CONTRIBUTIONS TO THE SYNTESIS AND EVALUATION OF BIOLOGICAL ACTIVITY OF HETEROCYCLIC COMPOUNDS

Smaranda Dafina Oniga

This habilitation thesis presents, in a synthetic manner, the main scientific, professional, and academic achievements in the period 2006-present after obtaining the PhD degree The thesis is structured in several distinct parts, namely: scientific, professional and academic achievements, career development plan, general conclusions and bibliographical references.

The scientific achievements chapter describe the main research directions addressed and the obtained results. Thus, the scientific research following the doctoral studies period has focused on the design, chemical synthesis, and evaluation of the biological potential of some synthetic heterocyclic compounds, mostly derivatives with thiazole structure, but also some fluoroquinolone derivatives. The synthesized thiazole derivatives belong to different classes: thiazolyl-1,3,4-oxadiazoline and thiazolyl-1,3,4-oxadiazoles, thiazolyl-2,5-diones, aryl-2-phenyl-thiazolyl-phenyl-thiazoles, trimethoxy-phenyl-thiazoles or bistiazole compounds, while quinolone derivatives are various substituted derivatives of norfloxacin. The structure of the new compounds proposed for synthesis was based on the concept of molecular hybridization, a rational strategy to design new bioactive molecules. The synthesized compounds were evaluated for the antimicrobial and anti-inflammatory action. In the case of antimicrobial potential assessment, the mechanisms of the emergence of bacterial resistance to chemotherapeutics, in particular microbial film formation, were also considered. As a result, one of the research directions aimed to evaluate the antibiofilm potential of the synthesized compounds. The design of compounds that can inhibit microbial biofilm formation and, as a result, favors the action of antibiotics on potentially resistant germs, is a highly relevant research direction in medicinal chemistry. Furthermore, using molecular docking techniques, we have attempted to elucidate the mechanism of action for the synthesized compounds. Also, by applying the ADMET method, the pharmacokinetic properties of the new molecules were proposed.

In the case of thiazolyl-oxadiazoline or thiazolyl-oxazole derivatives, four series of compounds were obtained. Regarding the antimicrobial potential of thiazolyl-oxadiazoline derivatives, it can be concluded that it is relatively modest, except for the antifungal activity. The first two series of compounds (pyridyl-thiazolyl-oxadiazoline) showed good antibiofilm potential, particularly on S. aureus strain. Using in silico molecular docking techniques, it was proposed a mechanism of action: the inhibition of StrA, a key enzyme in bacterial biofilm formation. In the case of compounds with thiazolyl-methylene-oxadiazole structure, a good antifungal potential (against C. albicans) was established. A mechanism of action has also been proposed for these compounds: the non-competitive inhibition of fungal lanosterol demethylase.

The thiazolidine-dione derivatives have different biological potential depending on the substituents of the thiazolidine-dione nucleus. In the case of thiazolidine-dione-PABA hybrid structure derivatives, they have direct antimicrobial potential, but they also inhibit microbial biofilm formation for *S. aureus* ATCC 25923, *C. albicans* ATCC 10231 and *C. parapsilosis* ATCC 22019 strains. In the case of N-oxazolylmethylthiazolidine-2,4-dione derivatives, they do not show direct antimicrobial potential, but they have good antibiofilm potential expressed against some fungal strains. The proposed mechanism of action, identified by in silico molecular

docking studies, is inhibition of Als family proteins, known to be key elements in adhesion, biofilm formation and virulence for *Candida* spp.

In the case of the aryl-2-phenyl-thiazolyl-phenyl-thiazole derivatives, good activity against *E. faecalis* biofilm formation was shown, with most compounds acting as non-competitive inhibitors of *E. faecalis* sortase.

Compounds with trimethoxy-phenyl-thiazole structure, evaluated for both anti-inflammatory (in vitro and in vivo) and antimicrobial/antibiofilm potential, showed much better anti-inflammatory potential than antimicrobial, with compounds showing a superior effect to meloxicam (used as reference molecule) and a similar selectivity index on COX isoforms. For most compounds, a very good correlation between COX inhibition in vitro and anti-inflammatory effect in vivo was established.

Regarding the biological potential of the synthesized 4,2- and 5,2-bistiazole derivatives, the anti-inflammatory potential of the compounds is superior to the antimicrobial one (both direct and inhibition of microbial biofilm). Regarding the anti-inflammatory activity, it can be noticed that there are differences between the two series of compounds which differ from each other by the binding positions of the two thiazole cores. Compounds in the 4,2-bistiazole series generally show modest anti-inflammatory action, while compounds in the 5,2-bistiazole series have been identified that show significantly better or at least similar anti-inflammatory action to meloxicam, the anti-inflammatory standard.

Two series of norfloxacin derivatives have also been synthesized, most of them substituted at the piperazine moiety of the norfloxacin molecule. These derivatives generally have lower or at most equal antimicrobial potential to norfloxacin (used as standard), but compounds with superior antimicrobial activity against K. pneumoniae or compounds with superior bactericidal activity compared to norfloxacin (lower CMB values) have also been identified. Also, some of the compounds obtained have varying degrees of inhibition of microbial biofilm, in particular that produced by S. aureus ATCC 25923. Molecular docking studies revealed that these compounds bind bacterial DNA-gyrase in a different way than norfloxacin.

The research activity has been supported by a series of research projects won by competition: internal competition (2 grants as director) and national competition (1 Romania-Moldova cooperation grant as director and 2 national grants as member). The results have been presented at national and international scientific events.

The results of the scientific and post-doctoral research activity are materialized in 34 scientific articles as main or co-author, published in scientific journals (31 of the articles are published in ISI indexed journals, 17 of them as main author). The visibility and relevance of the research is confirmed by the Hirsch index of 11, the 239 ISI citations, the cumulative lead author impact factor of more than 30 and the 6 UEFSCDI awards in the PRECISI research results competitions.

Another chapter of the habilitation thesis is dedicated to the results of the teaching and academic work carried out. All my teaching activity was carried out at the Therapeutic Chemistry discipline of the Faculty of Pharmacy, UMF Cluj-Napoca and consisted in the designing, planning and delivering lectures and practical work of Therapeutic Chemistry to undergraduate student, (Romanian and French sections), to post-graduate students (master and residency programs) and to pharmacists. As main achievements of my teaching activity, I have contributed to the conception of teaching materials for students (2 lab workbooks, a lecture material), and I am also co-author of 9 books.

An important and relevant part of the academic career is the academic career development plan which has several directions, namely teaching activity, increasing teaching skills and scientific research activity. For each direction the proposed future objectives are presented.

For the research activity I propose to continue the research themes we have in progress but also new direction of research. In this regard, I intend to continue the direction already started to obtain norfloxacin derivatives with improved antimicrobial action, but I want to expand the area of interest to obtain norfloxacin derivatives, with anticancer action, using as a model the molecule of vosaroxin, a compound in clinical trials for the treatment of acute myeloid leukemia. Another direction of research already underway is the incorporation of compounds with anti-biofilm activity into composite materials or polymers used for the manufacture of medical devices, followed by testing the ability of the finished products to prevent the growth of biofilm on their surface. Also, given the current pandemic context, an interesting direction of research would be to obtain thiazole derivatives, analogues of camostat, as potential inhibitors of the TMPRSS2 enzyme, a key enzyme that promotes SARS-CoV2 entry into the cell.