UNIVERSITATEA DE MEDICINĂ ŞI FARMACIE
„CAROL DAVILA" BUCUREŞTI
ŞCOALA DOCTORALĂ
DOMENIUL FARMACIE

FROM QUALITY CONTROL OF DRUGS APPLIED IN THE PHARMACEUTICAL FIELD TO OMICS TECHNOLOGIES FOR DETERMINING THE BIOCHEMICAL MARKERS IN PATHOLOGY

SUMMARY OF THE HABILITATION THESIS

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The Habilitation thesis entitled “From quality control of drugs applied in the pharmaceutical field to omics technologies for determining the biochemical markers in pathology” is a synthesis of the most representative research directions approached during the post-doctoral period.

The thesis presents the research activity carried out after the presentation of the doctoral thesis, entitled “*In vitro* tests as quality control procedures and biowaiver methodologies for different formulations containing ibuprofen”, including the main perspectives of academic, professional and research career development.

Quality control is an essential operation in the pharmaceutical industry. Pharmaceutical drug products must be marketed as safe and therapeutically active formulations with consistent and predictable properties and performance. As new and better medicinal agents are rapidly evolving, so are there more exacting and sophisticated analytical methods being developed for their evaluation.

According to the World Health Organization (WHO), the term quality control refers to the sum of all procedures undertaken to ensure the identity and purity of a particular pharmaceutical.

Quality control in particular is the area of good manufacturing practices (GMP) which deals with processes involving sampling, specifications and testing, and with the organization, documentation and release procedures. These help ensure that the necessary and relevant tests are executed and that materials are not released for use, nor products released for sale or supply, until their quality has been confirmed to comply with international standards.

This attribute of a pharmaceutical preparation cannot be easily measured and assured during in-process inspection and finished-product testing, since every stage of the manufacturing process affects the properties of the drug. This key fact opens up the necessity to properly train all personnel - from the basic concepts of assuring quality like donning on their personal protective equipment (PPE) prior to entering production sites; up to the complex protocols of aseptic technique.

Additionally, a consistent yield of products of the highest calibre will not only depend on the operators, rather, even the material of construction of the cleanrooms and various equipment used for handling and preparing the final products can be of influence. As such, a keen sense of attention to detail is essential.

It is also important to note that quality control is not confined to laboratory operations but must be involved in all decisions concerning the quality of the product; from the purchasing and storage of raw materials, to the in-process quality control testing, up until the labelling and packaging of the final product.

The use of organic reagents has led to the development of new methods of analysis which also benefited from the field of physical-chemical control of the drug. A newly created analytical method must be validated, and the validation methodology applied is meant to demonstrate that the proposed method corresponds to the intended use. Statistical processing of the results of an analysis offers the possibility to obtain the most probable value that is as close to reality as possible.

The analysis methods presented have been validated in accordance with conventional regulations, such as those of the Food and Drug Administration (FDA), European Medicines Agency (EMEA) and International Conference of Harmonization (ICH).

An important problem in pharmaceutical research is the optimization of the bioavailability of substances with low solubility and high permeability of the second class of biopharmaceuticals, which can be achieved through various methods such as complexation with carbohydrate derivatives such as cyclodextrins when inclusion complexes are obtained, encapsulation in lipo- or water-soluble systems for administration and transport or formulation in oral pharmaceutical forms with modified release.

The Habilitation thesis is structured into three sections. The first one describes the personal, professional and academic achievements, the second section is presented in the form of a detailed presentation of the main scientific achievements after the doctoral dissertation, and the third section presents the main future plans regarding the evolution and development of the professional, scientific and academic profile.

In the chapter entitled ”Professional and Academic Achievements” I presented the entire academic and professional career during my 14 years of activity at the Faculty of Pharmacy of the ”Carol Davila” University of Medicine and Pharmacy - Bucharest, and in 2014 I defended the PhD thesis in the field of Pharmaceutical Sciences. Also, during this period, I passed the exams for obtaining the academic titles and various degrees specific to the profession of pharmacist: resident, specialist, mayor. Professional training has been one of my main concerns with a multidisciplinary educational path adapted to the nature of the subjects taught. Throughout

this period, I have been concerned with motivating and supporting pharmacy students to deepen the scope of the drugs quality control. In this regard, I have helped, first of all, to improve the didactic material. Thus, as a co-author, I have participated in the development of 2 Laboratory Books required for pharmaceutical practical applications in the field of drug control. I have also been involved in the development of the didactic material - support for the Drug Control courses. The aspect of professional pharmaceutical activity completes, necessarily and elegantly, the academic and scientific activity. To that end, I have informed, prepared and supported numerous training courses and continuous training for pharmacists across the country. I have also participated, as a member of the Scientific Council and Organising Committee, in organizing numerous events within the Romanian pharmaceutical community.

