X Konwersatorium Chemii Medycznej

Book of Abstracts

3-5 września 2021

Lublin

Polskie Towarzystwo Chemii Medycznej

Uniwersytet Medyczny w Lublinie

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SCIENTIFIC PROGRAM

FRIDAY, 3.09.2021

15.00 – 17.00 Registration Hotel Victoria

17.00 – 17.10 Opening Ceremony

17.10 – 18.10 Inaugural Lecture (IL) Hotel Victoria

Rector of the Medical University of Lublin

Professor, MD, PhD Wojciech Załuska

18.10 Concert

19.00 Welcome reception

SATURDAY, 4.09.2021 Hotel Victoria

9.30 – 10.15 Lectures L1 (45')

10.15 – 11.15 Communications C1 – C3 (20')

Session moderators: Jadwiga Handzlik & Dariusz Matosiuk

L1

Lhassane Ismaili, Neurosciences EA, University Bourgogne Franche-Comté

„Environmentally friendly development of Multi-Target-Directed Ligands for Alzheimer's Disease"

C1

Dawid Panek, Jagiellonian University Medical College, Cracow, Poland

„Modulation of cholinergic and GABAergic neurotransmissions as a new strategy in Alzheimer's disease treatment"

C2

Tomasz Wichur, Jagiellonian University Medical College, Cracow, Poland

„Translating efficacy of combination therapy with 5-HT6R antagonist and cholinesterase inhibitor into novel multifunctional ligands against Alzheimer's disease"

C3

Katarzyna Szczepańska, Jagiellonian University Medical College, Cracow, Poland

„Dual histamine H3 and sigma-1 receptor ligands as novel pharmacological tools in the treatment of central nervous system disorders with the focus on neuropathic pain"

11.15 – 11.45 Coffee break

11.45 – 13.25 Communications C4 – C8 (20')

Session moderators: Katarzyna Kieć-Kononowicz & Lucjusz Zaprutko

C4

Wojciech Płaziński, Jerzy Haber Institute of Catalysis and Surface Chemistry
Polish Academy of Sciences, Cracow, Poland
„Chirality-related effects in biomolecular systems: methodologies for calculating the relative free energies by molecular dynamics simulations"

C5

Wojciech Pietruś, Maj Institute of Pharmacology, Polish Academy of Sciences,Cracow, Poland
„The role and significance of fluorine in medicinal chemistry - a review of in silico research"

C6

Beata Kolesińska, Lodz University of Technology, Poland
„Hybrid polysaccharide-peptide materials useful in the treatment of difficult-to-heal wounds"

C7

Michał Marszałł, Nicolaus Copernicus University in Torun, Collegium Medicum in Bydgoszcz, Poland
„Implementation of medical and pharmaceutical research to clinical practice"

C8

Katarzyna Wójcik-Pszczoła, Jagiellonian University Medical College, Cracow, Poland
„Novel 8-alkoxy- and 8-aminopurine-2,6-dione-based phosphodiesterase inhibitors as promising anti-asthmatic agents"

13.25 – 15.00 Lunch

15.00 – 16.30 Lecture L2 & L3 (45')

Session moderators: Barbara Malawska & Paweł Zajdel

L-2

David Nichols, Purdue University College of Pharmacy, West Lafayette,
LA, USA
„Overview of Medicinal Chemistry Research on Psychedelics"

L-3

Steffen Pockes, Institute of Pharmacy, University of Regensburg
„Development of selective caspase-2 inhibitors for the treatment of neurodegenerative disease"

16.30 – 17.00 Coffee break

17.00 – 18.10 Poster oral presentations PP1 – PP7 (10')

Session moderators: Zofia Mazerska & Marcin Mączyński

PP1

Michał Kosno, Gdansk University of Technology, Poland
„Unique physicochemical properties of the unsymmetrical bisacridines and their importance"

PP2

Tomasz M. Wróbel, University of Copenhagen, Denmark; Medical University of Lublin, Poland

„Non-steroidal CYP17A1 inhibitors”

PP3

Marek Jamrozik, Jagiellonian University Medical College, Cracow, Poland

„Molecular modeling of carbonyl reductase 1 and aldo-keto reductase 1C3 as new molecular targets in supporting cancer therapy”

PP4

Aleksandra Czerchawy, Lodz University of Technology, Poland

„Searching of biologically active fragments of hACE2 protein able to interact with S1 protein subunit of the SARS-CoV-2 virus”

PP5

Natalia Kocot, Collegium Medicum in Bydgoszcz, Nicolaus Copernicus University in Torun, Poland

„Application of immobilized lipases on Octyl-Sepharose CL-4B carrier as potential biocatalysts in obtaining pure enantiomers of NSAIDs”

PP6

Paweł Nowak, Nicolaus Copernicus University in Torun, Poland

„Interaction of HSA immobilized on magnetic nanoparticles with ketoprofen under artificially induced oxidative stress conditions”

PP7

Magdalena Dziurzyńska, Cracow University of Technology, Poland

„Activation of the dopamine D4 receptor – theoretical study”

17.30 – 18.30 Poster session

20.00 – 24.00 Get-together Party (Hotel Victoria)

SUNDAY, 5.09.2021 Hotel Victoria

9.15 – 10.35 Communications C9 – C12 (20’)

Session moderators: Dorota Piotrowska & Tomasz Gośliński

C9

Paweł Kafarski, Wroclaw University of Science and Technology, Poland

„Phopshonic acid analogues of phenylalanine as inhibitors of amino peptidases”

C10

Wojciech Szymanowski, Medical University of Bialystok, Poland

„Mucin 1 as a molecular target of a novel diisoquinoline derivative combined with anti-MUC1 antibody in MCF-7 breast cancer cells and AGS gastric cancer cells”

C11

Anna Szymanowska, Medical University of Bialystok, Poland

„Synthesis of novel sulfonamide pyrazolo[4,3-e]tetrazolo-[4,5-b][1,2,4]triazine derivatives and their proapoptotic effect on colon cancer cell lines DLD-1 and HT-29”

C12

Kamila Buzun, Medical University of Bialystok, Poland

„Anticancer activity of a new 2-thioxo-4-thiazolidinone derivative Les-3331 against MDA-MB-231 cancer cell line”

10.35 – 11.05 Coffee break

11.05 – 12.45 Communications C13 – C17 (20’)

Session moderators: Anna Bielawska & Krzysztof Bielawski

C13

Tomasz Koczorowski, Poznan University of Medical Sciences, Poland

„The power of a conjugated macrocyclic system and a metal ion complexation - porphyrazines as catalysts”

C14

Jacek Dulęba, Collegium Medicum in Bydgoszcz, Nicolaus Copernicus University in Torun, Poland

„The application of lipase Amano PS from Burkholderia cepacia as the potential biocatalyst in the reactions with pharmaceutical significance”

C15

Aleksandra Murzyn, Jagiellonian University, Cracow, Poland

„Multi-module pancreatic cancer therapy with the novel gold nanorod-based nanotherapeutic”

C16

Marek Staszewski, Medical University of Lodz, Poland

„Guanidine derivatives: From Broadened Bio-activity Profiling of Histamine H3R Antagonists to the Discovery of Potent Muscarinic M2R/M4R Antagonists”

C17

Przemysław Kołodziej, Medical University of Lublin, Poland

„Assessment of the nematicidal effect of some newly synthesized triazole derivatives”

13.00 – 14.30 Lunch

14.30 – 15.30 Communications C18 – C20 (20’)

Session moderators: Beata Morak-Młodawska & Łukasz Popiołek

C18

Kinga Mylkie, Academia Scientiarum Thoruniensis, Doctoral School of Exact and Natural Sciences, Poland

„Magnetic nanoparticles coated with modified starch for protein immobilization”

C19

Tomasz Laskowski, Gdansk University of Technology, Poland

„NMR studies on the antitumor acridine derivatives and their interactions with nucleic acids”

C20

Prezentacja firmy ABL&E-JASCO Polska Sp. z o.o., sponsora Konwersatorium

15.30 – 16.00 Coffee break

16.00 – 17.00 Poster oral presentations PP8 – PP13 (10’)

Session moderators: Monika Wujec & Roman Lesyk

PP8

Łukasz Balewski, Medical University of Gdansk, Poland

„Synthesis, structure and cytotoxic evaluation of novel imidazo[2,1-c][1,2,4]triazole derivatives”

PP9

Tadeusz Karcz, National Institute of Environmental Health Sciences, NIH, Research Triangle Park, North Carolina, USA; Jagiellonian University Medical College, Krakow, Poland

„Identification of purinergic receptor P2Y14 as a potential therapeutic target in allergic asthma”

PP10

Piotr Stępnicki, Medical University of Lublin, Lublin, Poland

„Synthesis and affinity studies of novel piperazine-based compounds as potential antipsychotics”

PP11

Damian Kułaga, Cracow University of Technology,Poland

„Design and synthesis of new potent 5-HT7 receptor ligands as a candidates for the treatment of central nervous system diseases”

PP12

Justyna Górska, Gdansk University of Technology, Poland

„Aromatic analogues of amphotericin B. How to isolate this promising agents for antifungal therapy?”

PP13

Valery Lutsyk, Jerzy Haber Institute of Catalysis and Surface Chemistry Polish Academy of Sciences, Cracow, Poland

„Conformational properties of glycosaminoglycan building blocks: molecular dynamics simulations”

16.00 – 17.00 Poster session

17.00 Closing of the conference

Lectures L1 - L3

L1

Environmentally friendly development of Multi-Target-Directed Ligands for Alzheimer's Disease

Lhassane Ismaili

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Dementia has become a global challenge. Recent reports highlight that as the world's population and life expectancy increase, the number of patients with dementia - Alzheimer's disease being the most common - will rise from 35 million to an alarming 135 million by 2050.

Alzheimer's disease is considered a highly complex, multifactorial neurodegenerative disorder for which there are no effective drugs despite unprecedented amounts of scientific publications (more than 100K in Pubmed). Polypharmacology could be a therapeutic opportunity to address this complex scenario[1]. We believe that rationally designed multi-targeted ligands (MTDLs), i.e. single molecules that simultaneously modulate several targets involved in the neurodegenerative cascade, can have disease modifying effects. Our approach is not only to develop drugs better suited to the complexity of Alzheimer's disease, but also to do so in a sustainable and more environmentally friendly way, preferring multicomponent reactions for atom economy, convergent synthesis to linear synthesis, microwave technology to conventional heating and less toxic solvents.[2].

In this talk, we will present these ideas and give an overview of our recent developments[3,4].

References

[1] Ismaili et al, Multitarget Compounds Bearing Tacrine- and Donepezil-like Structural and Functional Motifs for the Potential Treatment of Alzheimer's Disease. *Prog. Neurobiol.* **2017**, *151*, 4–34.

[2] Ismaili, L.; do Carmo Carreiras, M. Multicomponent Reactions for Multitargeted Compounds for Alzheimer`s Disease. *Curr Top Med Chem* **2018**, *17* (31), 3319–3327.

[3] Malek et al, New Dual Small Molecules for Alzheimer's Disease Therapy Combining Histamine H3 Receptor (H3R) Antagonism and Calcium Channels Blockade with Additional Cholinesterase Inhibition. *J. Med. Chem.* **2019**, *62* (24), 11416–11422.

[4] Ismaili et al,. (±)-BIGI-3h: Pentatarget-Directed Ligand Combining Cholinesterase, Monoamine Oxidase, and Glycogen Synthase Kinase 3β Inhibition with Calcium Channel Antagonism and Antiaggregating Properties for Alzheimer's Disease. *ACS Chem. Neurosci.* **2021**, *12* (8), 1328–1342.

L2

Overview of Medicinal Chemistry Research on Psychedelics

Nichols David

Purdue University, College of Pharmacy, 575 Stadium Mall Drive, West Lafayette, IN 47907, USA

This talk will begin with a brief review of the natural sources of psychedelics that have been used for millennia. That will be followed by a discussion of the 5-HT2A receptors that are targets for the classic psychedelics; where they are located and brief comments on the nature and function of g protein-coupled receptors (GPCRs). There will be brief discussion of how psychedelics are tested in animals and then discussion of some of the rigid analogue studies from the Nichols lab, including work to elucidate the conformation of the diethylamide function of LSD. In particularly, Leucine 229, in extracellular loop 2 of the receptor was shown to be a critical residue in the receptor kinetics of LSD binding to the receptor. Brief comments will be made on the crystal structure of LSD bound in the 5-HT2B receptor, published in 2017, and of LSD and 25CN-NBOH structures published in 2020. Some of the projects supported by the Heffter Research Institute, founded by the speaker in 1993, will be highlighted. A brief conclusion will note the effect of psychedelics on brain dynamics as a possible mechanism for their therapeutic potential.

L3

Development of Selective Caspase-2 Inhibitors for the Treatment of Neurodegenerative Diseases

Pockes Steffen [a,b,c]

[a] *Institute of Pharmacy, University of Regensburg, Universitätsstraße 31, 93053 Regensburg, Germany*
[b] *Department of Neurology, University of Minnesota, 2101 6th Street SE, Minneapolis 55455, USA*
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The group of caspases (cysteine-dependent aspartate-directed proteases) are protease enzymes cleaving target proteins after an aspartic acid residue.[1] They are traditionally related with programmed cell death (e.g. apoptosis) and can be subdivided in three groups: initiator caspases (e.g. caspase-2, -8, -9, -10), effector caspases (e.g. caspase-3, -6, -7) and inflammatory caspases (e.g. caspase-1, -4, -5). The caspase-2 (Casp2) enzyme plays a crucial role in neurodegeneration due to ischemic damage to the brain [2,3] and regulation of the decrease in spine density found in neuronal cultures and a mouse model of Alzheimer's disease (AD).[4] The cleavage of the tau protein by caspases is mainly attributed to Casp2 and Casp3, which leads to tau fragments inducing neurodegeneration in AD mice.[5] These results suggest that both tau and caspase-2 seems to forge a toxic partnership as the truncated non-fibrillar Δtau314, together with full-length tau, enter the dendritic spine, and thus contribute to neuronal disfunction.[6,7] For this reason, blocking the truncation of tau can be a promising therapeutic approach for AD or other tauopathies, like Huntington's disease (HD).
In this study, the long-term goal is to develop of new therapies for the treatment of cognitive disorders such as Alzheimer's disease and Huntington's disease. Thus, we propose to base on our ongoing studies of three, orthogonal approaches: (1) A brain-penetrant, selective Casp2 inhibitor, (2) Structure-activity relationships (SAR)-based modifications of candidate Casp2 inhibitors obtained in high-throughput screening (HTS) and (3) the native Casp2 cleavage site of different proteins including tau, which is YKPV**D314|L** (**D314|L** = cleavage site). On the basis of these approaches, there have already been tested more than 20,000 compounds, which are discussed in terms of their caspase-2 activity, their selectivity within the caspase family, and their pharmacokinetic properties.

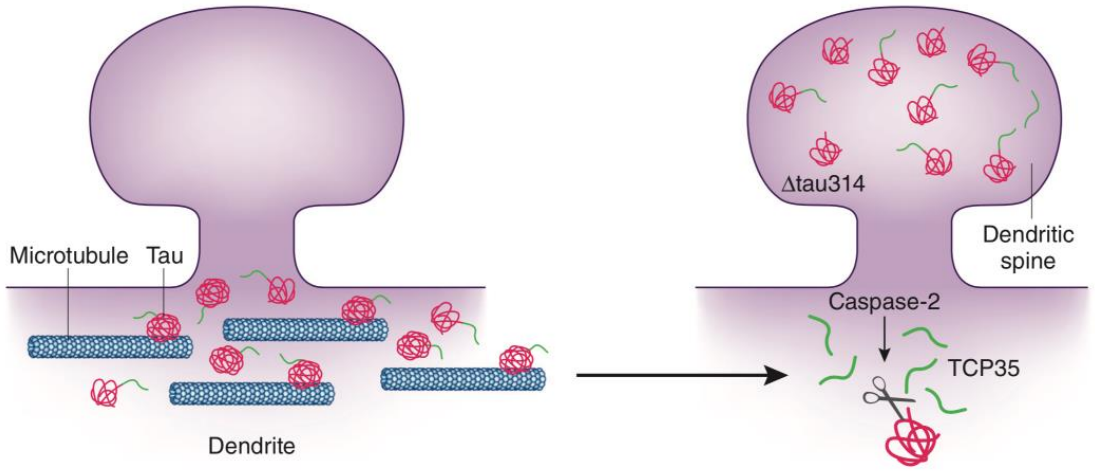

Figure 1: Caspase-2 cleavage of tau. Figure adapted from Troy et al. [7]

[1] Miles, M. A. et al.: Int. Rev. Cell Mol. Biol. 2017, 332, 155-212.
[2] Niizuma, K. et al.: Proc. Natl. Acad. Sci. 2008, 105(42), 16368-16373.
[3] Carlsson, Y. et al.: Pediatr. Res. 2012, 71(5), 566-572.
[4] Pozueta, J. et al.: Nat. Commun. 2013, 4(1), 1-12.
[5] Wang, Y. et al.: Biochem. Soc. Trans. 2010, 38(4), 955-961.
[6] Zhao X.: Nat. Med. 2016, 22(11), 1268-1276.
[7] Troy, C. M. et al.: Nat. Med. 2016, 22(11), 1207-1208.

Communications C1 - C20

C1

Modulation of cholinergic and GABAergic neurotransmissions as a new strategy in Alzheimer's disease treatment.

Dawid Panek[a], Anna Pasieka[a], Gniewomir Latacz[a], Jakub Jończyk[a], Justyna Godyń[a], Natalia Szałaj[a], Jaroslav Pejchal[b], Martin Mzik[c], Vit Sestak[c], Marketa Jerabkova[b], Jana Zdarova Karasova[b,c], Jan Korabecny[b,c], Ondrej Soukup[b,c], Fabien Chantegreil[d], Xavier Brazzolotto[d], Georg Höfner[e], Klaus Wanner[e]. Kinga Sałat[a], Anna Więckowska[a], Barbara Malawska[a]

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Alzheimer's disease (AD) is a progressive neurodegenerative disorder affecting around 35 million people worldwide. Complex and not fully explained etiopathogenesis, as well as asymptomatic development of the disease for several years , make it particularly difficult to search for an effective therapy.

Progression of AD leads to disturbances in neurotransmission systems, particularly cholinergic and GABAergic. Observed in AD decline in cholinergic neurotransmission may be compensate by inhibition of acetylcholinesterase (AChE) and/or butyrylcholinesterase (BuChE), that are enzymes hydrolysing acetylcholine. Overexpression of γ-aminobutyric acid transporter subtype 3 (GAT-3) observed on the astrocytes and microglial cells of hippocampus in animal models of AD leads to overloading of astrocytes with GABA, tonic inhibition and finally memory impairment [1]. We hypothesize that compounds blocking BuChE and GAT-3 at the same time may improve memory deficits in AD. Thus, we developed new multi-target-directed ligands simultaneously modulating both cholinergic and GABAergic neurotransmission by inhibition of BuChE and blocking GAT-3.

Based on the biological screening towards GAT-3 of our in-house library of BuChE inhibitors we selected a "hit" compound [2] that we optimized in terms of activity and drug-likeness. It led to the discovery of compound JT-3 inhibiting BuChE (IC_{50} = 0.22 μM) selectively over AChE and GAT-3 (IC_{50} = 7.76 μM). We tested its pharmacokinetics, safety profile as well as the effects on memory in animal models.

Acknowledgments: This work was supported by JUMC grant N42/DSB/000177.

[1] Wu Z. et. al. *Nature Communications* 5 (2014) 1 – 13
[2] Pasieka A et. al. *Eur. J. Med. Chem.* 218 (2021) 113397

C2

Translating efficacy of combination therapy with 5-HT$_6$R antagonist and cholinesterase inhibitor into novel multifunctional ligands against Alzheimer's disease

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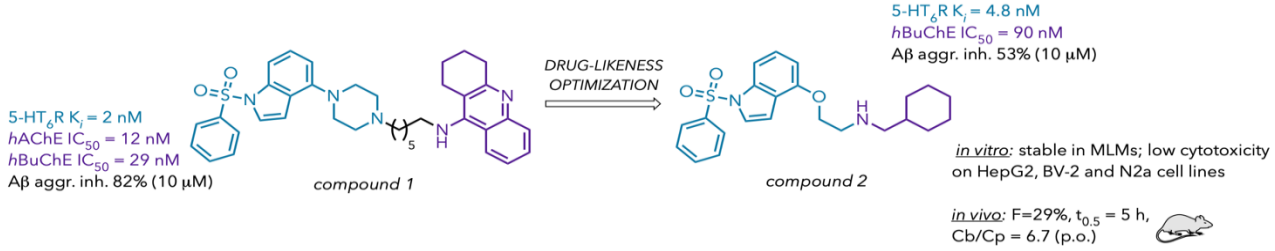
Alzheimer's disease (AD) is a complex and fatal neurodegenerative disorder mainly underlined by impaired neurotransmission, aggregation of amyloid-β (Aβ), and inflammation [1]. Currently available anti-AD drugs show limited efficacy [2] and a breakthrough may be brought by multifunctional ligands (MLs). Clinical trials of the combination therapy with the 5-HT$_6$R antagonist and the ChE inhibitor prompted us to search for MLs endowed with both activities. Following this idea, in our previous study, we have developed the first-in-class multifunctional 5-HT$_6$R antagonists–ChE inhibitors that were also able to inhibit Aβ aggregation as exemplified by compound **1** [3]. Herein we present the design, synthesis, and biological evaluation of a new set of multifunctional compounds with improved drug-like properties. Structure-activity relationship analyses supported by crystallography and docking studies led to the discovery of compound **2**, a balanced and potent multifunctional BuChE inhibitor (IC_{50} = 90 nM), 5-HT$_6$R antagonist (K_i = 4.8 nM), able to inhibit Aβ aggregation (53% at 10 μM). We carried out *in vivo* pharmacokinetic studies, preceded by positive results of the evaluation of metabolic stability on mouse liver microsomes (MLMs), cytotoxicity on HepG2 (human liver), BV-2 (mouse microglial) and N2a (mouse neuroblastoma) cell lines. Compound **2** was characterized by a half-life of ca. 5 h, absolute bioavailability of 29% and the brain to plasma ratio of 6.79 after p.o. administration, thus making it a promising candidate for *in vivo* pharmacology studies. Summing up, the determined structure-activity relationship within the obtained multifunctional ligands allowed for the identification of new, interesting scaffolds that may serve as a solid foundation for further research on effective anti-AD therapies.



Acknowledgements. This work was supported by the National Science Centre Poland, grant № 2016/23/D/NZ7/01328.

[1] Nesi, G. et al. Curr. Top. Med. Chem., 2017, 17, 3062–3079.
[2] H. Hampel et. al. Brain, 2018, 141, 1917–1933.
[3] Więckowska, A. et al. Eur. J. Med. Chem., 2016, 124, 63–81.

