

Course name: A	Modern research methods and techniques (pharmaceutical sciences)			
Name of School	Doctoral School of the Medical University of Silesia in Katowice			
Year 2	Course status	obligatory	Language of the cours	
Form of classes	Number of hours	Methods of evaluation	ECTS	
lectures		credit	3	
exercises	30			
seminars				
together	30			
Course coordinator	Dr hab. n. med. Ilona Bednarek, MD, prof. SUM - subject coordinator (Department of Biotechnology and Genetic Engineering) - Topic block no. 1 Dr. hab. n. med. Monika Paul-Samojedny (Department of Medical Genetics) -Topic block no. 2 Dr. n. pharm. Jakub Rok (Department of Pharmaceutical Chemistry) - Topic block no. 3 Dr. hab. n. pharm. Andrzej Zięba prof. SUM (Department of Organic Chemistry) - Topic block no. 4 Dr. hab. n. pharm. Krzysztof Marciniec (Department of Organic Chemistry) - Topic block no. 5			
Name of Department	Faculty of Pharmaceutical Sciences in Sosnowiec			
Course objectives	Upon completion of the course, the doctoral student has knowledge and skills related to the use of modern research techniques and tools used in various sub-disciplines of medical biology. The doctoral student is able to plan the research steps and interpret the results of the conducted research/assays. He/she is able to identify alternative methods of verification of research results and propose alternative methodology solutions for conducted research/assays.			
Prerequisites	graduate studies completed			
LEARNING OUTCOMES				
Category	Description of the outcome		Relationship to the outcomes for the programme	
knowledge	Knows the theoretical basis and application of the research techniques covered		P8S_WG	
	Knows the main trends in the development of the techniques discussed (including possible directions of the applicability of the techniques)		P8S_WG	
skills	Can plan a research project experiment using the subject techniques.		P8S_UW P8S_UU	
	Can select research tools from the group of techniques discussed and apply them to the goal of own research		P8S_UO	
social competencies	He is ready to perform a critical evaluation of the achievements in the research techniques discussed		P8S_KK	
	He is ready to make a substantial and critical assessment of his own contribution to the development of the techniques discussed , without violating the rules of intellectual property rights and with respect for the rule of public property of the results of scientific activity		P8S_KR	
CURRICULUM CONTENT				
Course description, range of subjects, raised issues and topics				
Block 1: Interaction of cells with the microenvironment. Phenotypic plasticity of cells and its modification (10 hours).				
"Gene transfer as a research tool"				

***Mechanisms of regulation of gene expression and opportunities for verification of phenotypic plasticity of cells and transgenesis***

***Interactions of cells with the microenvironment - control of migration and invasiveness of tumor cells***

***Cellular stress, apoptosis, cell differentiation - visualization techniques***

***Block 2: Flow cytometry - basics of experiment design and cytometric analysis in research (5 hrs):***

***Basics of flow cytometer construction and operation (startup procedure, setup, experiment design, setting voltages on detectors, shutdown procedure). Designing a flow cytometer experiment (type of material to be tested and conditions it must meet; selection of fluorochromes and lasers for multicolor analysis, types of controls, principles of cell labeling - selection of antibodies, principles of detection of surface and intracellular antigens; one- and two-step labeling). Types of measured parameters (FSC and SSC settings, population gating, optimization of settings for fluorescence). Principles of analysis and interpretation of results (type of graphs, interpretation of example results). Principles of cell cycle analysis, apoptosis and autophagy by flow cytometry.***

***Block 3: Application of confocal microscopy in research projects (5 hours):***

***"Basics of confocal microscopy".***

***Discussion of the basics of confocal microscopy technique: preparation of material for research, obtaining and analyzing images, interpretation of the obtained results***

***Discussion of the possibility of using confocal microscopy in quantitative analysis of biological preparations***

***Presentation of the possibilities of using confocal microscopy in biomedical research and pharmaceutical***

***Block 4: Introduction to the method of NMR spectroscopy, the possibilities of its use for the analysis of the structure of organic compounds and its application in pharmacy and medicine (5 hours) :***

***Introduction to nuclear magnetic resonance methods. Principles of recording NMR spectra on Bruker spectrometers.***

***Interpretation of <sup>1</sup>H, <sup>13</sup>C, <sup>31</sup>P NMR spectra.***

***Two-dimensional 2D-NMR nuclear magnetic resonance spectroscopy. Principles of registration of two-dimensional spectra.***

***COSY, HSQC, HMBC, NOESY techniques.***

***Solving structural problems concerning the structure of organic compounds and drugs by means of spectroscopic methods.***

***Block 5: Fundamentals of mass spectrometry (5 hrs):***

***"Fundamentals of mass spectrometry"***

***Analysis of mass spectra of low molecular weight organic compounds.***

***Types of ion sources and ionization mechanisms. Mass analyzers and ion detectors.***

***Construction of the QToF high-resolution mass spectrometer and introduction to its operation.***

***Methodology for performing HR MS analysis with a guide to the operation of the graphical interface of the program controlling the operation of the high-resolution mass spectrometer.***

