prof. Dr. Malinka Stoyanova, PhD † from IOCCP.

The set of electronic materials submitted by Zhanina Petkova is in compliance with the Regulation on the Terms and Procedure for Acquisition of Academic Degrees and the Occupation of Academic Jobs at IOCCP.

Brief Biographical Information about the PhD student

Zhanina Petkova graduated from the Faculty of Chemistry and Pharmacy at Sofia University "St. Kliment Ohridski ”, bachelor's program“ Chemistry ”, in 2009. In 2011 she graduated with honors the master's program in “Modern methods for synthesis and analysis of organic compounds” at the same faculty of the same university with the defense of a thesis on "Preparation and characterization of new microfiber materials from quaternized chitosan derivatives and co-polymers of styrene with maleic anhydride". Since 2011 she has been employed as a chemist in the laboratory "Organic Synthesis and Stereochemistry" of IOCCF-BAS. She is a full-time PhD student from 2011 to 2014, and since 2014 he has been an assistant professor of organic chemistry in the same laboratory. She completed his education as a full-time PhD student and she was given rights of defense.

The Relevance of the Doctoral Thesis and the Aims and Objectives Set in Thesis

The thesis submitted for review includes research material on a topic in the field of organic synthesis and is aimed at developing approaches for obtaining chiral compounds for use in asymmetric synthesis, as well as to create libraries of compounds with potential biological activity. It is necessary to emphasize that asymmetric catalysis is one of the fastest growing directions of asymmetric synthesis. An important advantage is that it provides the possibility of obtaining enantiomerically pure compounds by applying a small amount of the chiral information carrier (catalyst). This justifies the need to develop new approaches for the synthesis of chiral auxiliaries, reagents, ligands and catalysts to achieve high stereoselectivity in carrying out otherwise well-known chemical transformations. Asymmetric palladium-catalyzed allyl substitution

occupies an important place in organic synthesis as one of the effective methods for obtaining products with high enantiomeric excess.

The use of appropriate chiral ligands plays a crucial role in achieving a high enantiomeric excess. Chiral phosphine ligands containing additional heteroatomic functionality are widely used and effective. This type of structures facilitates the creation of a regulated catalytic system and the achievement of high enantioselectivity in certain transformations. In the present thesis the PhD student has focused on the application of asymmetric synthetic approaches for the preparation of chiral sulfonamide compounds further modified with a phosphine substituent to be used as ligands in palladium-catalyzed allyl substitution. In this regard, the following two goals are set:

1) preparation of new diphenylphosphine derivatives of chiral phenylsulfonamides to be used as P,O-ligands in palladium-catalyzed allyl substitution;

2) preparation of a series of enantiomerically pure β-aminoalcohols with a camphane skeleton and study of their *in vitro* antitubercular activity.

From the point of view of organic synthesis, these goals stimulate the use of new synthetic procedures, reagents, solvents and catalysts, the development of new isolation and purification techniques, as well as instrumental methods for characterization and structural elucidation of organic compounds. The main six tasks of the thesis are focused on:

- Development of a synthetic approach for the synthesis of chiral diphenylphosphine substituted sulfonamides;
- Study of the catalytic properties of the synthesized chiral compounds as P,O-ligands in palladium-catalyzed allyl substitution;
- Study of the catalytic activity of chiral P,O-ligands in *Suzuki-Miyaura* reaction;
- Synthesis of a vinyl-substituted compound with a camphane skeleton, using readily available (+)-camphor-10-sulfonyl chloride, as the main intermediate for the synthesis of diastereoisomeric oxiranes with a camphane skeleton, of which by aminolytic opening of the epoxy ring with suitable secondary amines for the preparation of series of novel chiral β-aminoalcohols;
- Investigation of the obtained β-aminoalcohols for *in vitro* antitubercular activity;
- Use of computer methods to find a quantitative relationship between structure and antimycobacterial activity (QSAR) of the obtained β-aminoalcohols.

From the above it is clear that the goals and tasks of the thesis are undoubtedly in a contemporary, current and perspective field of organic synthesis.

Knowledge of the Problem

In accordance with the goals and tasks of the thesis, the literature review is based on the reaction of directed *ortho*-metalation (D*o*M) as a tool for regioselective creation of complex and diverse polysubstituted aromatic and heteroaromatic structures. It has been explained that the D*o*M reaction is widely used in asymmetric synthesis due to the possibility of producing chiral compounds. Sufficiently briefly, but extremely informatively, the reagents for carrying out and the mechanism of the D*o*M reaction are considered. Attention is paid to *ortho*-lithiation as a powerful method for introduction of phosphine functionality within the aromatic structure. The main types of directed metalation group (DMG) are mentioned. *ortho*-Lithiation using one of the most effective targeting groups, namely the sulfonamide function, is discussed in detail. The combination of a DoM reaction with palladium-catalyzed carbon-carbon bonding reactions is a useful approach in organic synthesis that is widely used in the preparation of a variety of aromatic and heteroaromatic compounds. The following section briefly but competently discusses the

known literature on the features and mechanisms of known palladium-catalyzed coupling reactions, such as the *Heck* reaction, *Suzuki-Miyaura* reaction, *Negishi* reaction, *Stille* reaction, *Hiyama* reaction, *Kumada* reaction and *Sonogashira* reaction, as well as the application of these reactions for the total synthesis of a number of natural products and biologically active compounds.