C3

Dual histamine H_3 and sigma-1 receptor ligands as novel pharmacological tools in the treatment of central nervous system disorders with the focus on neuropathic pain

Katarzyna Szczepańska [a,b], Sabina Podlewska [a,b,], Maria Dichiara [c], Vincenzo Patamia [c], Niklas Rosier [d], Denise Mönnich [d], M. Carmen Ruiz-Cantero [e], Tadeusz Karcz [a], Dorota Łażewska [a], Steffen Pockes [d], Enrique J. Cobos [e], Holger Stark [f], Antonio Rescifina [c], Andrzej J. Bojarski [b], Emanuele Amata [c], Katarzyna Kieć-Kononowicz [a]*

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Since central nervous system disorders are characterized by diverse physiological dysfunctions and deregulations of a complex network of signaling pathways, optimal multipotent drugs should simultaneously and peculiarly modulate selected groups of biological targets. Interestingly, very recent studies have shown that some clinically evaluated histamine H_3 receptor (H_3R) antagonists possess nanomolar affinities at sigma-1 receptor (σ_1R) binding sites, suggesting that this property might play a role in their overall efficacy [1]. Therefore, we selected twenty representative structures among our previously reported H_3R ligands to investigate their affinity at σRs. Most of the tested compounds interact with both sigma receptors (σ_1R, σ_2R) to different degrees. Moreover, all these ligands share a common structural feature: the piperidine moiety as the fundamental part of the molecule. It is most likely a critical structural element for dual H_3/σ_1 receptor activity. Using molecular modeling techniques we identified the putative protein-ligand interactions responsible for their high affinity and then, selected compounds **KSK68** and **E377** as lead structures for further evaluation. Interestingly, both ligands turned out to be high-affinity histamine H_3 and σ_1 receptor antagonists with negligible affinity at the other histamine receptor subtypes and therefore promising antinociceptive activity *in vivo*. Considering that literature data clearly indicate high preclinical efficacy of individual selective σ_1R or H_3R ligands in various pain models, our research might be a breakthrough in the search for novel, dual-acting compounds that can improve existing pain therapies [2]. Whether such ligands are now more effective than single selective drugs will be the subject of our future studies.

We are pleased to acknowledge the generous support of the National Science Center, Poland granted on the basis of decision No. 2020/36/C/NZ7/00284.

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C4

Chirality-related effects in biomolecular systems: methodologies for calculating the relative free energies by molecular dynamics simulations

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Chirality plays an essential role in chemical and biological sciences. At the molecular level, the effects associated with this phenomenon can be studied by using well-established technique of molecular dynamics simulations. We have proposed several theoretical methods designed to study the chirality effects in biomolecular systems. They have been validated in the context of molecular dynamics simulations within classical, molecular mechanics force fields for selected model systems (CHFClBr, fenoterol, fructofuranose). All proposed methods rely on using enhanced-sampling methods to enable for interconversion between stereoisomers and on calculating the corresponding, unbiased populations of particular stereoisomers.

We have proposed and tested the following strategies: (i) biased sampling in the two-dimensional space, along the coordinates defined by the values of the selected torsional angles; (ii) biased sampling in the one- or two-dimensional space, along the path-based coordinate(s); (iii) rational alteration of the system's Hamiltonian in order to enable the interconversion between stereoisomers and reweighting the biased distribution of configurations; (iv) using the free energy landscape generated within approaches (i) or (ii) as time-independent bias in order to further improve sampling efficiency and simultaneously account for multiple chiral centers. All approaches demonstrated the good performance for at least one among model systems but some serious differences in the range of their applicabilities have been found.

We believe that the developed strategies can serve as starting points, allowing for their further refinements and optimizations with regard to the subject of interest. In particular, they can be generalized to the case of stereoselective binding of ligand-biomolecule by performing separate simulations for bound and unbound ligand and constructing the appropriate thermodynamic cycle to obtain the relative binding free energies

This research was funded by the statutory research fund of ICSC PAS. It was also partly funded by the National Science Centre, Poland (contract financed in 2019–2021 under Project Opus, No. 2017/25/B/NZ7/02654).

C5

The role and significance of fluorine in medicinal chemistry - a review of *in silico* research

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Fluorine is the most electronegative element found in nature, which in comparison to the other halogens (Cl, Br, or I) induces a change bioavailability, lipophilicity, metabolic stability, acidity/basicity, and toxicity [1]. Recently, the fluorination of chemical molecules has become the commonly used modification in the development of new drugs. Since the approval by the Food and Drug Administration (FDA) of the first drug containing fluorine substituent (1950), over 200 fluorine-containing drugs have been released to the market (more than 400 in clinical trials), which now makes up approximately 20% of all pharmaceuticals [2]. In recent years, the number of fluorinated drugs has increased to about 30% of all newly approved drugs and according to the latest statistics, drugs containing fluorine atom are among the most recommended and, at the same time, the most profitable in the US pharmaceutical market [3]. Surprisingly fluorine, as the most electronegative element, is a weak acceptor of hydrogen bonds (HB) and in contrast to other elements in its group, it cannot create halogen bonds (XB) in organic molecules [4,5].

Despite the importance of fluorine in medical chemistry, it is still not fully explained how this element affects both physicochemical properties of compounds, and changes in their biological activity. Quite often the search for new fluorinated derivatives takes place through trial and fault because the available software for rational drug design does not have adequate information to predict changes caused by the substitution of hydrogen (or other functional groups) by fluorine at different places in the molecule.

The results show that fluorine strongly influences the strength and frequency of the formation of intermolecular interactions of strong donors and acceptors of HB. Additionally, thanks to fluorine substitution, the energy of HB and XB can be tuned

, and in difluorinated compounds, fluorine becomes an attractive and competitive acceptor of HB and XB. In biological systems, HBs with fluorine show differences from the classic hydrogen bonds (O-H···O, N-H···O) - they slightly depend on the angle and prefer hydrophobic parts of the binding pockets. In aminergic GPCR receptors, fluorine substitution of aliphatic parts more often leads to a decrease in biological activity, while the favored position of fluorine to elicit a positive effect on biological activity is the *ortho* position in the aromatic ring.

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C6

Hybrid polysaccharide-peptide materials useful in the treatment of difficult-to-heal wounds

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Dressing materials are used in regenerative medicine to treat wounds, as well as materials supporting the regeneration of damaged tissues[1]. As a part of the research work carried out at the Institute of Organic Chemistry of the Lodz University of Technology, we conduct research on new, biocompatible, multi-component hybrid materials useful in the treatment of non-healing wounds, including diabetic foot syndrome[2,3]. The final hybrid materials stimulate all stages of the wound healing process, thereby providing the basis for a holistic wound treatment tool. The obtained multicomponent hybrid materials meet all the criteria of a uniform concept for treating chronic wounds - TIME strategy (Tissue debridement, Infection and inflammation control, Moisture balance, Epidermation stimulation). In the group of diseases occurring with the formation of difficult-to-heal wounds, the most common changes include wounds of the lower limbs. These include vascular wounds, diabetic foot syndrome, trophic ulcers, haematological wounds, wounds in purulent gangrenous dermatitis, cancer ulcers and wounds accompanying congenital vascular malformations. The problem of diabetes could affect up to 3 million people in Poland. The diabetic foot syndrome is one of the most common causes of hospitalization among diabetic complications and affects about 4-10% of patients. This is responsible for about 5% of chronic wounds; in Poland this problem affects 10% of people with diabetes. The risk of ulceration in a diabetic person ranges from 12 to 25%. It is also the most common non-traumatic cause of amputation in the lower limbs. The risk of amputation in the population of people with diabetes is even 30-40 times higher than in the general population.

As a result of the synergy between the selected set of biologically active peptides and polysaccharides, it has been found that it is possible to obtain a hybrid material useful in the treatment of difficult-to-heal wounds that affect (i) hemostasis (fragments: α- and γ-chain of fibrinogen, lactoferrin, fibronectin, factor XIII); (ii) inflammation (fragments of pro and anti-inflammatory cytokines); (iii) proliferation (fragments of ECM components: collagens, elastin, fragments of growth factors); and (iv) remodeling of tissue and protection against bacterial infection (fragments of peptides with antibacterial activity). Additionally, in the biodegradation process, these materials are decomposed only into natural compounds.

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C7

Implementation of medical and pharmaceutical research to clinical practice

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Collaboration between scientists and medical professionals is essential. The last decade has been very successful in the polish and international scientific area. This is evidenced by the implementation of many joint research, research and development projects as well as publications. These projects often cover the area of translational medicine – the concept of which is to combine laboratory and medical practice and transferring the new knowledge to clinical practice. It turns promising laboratory discoveries into clinical applications. It facilitates the prognosis, prevention, diagnosis and treatment of diseases. Undoubtedly, its most importnt element is bidirectional - it involves stimulating the transfer of information from laboratory to clinic and conversely. Its positive effect are already visible today. Willingness to cooperate and striving to achieve the set goal - create reserach teams with great interdisciplinary potential. Such teams are able to react quickly in treatment of civilization diseases.

C8

Novel 8-alkoxy- and 8-aminopurine-2,6-dione-based phosphodiesterase inhibitors as promising anti-asthmatic agents

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Bronchial asthma is a chronic lung disease affecting over 260 million people worldwide. Asthma is commonly classified as an inflammatory disease, but it is now known that airway remodeling associated with inflammation represent a serious clinical problem. While modern pharmacotherapy copes well with inflammation and the resulting exacerbations of the disease, chronic changes in the structure of the respiratory tract constitute a major therapeutic challenge. Recently, it has been demonstrated that selective inhibitors of phosphodiesterases (PDEs), enzymes that increase the intracellular level of cyclic nucleotides, may lead to an inhibition of both immune and lung resident cells' proinflammatory response. As it has also been shown that some PDE inhibitors can affect airway remodelling, there is now a growing interest in searching for new, effective PDE inhibitors. Here, we designed and synthesized a series of hydrazide and amide derivatives of 8-alkoxy- and 8-aminopurine-2,6-dione and characterized them as prominent pan-PDE inhibitors. Subsequently, we examined their influence on inflammation and airway remodelling related features in lung structural cells including airway smooth muscle cells, lung fibroblasts, bronchial epithelial cells and alveolar epithelial type II cells [1-3]. The obtained results prompted us to conclude that 8-alkoxy- and 8-amino-purine-2,6-dione derivatives represent promising agents for use in asthma therapy and need further detailed research in *in vivo* chronic asthma model.

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C9

Phopshonic acid analogues of phenylalanine as inhibitors of amino peptidases

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Inhibition of the activity of enzymes, which catalyse chemical reactions important for the development of pathogenesis is one of the major means of searching for new medicines. Small molecules able to bind to catalytic centres of the enzymes might be considered as lead compounds and/or building blocks in the process of designing of candidates for novel drugs.

The discovery of the first inhibitor belonging to aminophosphonate family - inhibitor of glutamine synthesise [1], initiated the development of chemistry of compounds with C-P bond, especially on their application as inhibitors of various enzymes metabolising amino acids and peptides [2]. These mimetic of amino acids and their small peptides appeared to be the most potent inhibitors of amino peptidases, with special attention given to leucylaminopeptidase and aminopeptidase N, both being considered as molecular targets for anticancer agents.

Phosphonic acid analogue of phenylalanine is the most potent amongst simple analogues of amino acids. It is quite commonly used as a building block in the search for novel, more potent inhibitors. Therefore, it seems to be of interest to identify how the modification of the structure of this mimetic influence its inhibitory potency. Recently we have synthesised series of such analogs of phenylglycine, phenylalanine and homophenlyalanine substituted with halogens in various positions of their phenyl rings. This resulted in series of interesting inhibitors. Their interactions with the chosen amino peptidases was further studied by means of molecular modeling. This, in turn, enabled to analyse their interactions with amino peptidases and to design novel inhibitors of more complex structure. Good results of inhibitory activities of the latter ones indicated the utility of molecular modeling in such studies.

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C10

Mucin 1 as a molecular target of a novel diisoquinoline derivative combined with anti-MUC1 antibody in MCF-7 breast cancer cells and AGS gastric cancer cells

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One of the strategies used in therapy aimed at specific molecular targets is the use of monoclonal antibodies with chemotherapeutic agents, which leads to an increase in their anticancer properties. A promising and attractive molecular target is mucin 1 which shows increased expression in many types of cancer. MUC1 may also contribute to the activation of signaling pathways responsible for tumour progression, as well as interact with many transcription factors [1].

The effect on the cytotoxic activity and the process of DNA biosynthesis of the OM-86II compound and the anti-MUC1 antibody used both in monotherapy and in combined treatment was checked in MCF-7 breast cancer and AGS gastric cancer cells. A key element of the presented study was to evaluate the effect of the compounds on molecular mechanisms leading to the induction of apoptosis. The effect of monotherapy and combined treatment of the compounds on the mitochondrial membrane potential, as well as on the concentration of selected signalling proteins: Bax, mTOR kinase, sICAM-1, MMP-2 and MMP-9 were compared. It was also checked how the use of the compound OM-86II together with the anti-MUC1 antibody affects the expression of p53 protein in the tested cell lines.

The study showed that the new derivative OM-86II in combination with anti-MUC1 antibody possesses higher cytotoxic and antiproliferative activity as compared to monotherapy and therapy with etoposide used with anti-MUC1 antibody. The molecular mechanism of anticancer action of the OM-86II and anti-MUC1 antibody is based on induction of apoptosis both in MCF-7 breast cancer and AGS gastric cancer cells. The process is related to activation of p53 protein in the tested cell lines and follows the intrinsic pathway. The study also showed that the incubation of MCF-7 cancer cells with the OM-86II compound used with anti-MUC1 antibody leads to an increase in the concentration of the proapoptotic protein Bax and a decrease in the concentration of signal transmitting proteins such as mTOR, sICAM-1, MMP-2 and MMP-9. In contrast, the incubation of AGS gastric cancer cells with OM-86II used with anti-MUC1 antibody leads to a decrease in the concentration of mTOR, sICAM-1 and MMP-9, as well as an increase in the concentration of the proapoptotic protein Bax and MMP-2.

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C11

Synthesis of novel sulfonamide pyrazolo[4,3-*e*]tetrazolo-[4,5-*b*][1,2,4]triazine derivatives and their proapoptotic effect on colon cancer cell lines DLD-1 and HT-29

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Colorectal cancer (CRC) represents a major health problem worldwide because of its mortality. There is a list of predisposing risk factors that play an important part in CRC development. A positive family history and environmental lifestyle factors such as alcohol intake, smoking, bad dietary habits, including low intake of vegetables and fruits as well as high red meat intake, affect disease pathogenesis [1].

In the last ten years a huge number of 1,2,4-triazine derivatives have been synthesized. Currently novel compounds are at different stages of *in vitro*, *in vivo*, and clinical trials. The observed increased interest in this group of derivatives is related to their wide biological activity [1-3].

The presented research involved synthesis of novel sulfonamide pyrazolo[4,3-e]tetrazolo-[4,5-b][1,2,4]triazine derivatives and evaluation of their effect on the apoptosis in colon cancer cell lines DLD-1 and HT-29.

Studies of molecular mechanism of action revealed that all synthesized compounds lead to initation of apoptosis in both colon cancer cell lines, which was confirmed by cytometric method using annexin V-FITC and by fluorescence microscopy using acridine orange and ethidium bromide staining. These compounds lead to a decrease of mitochondrial membrane potential and an increase in caspase 9 activity, tentatively confirming the activation of apoptosis via intrinsic pathway. Moreover these derivatives enhance the activity of caspase 8 and 10 indicating that the programmed cell death in DLD-1 and HT-29 cell lines is also activated via extrinsic pathway.

Obtained results provide a reason to continue the research work aimed at identifying molecular mechanism of action of the derivatives against colon cancer cells. Moreover functionalization of pyrazolo-[4,3-*e*]tetrazolo[4,5-*b*][1,2,4]triazine scaffold may contribute to design structures with high cytotoxic activity against cancer cells without effect on normal cells. In this context, it also seems reasonable to evaluate the activity of the synthesized sulfonamide derivatives against other types of cancer.

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C12

Anticancer activity of a new 2-thioxo-4-thiazolidinone derivative Les-3331 against MDA-MB-231 cancer cell line

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One of the promising direction for designing new structures of "drug-like" molecules, is the "hybrid-pharmacophore" approach. More specifically, this design approach involves combining different fragments in a new molecule that may be biomimetics and/or bioisosters, or part of a biologically active molecule or drug. The application of such a strategy at the structure design stage allows for the emergence of new therapeutic effects or the enhancement of a desired compound action. This constitutes a very important aspect in the process of searching for new, highly active 4-thiazolidinones derivatives, which constitute effective biophores. Modern studies on the biological activity of thiazolidinones resulted in the discovery of new properties of this group of compounds, including their anticancer, antiviral, antifungal, antibacterial or antiparasitic activity [1].

The aim of this study was to determine the molecular mechanism of anticancer potential of a new 2-thioxo-4-thiazolidinone derivative Les-3331 in MDA-MB-231 cancer cells. Cytotoxic and antiproliferative activity was measured using MTT assay and [^{3}H]-thymidine incorporation assay. Flow cytometry was used to examine the influence of a novel derivative on mitochondrial membrane potential and induction of apoptosis. Concentrations of caspase-8, caspase-9, topoisomerase II, LC3A, LC3B and Beclin-1 were determined using the ELISA method.

The obtained results showed that the novel 2-thioxo-4-thiazolidinone derivative Les-3331 exhibits a stronger cytotoxic and antiproliferative activity compared to the reference drug – etoposide. Les-3331 induces apoptosis and decreases mitochondrial membrane potential in human breast cancer cells. Furthermore, Les-3331 leads to a decrease in caspase-8, caspase-9 and topoisomerase II concentration as well as proteins involved in autophagy process.

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C13

The power of a conjugated macrocyclic system and a metal ion complexation - porphyrazines as catalysts

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Porphyrazines (Pzs) along with porphyrins and phthalocyanines belong to porphyrinoid-like macrocyclic compounds. Pz macrocycle consists of four pyrollyl rings linked together by *meso* nitrogen atoms in place of methine bridges which are present in the porphyrin macrocyclic ring. There are two main possibilities to modify the properties of porphyrinoids. The first one is to substitute metal cation to the macrocyclic core, whereas the second one relies on peripheral modifications [1]. Pzs reveal many potential applications in medicine and technology due to their spectral and electrochemical properties. For the last 30 years, porphyrazines substituted at their peripheral positions with nitrogen, oxygen, or sulfur groups have been synthesized and subjected to various physicochemical as well as biological studies. Pzs have been researched as potential photosensitizers for biomedical, analytical applications and as compounds for materials chemistry. Conjugated double bonds within Pz macrocyclic ring influence its strong electron-withdrawing effect [2]. For many years of research on porphyrazines, their cores have been equipped with various transition metal cations i.e., iron, zinc, cobalt, manganese, nickel, and copper, which impacted their electrochemical properties and allowed to use in various redox reactions [2].

I will present recent studies on the synthesis, physical- and electrochemical characterization of various peripherally substituted metallic porphyrazines applied in electrocatalytic as well as photocatalytic batch measurements with the use of selective chemicals or active pharmaceutical ingredients, like 1,3-diphenylisobenzofurane, L-cysteine, NADH, diclofenac sodium salt and ibuprofen as substrates [3-5].

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C14

The application of lipase Amano PS from *Burkholderia cepacia* as the potential biocatalyst in the reactions with pharmaceutical significance

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Lipases in free or immobilized form belong to the most common applied catalysts in the chemical and pharmaceutical industry. In this project, the kinetic resolution of (*R,S*)-1-phenylethanol catalyzed by Amano lipase PS from *Burkholderia cepacia* (APS-BCL) has been performed. The reaction was carried out in reaction media with different logP values (*t*-butylmethyl ether, dichloromethane, diisopropyl ether, toluene, cyclohexane, *n*-hexane, isooctane, and *n*-heptane). The high enantioselectivity was obtained for isopropenyl acetate as the acyl donor (*E* =308.5). The best solvent turned out to be diisopropyl ether, *C* =47.9%, ee_p =98%, ee_s =90% after 24 h of incubation. Furthermore, the effect of ω6/ω9 polyunsaturated fatty acids (PUFAs) and monounsaturated fatty acids (MUFAs) ratio in vegetable oils (from peanut, camelina, rape, pumpkin seed, walnut, sesame, avocado, rice, corn, black cumin, hemp, safflower, grape seed) was examined for the lipase activity. For the first time, the cut-off limit was suggested. The ratio equal to or higher than 2.3 allows receiving higher lipolytic activity.

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C15

Multi-module pancreatic cancer therapy with the novel gold nanorod-based nanotherapeutic

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Pancreatic cancer is one of the worst neoplastic human diseases. It is caused by difficult, too late diagnostics and poor treatment effectiveness [1]. A possible chance to increase the capability of treatment is the use of multi-module therapy and modern imaging techniques. Currently, gemcitabine is a commonly used chemotherapeutic agent in the anti-cancer treatment of pancreatic tumors [2]. For this purpose, a new nanotherapeutic was designed based on traditional gemcitabine combined with treatment in mild hyperthermia using gold nanoparticles [3,4].

The study aimed to check the impact of a new nanotherapeutic and its mechanism of action in the in vitro model.

The synthesis of the gemcitabine conjugate with carboxylated gold nanorods was performed using the water-soluble carbodiimide (EDCI /NHS) method, combining gemcitabine amino groups with the carboxyl coating of the gold nanorods. The new nano-therapeutic has been characterized by transmission electron microscopy, DLS, IR, HPLC, and XRF techniques. Construct properties such as solubility, stability, and toxicity were tested in vitro. MTT and long-term microscopic (JuliStage®) cell survival assays were performed in Panc-1, Pan_02. To obtain the hyperthermic effect, cells were irradiated for 10 min with 808 nm light (dose: 26.46 J / cm2).

An intense absorbance spectrum of the construct was observed at a wavelength of 808 nm. Additionally, it has been shown that the effect of hyperthermia is strongly dependent on the power density and the wavelength of the light source. Nanoconjugate increases toxicity and lowers the level of metabolic activity of cells.

To summarize, the new drug, which is a combination of a traditional chemotherapeutic agent with a modern approach to gold nanoparticles and hyperthermia, can be a very good drug carrier and makes it possible to effectively treat neoplasms.

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C16

Guanidine derivatives: From Broadened Bio-activity Profiling of Histamine H_3R Antagonists to the Discovery of Potent Muscarinic M_2R/M_4R Antagonists

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This work presents the discovery of novel potent muscarinic receptor antagonists identified during the searching for more active H_3R ligands[1]. Based on the previously obtained data of guanidine series the lead H_3R antagonist - ADS1017 [2], was selected for further structural optimization. The idea was to replace the flexible seven methylene linker with a semi-rigid 1,4-cyclohexylene or p-phenylene substituted group. These simple structural modifications of the H_3R antagonist led to the emergence of additional, unexpected pharmacological effects. During the *ex vivo* assay on the guinea pig ileum, decreasing the electrically-evoked contractility of ileum smooth muscles was noticed. This effect could be related to H_3R agonism as well as on the action on the muscarinic receptors present in the tested tissue. The tests carried out on the guinea pig ileum showed that the rigidity of the seven-carbon alkyl chain, not only decreases affinity at the histamine H_3R, but above all significantly increases activity at muscarinic receptors. To clearly explain on which one of the muscarinic receptor subtypes selected compounds act on, the hM_1-hM_5 radioligand binding experiments were performed in the membrane fractions of Chinese hamster ovary cells (CHO) stably expressing human variants of muscarinic receptors. To understand the molecular basis of the unexpected muscarinic activity, *in silico* studies were conducted.