Another aspect I would like to highlight relates to a special field of activity, which I have voluntarily assumed, through the involvement in the activity of Managing Editor at Farmacia journal, the only publication in the Romanian pharmaceutical scientific field indexed and quoted on the basis of ISI Thomson Reuters data (IF / 2021 = 1.550).

In the second chapter of the thesis, entitled ”Scientific Achievements”, I have detailed the results obtained and published in the form of articles quoted ISI or indexed by BDIs, as well as in books/chapters of books published nationally or internationally.

The main research directions developed in the scientific field are in line with the Development Plan of the faculty, the university and respectively the Research and Development-innovation strategies elaborated at national and international levels and can be grouped as follows:

- ✓ Development and validation of analytical and bioanalytical methods applicable in drug quality assurance and bioequivalence/bioavailability studies,
- ✓ *Omic* technologies for the development and validation of biomarkers with predictive potential of response to standard therapy.

Within the first research approach I have conducted several analyses, mainly *in vitro* dissolutions studies, but also HPLC, TLC and spectrometry method for establishing the quality of several pharmaceutical products, raw active substances or finished products, single or in mixtures. The dissolution test was used as the most performant tool for estimating the performance of an medicinal product, as it has acquired new values, being the most important guide in the development of pharmaceutical research and an essential tool for estimating bioavailability. These studies were performed on mesalazine *in house* tablets, a modified release

formulation containing *Silybum marianum* extract, five formulations prepared as hidroalcoholic gels with ketoprofen, industrial pentoxifylline tablets, metoprolol succinate tablets, tramadol tablets and trimetazidine tablets, each time establishing the biopharmaceutical impact starting from the pharmacokinetic modeling of plasma concentration profiles. I presented also the results of *in-vitro* release tests applied for assessing the impact of mechanical splitting on the performance of sustained release oral solid formulations containing metoprolol succinate and for underlying the influence of ethanol content in acidic media on the *in vitro* release of tramadol hydrochloride from controlled release tablets. For the development of the chromatographic method for piroxicam, a design of experiments (DOE) approach was used, along with a Box-Behnken design in order to identify the significant parameters for the optimization, by simultaneously taking retention time, peak symmetry, resolution of the chromatographic separation, number of theoretical plates and capacity factor of the first eluting peak as responses. Also, studies were performed on carvedilol tablets, furosemid tablets and injectable solution. The validation, where was performed, was carried out in accordance with the current ICH guidelines in terms of specificity, linearity, precision, accuracy, limits of detection/quantitation, and system suitability. Nevertheles, analitical methods were applied for the quality control of essential oils, in order to evaluate antimicrobial and antioxidant profiles for these candidates.

The second chapter's main focus is correlated with the purpose of finding new cancer treatments. For this, the research aimed to develop a circulating colorectal tumour cells detection protocol by using flow cytometry, by comparing the outcomes of the positive and negative selection strategies after fluorescent staining with the appropriate antibodies and flow cytometry analysis of the samples. Also, studies were performed in order to investigate the potential intra-nuclear signalling mechanism of the endogenous bioindoles: melatonin, serotonin and tryptophan, through their direct interaction with the DNA, determining the intensity of the fluorescent signal issued by the indole molecules, natively fluorescent, in the presence of the DNA, molecule which lacks fluorescence. The effects of methadone (MTD) on the proliferation of splenic lymphocytes from Wistar rats treated *in vivo* with methotrexate (MTX) after induction of chronic inflammation arthritis were investigated. The studies regarding the rheumatoid arthritis were continued with an analysis whose objective was to assess the possibility of spacing biological therapy while maintaining the disease under control to ensure that patients benefit as completely as possible from a clinical outcome with less medication.

The last chapter of the thesis presents the main future plans for the evolution and development of the professional, scientific and academic profile. The academics must have the ability to tirelessly combine teaching and scientific research, giving equal weight to them. The results achieved both in didactic and research activity are innovative and ensure the observance of the fundamental principles of quality assurance in the national and European space of research and higher education.

On the principle of continuity, I wish to build my professional profile in the coming years. In this respect, I intend to improve (in fact periodically update) the experimental methodology in the practical work with the students as a result of the acquisition of modern equipment, based on the ongoing and future research contracts of the discipline. I propose developing and introducing new experimental works within the Drug Control Laboratory. These new protocols will be modern, aligned with laboratory homologous curricula in the countries of the European Union.

As far as the development of scientific research activity is concerned, I have proposed that it should be oriented mainly on the same research directions in which I have achieved significant results, but I will also consider the development of related directions. The objectives of the research activities are to increase its competitiveness, to develop partnerships in the priority areas for the design of new technologies, innovative products and implementation mechanisms to solve complex problems associated with the research areas, and to disseminate the results of the studies accordingly.

In conclusion, the main objective of my career was, is and will be that of professional self-refurbishment so that it can carry out educational and research activities at a high-quality level, respecting both its own principles, and the qualitative and ethical ones adopted by the Charter and the Strategic Plan of “Carol Davila” University of Medicine and Pharmacy Bucharest, Romania.