Particular attention is paid to the literature data on the reaction of palladium-catalyzed enantioselective allyl substitution as an effective reaction to create a carbon-carbon bond and the formation of a new center of chirality. Its features and mechanism are competently presented. Serious attention is paid to the formation of (π-allyl) metal complexes, more precisely to their stereodynamics, as well as to the various factors influencing the regioselective course of the process, such as the steric and electronic effects, the substituents in the allyl substrate, the functional groups, the type of transition metal, the nature of the nucleophile, the ligands used, the solvent, the nature of the leaving group.

The following section justifies the need to develop effective methods for preparing enantiomerically pure medicinal forms in the manufacture of chiral therapeutic agents, due to the often different physiological activity of different isomers of the same compound. Attention is paid to the biologically active structures with antitubercular activity and the classification of the used antitubercular preparations is described. Finally, the literature data on the antitubercular activity of ethylenediamine derivatives and aminoalcohols are summarized with special attention to the previous studies on the synthesis of compounds with antitubercular activity of the group of Prof. Vladimir Dimitrov, to which PhD student Zhanina Petkova belongs.

The literature review discussed above leaves a very good impression with the concise and competent exposition of the cited works and characterizes the PhD student as a well-formed specialist in the field who is able to find, systematize and analyze critically the literature data. It is impressive to cite mostly original articles, but when based on secondary sources, monographs. The cited literature is a significant number (437 issues), they are precisely recorded according to the established method. Extremely good impression is made by the fact that in the list of cited literature there are 11 articles published after 2015.

Research Methodology

The selected two approaches for:

- synthesis of chiral diphenylphosphine-substituted sulfonamides, on which the catalytic activity to be studied as chiral P,O-ligands in palladium-catalyzed allyl substitution and in the *Suzuki-Miyaura* reaction; as well as
- to obtain a vinyl-substituted compound with a camphane skeleton as the main intermediate for the synthesis of diastereoisomeric oxiranes with a camphane skeleton, of which by aminolytic opening of the epoxy ring with suitable secondary amines,

have proved to be particularly productive for the production of chiral compounds for use in asymmetric synthesis, as well as for the creation of libraries of new chiral β-aminoalcohols to be tested for in vitro antituberculous activity, i. e. to achieve the set goals by performing the main tasks of the thesis are derived. This is achieved through the use of new synthetic procedures, reagents, solvents and catalysts, the development of new isolation and purification techniques, and instrumental methods for the characterization and structural elucidation of organic compounds.

Characteristics and Evaluation of the Thesis

The thesis is structured in accordance with the established requirements and is written on 198 pages and includes 79 diagrams, 38 figures and 8 tables. It is structured in the following sections: Introduction - 3 pages, Objectives and tasks - 2 pages, Literary review - 75 pages, Results and

discussion - 39 pages, Conclusions - 2 pages, Experimental part - 36 pages, List of newly synthesized compounds - 3 pages, Literature - 13 pages.

The thesis is constructed clearly, logically and is illustrated with appropriate schemes, figures and tables and contains original contributing research and results. Scientific results have a certain value and are achieved through appropriate methods and approaches. The impression of the precisely conducted experiment, of the correctly described synthesis procedures and of the detailed spectral characterization of the compounds is very good.

Particularly original and conducive to better clarity is the listing at the end of the thesis of the newly synthesized secondary and tertiary sulfonamides, *ortho*-phosphine-substituted tertiary sulfonamides and chiral β-aminoalcohols with a camphane skeleton, together with their exact names, as a way also contributes to emphasizing the merits of the work.

It is obvious that Zhanina Petkova has knowledge and successfully applies in his work modern methods for the synthesis and research of organic compounds, this is the educational goals of the doctoral program have been fulfilled.

Contributions and Significance of the Development for Science and Practice

Zhanina Petkova' thesis is rather fundamental in the field of organic synthesis, albeit with practical focus. The studies carried out have scientific and applied contributions with the enrichment of the existing knowledge in the field of organic synthesis - in the direction of synthesis of new enantiomerically or diastereoisomerically pure organic and organometallic compounds with center of chirality, research of enantioselective catalytic variants of organic reactions and study of their biological activity. In my opinion, the main scientific contributions and merits of the thesis are the following:

- An efficient approach for synthesis of a series of chiral phenylsulfonamides with diphenylphosphine substituent has been developed, introduced by means of *ortho*-lithiation directed with high efficiency and selectivity from the sulfonamide functionality;
- The catalytic properties of the synthesized diphenylphosphine-substituted chiral phenylsulfonamides as P,O-ligands in the reaction of palladium-catalyzed allyl substitution were studied and after optimization of the reaction conditions the enantioselectivity up to 83% ee was achieved. Some of the synthesized compounds were used as P,O-ligands and in the palladium-catalyzed *Suzuki-Miyaura* reaction, but under the tested conditions the desired coupling products were not isolated;
- A series of chiral β-aminoalcohols with a camphane skeleton was obtained by performing an aminolytic opening of the oxirane ring and introducing various pharmacophore substituents;
- The *in vitro* antituberculous and cytotoxic activity of some of the synthesized compounds was determined. Some of the compounds show excellent efficacy against the multidrug-resistant strain, one of which has low cytotoxicity;
- A computer model for estimating the contribution of each structural fragment to the activity of camphane-substituted β-aminoalcohols (QSAR) was demonstrated. The test compounds are promising and leading structures for the preparation of products with optimized biological activity.

Assessment of Thesis Publications

The scientific results have been published in three articles published in *Eur. J. Med. Chem.* (Q1), *Monatsh. Chem. (Chemical Monthly)* (Q2) and *Tetrahedron Lett.* (Q2). In two of the articles Zhanina Petkova is the first author, and in the third - the second. There are 17 citations to all three articles, with the most (13) citations to the article in *Eur. J. Med. Chem.*

The results of the thesis have been reported in 13 scientific forums, 7 of which are international and 4 of them have been conducted abroad. In 10 of the reports the PhD student is the first author, and in three - the second.

Personal Participation of the PhD student

I have no doubt in the significant contribution of the thesis in the implementation of the goals and tasks, as well as that the results obtained and scientific contributions are the personal merit of PhD student Zhanina Petkova, of course, with the significant mentoring of their supervisors. For the article in *Monatsh. Chem.* a separation protocol with Martin Ravutsov was correctly applied.

Dissertation Abstract

The author's extended summary (dissertation abstract) accurately and correctly reflects the content of the thesis and is written in accordance with the established rules.

Critical remarks

In general, the thesis and accompanying materials submitted to me for review meet the requirements of the normative documents. Some critical remarks can be made:

- Very general title of the thesis. In this form it is more suitable for a monograph on this subject;
- In "Results and Discussion", in some cases, experimental details of the reactions are described in some detail. This is completely unnecessary - their place is only in the "Experimental part";
- The same applies to the dissertation abstract - the description of the experimental procedures is superfluous. In addition, in my opinion, it is not necessary to make a brief literature review at the beginning of each section, citing a large number of literature sources - this has already been done in the dissertation itself. Obviously, this is the main reason for the rather extensive abstract. The list of abbreviations is missing;
- At the end of the literature review there is no summary and conclusions, which would better outline the ideas of the dissertation, as well as to better motivate the goals and tasks for their achievement. Nevertheless, the literature review is sufficiently informative and shows the high competence of the PhD student in the field of the topic of the thesis.

The critical remarks made do not change the overall excellent impression of the conducted research and their presentation in the dissertation.

Personal Impressions

Although I know Zhanina Petkova vaguely (mostly from our participation in conferences), I have no personal impressions of her qualities, but having in mind his thesis, she undoubtedly has a very good theoretical and experimental background, based on which I can state, that they were productive in the implementation of her research work, carried out under the leadership of Prof. Dr. Vladimir Dimitrov, PhD, DSc and Assist.-Prof. Dr. Malinka Stoyanova, PhD †.

CONCLUSION

Zhanina Petkova's thesis is an in-depth and systematic study in the current field of organic synthesis and contains original scientific studies for the synthesis of chiral compounds used in asymmetric synthesis, as well as for the creation of libraries of enantiomerically pure β-aminoalcohols with a camphane skeleton for their study *in vitro* antitubercular activity. The work performed is considerable in scope and diverse in nature, which suggests that the PhD student has acquired sufficient experience and ability to perform independent scientific work in the field of organic synthesis.

The thesis contains scientific and applied scientific results, which represent an original contribution to science and meet all the requirements of the Law for the Development of the Academic Staff in the Republic of Bulgaria (LDASRB), the Rules for the implementation of the LDASRB and the Rules for the acquisition of scientific degrees and academic positions at IOCCF-BAS. The thesis shows that the PhD student Zhanina Petkova has in-depth theoretical knowledge and professional skills in the field of organic chemistry, demonstrating qualities and skills for conducting independent research.

All the above motivates me to give without hesitation *a positive assessment* of the study, presented by the above peer-reviewed thesis, abstract, results and contributions, I would like to suggest the Honorable Scientific Jury to award *the educational and scientific degree* ***doctor (PhD)*** to **Zhanina Stoyanova Petkova** in the field of higher education: **4. Natural Sciences, Mathematics and Informatics**, professional field **4.2. Chemical Sciences**, **Organic Chemistry** Doctoral Programme.

15. 11. 2020
Sofia

Reviewer:
(Prof. Dr. Valerij Christov, PhD, DSc)