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C17

Assessment of the nematicidal effect of some newly synthesized triazole derivatives

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Parasitic invasions pose a serious threat to human and animal health and cause very large economic losses worldwide. Excessive and inappropriate use of anthelmintic drugs leads to ever increasing drug resistance. There is an urgent need to find new, alternative nematicides for use in the treatment of human and animal parasites. Triazole derivatives are increasingly used in human and veterinary medicine. Research in recent years shows that triazole derivatives interact with various biological targets. The above mentioned derivatives show, inter alia, antitumor, antibacterial, antiviral and anthelmintic activity [1-4]. The aim of the research was to analyze the nematicidal activity of new triazole derivatives. The nematocidal activity of the newly synthesized derivatives was assessed according to the proprietary procedure [5]. The obtained results showed that the tested triazole derivatives showed a nematicidal effect.

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C18

Magnetic nanoparticles coated with modified starch for protein immobilization

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Albumin (HSA) and α_1-acid glycoprotein (AGP) are the most important proteins in human blood serum. One of their key functions is to binding and transporting exogenous and endogenous substances such as drugs [1]. HSA is composed of 585 amino acid with six binding site of which two are best characterized (the Sudlow site I and the Sudlow site II) [2]. The α_1-acid glycoprotein molecule contains a single polypeptide chain composed of a protein core (183 amino acids) and five branched glycan chains. After albumin, AGP is the main protein binding and transporting drugs, mainly of an alkaline nature. The serum concentration of HSA is about 50 times higher than that of AGP, therefore in the case of drugs that bind to both albumin and AGP, quantitatively more of the drug is bound to albumin. The study of drug-protein interactions is of key importance in pharmacology, especially in the case of newly synthesized active substances with the expected pharmacological activity. Protein-bound drugs are pharmacologically inactive - only the free, unbound fraction of the drug is active. Traditional methods of determining the basic pharmacokinetic parameter, which is the degree of drug binding, are characterized by high difficulty of implementation and low repeatability due to the complicated process of separating the protein from the supernatant. An alternative method is to immobilize the protein on a support that would be easy to separate from the reaction mixture and at the same time allow the protein to remain active [3].

In this work, magnetic nanoparticles were synthesized by standard co-precipitation reaction and coated with modified starch. Because of different chemical properties of HAS and AGP two methods of starch modification were used to immobilize proteins. For the immobilization of human serum albumin, starch was oxidized and then enriched with amino groups. In order to immobilize the α_1-acid glycoprotein, starch was enriched with dihydroxyboryl groups. Magnetic nanoparticles obtained as carriers for plasma protein immobilization were fully characterized by ATR-FTIR, TEM, SEM and thermal analysis. The amount of protein immobilized was determined by the Bradford method.

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C19

NMR studies on the antitumor acridine derivatives and their interactions with nucleic acids

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Acridine has a long tradition in service as an attractive basis for rational design of potential antitumor agents. At our Department, faith in acridine as structural basis was so strong that it resulted in the introduction of first, Polish antitumor drug – Ledakrin (Nitracrine). While novel and potent antitumor acridine derivatives are still being designed in Gdańsk, the most successful compounds belong to the families of imidazoacridinones, triazoloacridinones and nitroacridines.

Triazoloacridinone C-1305 and imidazoacridinone C-1311 (Symadex) are topoisomerase II poisons, which were recommended for Phase I and Phase II clinical trials, respectively. Both display high activity towards breast cancers and colon carcinomas. Although they are classic minor-groove dsDNA intercalators, recent NMR studies have proven that they exhibit some unusual sequence specificity, targeting 5'-TA-3' dinucleotide step whenever available. Also, dsDNA-sequence screening has shown that sequence specificity of C-1305 and C-1311 should be considered in terms of tetranucleotide steps, while the 5'-CTAG-3' sequence seems to be the most favorable so far[1]. On the contrary, another promising antitumor agent, nitroacridine C-1748 turned out NOT to be an efficient dsDNA intercalator. It exhibited some unspecified interactions with dsDNA, most likely binding to the minor groove.

Ongoing studies have recently revealed that imidazoacridinones are also targeting DNA G-quadruplexes. While proton NMR spectra of all G-quadruplexes studied to this date were significantly altered by the presence of C-1311, formation of structurally well-defined complexes seems to be reserved exclusively for parallel G-quadruplexes, such as c-MYC Pu22[2] and K-RAS 22RT[3]. C-1311 provides substantial, thermal stabilization of those two structures by formation of DNA/drug 1:2 mol/mol complexes, which was revealed by detailed NMR investigations. Complete structural studies on these two non-covalent adducts are work-in-progress. It should also be noted that Pu22/C-1311 complex is of special interest, since titration of pure, $Na^+/K^+/NH_4^+$ cation-free solution of unstructured Pu22 with a similar solution of C-1311 results in the formation of parallel G-quadruplex/ligand complex. Such a ligand-forced G-quadruplex formation in the presence of trace amounts of lithium ions is quite rare end even more exciting.

Moreover, it was revealed that nitroacridine C-1748 also targets G-quadruplexes, but, interestingly, not by π-π interactions between guanine/ligand ring systems. Finally, triazoloacridinone C-1305, supposedly working in a similar fashion to C-1311, is yet to be studied.

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C20

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Poster oral presentations PP1 - PP13

PP1

Unique physicochemical properties of the unsymmetrical bisacridines and their importance

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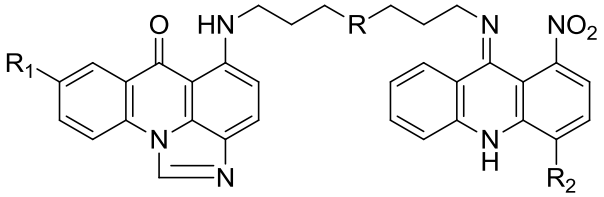
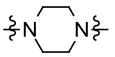
Unsymmetrical bisacridines, UAs, belong to the novel and prosperic class of chemical compounds and exhibit high antitumor activity towards tumor cell lines, e.g. prostate tumor DU-145, colon HCT-116, lung H-460 and breast MDA-MB-231. Particularly noteworthy is the fact that these compounds are active towards pancreatic cancer cells. Considering that there are almost no effective drugs against pancreatic cancer, UAs seem to be very promising as a potential drugs.



	C-2028	C-2041	C-2045	C-2053
R				
R_1	-	-		-
R_2	-	-		

To investigate properly the metabolic transformations or interactions with DNA, the exact structure of a studied compounds must be known. There are hardly any reports regarding the protonation of acridine monomers, hence we've decided to investigate the proton dissociation constants equilibrium of the dimer UAs studied. Using UV-Vis spectra with the advanced chemometric analysis, we've discovered that there are five pKa values for C-2028 and most probably six for C-2045. Order of proton dissociation was precisely determined by NMR spectroscopy. All conducted research indicated that there is a significant change in the electron density between the two forms, which can be found under physiological conditions. Additionally, the individual UA forms associated with protonation were shown to have various tendencies to self-association and probably can exist in a monomeric, dimeric or even micellar form. This discovery sheds new light on the nature of these agents, which can be metabolised differently, depending on their protonation and aggregation state. From the metabolic point of view, oxidative enzymes, other than CYPs, turned out to be particularly interesting. The representative class considered is flavin-containing monooxygenase (FMO). Our previous studies suggest that UAs are poorly metabolised by FMO. Recent findings allowed us to better understand this result and help us better simulate the physiological conditions during further *in vitro* investigations. Furthermore, UAs are being transformed in an uncommon fashion by many other classes of enzymes and, due to our recent findings, we may finally understand, why.

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PP2

Non-steroidal CYP17A1 inhibitors

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Prostate cancer is responsible for 7.1% of all cases of cancer considering both sexes. In males alone, prostate cancer is the second most frequently diagnosed form of cancer [1]. Cytochrome P450 17A1 (CYP17A1) is a well-established target in prostate cancer. However, even though it has been known for over a decade, only one CYP17A1 inhibitor (Abiraterone, approved in 2011) has so far been approved for clinical use. Abiraterone prolongs a patient's life by only 4 months and mainly because of its steroidal scaffold it is burdened with many side effects [2]. CYP17A1 is an enzyme involved in the synthesis of steroids. It is therefore a pivotal target in the treatment of hormone-dependent tumors such as prostate cancer [3]. Because androgenic signaling has been proposed as the primary driver of castration-resistant prostate cancer, it remains an important area of research as there are, yet no effective treatments for true hormone resistance [4]. Moreover, the area of non-steroidal inhibitors of CYP17A1 is underexplored. There are only handful of nonsteroidal CYP17A1 inhibitors that either failed clinical trials (e.g., Orteronel) or have not yet completed them (e.g., Seviteronel). A current view and recent advances of non-steroidal CYP17A1 inhibitors will be presented.

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PP3

Molecular modeling of carbonyl reductase 1 and aldo-keto reductase 1C3 as new molecular targets in supporting cancer therapy.

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Searching for new methods of treatment of cancer, as well as improvement of currently used methods, remain one of the biggest challenges in modern medicine and pharmacology. Anthracycline antibiotics (ANTs), the main representatives of which are doxorubicin (DOX) and daunorubicin (DNR), are still widely used in the treatment of several types of cancer, including leukemias, lymphomas and solid tumors. However, their use is limited due to cardiotoxicity, which may occur even many years after the end of treatment. It is postulated, that this side effect is caused by ANT-reduced metabolites, mainly formed as a result of the activity of 2 enzymes: carbonyl reductase 1 (CBR1) and aldo-keto reductase (AKR1C3). The metabolites additionally do not posses anticancer activity typical for ANTs [1,2].

The aim of the study was to apply computational methods in the optimization of crystal structures of CBR1 and AKR1C3, to obtain high-quality models, which can then be used in the search for compounds that inhibit the activity of both major ANTs reductases. Such compounds could improve ANTs effectivity and decrease the risk of cardiotoxicity while administered together with the drugs.Optimization of selected CBR1 and AKR1C3 crystal structures was performed using Small-Molecule Drug Discovery Suite (Schrödinger, Inc). 20 CBR1 ligands and 13 AKR1C3 ligands were selected to Induced-Fit docking procedure, leading to the initial models. The models were then evaluated in retrospective virtual screening. The best models were additionally evaluated in molecular dynamic simulations. All steps resulted in the selection of one model for each CBR1 (model 11E3: $BEDROC_{\alpha=20}$: 0.698; $EF_{1\%}$: 36) and AKR1C3 (model 74H1: $BEDROC_{\alpha=20}$: 0.643; $EF_{1\%}$: 22). These models are now planned to be used in prospective virtual screening to select new potential inhibitors of main ANTs reductases.

This study was funded by National Science Centre's grant 2020/37/N/NZ7/01097 and Jagiellonian University Medical College grant N42/DBS/000188

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PP4

Searching of biologically active fragments of hACE2 protein able to interact with S1 protein subunit of the SARS-CoV-2 virus

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For over a year, the world has been facing the COVID-19 pandemic caused by the SARS-CoV-2 virus infection. A huge problem is the lack of scientific data allowing to understand at the molecular level the pathophysiology of the disease, its progression and the impact on the body during the recovery period or later. One of the ideas to suppress the virus transmission, beyond the vaccination program, is to develop a new class of biomedical tests indicating the presence of characteristic biomarkers for SARS-CoV-2 infection in physiological fluids e.g. blood, urine or exhaled air with saliva particles. One possible solution is test that should act as a bioelectronic nose and mimic the action of natural receptors to enable simple, fast analysis performed in a non-invasive way with high sensitivity on ppb or ppm scale [1]. One of the receptors, for which high binding ability the S1 subunit of SARS-CoV-2 spike protein was observed, is human angiotensin-converting enzyme type 2 [2]. Nevertheless, the use of the whole receptor during the test preparation would be a challenging task considering the difficult, expensive and time-consuming handling of the protein membrane. A good alternative is an application of peptides (fragments of receptor protein) responsible for the virus and biomarkers recognition due to their smaller size, stability in higher temperatures, facilitated sterilization and storage [1].

Studies conducted in the Institute of Organic Chemistry of Lodz University of Technology concern the search for potentially biologically active fragments of proteins based on the dot-blot methodology [3]. The method included the SPOT synthesis of library of overlapping fragments of human receptor ACE2 immobilized on a cellulose membrane by using DMT/NMM/$TosO^-$ as a coupling reagent [4]. The incubation of 399-element peptide library with the HRP labelled S1 subunit of SARS-CoV-2 spike protein enabled the detection of peptides that would reconstruct the outer sphere of the protein and be involved in protein-protein interaction. Based on the colour intensity of received complexes, 16 fragments of hACE2 strongly interacting with S1 subunit of SARS-CoV-2 spike protein and 16 moderate affinity fragments, of various lengths of peptide chains were selected. In the next step, the synthesis of selected fragments will be performed to confirm their binding ability with the S1 subunit of the SARS-CoV-2 virus.

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PP5

Application of immobilized lipases on Octyl-Sepharose CL-4B carrier as potential biocatalysts in obtaining pure enantiomers of NSAIDs

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Lipases are enzymes belonging to the class of acylglycerol hydrolases, subclasses of esterases (EC 3.1.1.3). Their catalytic activity is manifested, among others, by the hydrolysis of triglyceride ester bonds taking place in an aqueous medium and the reactions of esterification or transesterification in a nonaqueous medium. The mechanism of catalyzing by lipases is based on their activation at the interface, i.e. water and oil phase. Therefore, also a synthesis reaction in an organic medium requires the presence of a small amount of water, the so-called *essential water* [1]. Lipases are widely used in the chemical and pharmaceutical industries and for these purposes, they are often immobilized. This process consists of immobilizing the enzyme on support, aiming to increase its activity, stability and to enable its recovery after the reaction is completed. The area of lipase application is often kinetic resolution of racemic mixtures of drugs or compounds, which are building blocks in the reactions of obtaining drug enantiomers [2]. Lipase from *Candida rugosa* (CRL) is a lipase that has a "lid" in its structure, while lipase B from *Candida antarctica* (CALB) is characterized by its absence. Moreover, in the kinetic resolution of racemates, the CRL lipase is directed to carry out the esterification of the *S* enantiomer, and CALB lipase shows enantioselectivity toward the *R* enantiomer [3].The aim of this study was to optimize the catalytic systems to obtain pure enantiomers of (*R*,*S*)-flurbiprofen by esterification with methanol in organic solvents differing in the log P value. It was the first time, lipases immobilized on Octyl-Sepharose CL-4B have catalyzed the kinetic resolution of racemate by esterification in organic solvents with a good yield and enantioselectivity. Studies have shown that CALB has high enantioselectivity (eep = 90.48%) when the kinetic resolution of (*R*,*S*)-flurbiprofen is performed in dichloromethane, characterized by a low log P value. In contrast to CALB, CRL catalyzed the reaction to a very small extent and only in dichloroethane (C = 2%, C=3%). Comparing the activity of the free form and that of the immobilized CALB, it was proved that immobilization on the Octyl-Sepharose CL-4B support increased the activity of the tested lipase by about 12 times in dichloromethane and about 45 times in dichloroethane. Moreover, it was observed that the choice of the reactor with regard to the material from which it is made is important. The higher values of enantioselective reaction parameters were obtained in polypropylene reactors than in glass reactors.

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PP6

Interaction of HSA immobilized on magnetic nanoparticles with ketoprofen under artificially induced oxidative stress conditions

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Magnetic nanoparticles are a material whose size should not exceed 100 nanometers. An important feature of these materials is superparamagnetism and the possibility of surface functionalization by covering them with low and high molecular compounds. One of the applications of functionalized magnetic nanoparticles is their use as carriers for the bioligands immobilization. Human Serum Albumin is one of the proteins more often immobilized on the nanoparticles surface. It plays an important role in the body and the interaction with HSA is one of the basic parameters for active substances. [1] The degree of drug binding to HSA is influenced by many factors, of which oxidative stress is a very significant one. Oxidative stress causes disturbances in the balance between the activity of free radicals and the biological abilities of proteins to neutralize their activity. Such a state is metabolically unfavorable because free radicals easily initiate into chemical reactions with cell components. The source of many radicals are metabolic processes naturally occurring in the body and several exogenous factors. [2]

As part of this work, magnetic nanoparticles with a surface able to binding bioligands were obtained, which were then used to immobilize albumin from human blood serum. The obtained material was tested for the interaction with NSAIDs, on the example of ketoprofen under normal conditions and artificially induced oxidative stress.

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PP7

Activation of the dopamine D_4 receptor – theoretical study

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The human dopamine D_4 receptor (DRD4), a member of the G protein-coupled receptor (GPCR) family, regulates signalling pathways in the brain's dopaminergic system. It is a therapeutic target in the treatment of mental and neurological disorders, including schizophrenia, Parkinson's disease or Alzheimer's disease [1]. In order to investigate the molecular mechanism of the dopamine D_4 receptor activation, the appropriate complexes of its agonists and antagonists were simulated by molecular dynamics. Initially, ligands with known dopamine agonist and antagonist functionality were docked to the receptor protein crystal. Then, the complexes were optimized using the molecular dynamics method. For representative structures, an analysis of the stabilization energy of active and inactive complexes was performed using the FMO/PIEDA method.

Using molecular dynamics simulation, a comprehensive analysis of the dopamine D_4 receptor complexes with agonist and antagonist activity was provided. Therefore, the applied in silico methods and the results obtained allowed for a more detailed analysis of the receptor activation mechanism, which is of key importance in the design of highly selective DRD4 ligands, and thus new effective drugs.

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PP8

Synthesis, structure and cytotoxic evaluation of novel imidazo[2,1-*c*][1,2,4]triazole derivatives

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Incomplete effectiveness and acquired resistance of cancer cells for commonly used chemotherapeutic agents remain a major challenge in searching for new synthetic, selective and safe anticancer drugs. Recently, special attention has been paid to the fused imidazo-triazole scaffold which constitutes a recurring motif of synthetic compounds of pharmacological interest due to their proven anticancer, antimicrobial, antifungal, or antiviral activities [1,2,3].

As a part of our research aimed at the synthesis of heterocyclic compounds with anticancer activity, we would like to describe a series of imidazo[2,1-*c*][1,2,4]triazole derivatives. In the first step, the reaction of the 1-arylhydrazinecarbonitriles **1** with 2-chloro-4,5-dihydro-1*H*-imidazole **2** afforded 7-(4,5-dihydro-1*H*-imidazol-2-yl)-2-aryl-6,7-dihydro-2*H*-imidazo[2,1-*c*][1,2,4]triazol-3(5*H*)-imines **3**, which subsequently upon treatment with acyl or sulfonyl chlorides were converted into the corresponding amides **4** or sulfonamides **5**. Finally, the reactions of **3** with aryl isocyanates or isothiocyanates gave rise to the formation of corresponding ureas **6** or thioureas **7**, respectively. The structures of **3-7** were confirmed by the use of spectroscopic methods (IR, NMR). Single crystal X-ray analysis was performed at the Department of Crystallography, Faculty of Chemistry, A. Mickiewicz University in Poznań (*Figure 1*). *In vitro* cytostatic potency was investigated at the Department of Medicinal Chemistry, University of Greifswald, Germany.

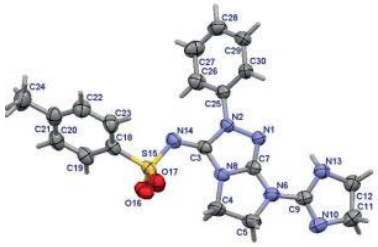

Figure 1. Crystallographic structure of sulfonamide derivative **5**

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PP9

Identification of purinergic receptor $P2Y_{14}$ as a potential therapeutic target in allergic asthma

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Human asthma represents a wide spectrum of pathologies that likely arise through distinct molecular pathways. Accordingly, a major therapeutic goal is to target individual therapies to specific forms, or endotypes, of this disease [1]. Here we studied the role of UDP-glucose (UDP-Glc), a nucleotide sugar which we found to be released into the airways of allergen-sensitized mice upon their subsequent challenge with allergen. UDP-Glc signals through the purinergic receptor, P2Y14R, and mice lacking this receptor, had decreased airway eosinophilia and airway hyperresponsiveness compared with wildtype mice in a protease-mediated model of asthma. P2Y14R was dispensable for allergic sensitization and for the production of type 2 cytokines in the lung after challenge. However, UDP-G increased chemokinesis in eosinophils and enhanced their response to the eosinophil chemoattractant, CCL24. In turn, eosinophils triggered the release of UDP-G into the airway, thereby amplifying eosinophilic recruitment [2]. Moreover, this positive feedback loop is sensitive to therapeutic intervention. Our studies revealed that administration of a small molecule antagonists of P2Y14R, reduces eosinophilic airway inflammation in the protease-mediated model of asthma [3, 4]. Together, these findings reveal a pathway that can be targeted therapeutically and suggest that antagonizing P2Y14R function might mitigate exacerbations in humans with eosinophilic forms of asthma.

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PP10

Synthesis and affinity studies of novel piperazine-based compounds as potential antipsychotics.

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In the previous studies aimed at searching for novel compounds with antipsychotic properties, structure-based virtual screening was conducted [1]. Among found dopamine D_2 receptor antagonists, the compound D2AAK3 with 115 nM affinity for D_2 receptor was identified. It shows nanomolar or low micromolar affinity also for D_1, D_3, 5-HT_{1A}, 5-HT_{2A} and 5-HT_7 receptors. Interactions of D2AAK3 with its molecular targets at the molecular level were studied *in silico* using molecular modeling methods. Behavioral studies performed for D2AAK3 revealed that it decreases amphetamine-induced hyperactivity measured as spontaneous locomotor activity in mice, improves memory consolidation after acute treatment in passive avoidance test and exhibits anxiogenic activity 30 minutes after acute treatment in mice in elevated plus maze (this effect was reversed 60 minutes after administration of D2AAK3) [2]. In the light of above outcomes, D2AAK3 may be considered as a promising starting point for further optimization toward obtaining molecules with more beneficial receptor profile as for potential antipsychotics. A series of derivatives has been synthesized. The designed modifications of the lead structure included the exchange of substituent at piperazine moiety and elongation of the alkyl linker. The obtained compounds were tested in radioligand binding assays in order to evaluate their affinities for main molecular targets in schizophrenia. These results will be next complemented with behavioral studies.

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PP11

Design and synthesis of new potent 5-HT$_7$ receptor ligands as a candidates for the treatment of central nervous system diseases

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Arylpiperazine derivatives are one of the most extensive examined potential CNS drugs, widely used for treating central nervous system diseases including the depression [1] or neuropathic pain.[2] In most cases they exhibit multifunctional properties acting on serotonin or dopamine receptors. On the other hand, it was reported that selective 5-HT$_7$ receptor ligands are crucial in treatment of depression [3] as well as triple-negative breast cancer [4] or non-small cellular lung cancer [5]. Herein we designed and synthesized novel 5-HT$_7$ ligands derived of 2,4,6-triamino-1,3-5-triazine with increased affinity to 5-HT$_7$ receptor and simultaneously decreased affinity to 5-HT$_{1A}$ receptor. All final compounds were obtained under solvent-free conditions supported by microwave irradiation with the yield more than 51%. The studied compounds showed high affinity and selectively towards 5-HT$_7$ receptor with the range of K_i = 8 – 109 nM (antagonist mode) showing good metabolic stability and moderate affinity to CYP3A4 isoenzyme. Selected compounds were submitted to HepG2 cell line MTT tests where exhibited moderate or good hepatotoxic properties.