### ***VERIFICATION OF LEARNING OUTCOMS***

***Method for the validation of learning outcomes***

*Enter how the doctoral student's knowledge is verified, e.g., oral colloquium, written test, written paper, presentation, other, etc.*

*Learning outcomes of the doctoral student verified by evaluation of activity in class and/or oral colloquium, and/or solution of subject problem tasks for the block of classes*

***Form and conditions of completing the course***

*The choice depends on the educational program: pass, pass/fail, exam. The course grade scale according to the Rules of the Doctoral School: very good 5.0, above good 4.5, good 4.0, fairly good 3.5, sufficient 3.0, fail 2.0.*

*Description of the requirements for passing the course, e.g. 100% attendance in classes, other, etc., guidelines for pass mark credit or exam According to the Training Program, the course ends with a credit.*

*Conditions for passing the course: 100% class attendance and a pass grade on the problem assignment and/or oral colloquium.*

**Primary literature**

1. Sambrook J, Russell D.W. Molecular Cloning: A Laboratory Manual, the third edition. Cold Spring Harbor Laboratory Press, Cold Spring Harbor, New York 4th ed. 2012.
2. Słomski R. (red.) Analiza DNA - praktyka. Wydawnictwo Uniwersytetu Przyrodniczego w Poznaniu, Poznań 2014.
3. Brown T.A. Genomy. Wydawnictwo Naukowe PWN, Warszawa wyd.3 2019.
4. Podstawy cytometrii. [w:] Wiktorowicz K, red. Ćwiczenia z immunodiagnostyki. Poznań: Akademia Medyczna im. Karola Marcinkowskiego w Poznaniu, 2001 str. 42-57.
5. Testy immunologiczne. [w:] Lydgard PM, Whelan A, Fanger MW. Krótkie wykłady. Immunologia. Warszawa: PWN 2001 str. 314-317.
6. Kawiak J. Cytometria przepływowa – badania immunocytochemiczne. [w:] Zabel M, red. Immunocytochemia. Warszawa: PWN, 1999 str. 260-284.
7. Kaczmarek A., Osawa T., Leporowska E. Mackiewicz A. Rola i miejsce cytometrii przepływowej w diagnostyce klinicznej. Wapółczesna onkologia 2002, 6(6):366-373.
8. Sędek Ł., Szczepański T., Mazur B. Techniczne aspekty cytometrii przepływowej. Journal of Laboratory Diagnostic 2010, 46(4):415-420.
9. Skotny A., Pucińska J. Współczesna cytometria przepływowa. Acta Bio-Optica et Informatica Medica 2013, 19(1):3-11.
10. Pawley J.B. Handbook Of Biological Confocal Microscopy. 3<sup>rd</sup> ed. Springer 2006, Boston
11. R.M. Silverstein, F.X. Webster, D.J. Kreml, „Spektroskopowe metody identyfikacji związków organicznych”, Wydawnictwo Naukowe, PWN, Warszawa, 2008.
12. "Spektroskopowe metody badania struktury związków organicznych" praca zbiorowa red. A. Rajca, W. Zieliński, WNT, Warszawa, 1995 lub 200
13. M. Szafran, Z. Dega-Szafran, „Określanie struktury związków organicznych metodami spektroskopowymi”, Tablice i ćwiczenia, PWN, 1998

**Supporting literature**

Wybrane aktualne artykuły naukowe wskazywane przez Prowadzącego.

1. Pygall SR, Whetstone J, Timmins P, Melia CD. Pharmaceutical applications of confocal laser scanning microscopy: the physical characterisation of pharmaceutical systems. Adv Drug Deliv Rev. 2007, 59, 1434-52. doi: 10.1016/j.addr.2007.06.018. Epub 2007 Aug 25. PMID: 17945376.
2. Zou Y, Celli A, Zhu H, Elmahdy A, Cao Y, Hui X, Maibach H. Confocal laser scanning microscopy to estimate nanoparticles' human skin penetration in vitro. Int J Nanomedicine. 2017, 12, 8035-8041. doi: 10.2147/IJN.S139139. PMID: 29184403; PMCID: PMC5673047.
3. Bernaś T. Mikroskopowe techniki korelacji fluorescencji. KOSMOS 2013, 62, 205-211
4. Brutkowski W. Mikroskopia konfokalna a mikroskopia szerokiego pola — dwa podejścia do badań przyżyciowych. KOSMOS 2013, 62, 171-180
5. R.J. Abraham, J. Fisher, P. Loftus "Introduction to NMR Spectroscopy" John Wiley and Sons, Chichester, 1988.
6. R. M. Silverstein, G. C. Bassler "Spektroskopowe metody identyfikacji związków organicznych" PWN, Warszawa, 1970.

