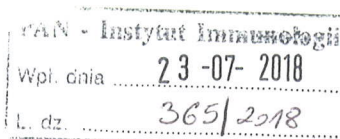




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Wydział Farmaceutyczny
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REVIEW

doctoral dissertation Mrs. MA Fairoz Ali Al-Wrafiy, entitled „Immunochemical and biochemical characterization of phage receptors in two clinical strains of *Pseudomonas aeruginosa*”

Nowadays, the global problem is infection with the etiology of multi-drug resistant or resistant to all antibiotics available in medicine. The consequences of this state are therapeutic failures, increased mortality and the spread of such strains in the human population and in the environment. This prompts scientists to look for new therapeutic strategy. For this reason, choosing the subject of the doctoral dissertation by Mrs. MA Fairoz Ali Al-Wrafiy is valid and important both from the cognitive point and practical activities.

The configuration of the thesis of 125 pages is typical for experimental thesis. The introduction is divided into subsections (29 pp.). The following parts include: Aim of the study (1 p.), Material and methods (16 pp.), Results (33 pp.), Discussion (5 pp.), Conclusions (1 p.), Author's list of publications (1 p.), Literature with 173 items (18 pp.), and at the beginning of the study Table of contents (4 pp.), Acknowledgments (1 p.), Summary in Polish and English (3 pp. each) and List of abbreviations (3 pp.).

In the Introduction, the author introduces the reader to the research subject in a logical order. Widely familiarizes the reader with *Pseudomonas aeruginosa*, with their characteristic, virulence factors, antibiotic resistance mechanisms and phages, taking into account their therapeutic potential as a new strategy in the treatment of infections.

PhD student presented the aim of the study at three points, and they were:

- immunological studies of surface polysaccharides and protein *P. aeruginosa* to reveal the nature of phage protein receptors on bacteria,
- isolation and purification of phage proteins to examine how they are affecting bacteria in order to identify phage proteins recognizing bacterial receptors,
- further investigations on the effect of phage proteins as a promising approach for treatment with both alone or in combination with antibiotics.



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Mrs. MA Fairoz Ali Al-Wrafy in her research included two strains of *P. aeruginosa* PAR21 and PAR50 from foot ulcers of diabetic patients, *P. aeruginosa* PCM 2720 as a phage host and two phages which were isolated from Wrocław sewage. Noteworthy is the comprehensive characterization of examined strains (identification, sensitivity to antibiotics, extraction and purification of extracellular polymeric substances [EPS], determination of total carbohydrate and of glucuronic acid contents in purified EPS fractions, isolation outer membrane proteins [OMPs], immunological study) and bacteriophage (determination their activity, identification, concentration, characterization and activity phage protein, enzymatic and proteolytic activity) through the skillful use of many diverse methods, both classical and modern. In well-designed experiments, an ingenious direction of research was the selection of EPS and OMPs as target to define of phage receptors on the surfaces of cells two of *P. aeruginosa* strains and treatment bacterial strains with phage protein, analysis of the protein receptor.

PhD student obtained a number of valuable research results, which she presented in a legible way, documenting them on 30 drawings and in 10 tables. The author introduces the reader to the essence of the conducted research, which took place in several stages.

An important area of research was to determine the activity two phages against multi-drug resistant (MDR) *P. aeruginosa* PAR50 strain but not against second examined *P. aeruginosa* PAR21 strain. A valuable result is to define of high efficacy of two proteins PA-PP1 and PA-PP2 isolated from both phages against *P. aeruginosa* PAR50 strain.

Another interesting research direction chosen by Mrs. MA Fairoz Ali Al-Wrafy was to explain, that the phage receptor on the surface of this bacterial strain has appeared as one of the OMPs porin. The utilizing this protein as component multidrug efflux pump MexAB and MexXY – very important mechanism in antibiotic resistance – by using phage contributes to changes the efflux pump works and makes increased sensitivity to several antibiotics in MDR *P. aeruginosa* PAR50 strain.

Demonstration by Mrs. MA Fairoz Ali Al-Wrafy bactericidal activity of phages against MDR *P. aeruginosa* PAR50 strain at the molecular level and additionally documenting their effect in the form



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of defects in bacterial cells using scanning electron microscopy, as well as through synergy between phage protein and piperacillin against this strain, is an innovative element of study.

PhD student demonstrates knowledge of the subject matter of research. This is indicated by a skillfully written introduction and a discussion of own results on the background of the literature included. Analyzing the results of multidirectional own research, she formulated 7 conclusions that are a response to the set goals.

Notes:

The author does not use a uniform abbreviation. They should be placed according to their origin along with the first use of the full name, in the same way for all in order to use them consistently in the further part of the study. Fig. 31 and 32 does not contain information about the used magnification size. The author does not provide information about the reference strains used for the susceptibility assessment carried out according to the CLSI (Clinical and Laboratory Standards Institute) recommendations.

Conclusions:

In conclusion, I would like to emphasize that the study addresses an important and current scientific subject. Its author has undertaken multidirectional research in the implementation of the intended goals, demonstrating knowledge of various research methods, from the area of microbiology, immunology, biochemistry, mass spectrometry, scanning electron microscopy, thus indicating that it has a modern research workshop and maturity to solve the problems. In experiments designing, she showed her knowledge of the subject and the ability to connect knowledge in the field of methods, theory and practice. The PhD student externalizes the critical standpoint to the obtained research results and the ability to interpret them as phenomena in the background of the literature.

I would like to emphasize that the doctoral dissertation Mrs. MA Fairuz Ali Al-Wrafy is a valuable and original scientific achievement. It has revealing values and serious research and clinical implications. It is a valuable scientific study, constituting a real contribution to the progress of knowledge. The presented methods and the promising results obtained thanks to them are valuable



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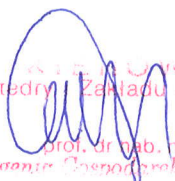
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research information and constitute the basis for further research to use the applications with a potential therapeutic strategy against MDR strains of *P. aeruginosa* containing a porin protein as a receptor for PA-PP1 and PA-PP2 proteins.

I state that doctoral dissertation **MSc MA Fairoz Ali Al-Wrafy, entitled „Immunochemical and biochemical characterization of phage receptors in two clinical strains of *Pseudomonas aeruginosa*”** reviewed by me, corresponds to the conditions set for the doctoral dissertations presented in the Act of 14 March 2003 (On academic degrees and academic title, and degrees and title in the field of art and late changes) and therefore I apply to the High Scientific Council Institute of Immunology and Experimental Therapy Ludwik Hirsfeld of the Polish Academy of Sciences in Wrocław for admission to further stages of the doctoral program.

The entirety of the scientific argument, as well as the quality of experimental research carried out by PhD student in Poland are pioneer and unique. Taking into account the importance of the subject taken, the research methods used, the original and innovative nature, contribution to the development of medical science and the very careful formal side of the reviewed dissertation, lead me to apply for its distinction with an appropriate reward.


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