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PP12

Aromatic analogues of Amphotericin B. How to isolate this promising agents for antifungal therapy?

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Systemic mycoses are a very common diseases nowadays. Paradoxically, the increase in incidence is caused by development of medicine. It can be a side effect of anticancer chemotherapy, HIV infection or administration of immunosuppressive drugs after organ transplant — the infections affect immunocompromised patients. Moreover, the resources of antifungal agents used in human antifungal therapy are limited. Thus, the need for new treatment methods.

The leading and prospective antibiotics are polyenes macrolides. This is due to their exceptional properties comprising: strong fungicidal activity, broad antifungal spectrum and ability to overcome multidrug resistance (MDR) of fungi. This large group of agents is characterised by a large macrolactone ring with the series of conjugated double bonds (polyene chromophore) and specific chemical groups. Based on the chromophore size, heptaenes deserve special attention; it is the most promising group in antifungal treatment.

So far, Amphotericin B (AmB) has been commonly used in medicine as a „gold standard". However, there is also an interesting subclass of heptaene macrolides – aromatic heptaenes that are structurally similar to the Amphotericin B. The major difference is the presence of an aromatic moiety in the structure. The aromatic heptaene macrolides exhibit high antifungal activity, up to two orders of magnitude higher than AmB.

However, their potential has not been fully exploited yet due to the essential technical drawback. These antibiotics are synthesised by *Streptomycetes* as multicomponent complexes of closely related compounds. This characteristic makes it very difficult to isolate in preparative scale, hence the individual components are not available commercially. Moreover, there are many dissonances in the literature regarding the composition of the AHMs complexes, as well as most of the structures of their individual components, which are still unknown.

The aim of this work was to develop the procedure of isolation of aromatic heptaene macrolides from their complexes by the usage of chromatographic methods.

The undertaken studies concern Aureofacin and Candicidin complexes as the main representants of AHMs. The composition of Aureofacin includes two antibiotics - Vacidin and Gedamycin, whereas Candicidin consists of three major antifungal agents - Candicidin A1, D and A3. The isolation process was performed mostly using chromatography techniques such as Centrifugal Partition Chromatography (CPC) and optionally - preparative high-performance liquid chromatography (prep-HPLC).

The work introduces the isolation method of AHMs of a high purity level.

PP13

Conformational properties of glycosaminoglycan building blocks: molecular dynamics simulations

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Glycosaminoglycans (GAGs) are linear, negatively charged and structurally diverse polysaccharides. They are present in the extracellular matrix as well as on and within cells as a part of proteoglycans, i.e. the group of protein-carbohydrate conjugates which are a fundamental component of tissue structure in animals. GAGs, interacting with proteins, are capable to induce numerous responses in the biological machinery. At the molecular level, the mechanism of GAGs-proteins and, more generally, GAGs-molecular environment interactions, relies on the mutual shape and charge complementarity. One of the most fundamental issues in determining the conformational properties of GAGs is to find the dependence of the conformation of the given linkage on its type, the type of adjacent residues and their possible functionalization patterns. This general issue has never been considered in a systematical manner, by taking into account the complete set of 106 disaccharides building naturally-occurring GAGs. Here we describe the results of such investigation, relying on the all-atom molecular dynamics simulations.

The results allowed to draw some general conclusions about the factors that influence the conformational preferences of GAGs as well as to determine the significance of various functionalization patterns: (1) The 'local' conformational preferences of a long GAG chain can be predicted on the basis of known behavior of the small, disaccharide fragments. (2) A limited influence of sulfation on the dominant geometry of glycosidic linkages has been found. For 102 out of 106 studied disaccharides, the optimal geometry is determined by the topology of the glycosidic linkage. (3) Residue-residue hydrogen bonding does not influence either the optimal geometry of the glycosidic linkage nor its flexibility. (4) The effect of sulfation is the most pronounced in the case of heparan sulfate where it influences the conformation of the glycosidic bond to the largest extent. The conformation of chondroitin and dermatan is altered to a significantly smaller degree. Finally, sulfation affects the conformation of keratan only to a minor extent. (5) The most important functionalization patterns include: the sulfation at C6 and C3 of GlcNAc.

The additional result of our study is the complete library of free energy maps, quantitatively describing the conformational properties of the GAG-related glycosidic linkages. They can serve as a source of data for predicting the possible structures of heterogeneous, long GAG chains or as a benchmark for comparative studies involving chemically-altered GAG derivatives.

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Posters P1 – P96

P1

New hybrid pirrolidyne-2,5-dione derivatives as candidates for new potent and wide spectrum anticonvulsants

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Hybrid compounds are of great interest for the development of new drugs effective in diseases with complex pathomechanisms, such as Alzheimer's disease, Parkinson's disease, cancer, or epilepsy. Therefore, according to the molecular hybridization method and to obtain new highly effective and broad-spectrum anticonvulsants, in the previous study we have developed integrated hybrid molecules. These compounds were designed by applying the fragment-based approach, thus they overlap on the common structural framework the chemical fragments of three chemically and pharmacologically diversified ASDs such as ethosuximide, levetiracetam, and lacosamide. The hybridization process yielded substances with potent and broad-spectrum anticonvulsant activity in the most widely employed animal seizure models (MES, *sc*PTZ, 6 Hz). Notably, these compounds joined pharmacological properties of all ASDs creating hybrid structures. [1,2] Considering beneficial anticonvulsant properties of the hybrids reported previously, in the current studies we have developed a new series of hybrid molecules based on the pyrrolidine-2,5-dione core fragment. These compounds revealed potent anticonvulsant activity in the MES, *sc*PTZ, and 6 Hz (32 mA) seizure models as well as in the 6 Hz (44 mA) test, which is widely recognized as a model of pharmacoresistant epilepsy. The *in vivo* studies enabled to select the most potent compound **AS-34**, which was characterized by beneficial pharmacological and toxicological properties. This molecule was active in several animal seizure models (i.e. ED_{50} [MES] = 40.5 mg/kg, ED_{50} 6 Hz [32 mA] = 7.6 mg/kg; ED_{50} [*sc*PTZ] = 50.2 mg/kg). In addition, this substance was also effective in the 6 Hz (44 mA) model of drug resistant epilepsy (ED_{50} = 69.5 mg/kg) and revealed a high safety profile in the rotarod test (TD_{50} > 500 mg/kg).

The studies were supported by the Polish National Scientific Centre grant 2018/29/N/NZ7/01966.

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P2

Epigallocatechin gallate and its applications in medicine and pharmacy

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Green tea is one of the most popular beverages in the world. Epigallocatechin gallate (EGCG), as the most abundant flavonoid found in the green tea extract, reveals many beneficial effects on human health, most importantly antioxidant, anti-inflammatory, anti-fibrotic, antibacterial and antiviral, proosteogenic, as well as protective against excess UV radiation [1, 2]. These activities predispose EGCG to be used as an active pharmaceutical ingredient in the treatment of metabolic, neurodegenerative, and neoplastic diseases [3, 4, 5]. Unfortunately, despite having so many advantages, some obstacles hamper the widespread use of EGCG for medical purposes. The most crucial hindrance is the poor stability of EGCG both in the solid-state and solution. In addition, EGCG's stability depends on various factors, including temperature, pH, presence of oxygen, and light [2, 6].

Herein, a summary of EGCG's stability was provided and discussed along with the possibilities of its development using (i) chemical transformation to its prodrugs (molecule modification) and/or (ii) preparation of nanoformulations (e.g. liposomes). Initial synthetic experiments focusing on modifications of EGCG's hydroxyl groups, with different moieties, such as acetyl, methyl, or tert-butyldimethylsilyl groups, are also presented [7,8]. Finally, the preparation of liposomal formulations with EGCG derivative, using the thin film hydration method of the phospholipid bilayer, are demonstrated along with stability study of EGCG alone and in liposomal formulations [9].

This research was funded by National Science Centre, Poland, grant number *2019/35/B/NZ7/01165.*

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P3

New water-soluble lead structure based on isoxazole-linked 1,3,4-oxadiazole derivative with delocalized positive charge

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The synthesis of heterocycles is possibly one of the oldest and at the same time one of the youngest branch of organic chemistry. Chemistry of heterocycles is an ever-expanding subject of organic chemistry and plays a crucial role in drug design. Among all known pharmaceuticals a predominant number of them are small molecules with heterocyclic moieties therefore there is a strong need to design a new, rapid and cost-efficient synthetic protocols. Isoxazole/oxadiazole- based molecules have been characterized as compounds possessing anti-inflammatory, antibacterial, anti-viral or anti-cancer activities [1-2]. Despite significant progress that has been achieved in many fields of medicine including anticancer or anti-inflammatory therapy, the development of new potential drugs represents a major challenge to medicinal chemist researchers. Taking into account, our efforts have continued in the last years studies on isoxazole and oxadiazole derivatives in searching for new biological active derivatives [3]. Although, there are known molecules with positive charge localized at the nitrogen atom [4-5] in the literature there is less mentioned about compounds possesing local positive charge at the carbon atom, and they are limited to small molecules [6]. Motivated by the aforementioned biological and pharmacological importance of the heterocyclic compounds, especially in those with directly directed rings, and as a continuation with our previous research on isoxazoles, herein we report the synthesis of new stable and water-soluble 5-amino-2-(5-amino-3-methyl-1,2-oxazol-4-yl)-3-methyl-2,3-dihydro-1,3,4-oxadiazol-2-ylium bromide (ED) with the unusual electron charge delocalization owing the local positive charge at the carbon atom of oxadiazole moiety. X-ray single crystal of $C_7H_{10}N_5O_2 \cdot Br^{(-)}$ showed the molecule crystalized in monoclinic, space group P21/c. Both five membered rings are planar and twisted forming the ring motif with the counter ion where H···Br interactions are one of the dominant. The presented compound is characterized by high ionization efficiency in ESI-MS mode and undergoes dissociation within oxadiazole moiety under ESI-MS/MS conditions even under low collision energies. The presented compound is an interesting example of heterocyclic stable carbocation which may serve as a new lead structure.

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P4

Moxifloxacin-mediated growth inhibition and apoptosis induction of human breast cancer MDA-MB-231 cells

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Breast cancer is the most commonly diagnosed cancer and the leading cause of cancer death among females [1]. Triple-negative breast cancer (TNBC) lacks oestrogen receptors, progesterone receptors and HER2 overexpression and is characterized by the poorest overall survival of all breast cancer subtypes. Lack of effective targeted therapies and the poor prognosis encourage the intensive research to develop additional and better systemic treatment options for patients with TNBC [2]. Moxifloxacin belongs to IV generation of fluoroquinolones and is characterized by a broad spectrum of antibacterial activity [3]. Recent scientific research concerning the assessment of fluoroquinolones properties focus on their potential anticancer activity. However, scientific literature describes only a few cases of potential use of moxifloxacin as an anti-cancer drug [4].

In order to provide strong cellular and molecular evidence for moxifloxacin-mediated anti-cancer effect towards breast cancer cells, the cell viability (WST-1 assay) and apoptosis induction (mitochondrial membrane potential) were determined. MDA-MB-231 cell line was used as an *in vitro* experimental model.

The cells were treated with the fluoroquinolone derivative in the concentration range from 0.001 mM to 1.0 mM for 24, 48 and 72 h. After incubation of cells with lower moxifloxacin concentrations (from 0.001 mM to 0.01 mM) for 24 h the loss in the cell viability was not statistically significant. Exposure of cells to the drug in concentrations of 0.05 mM, 0.1 mM, 0.5 mM and 1.0 mM resulted in the decrease of cell viability by 17 %, 20 %, 40 % and 61 %, respectively. Interestingly, the cytotoxic response intensified when MDA-MB-231 breast cancer cells were exposed to moxifloxacin for 48 h and 72 h in all the studied drug concentrations. Under these conditions the viability of cells decreased by 8 % to 89% for 48 h and 11 % to 98 % for 72 h incubation time. Following image cytometric analysis the percentages of mitochondrial membrane depolarized breast cancer cells exposed to moxifloxacin in concentration of 1.0 mM for 24 h, 48 h and 72 h significantly increased and was determined to be 22 %, 74 % and 94%, respectively, while the value determined for the controls was about 7 %. The use of the drug in lower concentration (0.5 mM) had no impact on the loss of mitochondrial membrane potential in MDA-MB-231 breast cancer cells.

This is the first study that characterized the anti-cancer effect of moxifloxacin towards TNBC cells, opening the possibility to use of this drug as a potential agent for the treatment of breast cancer.

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P5

Searching for biological active peptides important in wound healing process

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The main goal of research is to design, synthesize and investigate the properties of new, biocompatible, multi-component hybrid materials useful in the treatment of non-healing wounds, including diabetic foot syndrome, based on immobilized, on the polysaccharide matrix, a set of selected biologically active peptides derived from proteins affecting all stages of the process of healing wounds. It has been assumed that peptide fragments exposed on the outside protein should guarantee their biological activity (Figure 1) [1-2].

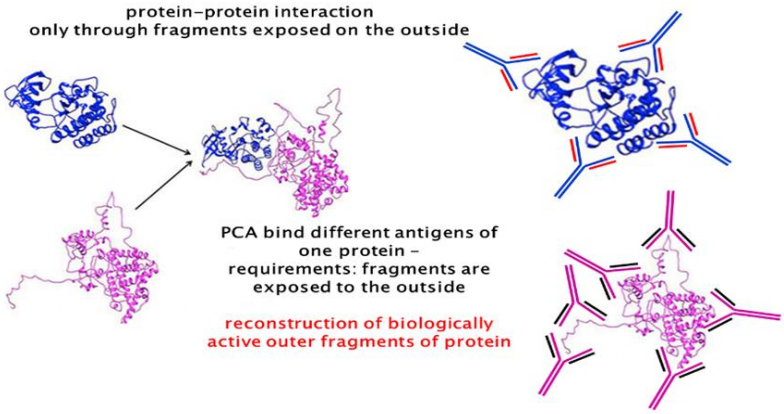

Figure 1. Proposed method for selecting a SET of biologically active peptides.

Biologically active peptides potentially useful in wound healing process were selected from lactoferrin and bovine κ casein. Lactoferrin affecting hemostasis and promotes the healing of skin wounds by enhancing the initial inflammatory phase, while bovine κ casein inhibit bacterial and viral adhesion, suppress gastric secretions, promote bifidobacterial growth and modulate immune system responses. To identify the exposed fragments of both proteins, SPOT synthesis technique and the dot-blot assay was used. The libraries of peptides being fragments of both proteins were obtained by using triazine coupling reagent [3]. Based dot-blot assay, 11 fragments of κ casein and 6 fragments of lactoferrin were selected. All fragments were synthetized and their biological activity will be tested.

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P6

Prediction of lipophilicity and ADME properties of betulinic aldehyde derivatives

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In recent years, pentacyclic triterpenes of the lupane type have gained attentions as anticancer agents. The effectiveness of betulin and betulinic acid derivatives as therapeutic substances is related to their bioavailability and solubility in aqueous media. The search for novel triterpenoids is associated with carrying out chemical modifications of the parent structures in order to obtain compounds with an improved pharmacokinetic properties [1-2].

Determination of the physicochemical properties of new medicinal substances most commonly requires to take into account solubility and permeability thorough biological membranes. Conducting research on a new drugs, the substances candidates are selected in such a way that they fulfill the Lipinski's rule-of-Five. According to Lipiński's rule, a drug substance should meet at least three of the following criteria: molecular weight below 500, logP value not higher than 5, presence of not more than 5 donors and not less than 10 hydrogen bond acceptors [3].

The experimental $logP_{TLC}$ lipophilicity values for the new betulinic aldehyde derivatives were determined by the reverse phase thin layer chromatography (RP-TLC). In addition, the parameters related to ADME and theoretical values of lipophilicity were calculated using computer programs. An important stage of the research was to determine the relationship between the lipophilicity of betulinic aldehyde derivatives and their antitumor activity against the human leukemia (MV-4-11) cell line [4].

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P7

Anti-HER2 monoclonal antibodies intensify the susceptibility of human gastric cancer cells to etoposide by promoting apoptosis

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Gastric cancer (GC) is a multifactorial disease with high mortality. Anti-HER2 therapy is a promising strategy in GC treatment and trastuzumab was approved by Food and Drug Administration as the first and the second line of treatment of the disease. Etoposide as a single agent was proved in several malignancies including small cell lung cancer, lymphomas, ovarian and testicular cancer. Etoposide is a component of two treatment regimens for patients with gastric cancer and it is used with doxorubicin and cisplatin or in combination with calcium folinate and 5-fluorouracil. Number of studies showed that combining monoclonal antibodies against specific targets with chemotherapeutic agents play a major role in treating patients with cancer [1].

The aim of the study was to examine the effectiveness of a combination of etoposide with trastuzumab or pertuzumab in AGS gastric cancer cells. The cytotoxic effects of the tested compounds against gastric cancer cells were checked by MTT assay. Fluorescent microscopy was used to demonstrate the effect of the compounds on apoptosis. The mitochondrial membrane potential, and the activity of caspase-8 and caspase-9 were assessed.

The results from our study proved that the combination of etoposide with anti-HER2 antibodies decreased viability of gastric cancer cells. The interaction of etoposide with pertuzumab or trastuzumab induced programmed cell death via extrinsic and intrinsic apoptotic pathways in AGS gastric cancer cells, where increased activity of caspase-8 and caspase-9 was demonstrated.

The study demonstrated that etoposide with anti-HER2 antibodies represent a promising strategy in gastric cancer treatment, but further *in vivo* examinations are also required.

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P8

The influence of anti-HER2 monoclonal antibodies with etoposide on autophagy in gastric cancer cells

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Autophagy is a process of self-degradation that plays an important role in removing damaged proteins, organelles or cellular fragments from the cell. Under stressful conditions such as hypoxia, nutrient deficiency or chemotherapy, this process can also become the strategy for cell survival. Autophagy can be nonselective or selective in removing specific organelles, ribosomes, and protein aggregates, although the complete mechanisms that regulate aspects of selective autophagy are not fully understood. Intensive studies have revealed a whole range of novel compounds with an anticancer activity that inhibit or activate regulatory pathways involved in autophagy [1].

The aim of the study was to examine the effect of etoposide with trastuzumab or pertuzumab on autophagy in AGS gastric cancer cells. The influence of etoposide alone and in combination with pertuzumab or trastuzumab on autophagosomes and autolizosomes formation was conducted by flow cytometry. The concentrations of Beclin 1, LC3A and LC3B were assessed by ELISA.

We demonstrated that Beclin-1 concentration was dependent on the chemotherapeutic agent dose. The higher dose of etoposide in combination with trastuzumab or pertuzumab led to a lower concentration of the analyzed protein in cell lysates. All the studied compounds decreased the levels of LC3A and LC3B after 24 hours of incubation in comparison with the untreated cells. Flow cytometric analysis confirmed that autophagy was not induced after treatment with the monotherapy as well as combination strategy. Our data supports the thesis that the inhibition of autophagy increases the susceptibility of cancer cells to the treatment. We proved that such a dual treatment regimen allowed to achieve relevant efficacy, but further studies are required.

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P9

Searching for highly selective 5-HT$_7$ receptor biased ligands in the group of arylsulfonamide derivatives of alicyclic amines

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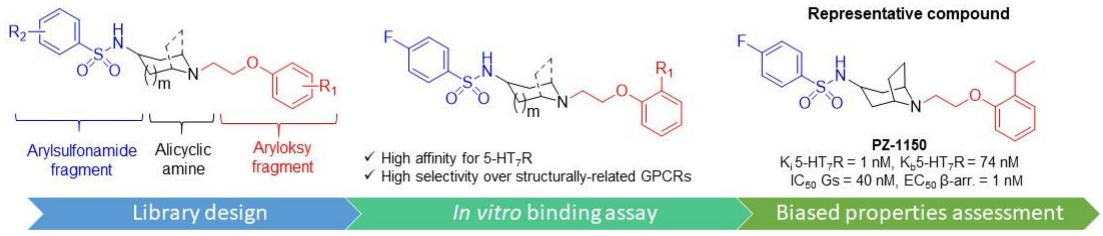
The 5-HT$_7$ receptor (5-HT$_7$R) is a member of the family of serotonin G protein-coupled receptor (GPCR) subtype and it represents a promising pharmaceutical target for the treatment of neuropsychiatric disorders [1]. Independently from modulating the canonical G protein ($G_{\alpha s}$), the 5-HT$_7$R promotes the recruitment of β-arrestin, which is involved in the regulation of alternative G-protein-independent signaling pathways [2].
In an attempt to develop specific ligand (biased ligand) which might preferentially and/or selectively modulate G-protein or β-arrestin signaling pathways, a focused series of structural analogs of arylsulfonamides of (aryloxy)alkyl alicyclic amines (a known class of known 5-HT$_7$R antagonists) [3,4] has been designed, synthesized and pharmacologically evaluated using *in vitro* methods.



Tested compounds displayed high affinity for 5-HT$_7$R (K_i < 30 nM) in radioligand binding assay and showed high selectivity over 5-HT$_{1A}$R subtype and other structurally related GPCRs. Moreover, evaluated compounds decreased the constitutional $G_{\alpha s}$-mediated cAMP production acting as potent inverse agonists in this cellular setup. Selected derivatives were found to recruit β-arrestin pathway in BRET-based assay and thus might be classified as potent agonist at this G-independent signaling pathway.
Preliminary functional evaluation for biased properties identified some derivatives, which displayed preference for the β-arrestin-dependent signaling (up to 20-folds) over the canonical $G_{\alpha s}$.
Further studies focused on this class of biased 5-HT$_7$R ligands might provide a valuable chemical probe to better understand the relationship between G protein/β-arrestin signalling pathways and *in vivo* pharmacological effects of 5-HT$_7$R.

The project was financially supported by the National Science Center, Poland grant no *2019/33/B/NZ7/02822.*

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P10

Homology modeling of GPCRs – optimization of the 5-HT$_7$ receptor model for virtual screening

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Serotonin 7 receptor is thought to be a possible target for new CNS drugs which could be used in such disorders as depression, epilepsy, cognitive deficits. Currently, there is no approved selective 5-HT$_7$ drug, however, a few available clinically drugs, such as vortioxetine and amisulpride, have a noticeable affinity for this receptor [1].

The lack of crystal structures forced the use of homology modeling to generate a 3D structural model of the 5-HT$_7$ receptor. The initial model was built by SwissModel, using the crystal structure of 5-HT$_{1A}$ receptor (PDB code: 7E2Z) as a template. Optimization of the binding site was performed using the package of SCHRÖDINGER tools, including Induced Fit Docking and Desmond Minimization. Application of retrospective virtual screening, with the use of various chemotypes of 5-HT$_7$ ligands and DUD-E, DUDE-Z datasets as decoys, helped validate the models.

The study showed that the application of IFD twice, followed by Desmond Minimization can increase the quality of the model compared to the non-optimized model (all active ligands docked successfully and BEDROC$_{\alpha=20}$ parameter about 2-4 times better). Moreover, the optimized amino acid side chains conformation resulted in effective binding mode prediction of the reference ligands. Besides the main interaction with Asp$^{3.32}$, the active ligands also interacted with Phe$^{3.28}$, Phe$^{6.52}$, Arg$^{7.36}$ [2].

This study can help to refine a universal optimization procedure to gain the best-performing conformational models for prospective virtual screening.

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P11

Medicinal mechanochemistry, a new method for an efficient and sustainable synthesis of biologically active compounds targeting α_2/5-HT_7 receptors

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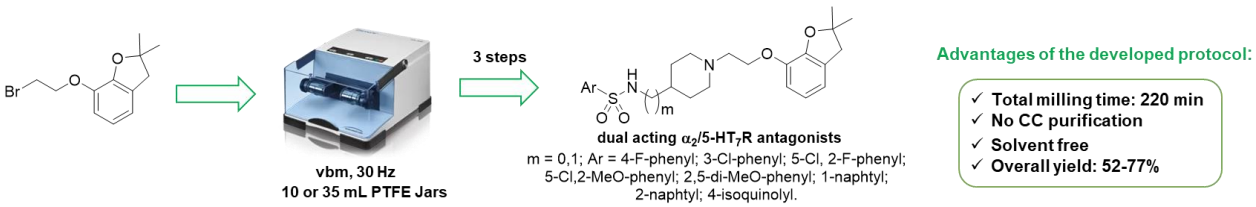
Since many years mechanochemistry has been used in the drug technology field to impact the solid-state properties of pharmaceutical products (i.e. salts, solvates, co-crystals) [1]. In addition to its efficiency and versatility, mechanochemical processes are also recognized for their sustainability, as they satisfy the green chemistry principles [2]. Recently, a growing number of mechanochemical procedures for generating active pharmaceutical ingredients (APIs) [3], and for a greener synthesis of lead structures [4] have been reported so far and led to coining the term “medicinal mechanochemistry”.
Herein we present an application of the medicinal mechanochemistry approach to study structure-activity relationships within a focused library of arylsulfonamides of (aryloxyethyl)piperidines as potential dual acting α_2/5-HT_7 receptor antagonists.



The newly employed multi-step mechanosynthesis provided designed compounds in a high overall yields (52-77%) and purities (> 96%) with a required milling time of 220 min in a total absent of organic solvents [4]. In addition, reaction conditions and work-up procedures were simplified as intermediates and final compounds were isolated without the need of column chromatography purification.
Synthesized compounds displayed high-to-low affinity for α_2-AR (K_i = 80–1194 nM) and for 5-HT_7R (K_i = 30–727 nM) in radioligand binding experiments, while selected derivatives were further classified as potent α_2/5-HT_7R antagonists in fluorescence-based and cAMP-based cellular assays, respectively.
These findings might further confirm the power of medicinal mechanochemistry as a widely accepted sustanaible alternative for organic synthesis and might facilitate the integration of the mechanochemical approach as a key component of lead discovery programs in both academic and industrial research.

Fundings: The project was financially supported by the National Science Center, Poland grant no *2019/33/B/NZ7/02822.*

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P12

Prolidase - dependent resistance to chemotherapy in breast cancer cells

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Background: Contemporary anti-cancer therapy is focused on the activation of apoptosis in cancer cells. One of the most important activators of this process is the p53 protein [1,2].
Recent studies shown that prolidase plays a regulatory role in the level and activity of p53. Prolidase binds more then 50% of intracellular amount of p53 protein [3]. This study was undertaken to establish the role of prolidase in the regulation of p53-dependent apoptosis.
Material and Methods: Model of breast cancer cells MCF-7 (wild type) and prolidase overexpressed breast cancer MCF-7 (MCF-7^{PL}) were used. Daunorubicin (direct p53 activator) and Indometacin (indirect p53 activator) were used to induce apoptosis in both model cells. Cell apoptosis was determined using Nucleo-Counter NC-3000. DNA biosynthesis was determined by radiometric method. Expression and translocation of selected proteins were assessed by fluorescent microscopy and Western Immunoblot.
Results: The used substances increase the apoptosis and inhibit DNA biosynthesis more in MCF-7 cells (wild type) than in cells with prolidase vector MCF-7^{PL}. The proapoptotic effect of studied compounds in MCF-7 cells were accompanied by increase the expression and translocation to the nucleus of p53 protein and activation of caspase-9. Prolidase overexpression in MCF-7^{PL} protects cells against the pro-apoptotic effects of direct and indirect activation of p53 and contributed to increasing the expression of Atg7 and Beclin-1, which may provide to pro-survival effect of cancer cells.
Conclusions: The data suggest that overexpression of prolidase in breast cancer cells contributes to decrease response to activation of p53-dependent apoptosis. This may be helpful in choosing the type of cancer therapy, especially in tumors characterized by overexpression of prolidase.

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P13

Novel purine-2,6-dione-based multifunctional PDE inhibitors and TRPA1 antagonists as a new approach for treating chronic airway diseases

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The current therapeutic management of chronic respiratory diseases, such as asthma and COPD, does not affect airway remodeling, therefore, the need for the specific therapy aimed at counteracting or preventing processes associated with lung fibrosis is very urgent. Our recent studies revealed that 7,8-disubstituted purine-2,6-dione derivatives may effectively diminish airway smooth muscle cells remodeling and lung fibroblast to myofibroblast transition as a result of concomitant PDE inhibition and TRPA1 antagonism [1, 2]. Based on these findings, a new series of 8-aminopurine-2,6-dione derivatives were designed and synthesized as multifunctional ligands acting as PDE1/3/4/7/8 inhibitors and TRPA1 antagonists. Molecular modeling studies explained the multitarget-directed activity exhibited in vitro. Pharmacological evaluation of the selective compounds revealed their anti-inflammatory and anti-fibrotic properties in the LPS-induced mouse macrophages RAW264.7 cell line and in the TGF-β_1-induced human airway smooth muscle cells, respectively. The results of pharmacokinetic study performed for the most active multifunctional ligand showed its favorable serum concentration versus time profile following intravenous administration to mice. In addition, it was vividly distributed to important organs, which makes it suitable for further in vivo investigation, e.g. using animal models of COPD or asthma. The presented results open novel perspectives for this class of compounds as anti-inflammatory and anti-fibrotic agents in chronic respiratory diseases.

Acknowledgements: This study was supported by the National Science Centre, Poland, funded grant No. 2018/29/B/NZ7/00285.

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P14

Lipophilicity and pharmacokinetic properties of phosphate derivatives of betulin

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Research on bioactive substances, aimed at discovering a new drug with the right properties, is associated with high risks, costs and time. A key challenge to successful drug discovery is finding the right balance between the limitations of the physicochemical properties of drug candidates and maintaining the appropriate potency to ensure an effective dose.

Triterpenes are an extremely diverse group of chemical structures that are widely distributed in nature. The vast majority of them are produced by plants. Among the various substances from this group, betulin, belonging to the pentacyclic triterpenes of the lupane type, stands out due to its easy availability and interesting biological properties. It is also subject to numerous chemical modifications leading to new derivatives [1]. Studies on the antitumor and antiviral activity of these substances provide promising results [2, 3]. An important part of this research is also the development of various elements of the physicochemical properties profile for the synthesized compounds as potential pharmaceutical preparations.

Lipophilicity expressed as logP parameter, as well as molecular weight (M), number of hydrogen bond acceptors (nHA) and donors (nHD), number of rotational bonds (nROT) and topological polar surface of the molecule (TPSA) are analyzed early in the study of candidates for drugs. Determining the theoretical values of the logP parameter is one of the elements of the optimization and evaluation of pharmacokinetics of new chemical compounds as part of predicting their ADME profile [4].

The aim of the study was to experimentally determine the lipophilicity of betulin phosphate derivatives by reversed-phase thin layer chromatography (RP-TLC) as well as to determine the pharmacokinetic parameters using the *in silico* method.

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P15

Searching for biologically active peptides derived from growth factors

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Growth factors are proteins that are involved in regeneration processes. Disruption of skin permanence results in cascade of reactions that lead to growth factor migration to the milieu of the wound. Depending on the type, they stimulate proliferation of different cells, for example epidermal growth factor (EGF) helps in multiplication of epidermal cells, vascular endothelial growth factor (VEGF) induces regeneration of blood vessels endothelium, nerve growth factor promotes formation of neural cells. Combination of variety of growth factors is essential for every regeneration process in living organisms, what makes them really important object for the regenerative medicine. Growth factors are also involved in many undesirable events, like neoplasm, also proteins may be immunogenic, therefore they may have limited uses in clinical practice as a full-length proteins. The aim of the studies was to find short fragments of selected proteins involved in the regeneration process with maintained biological activity. Research was performed for growth factors as epidermal growth factor (EGF), transforming growth factor α (TGF-α), vascular endothelial growth factor A (VEGF-A), fibroblast growth factor 1, 2, 7, 10 (FGF-1, 2, 7, 10), tumor necrosis factor (TNF-α), fibronectin and coagulation proteins like coagulation factor XIII or fibrinogen (FG). Protein sequences were divided into decapeptides and libraries of fragments of proteins were synthesized by using SPOT technique in order to receive full map of selected structure. DMT/NMM/$TosO^-$ was used as a coupling reagent [1]. Peptide reactivity was examined in dot blot tests using specific polyclonal antibodies. Selected active fragments were synthesized for further proliferation tests. It has been found that for EGF peptides selected in blotting test were overlapping with EGF receptor (EGFR) binding fragments [2]. It has been found that EGF-derived peptides induced proliferation of fibroblasts on similar level to the native EGF protein. They were also examined to check their binding to EGFR in microscale thermophoresis (MST) assays, what showed ligand-receptor interaction with proper Kd levels. Results of the research show that short protein fragments may find potential use in regenerative medicine as substitutes of whole proteins.

Acknowledgements: The financial support from the National Science Centre, Poland, under project number UMO-2018/31/B/ST8/02760

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P16

Pharmacological Profile and Cytotoxic Evaluation of Novel 5-(3-Methylthiophen-2yl)hydantoin Derivatives with Arylpiperazinylalkyl Moiety

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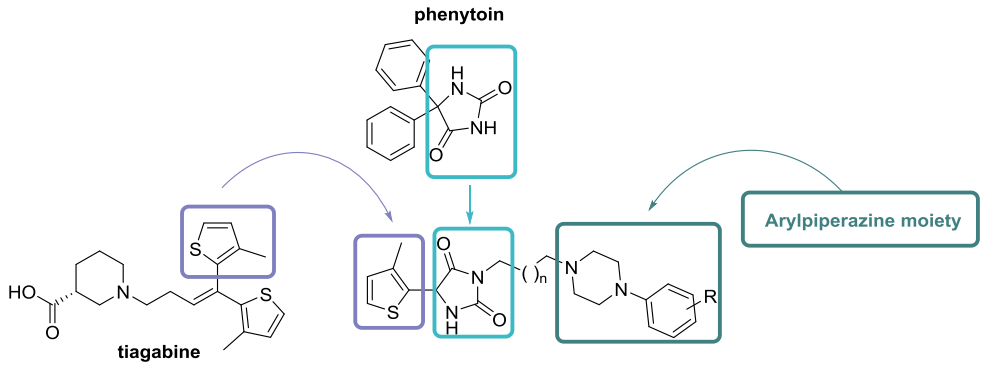
Apart from epilepsy, antiepileptic drugs (AEDs) are used in the treatment of central and peripheral nervous system disorders such as bipolar disorder, anxiety, depression, and neuropathic pain, etc. [1,2] This is due to the complex mechanism of action of AEDs, that interact with receptors i.e. GABAergic, glutamatergic and ion channels such as sodium or calcium. The aim of this study was to design the hybrid compounds based on AEDs structural fragmnets which will exhibit antidepressant- and/or anxiolytic- like activity. The hybrid compounds presented herein contain the 3-methyltiophene fragment (from the tiagabine structure), hydantoin core (from the phenytoin structure), and an arylpiperazine moiety, that can interact with serotonin receptors. Moreover, the designed compounds differ in the length of the linker between the hydantoin ring and arylpiperazine moiety.



The starting 5-(3-methyltiophen-2-yl)hydantoin was obtained in Bucherer-Bergs reaction, followed by alkylation with 1-bromo-3-chloropropane or 1-bromo-4-chlorobutane and condensation reaction with the appropriate arylpiperazine moiety. Preliminary affinity results revealed that the final compounds demonstrated high/average-to-low affinity for 5-HT_{1A} and 5-HT_7 serotonin receptors (K_i = 1 – 2400 nM and 167 – 2580 nM, respectively). Subsequently, for the most potent derivatives, the *in vitro* pharmacological profile will be assessed and cytotoxicity studies will be performed.

This study was financially co-founded by the National Science Centre (NSC) Poland grant No 2017/25/B/NZ7/01048 and Funds for Statutory Activity of Jagiellonian University Medical College (N42/DBS/000020).

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P17

Stop The Silent Killer - in search of a new efficient method of obtaining Olaparib, used in the fight against ovarian cancer

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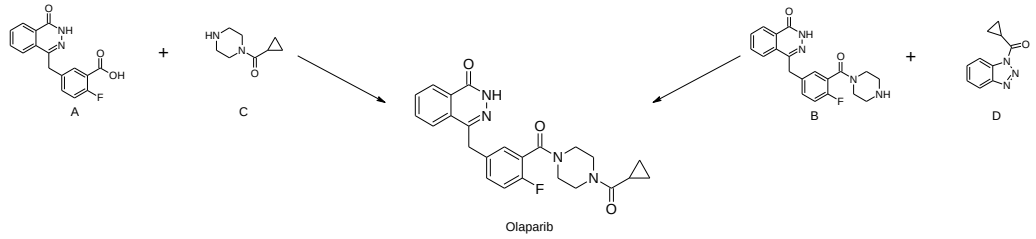
"The Silent Killer" is a name that has been given to ovarian cancer, which is one of the most common gynecologic cancers that has the highest mortality rate. It is predicted that, by the year 2040, these statistics will be even worse. Asymptomatic and secret growth of the tumor, delayed onset of symptoms, and lack of proper screening are the primary causes of the increase in mortality in this type of cancer. [1]

A new group of anti-cancer drugs are PAPR inhibitors, the use of which gives hope for improvement in the treatment of patients with ovarian cancer. This group of drugs includes Olaparib, which is their first representative [2]. The benefits of this drug include reducing the size of the tumor and increasing Progression-Free Survival (PFS), but treatment with Olaparib is extremely expensive. [3]

As part of the research, the synthesis of Olaparib was carried out by means of two synthetic pathways, which were carried out in the presence of microwave radiation.



The first method of synthesis involved the reaction between a 2-fluoro-5-[(4-oxo-3,4-dihydrophthalazin-1-yl)methyl]benzoic acid (A) and a piperazine derivative (C). The second method of synthesis involved the reaction between 4-{[4-fluoro-3-(piperazine-1-carbonyl)phenyl]methyl}phthalazin-1(2H)-one (B) and a benzotriazole derivative (D). The influence of the applied conditions and various parameters of the synthesis on the course of the reaction were examined - the pressure and the used solvent, catalyst, operating time and microwave power. The obtained results prove that one of the proposed optimized methods can be successfully applied in the preparation of Olaparib – it allows obtaining API with good efficiency comparable to that in the literature, but over a period of 75 seconds, which is faster than previously known methods.

"Innovation Incubator 2.0" The program financed by the non-competitive project "Support for the management of scientific research and commercialization of R&D results in scientific units and enterprises" , implemented under the Smart Growth Operational Program 2014–2020 (Measure 4.4).

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P18

The green highway for the synthesis of PZ-1388, 5-HT$_6$ receptor inverse agonist with neuropathic pain-alleviating activity

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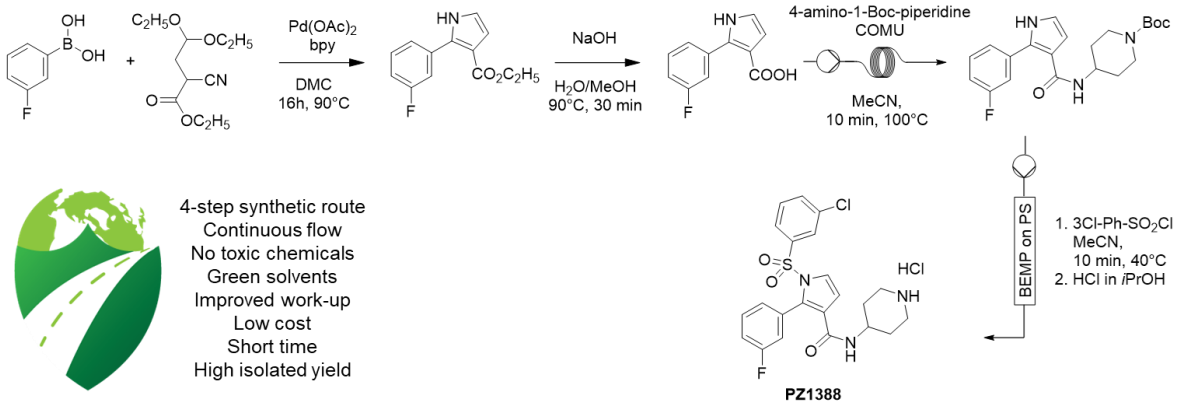
Although development of new drugs by organic synthesis has contributed to a revolution in pharmacotherapy over the last decades, its detrimental impact on environment remains a current global problem. In this context, application of **green chemistry principles** and **high-throughput technologies** has gained a particular interest in the early-stage development of bioactive molecules [1].

Our studies in the group **2-phenyl-1*H*-pyrrole-3-carboxamides** [2] identified compound **PZ-1388**, which behaves as a potent 5-HT$_6$ receptor inverse agonist at Gs, Cdk5, and mTOR signaling. The compound was subsequently used as a molecular probe to establish **new paradigm in neuropathic pain**. PZ-1388 suppressed the non-physiological mTOR activation by constitutively active spinal 5-HT$_6$Rs and thus alleviated painful symptoms in traumatic and toxic neuropathy as well as improved co-morbid cognitive deficits in rats [3].



Herein, we report the **eco-compatible synthesis** of PZ-1388 in the sequence of palladium-catalysed C−C bond formation, alkaline hydrolysis, followed by **continuous flow amide coupling and sulfonylation** with simultaneous formation of hydrochloride salt. As compered to previously used approach [2,4], the optimization process limited the synthetic steps, replaced toxic chemicals and solvents with green alternatives, decreased time and cost, and significantly increased the total yield.

The studies were financially supported by the National Science Centre, Poland (grants no *2019/33/N/NZ7/01875*, *2016/21/B/NZ7/01742*), The French National Centre for Scientific Research, French Government Scholarship, and Jagiellonian Interdisciplinary PhD programme.

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P19

Investigation of an antipsychotic compound D2AAK1 binding to human 5HT1A receptor by funnel metadynamics simulations

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Therapy of schizophrenia with currently available antipsychotics meets several difficulties. Available drugs are only efficient against positive symptoms of the disease, with little or no effect on negative and cognitive symptoms. They provide apparent improvement in only a half of schizophrenic patients. Furthermore, they have severe side effects of neurological and metabolic nature. Therefore, further research for novel antipsychotic drugs is inevitable.

D2AAK1 is a novel multi-target compound, showing affinity to dopamine D2, serotonin 1A and 2A receptors[1]. Interestingly, it is characterized by a complex effect on anxiety observed in vivo (data not published). To gain deeper insight into principles underlying its complex behavior, detailed binding kinetics data are necessary. Moreover, knowledge of binding site and binding mode would allow to plan further modifications on the basis of known protein-ligand interactions. Unfortunately, obtaining kinetic or structural experimental data on membrane proteins is more demanding than similar procedures performed on soluble proteins. Fortunately, recent inventions in computational approaches made it possible to gain reliable and valuable insights into ligand binding processes.

Funnel metadynamics is a method of calculating binding free energy and binding mode, introduced by Limongelli, Bonomi and Parrinello in 2013 [2]. It is a modification of the metadynamics approach, where during a molecular dynamics simulation artificial potentials are added along initially defined collective variables. The method is a modification of metadynamics, in which an additional funnel-shaped potential Is applied to reduce the volume non-productively sampled by the ligand during the simulation. It's recent edition [3] it includes graphical user interface module for preparation and analysis of the simulation results.

In the present study, we apply the Funnel Metadynamics method to the D2AAK1-5HT1AR complex to find its binding mode and gather binding kinetics data. The obtained free energy surface reveals a metastable binding site located above the orthosteric binding pocket, and provides information concerning the path connecting the metastable and orthosteric binding sites.

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P20

Multicomponent materials useful in the regeneration of cartilage/bone tissue

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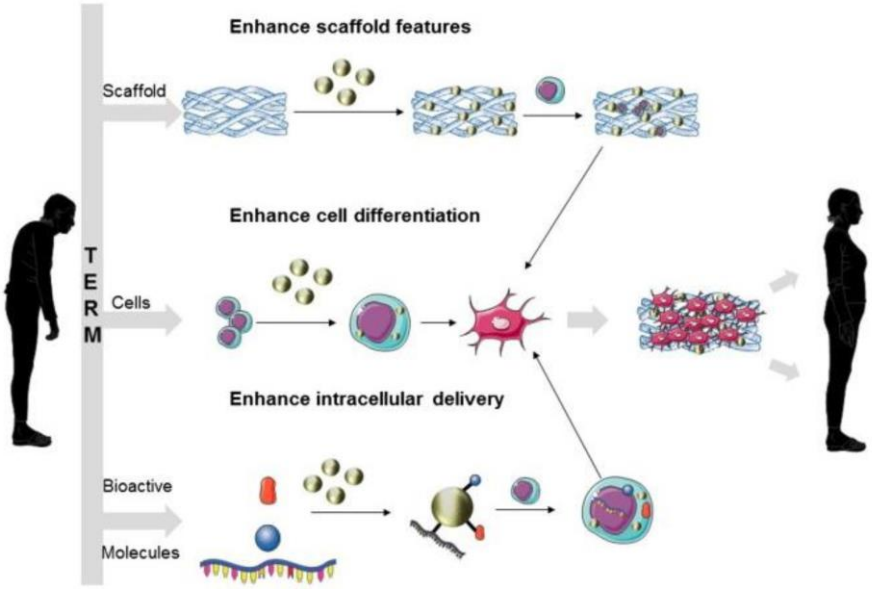
For years, there has been an intense development of regenerative medicine related to obtaining and using multicomposite materials to regenerate various tissues. The combination of tools used in medicine, biochemistry, and material engineering has resulted in the interdisciplinary field of Tissue Engineering and Regenerative Medicine (TERM), the use of which is shown in the figure below [1].



The Institute of Organic Chemistry TUL conducts research on the design and synthesis of materials useful in the regeneration of cartilage/bone tissue. The research is carried out in accordance to the assumptions of TERM. The developed materials can be described as multicomponent multicomposite materials. They contain carbon non-woven fabric that guarantees appropriate mechanical strength and also biomolecules that influence the regeneration of bone and cartilage tissue. Bone Morphogenetic Protein fragments and polysaccharides were used as biomolecules. There have been made attempts to develop a method of incorporating ferromagnetic nanoparticles into the obtained materials, which should enable imaging of the implanted material.

Acknowledgements: The financial support from the National Science Centre, Poland, under project number UMO-2018/31/B/ST8/02418.

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P21

Effect of selected acids on the viscosity of chitosan hydrogels

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Chitosan is a natural polymer obtained by deacetylating chitin. In its structure, it has reactive functional groups - hydroxyl and amino groups, which are involved in the processes of chemical modification of the compound. The primary amino groups in the molecule determine the ability of chitosan to dissolve only in an acidic environment with the formation of a polycation [1, 2].

In the conducted research, to enable the dissolution of chitosan, lactobionic, mandelic and shikimic acids were used. They belong to hydroxyacids, which are compounds containing hydroxyl and carboxyl groups in their molecule, so they have the properties of both – alcohols and carboxylic acids. In addition to their wide use in cosmetology as chemical peels, hydroxyacids can be used in the treatment of skin lesions. The obtained hydrogels combine the beneficial properties of hydroxyacids with the regenerating effect of chitosan on the skin and can be used in pharmaceutical preparations, dressing materials and in dermocosmetics [3].

The aim of this experiment was to compare the rheological properties of hydrogels formed by dissolving chitosan in solutions of the above-mentioned hydroxyacids. Chitosan of medium molecular weight (MMW) in the amount of 2% (w / v) and hydroxyacids in the amount of 0.002 mol each were used for the preparation of the samples. The prepared gels were incubated for 24 hours under refrigeration and then thermostated to 25°C. Measurements were made using a SMART series rotational viscometer (Fungilab) with a set of appropriate spindles.

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P22

Structure modeling of TRPA1 ion channel – determination of the binding site for antagonists

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TRPA1 is a transmembrane nonselective cation channel, one of the most promising and studied targets in preclinical pharmaceutical research, especially in the context of chronic and acute pain and respiratory diseases [1,2]. Its whole structure has already been experimentally resolved, but the binding site of TRPA1 antagonists, e.g. HC-030031, a model methylxanthine derivative, remains unknown.

The aim of the study was to determine the potential binding site of xanthine antagonists and to describe the binding mode of their representative, compound HC-030031, using molecular modeling approach.

To this end, an approach using the available molecular modeling tools predicting binding sites of biological targets, e.g. Sitemap, MetaPocket 2.0, DeepSite, CASTp 3.0 was applied. The TRPA1 channel model was prepared using a properly optimized 6PQQ three dimensional atomic structure provided by cryo-EM [3]. The obtained results were analyzed in terms of scoring functions, pocket size and frequency of occurrence. Compound HC-030031 was docked to the selected binding pockets using Induced Fit Docking procedure, and the obtained HC-030031-TRPA1 complexes served as a source of conformational models of the protein, and were further tested using retrospective virtual screening and molecular dynamics simulations.

The research showed that HC-030031 binds to a pocket formed by the TRP-like domain and the pre-S1, S4, S5 helices of one subunit and establishes crucial interactions: hydrogen bond with Asn-855 and π-π stacking with Trp-711, which might play an equally important role in recognition of other xanthine derivatives and their bioisosteres.

These results allow for a more specific search for new potential TRPA1 antagonists in the group of xanthine derivatives as well as their bioisosteres and hence new potential drugs for the treatment of neuropathic pain, asthma, and COPD. This study represents also the first attempt to determine the binding site and describe interactions of HC030031 - the approach that brings together reports on site-directed mutagenesis and the latest cryo-EM structure of the other antagonist bound to TRPA1.

The study was financially supported by the National Science Centre, Poland (grant no. 2020/37/N/NZ7/02365) and Jagiellonian University Medical College (grant no. N42/DBS/000187)

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P23

Benzophenone derivatives with histamine H_3 receptor affinity and cholinesterase inhibitory potency as multitarget-directed ligands for possible therapy of Alzheimer's disease

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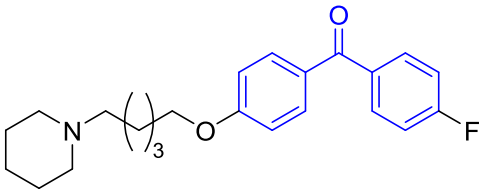
Multitarget-directed ligands (MTDLs) are compounds that affect more than one pathophysiological pathway [1], being promising alternative for effective future therapy of complex diseases like Alzheimer's disease (AD). Among pharmacological targets linked to the pathogenesis of AD, neurotransmitters such as acetylcholine, histamine, dopamine, serotonin, noradrenaline, and glutamate are considered crucial, as their dysfunctions can lead to the progressive deterioration of cognitive functions [2]. Based on our previous findings [3] we have designed, synthesized, and evaluated through *in vitro* biological assays a series of MTDLs potentially affecting neurotransmission in the central nervous system. Novel derivatives of substituted benzophenone moiety linked by an alkoxyl chain to a (methyl)piperidine or an azepane residue were tested towards human histamine H_3 receptors (*h*H_3R) affinity along with cholinesterase inhibitory potency. Moreover, inhibitory activity to human monoamine oxidase B (*h*MAO B) was also evaluated. The research results revealed promising biological activity of tested compounds with obtained nanomolar range of K_i (for *h*H_3R), submicromolar/micromolar IC_{50} values (for butyryl- and acetylcholinesterase) and weak inhibitory potency of *h*MAO B (inhibition < 50% at 1 μM). The compound presented below (**E325**) was identified as the most interesting MTDL for further optimization for AD treatment.



hH_3R K_i = 8 nM
AChE$_{electric\ eel}$ IC_{50} = 2306 nM AChE$_{human}$ IC_{50} = 9585 nM
BuChE$_{equine\ serum}$ IC_{50} = 172 nM BuChE$_{human}$ IC_{50} = 1155 nM
*h*MAO B = 25% of inhibition at 1 μM

This research was funded by the National Science Centre, Poland grant: No *UMO-2016/23/B/NZ7/02327*.

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P24

Donepezil derivatives as new multifunctional anti-Alzheimer's ligands combining 5-HT$_6$R antagonism, cholinesterase inhibition with antioxidant and chelating properties

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The concept of multifunctional ligands - substances capable of interacting with at least two biological targets may be a breakthrough in the therapy of diseases with complex pathophysiology. One of them is Alzhiemer's disease (AD) - the most common form of dementia with over 35 million people diagnosed worldwide. The disease is marked by neurodegenerative changes in the brain tissue and disruption of many neurotransmitter systems due to accumulation of abnormal protein aggregates, oxidative stress, and metal dyshomeostasis. These processes lead to gradual deterioration of cognitive functions and behavioral impairments [1]. One of the possibilities of anti-AD therapy is to target cholinergic system enzymes - acetyl- (AChE) and butyrylcholinesterase (BuChE). Inhibition of their activity leads to increased level of acetylcholine in the brain and alleviation of symptoms. Another promising biological target is the serotonin 5-HT$_6$ receptor (5-HT$_6$R). Its blockade enhances neurotransmission and exerts anxiolytic and antidepressant activity [2]. In our research, we designed multifunctional ligands by combining pharmacophore elements of the cholinesterase inhibitor - donepezil and a known 5-HT$_6$R antagonist (Fig. 1). Compounds were evaluated in *in vitro* studies (Ellman assay and radioligand binding assay) which revealed lead structure with nanomolar activities - IC$_{50}$ of 930 nM and 16 nM towards human AChE and BuChE respectively and human 5-HT$_6$R with K_i = 19 nM. In the ABTS assay the compounds showed ability to scavenge the pre-formed radicals and finally, two ligands were revealed as selective Fe^{2+} chelators. Due to the multidirectional activity, we consider them as an excellent starting point for further development and optimization.

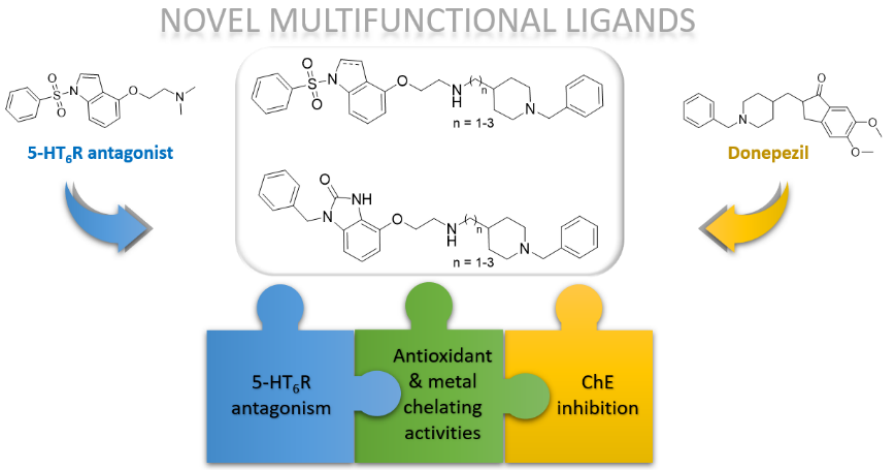

Figure 1. Designed multifunctional ligands – 5-HT$_6$ receptor antagonists and cholinesterase inhibitors with antioxidant and metal chelating properties.

The presented studies were financially supported by National Science Centre, Poland grant No *UMO-2016/23/D/NZ7/01328* and by Jagiellonian University grant *No N42/DBS/000177*.

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P25

Synthesis and cellular effects of novel 1,2,4-triazine derivative in human colon cancer

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Objectives: Ineffective therapy of colorectal cancer forces to look for new compounds, useful in treatment. **Methods:** The target compound MM-129 was obtained by a multi-step synthesis, starting from 1,2,4-triazine. the synthesis of 3-methyl-5-methylsulfonyl-1-phenyl-1H-pyrazolo[4,3-e][1,2,4]triazine was performed based on the reaction of nucleophilic substitution of hydrogen in 1,2,4-triazine and oxidation process under two-phase-transfer catalysis conditions. The antiproliferative activity of MM-129 and the reference drugs roscovitine and 5-fluorouracil were examined by [3H]thymidine incorporation assay and in a zebrafish embryo model. To explore the cellular mechanism, by which the synthesized compound triggers induction of apoptosis, we examined the alterations of the mitochondrial transmembrane potential, phosphatidylserine externalization and caspase activity by using flow cytometry analysis.

Results: Screening results revealed that MM-129 exhibited strong inhibition activity toward DLD-1 and HT-29. It showed potent antiproliferative effects against colon cancer cell lines with IC50 values 3.1 μM compared to 5-fluorouracil (5-FU) and roscovitine (ROS) with values above 10 μM. Flow cytometry analysis revealed that apoptosis was the main response of colorectal cancer cells to MM-129 treatment.

Conclusion: These preclinical results suggest that this novel heterofused 1,2,4-triazine derivative, due to high proapoptotic activity, should be tested in the clinic for treatment of solid tumors, such as colon cancer.

This research was funded by National Science Center, Poland grant number 2018/31/B/NZ7/00875.

P26

Study of the influence of dinucleoside polyphosphates on the catalytic activity of human adenylate kinase.

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Adenylate kinase (EC 2.7.4.3, AK) (Fig.1) is a small enzyme present in eukaryotes and prokaryotes. Its function is to catalyze the transfer of phosphate groups between nucleotides: $MgATP^{2-} + AMP^{2-} <=> MgADP^{-} + ADP^{3-}$. It regulates the concentration of adenine nucleotides and controls the rate of changes in metabolic pathways in cells. Disruptions in catalytic activity and mutations of the genes encoding this enzyme can lead to pathological conditions and diseases. [1] For this reason, there is a need to find new adenylate kinase modulators. The only known inhibitor of AK activity is P^1,P^5-diadenosine-5'pentaphosphate (Ap_5A), but its effect on the catalytic activity of the human adenylate kinase isoenzyme 1 (hAK1) has not yet been determined. [2]

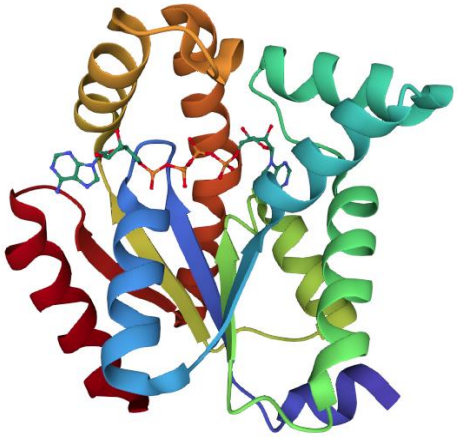

Fig 1. Crystal structure of human adenylate kinase isoenzyme 1 with Ap_5A (PDB code: 2C95).

For our study, we have selected three dinucleoside polyphosphates: (1) P^1,P^5-diadenosine-5'pentaphosphate [Ap_5A], (2) P^1-(5'-adenosyl) P^5-[5'-(8-iodo)-guanosyl] pentaphosphate [AP_5(8-Iodo-G)], and (3) P^1-(5'-Adenosyl) P^5-[5'-(2'-deoxy-thymidyl)] pentaphosphate [AP_5dT]. We determined that all investigated compounds inhibited hAK1 in both directions of the enzymatic reaction in a concentration-dependent manner, although with different efficiencies. Ap_5A was found to efficiently inhibit hAK1 at lower concentrations as compared to other tested dinucleoside.

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P27

Screening and quantitative evaluation of cinnamic acid derivatives as tyrosinase inhibitors

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Skin dies - melanins are synthesized in epidermal pigment cells (melanocytes) in a multistage process involving both enzymatic and chemical reactions. The two initial steps in this pathway, i.e. the hydroxylation of L-tyrosine to 3,4-dihydroxy-L-phenylalanine (L-DOPA) and then oxidation of L-DOPA to dopaquinone, are catalyzed by the enzyme tyrosinase. Further transformations of dopaquinone depend, among others, on presence in melanocytes of thiol compounds - cysteine and / or glutathione, and take place as chemical or enzymatic reactions [1,2].

Hyperpigmentation disorders result from an excessive amount and / or inappropriate distribution of melanins in the skin. They constitute a significant aesthetic and medical problem, and significantly affect the mental health of affected patients. The current therapeutic methods of hyperpigmentation disorders including external preparations and instrumental methods require long-term use and are characterized by insufficient effectiveness. Currently there is no substance that would completely and safely inhibit the formation of pathological hyperpigmentations, and moreover, the used cosmetic raw materials often cause multiple side effects. As a results, much effort is done to identify new, more effective and safer compounds for inhibition of melanin production and reducing existing hyperpigmentations [3,4].

The performed research led to identification of novel tyrosinase inhibitors among cinnamic acid derivatives. In the tested series of 20 compounds, 2 novel active derivatives were identified. In the concentration of 500μM they inhibited diphenolase activity of tyrosinase by 55.32±1.9% and 64.31±0.21%. Quantitative evaluation resulted in IC_{50}s of 265.34±33.41 and 152.87±14.00 μM. Kinetic studies showed that they could act as mixed-type enzymatic inhibitors. Moreover, two enantiomers of a previously identified racemate derivative were tested. Both enantiomers showed weaker tyrosinase inhibition than racemate (36.71% and 43.23% vs 55.38%). The most active compounds will be further tested in more advanced models like cell lines and pigmented human epidermis. They could be potentially used in cosmetic products for hyperpigmentation disorders.

The research was financed by National Centre for Research and Development within LIDER XI program (contract number LIDER/26/0094/L-11/19/NCBR/2020).

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P28

Influence of “point diversity”on the activity for serotonin receptor 5-HT$_6$R in the group of topologically similar triazine derivatives

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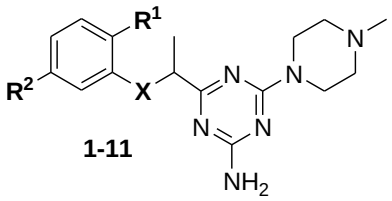
Recently, 1,3,5-triazine derivatives were discovered as original new chemical group of antagonists for the serotonin receptor 5-HT$_6$ [1], promising in search for new drugs of CNS diseases [2]. In this study, a new series of triazine derivatives (**1**-**11**, Fig.1) with common topology of 4-(4-methylpiperazin-1-yl)-6-(1-phenoxyethyl)-1,3,5-triazin-2-amine restricted by substituents at position 2 and 5 of the phenyl ring and changeable hetero-atom within the linker was under consideration.



1-11

R^1= H, F, Cl, *i*-Pr, CF_3

R^2= H, F, Cl, CF_3, CH_3

X= O, S, Se

Fig. 1

The compounds were obtained within 3-step synthesis, including two kinds of alkylation and cyclic condensations. The affinities for 5-HT$_6$R and off-targets: dopamine D_2 and serotonin 5-HT$_7$ receptors, were tested in the radioligand binding assays. Crystallographic X-ray analysis for a representative member and docking studies to the homology model of 5-HT$_6$ receptor have been performed. Based on the obtained results structure-activity relationship analysis indicated that a branched substituent at position 2 of Ph (*i*-Pr > CF_3) is beneficial “point-property” for the high affinity and selectivity towards 5-HT$_6$R in case of this topology of compounds.

Partly supported by by the National Science Centre (grant UMO-2018/31/B/NZ7/02160). M.P. and L.W. have performed synthesis within Student Medicinal Chemistry Club (Studenckie Koło Chemii Medycznej UJ CM w Krakowie).

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P29

New phenyl-glycinamide derivatives with hybrid structure as candidates on new effective anticonvulsants

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Epilepsy is recognized as one of the most common neurological disorders just after the stroke. It is characterized by a multifactorial pathogenesis, which reflects generally in a low clinical efficacy of drugs currently available in the pharmacotherapy. Notably, despite huge advances in epilepsy studies and approval of several new antiseizure drugs, in nearly 30% of patients pharmacotherapy does not produce expected improvement and they suffer from drug resistant epilepsy (DRE). In recent years development of new drugs for the treatment of multifactor diseases such as Alzheimer's disease, epilepsy and pain of various origin but also diseases with high risk of drug resistance is focused on the multifunctional compounds which possess predominantly hybrid structures. Hybrid molecules are compounds that contain several pharmacophores merged on one chemical scaffold. The combination of several structural domains in one chemical molecule gives the possibility of interaction with more than one molecular target through the use of one substance.[1,2] Bearing in mind the assumptions of multi-target strategy in the current studies the series of 22 new chemically original compounds have been obtained. These compounds were designed by applying the fragment-based approach, as they merge chemical fragments of well-known **TRPV1 antagonists** (e.g. BCTC)[3] and potent anticonvulsant, namely compound **KA-104** described in our earlier studies.[4] The compounds obtained possess wide spectrum of activity in the preclinical studies, as they are effective in the most widely employed animal seizure models, the maximal electroshock (MES) test, the psychomotor 6 Hz (32 mA) seizure model, and importantly also in the 6 Hz (44 mA) model of DRE. It is also worth mentioning that the obtained molecules beyond TRPV1 antagonism, also inhibits fast sodium and calcium currents in the *in vitro* studies. The most potent anticonvulsant **53** and **60** revealed the following pharmacological properties: ED_{50}=89.7 mg/kg (MES), ED_{50}=29.9 mg/kg (6 Hz, 32 mA), ED_{50}=68.0 mg/kg (6 Hz, 44 mA) for **53** and ED_{50}=73.6 mg/kg (MES), ED_{50}=24.6 mg/kg (6 Hz, 32 mA), ED_{50}=56.3 mg/kg (6 Hz, 44 mA) for **60**. Additionally both compounds, namely **53** and **60** were effective in *iv*PTZ seizure threshold test in mice. Furthermore, *in vitro* studies proved beneficial ADME-Tox properties. Thus, obtained molecules may be potentially effective in various human epilepsies including i. a. tonic-clonic, psychomotor and pharmacoresistant seizures.

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P30

Synthesis of novel derivatives of moxifloxacin and enoxacin with fatty acids and evaluation of their cytotoxic effect on cancer cells

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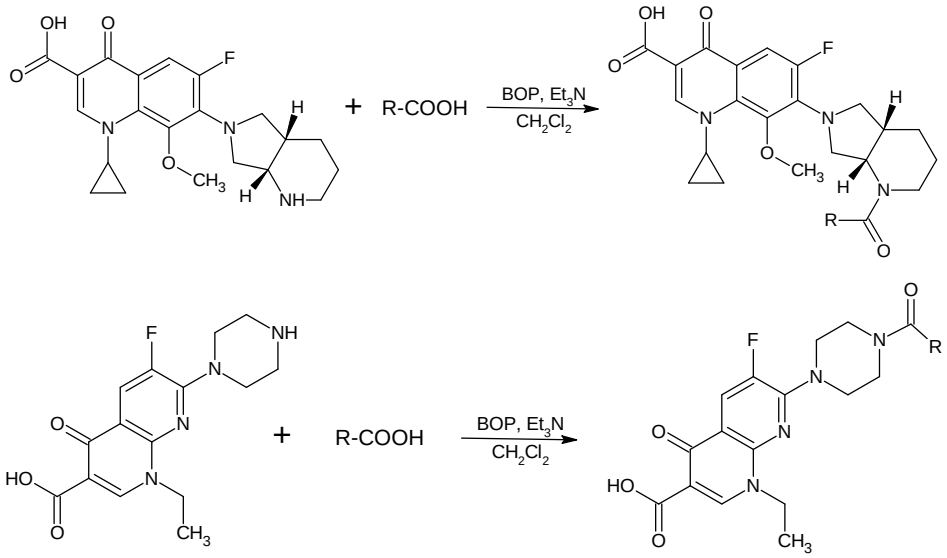
Even during COVID-19 pandemic cancer is still a leading cause of death worldwide causing in 2020 alone almost 10 million of deaths [1], while COVID-19 has been reported as cause of death of approx. 2 million people [2]. There is no doubt there is still a need for more effective and safer cancer treatment.

Both enoxacin and moxifloxacin (originally registered as antibiotics) have shown cytotoxic effects against cancer cells. Those effects include inhibition of cell proliferation, causing a cell cycle arrest and induction of apoptosis [3-4]. Fatty acids alone can also induce apoptosis in cancer cells [5] as well as enhance cell membrane penetration thanks to their lipophilic nature [6]. Our group previously has shown a synergistic anticancer effect of conjugates of fatty acids and another fluoroquinolone: ciprofloxacin [7]. Presented study verifies whether the same mechanism could be observed in conjugates of enoxacin or moxifloxacin.

The structures of derivatives were confirmed by H^1NMR, C^{13}NMR, and Mass Spectrometry. Cytotoxicity was evaluated by MTT assay.



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P31

The anticancer properties of antidepressants

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Cancer is a major public health problem and a complete cure for most cancer types is still unclear. The development of new anticancer agents is expensive and time-consuming. Drug repurposing has been proved to be an effective strategy to meet the urgent need for novel anticancer agents for treating or supporting cancer treatment. An important factor that can increase the risk of cancer and worsen the prognosis after a diagnosis is depression. Therefore, the strategy to repurpose and utilize modified antidepression medications as anticancer drugs seems to serve a double purpose.

The goal of our research was to investigate the mechanism of anticancer activity of serotonin partial agonist reuptake inhibitor (SPARI) using in vitro and in silico methods.

The cytotoxic activity of selective serotonin transporter inhibitors with partial 5-HT1A receptor activity (vilazodone, vortioxetine) have been examined in androgen-insensitive human PC-3 prostate cancer cell and human immortalized prostatic cell line PNT1A, and reference cells BALB/c 3T3 cells. The docking studies to the 5-HT1A receptor model based on the crystal structure of the serotonin 1B receptor were performed using ICM Pro software. The docking studies to the serotonin transporter are in progress.

Both compounds exhibited cytotoxic activity against PC-3 prostate cancer cells. As a result, the potential antitumor activity of SPARI antidepressants in late stage prostate cancer has been demonstrated for the first time. Preliminary testing of the anticancer properties of this group of compounds showed activity comparable to that of the reference compound used. The oncotoxic activity of vortioxetine is two times greater than that of vilazodone. Docking studies indicated some differences of binding to the 5-HT1A receptor that may have implication for understanding their mechanism of action.

Further studies on anticancer properties of antidepressants as well as their potential use in cancer prevention and treatment, is required and justified.

This study was funded by statutory grant of NMI.

P32

Sonochemical synthesis and preliminary evaluation of the activity of the group of alkyl arylpiperazines as an antitumor activity in the treatment of breast cancer

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Breast cancer is the most common cancer in women in the world today. In 2020, 2.3 million cases were diagnosed and 685,000 deaths were registered. It is estimated that in the world today live 7.8 million women diagnosed with breast cancer in the past 5 years. Modern therapy allows in many cases a complete recovery, especially if the disease is diagnosed quickly. However, new drugs in the context of breast cancer treatment are still widely sought because drug-resistant cases still occur or the disease is so advanced that the therapy focuses on extending life with maintaining an acceptable quality of life.[1]

Knowing that alkyl derivatives of arylpiperazines, in addition to their widely described effects on the central nervous system, are also considered in many studies as molecules with potential antitumor activity[2,3,4,5], we designed and synthesized a group of compounds for the assessment of activity towards MDA-MB-231 breast cancer cells. The compounds were obtained by a newly developed sonochemical phase-transfer catalysis (PTC) method in the presence of potassium carbonate. The most active substance among those obtained was the compound JJ-ON-5, which inhibited the viability of MDA-MB-231 cells by 50% at a concentration of only 6.5 μM.

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P33

New angularly condensed diquinothiazines as anticancer agents

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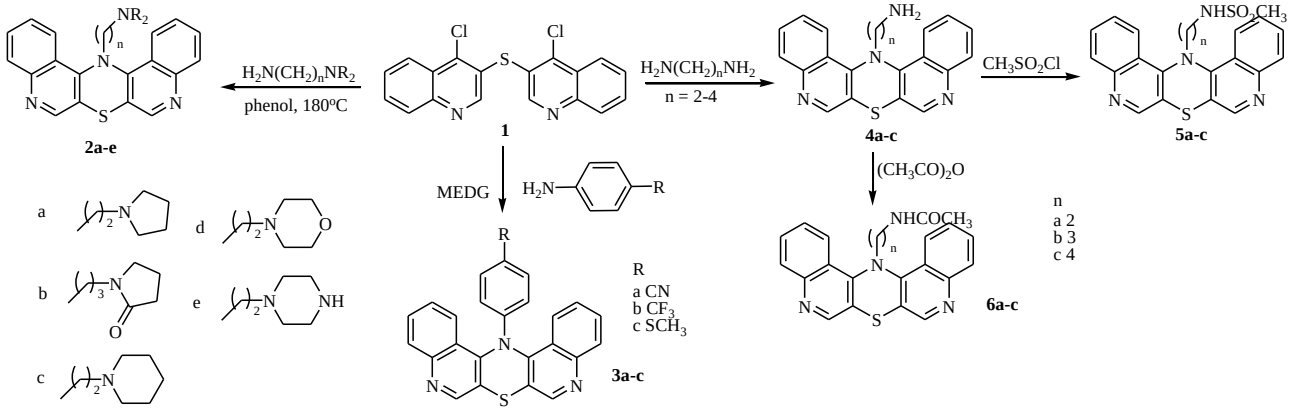
Replacing the benzene rings in the phenothiazine core with the quinoline ring leads to various types of quinobenzothiazines and diquinothiazines. N-substituted lineary condensed diquinothiazines exhibit strong action against tens of cancer cells derived from melanoma, leukemia, colon, CNS, ovarian, renal, breast, skin, prostate, and non-small cell lung cancers. The most promising lineary condensed diquinothiazine, 6-chloroethylureidoethyldiquinothiazine, exerted suppressive and anti-inflammatory activities in the mentioned above *in viv*o models, and showed inhibitory activity of IFNβ expression and IFNβ-dependent downstream genes and proteins involved in the pathogenesis of autoimmune diseases.
The aim of this study is to report the synthesis of double angularly condensed diquinothiazines with aminoalkyl, amidoalkyl, sulfonamidoalkyl, and substituted phenyl groups and the evaluation of their anticancer activity.



Sulfide **1** reacted with dialkylaminoalkylamines, diaminoalkanes, and substituted anilines in hot phenol or in boiling monomethyl ether of diethylene glycol to form 14-dialkylaminoalkyl-, 14-phenyl- and 14-aminoalkyldiquinotiazines **2a-e**, **3a-c** and **4a-c**. The last compounds were acetylated and methanesulfonylated to give 14-acetamido- and 14 methanesulfonamidodiquinothiazines **5a-c** and **6a-c**.
Using a cancer (A549, H1299) and normal (BEAS-2B) pulmonary cell lines we have performed a preliminary studies *in vitro*, with viability decreasing and cellular death (apoptosis and necrosis) induction. Using a 72 h MTT viability assay (Promega) and flow cytomertic analysis with Annexin-V (Becton Dickinson) we have found a novel anticancer potential of tested compounds. The promising results suggesting a selectivity against the cancer cell lines with toxicity connected to the compounds structure.

P34

New 1,3,5-triazine derivatives containing cell penetrating peptides and secondary amines as a substituents of 1,3,5-triazne ring, molecules with potential anti-tumor activity

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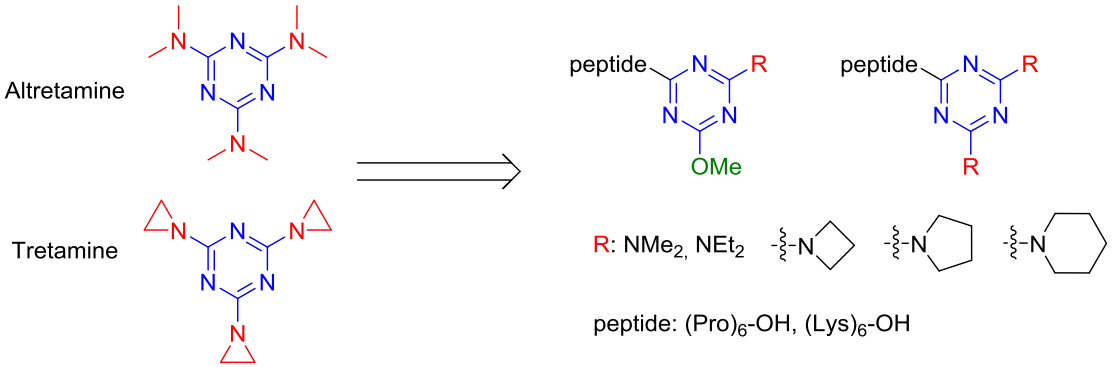
The search for and designing and preparing new and more effective medicinal active substances is one of the goals that have been pursued for many years. It can be accomplished by finding new compounds or modifying known molecules. One of the privileged molecules characterized by wide spectrum of biological activity is 1,3,5-triazine (*s*-triazine). 1,3,5-Triazine derivatives have antimicrobial, antimalarial or antiviral activity, furthermore *s*-triazine derivatives own anticancer activity, what is documented by an amine substituted *s*-triazine derivatives use in cancer treatment: Tretamine and Altretamine[1]. Our team conducts research on the search for new 1,3,5-triazine derivatives with anticancer properties. It was designed and synthesized conjugates contain 1,3,5-triazine core with peptides and amines attached to the ring. As peptide fragments were used hexaproline and hexalysine, because these peptides are cell-penetrating peptides[2] (CPPs) and should increase the permeability through cell membranes of 1,3,5-triazine derivatives. As amine fragments were used secondary alkyl amine groups (dimethylamine, diethylamine) and amine cyclic moiety (azetidine, pirolidyne and piperidyne)



Amine-peptide-triazine conjugates have been obtained in the multi-step solid phase synthesis strategy with high purity determined by LC/MS, and were evaluated *in vitro* on selected cancer cell lines.

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P35

Novel multi-target ligands of aminergic GPCRs for the treatment of schizophrenia

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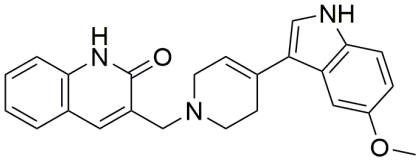
Schizophrenia is an important health issue, affecting almost 1% of the population, frequently with a strong social and economic impact, as patients often suffer from unemployment and are homeless. Antipsychotics used to treat schizophrenia are also prescribed in bipolar affective disorder, which has a prevalence of 2.3% in the population. As a consequence, about 16.5 million patients in the EU need antipsychotics on a daily basis. A modern approach to design bioactive compounds against diseases with a complex pathomechanism, such as schizophrenia, is searching for multi-target ligands.

As a part of our research on CNS agents we performed structure-based virtual screening to identify multi-target ligands of aminergic G protein-coupled receptors with affinity to different dopamine and serotonin receptors [1]. As a result we identified 10 active compounds, confirmed their affinity *in vitro* and four of them (D2AAK1, D2AAK2, D2AAK3 and D2AAK4) were studied as potential antipsychotics [2].

We obtained 88 derivatives of virtual hit D2AAK1 [3,4] and here we present a part of our optimization campaign. 17 selected derivatives were studied *in vitro* to determine their affinity and functional activity towards dopamine D_2, serotonin 5-HT_{1A} and 5-HT_{2A}, histamine H_1 and muscarinic M_1 receptors. Two most promising compounds were studied *in vivo* and decreased amphetamine-induced hyperactivity in mice at the studied doses. Molecular modeling allowed to rationalize the observed structure-activity relationships.



D2AAK1

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P36

Correlation between pharmacokinetic parameters and anticancer activity of betulin derivatives attached 5,8-quinolinedione moiety

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Nowadays, the search for potentially new drugs requires the application of computational chemistry, including experimental and *in silico* analysis of physicochemical properties, pharmacokinetic features, ADMET analysis, and quantitative structure-activity relationship (QSAR) research. The therapeutic potential of a drug depends on its distribution in the body [1].

Modifications of natural substances are one of the commonly used methods of searching for new bioactive substances. One of the first compounds obtained from the plant is betulin, which contains a hydroxyl group at C3 and C28 positions. Introduction of substituents at these positions influences the bioavailability and biological activity of derivatives [2]. Second sources of bioactive substances are fungi and bacteria. One of the group of substances obtained from *Streptomyces* bacterial were 7-amino-5,8-quinolinedione antibiotics, which exhibit a broad spectrum of biological activity. The 5,8-quinolinedione scaffold was one of the first organic compounds, for which structural modifications were correlated with anticancer properties and low toxicity [3]. The combination of these two active moieties caused hybrids with high anticancer activity [4].

The present study aimed to determine the lipophilicity of betulin-5,8-quinolinedione hybrids. Moreover, a correlation between lipophilicity and pharmacokinetic properties was also analyzed. The study was supplemented by analysis of the relationship between biological properties, like anticancer activity and NAD(P)H:quinone oxidoreductase (NQO1), and lipophilicity of hybrids.

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Method for determination of azidothymidine using copper(I) catalyzed alkyne-azide cycloaddition (CuAAC)

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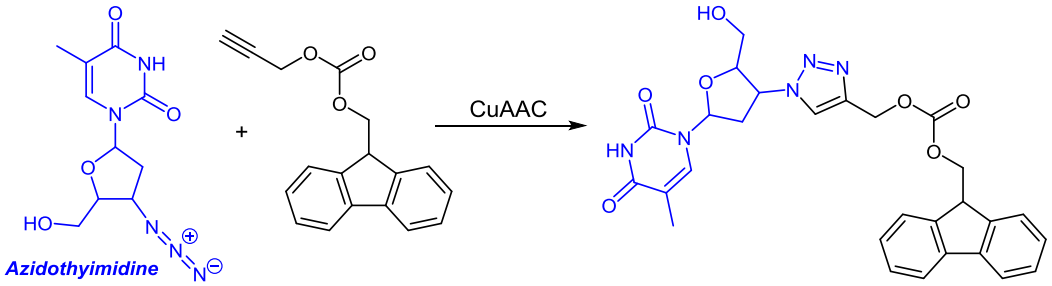
Compound labeling and/or derivatization are often used for the analysis of biologically active compounds in biological matrices and pharmaceutical formulations. In biochemical and ADME studies, labelling can help to determine drug distribution and their fate in a body.[1]
Herein, we report a method for labelling of azidothymidine using a *click chemistry* concept, which enables quantification of the active pharmaceutical ingredient in *Retrovir* using RP-HPLC with UV detection. Azidothymidine (AZT) is an antiretroviral drug used in the treatment of HIV infections. The azide group makes AZT a ready-to-use substrate for the copper(I)-catalyzed azide-alkyne cycloaddition (CuAAC), the main reaction of the *click chemistry* approach.[2] Earlier, only one study attempted to use CuAAC labelling of AZT for analytical method development, using a custom-made alkyne label.[3]



Scheme 1. Derivatization including labeling of azidothymidine with propargyl-Fmoc tag based on the CuAAC reaction.

Copper-catalyzed(I) azide - alkyne cycloaddition reactions (CuAAC) of AZT and other model compounds with the propargyl-Fmoc label, previously proposed in our research team[4], were studied. We used a propargyl-Fmoc label, easily made from common reactants, making the method much more accessible. Likewise, we optimized CuAAC labelling conditions and found that application of a copper-chelating ligand (AMTC) significantly improves labelling efficacy. The obtained results allowed to determine limits of detection and quantification (LOD and LOQ) for the AZT-labeled conjugate, according to the ICH guidelines. We also confirmed that, Fmoc label and AZT-labeled conjugate are devoid of cytotoxic activity guarantying laboratory *hazardous chemicals* safety policy.

The project was financially supported by National Science Center, Poland grant no 2018/29/N/NZ7/01918.

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P38

Curcuminoid Chalcones as New Therapeutic Substances for Use in Oxidative Stress-Dependent Diseases

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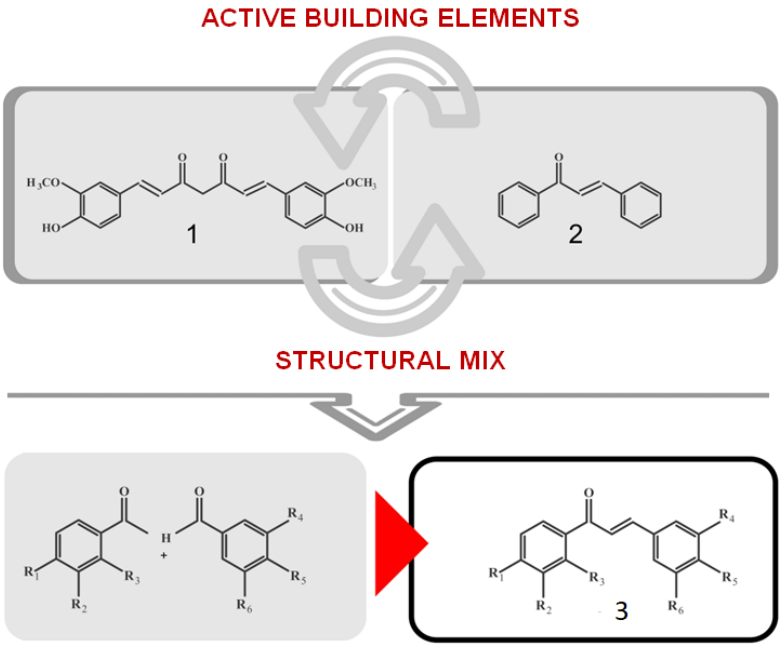
It is generally believed that the pathogenesis of a majority of multifactorial diseases like atherosclerosis, hypertension, Alzheimer's and Parkinson's disease, cancer and inflammatory conditions is related to the imbalance between pro-oxidant and antioxidant homeostasis. In light of the role of free radicals in the pathogenesis of these diseases, the issue of multifaceted activities of antioxidants as potential and versatile therapeutics has become very important [1]. The current trend of modern medicinal chemistry is the search for completely new entities e.g. hybrids, conjugates, etc. [2] in the area of so-called small-molecule drugs and natural origin compounds. The current priority, apart from designing effective active structures, is also to develop the concept of their rational synthesis. The main purpose of this study is to design and obtain a series of complex compounds - curcuminoid chalcones (3) - containing active moieties characteristic of the structure of the curcumin (1) and chalcone (2) type compounds (Scheme, R_1-R_6: -OH or -OCH_3 or -H). The proposed derivatives were obtained by the Claisen-Schmidt reaction [3] of selected acetophenones and aldehydes under various conditions, both classical in solutions and in solvent-free microwave or ultrasound variants, in order to optimize the method. The obtained curcuminoid chalcones will be substrates for further structural transformations.



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P39

Virtual screening for discovery of new thymidine phosphorylase inhibitors as anticancer agents

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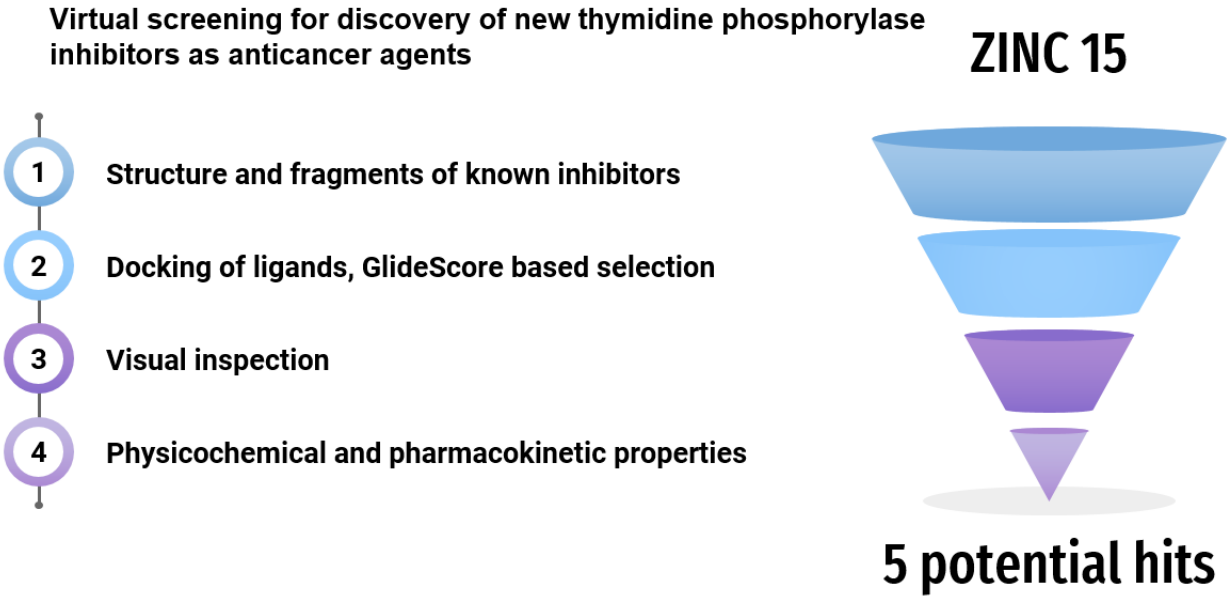
Excessive activity of thymidine phosphorylase (TP) can play an important role in cancer development and metastasis. Activity of TP elevates level of 2-deoxy-D-ribose which leads to promotion of angiogenesis, blockage of apoptotic pathway or activation of kinase pathway [1–3]. Since significance of TP in the course of the neoplastic disease was proved, extensive research has been conducted in order to find effective inhibitors able to restrain progression of cancer [4]. In our research we applied molecular modeling including structure-based virtual screening approach to find novel lead compounds with inhibitory activity against TP. As a result of our studies we obtained 330 ligands possessing favorable scoring function values. Further selection was carried out based on visual inspection and assessment of *in silico* predicted physicochemical and pharmacokinetic properties. Finally, we found several compounds which belong to uracil and hydantoin derivatives and fit to TP active site which can implicate significant interactions, resulting in beneficial inhibitory properties.



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P40

A new approach to encoding the chemical structure based on the FTIR spectra of compound

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Molecular fingerprints encoding the structure of a molecule in the form of a binary vector are well-known and have been used for many decades in chemoinformatics.[1, 2] They are commonly used for searching databases of compounds (virtual screening), or modeling of physicochemical properties of compounds (QSPR) or their biological activity (QSAR). However, it seems that the capabilities of describing the structure of compounds using molecular fingerprints have already been achieved, and obtaining more accurate prediction models is only joined with improving the machine learning algorithms.

To go beyond the current methodologies used to encode the 2D structure of compounds into binary vectors (e.g. MACCS, ECFP, FCFP, Molprint 2D, etc.) we proposed a new algorithm using information from the FTIR (Fourier–Transformed Infrared Spectroscopy) spectra of compounds (**fpf** – FTIR molecular fingerprint). In the first step, the individual positions of the bit sequence were defined using the FTIR frequency ranges (650–4000 cm^{-1}) of the specific vibrations of the functional groups in the molecules, and their corresponding SMARTS codes (101 positions). In the second step, concatenation of vectors indicating the presence of a particular group in the FTIR spectrum and its corresponding substructure in the compound was performed.

Initial tests of the **fpf** showed that significant factors influencing its properties are: the input FTIR spectra (experimental FTIR spectra, functionally-enhanced derivative spectroscopy (FEDS) transformed FTIR spectra, and theoretical spectra), and the cutoff level of the band intensity. Interestingly, results showed that theoretical FTIR spectra can be as good as experimental ones. Next, the set of 103 small molecules (commonly used reagents in medicinal chemistry) was used to build the prediction models for logP, logD, and logS. The **fpf** showed better performance than any used 2D binary fingerprint (MACCS, ECFP, Molprint 2D) as a reference. Preliminary results show that the proposed representation (**fpf**) extends the description of the structure of the chemical compound and can be successfully applied in cheminformatics.

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P41

SilicoPharm – AI-driven polypharmacological in silico screening platform for the next generation drugs

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In recent years, many drugs have either been withdrawn from the market or discarded in the late phases of clinical trials. The main reason for this situation is the side effects of drug action, which are mainly caused by the interaction of drug molecules with biological off-targets. To reduce the risk of this effect, it is recommended to address the chemical molecule interactions at the molecular level with biological on/off-targets at the same time in the early stages of drug design/searching – this is the main aim of the in silico polypharmacology.[1,2]

In response to this demand, a prototype of an in silico computational platform to support the design process of new drugs concerning the polypharmacology paradigm was developed. The platform SilicoPharm was developed in a system-as-a-service architecture (SaaS), and it combines three modules to evaluate potential ligands in terms of their interactions with multiple biological targets (i. e. PharmacoPrint, mt-QSAR, mt-SIFt) using advanced artificial intelligence algorithms and innovative compound representations. SilicoPharm enables to build of user-defined workflows for different polypharmacological profiles.

Acknowledgments: The work was supported by the Polish National Centre for Research and Development (grant LIDER/37/0137/L-9/17/NCBR/2018).

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P42

Polymeric systems as carriers for non-steroidal anti-inflammatory active pharmaceutical ingredients

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The aim of modern pharmaceutical technology research is to develop drug carriers to improve their bioavailability. The vast majority of new drug substances are hardly soluble in water, which is a limiting factor for their bioavailability and thus may limit the effectiveness of oral therapy. A promising way to improve drug bioavailability is to use polymeric nanoparticles as drug carriers. Polymeric carriers have been proven to increase drug solubility, improve drug penetration through biological membranes, and can also be used as drug carriers to modify the time or site of action. One of the popular polymers used is PLGA (poly(lactide-co-glycolide)), a biocompatible, non-toxic and biodegradable polymer approved for use as a drug carrier by the FDA (Food and Drug Administration) and EMA (European Medicines Agency). Rheumatoid arthritis (RA) is a chronic, systemic autoimmune disease. It is characterized by inflammation of the synovial membrane in symmetrically located joints. If ineffectively treated, RA is progressive, leading to disability and, in some cases, premature death.[1] One group of medications that relieve the symptoms of the disease are nonsteroidal anti-inflammatory drugs (NSAIDs). They reduce swelling and pain in inflamed joints, leading to improved joint function. NSAIDs are mostly characterized by poor water solubility and thus limited bioavailability. Increasing bioavailability, in turn, makes it possible to reduce the dose of drug needed to achieve an effective analgesic and anti-inflammatory effect, thereby reducing the risk of adverse reactions. The increased efficacy of PLGA-drug combinations over commercially available drug forms in the treatment of ocular inflammation has been confirmed [2,3]. Other drugs (non-NSAIDs) with increased accumulation within the inflamed area have also been studied and prolonged drug release has been obtained for the treatment of inflammatory bowel disease, rheumatoid arthritis and inflammatory lung diseases [4]. The aim of the study was to obtain a drug carrier loaded with a substance from the NSAID group (etoricoxib and sulindac), physicochemical characterization of the PLGA-drug combination and analysis of the release of the substance from the obtained carrier.

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P43

Evaluation of cytotoxic activity of *Scorzonera hispanica* seed extracts against MDA-MB-231 breast cancer cell line

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Scorzonera L. (Asteraceae) is a genus comprising approximately 180 species, spread in the Eurasia region and northern Africa. Species within the genus are reported as sources of various bioactive compounds, including flavonoids, phenolic acid, terpenoids, coumarins and others. Furthermore, *Scorzonera* species are present in folk medicine as remedies for various ailments, including inappetence, inflammation, hypertension, liver, kidney and pulmonary diseases. Aerial and subaerial parts of *S. hispanica* have been previously investigated for the biological activity of their constituents [1]. The study aimed to evaluate the cytotoxic and antiproliferative potential of *S. hispanica* seeds extracts and fractions. Moreover, this study is the first attempt to evaluate the biological activity of *S. hispanica* seed extracts.

The plant material (W. Legutko, batch no. 68347) was grounded and extracted in two separate processes [2]. First, crude methanolic (SH1), 50% methanolic (SH2), water (SH3) and 70% acetonic (SH8) extracts were obtained by sonification. The remaining extracts were obtained by extraction in a Soxhlet apparatus using n-hexane (SHH) and chloroform (SHCI). Then, the plant material was extracted with methanol and 50% methanol under reflux. Combined methanolic extracts were fractioned with solvents of increasing polarity: chloroform (SHCII), diethyl ether (SH4), ethyl acetate (SH5), and *n*-butanol (SH6). The solvents from extracts and fractions were removed, and the residue was suspended in water and freeze-dried. The extracts and fractions were then evaluated for their cytotoxic activity against MDA-MB-231 cancer cells. To assess the cytotoxicity the Carmichael's method with MTT was used. Antiproliferative activity of extracts and fractions were evaluated by measuring the incorporation of radioactive [3H]-thymidine into the DNA of cells.

The study has shown that SHH and SHCII exhibit the most potent cytotoxicity in the MDA-MB-231 cell line (IC_{50} 172.12±6.97 and 306.84±7.47 µg/mL, respectively). Furthermore, the most significant inhibition of [3H]-thymidine incorporation was observed for SHCII and SH8 (IC_{50} 101.84±2.4 and 114.27±4.11 µg/mL, respectively). However, further study needs to be performed to evaluate the phytochemical composition and the mechanism of cytotoxic activity of the analyzed extracts and fractions.

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P44

Unsymmetrical bisacridines target parallel G-quadruplexes, yet not double-stranded DNA

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Imidazoacridinone C-1311 is a potent dsDNA intercalator, which was recommended for Phase II clinical trials for breast and colon cancers. Nitroacridine C-1748 is another antitumor agent, targeting prostate cancers and adenocarcinomas. Although both are acridine derivatives, they exhibited different molecular modes of action and expressed different strengths and weaknesses. But… what if we combined their potential? What could we expect? This idea stands behind a novel and promising class of potential drugs, unsymmetrical bisacridines (UAs). They comprise linked ring systems of imidazoacridinones and nitroacridines, with C-2028 and C-2045 being among several leading compounds, selected for further investigations (Figure 1). They efficiently target pancreatic cancers, which are among the deadliest and most difficult tumors to treat.

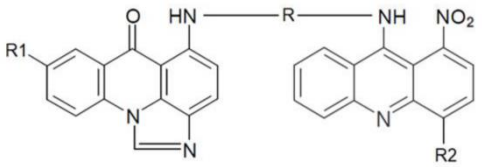

Figure 1. The general structure of UAs. C-2028: R1 = H, R2 = H. C-2045: R1 = OH, R2 = CH_3. In both cases, R = $(CH_2)_3NCH_3(CH_2)_3$.

As one of the oldest principles of science states, 1+1>2 – hence physicochemical and biochemical properties of UAs differ substantially from the sum of mother compounds' characteristics. Although imidazoacridinones are very efficient and sequence-specific dsDNA intercalators, we showed that UAs' interactions with dsDNA are rather weak and unspecific. This effect might be explained by the properties of nitroacridine moieties – their ring systems are not perfectly planar, despite the plain structure might suggest otherwise. Due to their positive charge under physiological pH and below, they are believed to bind to the minor groove of DNA helix. On the other hand, NMR studies have proven that UAs quite efficiently bind to parallel DNA G-quadruplexes – namely c-MYC Pu22 and K-RAS 22RT. Yet, the binding constant to DNA and even the specificity strongly depends on the ligand's structure. For instance, structured 22RT sequence binds both C-2028 and C-2045 at the same ratio, while structured Pu22 binds solely C-2045, utterly ignoring the presence of C-2028. Given the fact that C-2028 and C-2045 differ by only one hydroxyl and one methyl group (Figure 1), this result is currently under our detailed investigation.

Moreover, NMR titration also revealed that G-quadruplexes bind UAs in DNA/drug 1:2 mol/mol stoichiometry. This result – combined with knowledge gained on C-1311's and C-1748's interactions with G-quadruplexes (results not published) – strongly suggested that the imidazoacridinone ring system of the dimer binds to guanine tetrad planes, while nitroacridine moiety choses to reside within one of the grooves.

This study was supported by National Science Centre under grant No 2019/33/B/NZ7/02534.

P45

Design, synthesis, and *in silico* studies of new ciprofloxacin derivatives with potential antiproliferative activity

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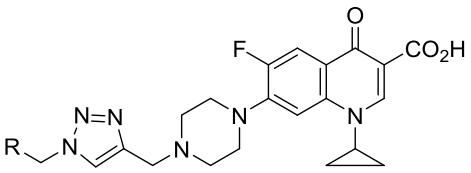
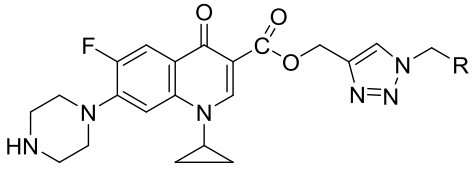
Ciprofloxacin is a second-generation fluoroquinolone, characterized by high antimicrobial activity, a wide range of indications, and rare adverse effects [1]. Despite good antimicrobial activity, ciprofloxacin is also known for its anticancer properties [2-4]. Moreover, the current research indicates that blocking of MITF and Mcl-1 proteins by ciprofloxacin could be considered as a potential target in malignant melanoma treatment [5].



1



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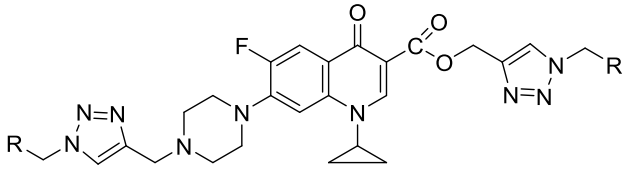

3

The aim of the present study was to discover novel MITF and Mcl-1 inhibitors among novel ciprofloxacin derivatives. From an in-house library of compounds, 120 molecules derived from the ciprofloxacin scaffold were chosen for virtual screening. Based on results of the docking procedure, 20 derivatives of ciprofloxacin were selected for synthesis. Novel MITF and Mcl-1 inhibitors, described by general formula **1**–**3**, were synthesized using the copper(I)-catalyzed azide-alkyne cycloaddition (CuAAC) protocol with $CuSO_4$/sodium ascorbate system as catalyst.

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P46

Aminoalkanol derivatives of 5-chloro-2- or 5-chloro-4-methyl-9*H*-xanthen-9-one and their anticonvulsant activity

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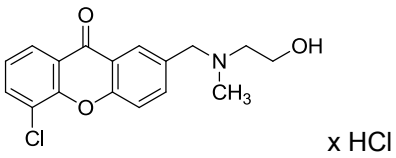
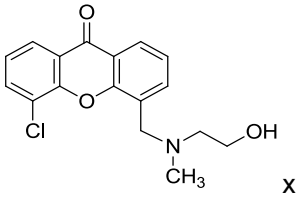
A series of aminoalkanol derivatives of 5-chloro-2-methyl-9*H*-xanthen-9-one or 5-chloro-4- methyl-9*H*-xanthen-9-one was synthetized. The first step of the synthesis was the Ullmann condensation of 2,3-dichlorobenzoic acid with *o*-cresol or *p*-cresol, respectively. In the next stage, the cyclization reaction was carried out in order to close the xanthone ring [1]. The final step was aminolysis of the appropriate bromo-substituted derivative by means of commercially available aminoalkanols, in order to obtain compounds with expected anticonvulsant activity [2]. The products were converted into hydrochlorides. The most active among obtained compounds were structures presented below, tested for: anticonvulsant activity (maximum electroshock seizures MES), toxicity (rotarod), and within pharmacokinetics evaluation - distribution. Crystallography analysis enabled evaluation of conformational analysis.



x HCl

AW18007

MES ED_{50}=42.78 mg/kg (mice, *i.p.*)



x HCl

AW18008

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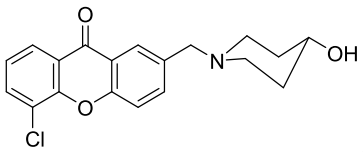
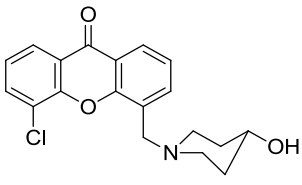


x HCl

AW18012

MES ED_{50}=25.76 mg/kg (mice, *i.p.*)



x HCl

AW18017

MES ED_{50}=52.50 mg/kg (mice, *i.p.*)

Jagiellonian University Collegium Medicum, Faculty of Pharmacy, Department of Organic Chemistry, Department of Bioorganic Chemistry, research financed by the subsidy number: *N42/DBS/000234*

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P47

Synthesis and antioxidant activity of novel diosgenin derivatives

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Diosgenin (a steroidal saponin) is present mainly in the *Dioscoreaceae* plant family, but also in some species of *Solanaceae* and *Fabaceae* families in combination with various sugars in the glycosides form, such as dioscin and gracilin. Diosgenin has been widely used as a starting material for the industrial production of steroid drugs and display various types of pharmacological activity, such as antibacterial, anti–inflammatory, anticancer and hypocholesterolemic [1][2]. It is therefore expected to become the key building block of new drugs.

A serie of novel diosgenin derivatives with steroid aglycone linked to various acids by an ester bond at the C3–oxygen atom of the steroid skeleton has been synthesized. Diosgenyl esters have been prepared by the esterification reaction (DCC/DMAP) of diosgenin with the corresponding acids. An in silico ADME (properties, absorption, distribution, metabolism, excretion) study was also performed to predict the pharmacokinetic profile of the synthesized compounds.

The obtained compounds were further tested for their antioxidant activity in an oil-in-water emulsion system. It was shown that PABA-Dios exhibited the highest activity. Moreover, in our study we also used human red blood cells to predict the cellular response to our derivatives. All synthesized compounds, except Dios-O-4-OH-Benz, displayed no toxicity, and can therefore be used in the pharmaceutical or cosmetic industry.

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P48

Anticancer and pharmacokinetic studies of 3,6-diazaphenothiazine

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The asymptomatic and high resistance of various line of cancer cell lines results poor treatment outcome and high mortality rate. Unfortunately the fundamental chemotherapy give the promising anticancer activities but associated with severe side effects. Therefore is need to discover and develop new more potent antitumor agents with better selectivity and reduced side effects [1].

Phenothiazines exhibited promising anticancer activity against several cancer cell line (breast, ovarian, lung, colorectal, prostate, leukemia, melanoma, and renal) [2,3]. Among of the diazaphenothiazines, 10*H*-3,6-diazaphenothiazine had been reported effective in killing breast cancer cells, glioblastoma, melanoma and ovarian cancer cell line instead less toxic towards normal human fibroblast cells [4]. This compound induces apoptosis through upregulation of pro-apoptotic genes such as BAX, p53 and CDKN1A and downregulate anti-apoptotic gene such as Bcl-2 and H3. Additionally the apoptosis studies suggested inducing apoptosis on A2780 cancer cells *via* mitochondrial-dependent and cell death receptor-dependent pathway by increased activities of caspase-9, caspase-8, caspase-10 and caspase-2, together with the downstream caspases, the caspase-6, caspase-3 and caspase-7.

Based on these promising results above, further studies were performed on the human lung carcinoma (A549) and non-small lung carcinoma (H1299) showing promising results. Using a 72 h MTT viability assay (Promega) we have observed a strong cytotoxic activity of tested compound, and calculated IC_{50} [17 μM] showed promising results in both cancer cell lines.

At the same time, preliminary pharmacokinetic studies were performed related to the possibility of binding the studied molecule with human serum albumin.

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P49

β-arrestin recruitment by histamine H_4R ligands from the group of analogues of JNJ7777120.

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Histamine H_4 receptor (H_4R), considered as potential drug target for treatment of inflammatory and immunological diseases, is a Gi-coupled receptor that can independently activate β-arrestin-2 signaling pathway[1,2].Having the regard of the varied response of the H_4R ligands in different native and culture cells, potential H_4R ligands should be tested for their $G_{\alpha i}$ as well as β-arrestin-2 activity [3]. JNJ77777120 – former "standard" H_4R antagonist was reported as neutral antagonist and inverse antagonist in $G_{\alpha i}$ dependent pathway [4-6] while partial agonist in β-arrestin pathway [7].

For testing beta-arrestin activity we have chosen and synthesized three H_4R ligands described in the literature (JNJ7777120,JNJ10181584 and Compound 75)[4-7] and their sulphur analogues synthesized in our Department (JSJ-Cl, MK-6 and MK-9). Compounds were tested in agonist and antagonist mode of the β-arrestin recruitment assay. Results, shown as IC_{50}, EC_{50} and % of maximal response, were compared.

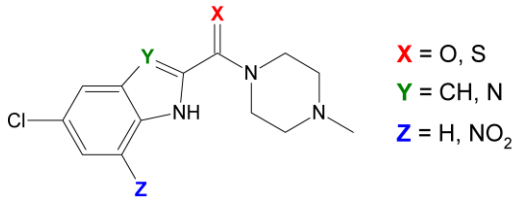

Figure 1. Scheme of the structure of tested compounds.

4 compounds showed some beta-arrestin activity in agonist mode, 5 in antagonist mode of the assay. One compound was inactive in both modes. We also observed differences in activity between ketones and thioketones.

β-arrestin recruitment was tested in LiveBLAzer™cell based assay (using Tango-H4-bla U2OS cells and LiveBLAzer™ technology.

The research was partially founded by Polish National Science Center grant Preludium no *DEC-2016/23/N/NZ7/01906* and byJagiellonian University Medical College grants no. *N42/DBS/000196, N42/DBS/000039*.

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P50

Design, synthesis and biological activity of new 3-acetyl-2,5-disubstituted-1,3,4-oxadiazoline derivatives.

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Antibiotics are one of the most important groups of medicines and their invention is considered as one of the most significant discovery in medicine. Without them, it would be impossible to treat bacterial infections as well as perform medical procedures such as surgeries or transplantations [1]. According to scientists and medical practitioners, the growing resistance of bacteria to antibiotics is a serious threat to the life and health of modern society. Due to this, it becomes necessary to search for novel molecules that will better cope with bacterial resistance and at the same time show less toxicity [2].

Our literature review showed that hydrazones obtained from carboxylic acids are interesting chemical compounds with high antimicrobial activity [3]. This group of compounds display also anti-inflammatory, anticancer and anticonvulsant properties and can be easily transformed into 3-acetyl-2,5-disubstituted-1,3,4-oxadiazolines [3]. The 1,3,4-oxadiazoline derivatives also show biological activity, especially antibacterial, antitubercular, antifungal and antitumor properties [4].

In our research, we used nicotinic acid as a starting compound, which was subjected to a series of condensation reactions with appropriate aldehydes. As a result of these reactions, we were able to obtain a series of 13 compounds, two of them showed promising activity against Gram-positive bacteria MIC = 1.95-15.62 µg/ml, especially against *Staphylococcus epidermidis* ATCC 12228 MIC = 1.95 µg/ml. Then, we subjected the entire series to a cyclization reaction in the acetic anhydride, thanks to which we were able to obtained 13 new 3-acetyl-2,5-disubstituted-1,3,4-oxadiazoline derivatives. Obtained 1,3,4-oxadiazolines were also tested for antimicrobial activity. The results showed high activity of the compound with a 5-nitrofuran substituent, which was active against all tested strains. The greatest activity was found against Gram-positive bacteria, in particular against *Bacillus subtilis* ATCC 6633 and *Staphylococcus aureus* ATCC 6538 (MIC = 7.81 µg/ml).

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P51

New, potent and selective butyrylcholinesterase inhibitors as promising anti-AD agents

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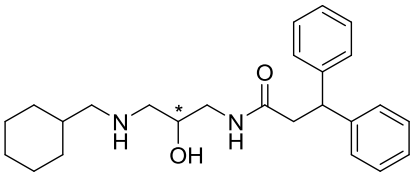
Forgetting is the main symptom of Alzheimer's disease (AD) – a progessive neurodegenerative disorder affecting millions people around the world. This memory loss is mainly caused by disturbances observed in cholinergic neurotransmission. Butyrylcholinesterase (BuChE) is one of the crucial enzymes responsible for maintaining of this homeostasis. BuChE is a serine hydrolase catalyzing the hydrolysis of choline and non-choline esters. For many years, the role of BuChE in the cholinergic neurotransmission was considered as irrelevant. Nowodays, more and more often scientists empathize its role in the hydrolysis of acetylcholine (ACh) - a neurotransmitter linked to cognitive functions when acetylcholinesterase (AChE) is absent or insufficient. It is observed that in the progression of AD the level of BuChE significantly increases (120% of normal values) and thus, the activity of AChE is reduced [1]. Therefore, BuChE should be considered as potential, biological target in the search of new anti-AD drugs.

Based on our previous results [2,3], we designed and synthetized a novel series of selective BuChE inhibitors containg 2-hydroxyaminopropyl scaffold. Among them, we identified cyclohexyl derivative **16** with a high inhibitory potency against *h*BuChE (IC_{50} = 68 nM) and optimal physicochemical properties reflecting in druglikeness. We performed a resolution of racemic mixture to pure enantiomers and determined the influence of chirality on the biological properties. We obtained the crystal of *h*BuChE with inhibitor **16** to study crucial ligand-protein interactions in the active site. Moreover, we conducted preclinical studies including ADME-tox *in vitro* assessment, as well as pharmacokinetic and pharmacodynamic *in vivo* studies.

Acknowledgments: This work was supported by JUMC grant N42/DBS/000061.



Compound 16
*h*BuChE IC_{50} = 68 nM

Fig 1. The structure of compound 16.

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P52

Benzophenone-3 and 4-Methylbenzylidene camphor biodegradation in *Cunninghamella species*

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Recently UV filters have been identified as important contaminants of the aquatic environment [1, 2]. Therefore, their environmental fate, degradation and removal are of great importance. Many environmental microorganisms including fungi play a key role in the bioconversion and biodegradation of organic contaminants [3, 4]. Filamentous fungi belonging to the genus of *Cunninghamella* found in soil and plant material are well known for their ability to metabolize various xenobiotics including pollutants [5-7].

In the present study microbial transformation of Benzophenone-3 (BP-3) and 4-Methylbenzylidene camphor (4-MBC), two *UV filters frequently used* in sunscreen formulations was carried out by using different strains of *Cunninghamella* species (*C. echinulata, C. blakesleeana,* and *C. elegans*). Biotransformation processes *were carried out* for 7 days. The progress of biotransformation was monitored using liquid chromatography coupled with tandem mass spectrometry (LC-MS/MS).

Biodegradation of BP-3 yielded four different products (P1-P4), all of which were detected with strain *C. blakesleeana*. As regards two other species, only the main product (P1) was identified. P1 was obtained through *O*-demethylation and hydroxylation of the aromatic ring with subsequent oxygenation to *orto*-quinone. The study indicated that *C. echinulata* and *C. blakesleeana* were able to degrade 4-MBC in a similar pattern giving seven degradation products (P1-P7), whereas in case of *C. elegans* five biodegradation products were identified. The major intermediate P1 was performed by the aliphatic carbonylation reaction.

To sum up, the present study demonstrated the ability of the fungal strains *Cunninghamella* to degrade pollutants such as some commercially used UV filters.

Acknowledgements The project was supported by the National Science Center Grant No 2020/37/B/NZ7/02546.

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P53

Isomeric activity cliffs (iAC) – a case study for the fluorine substitution for aminergic GPCRs

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Nowadays, almost 700 unique human proteins are drug targets, of which five of the most druggable target classes are: G protein-coupled receptors (GPCRs), ion channels, kinases, enzymes, and transporters. In turn, GPCRs are a target for 34% of the global market share of therapeutic drugs [1,2], with aggregated sales for 2011–2015 of ~US$890 billion [3]. Considering the therapeutic potential of GPCRs and fluorine introduction as one of the most frequently used modifications of the lead structure to improve its biological activity [4], we examined fluorine substituent in the context of a new approach of isomeric activity cliffs (*i*ACs). An activity cliff is generally defined as a pair of structurally similar compounds having a large difference in potency.

Herein, we report an analysis of fluorinated aminergic GPCR ligands in ChEMBL database. Ligand sets (F_iSAR) of pairs that differ only in the substitution site of fluorine atom(s) were determined and its potency variation was analyzed between different isomeric analogs found. In addition, substructure relationships between ligands pairs/sets were calculated (MMP) and extended the underlying structure-activity relationships (SARs).

An introduced new type of activity cliffs approach (*i*ACs) might support the rational introduction of the fluorine atom into molecules to improve its biological affinity. The analysis of the ChEMBL database showed that for aminergic GPCRs there are 931 sets of F_iSAR containing almost 2300 fluorinated compounds. However, only 33 compound sets had a 50-fold difference in biological activity (ΔpPot +/– 1.7) and thus represented *i*ACs. The most fluorinated compounds (over 200 per target) were found for the dopamine D_2 receptor (112 sets) and serotonin receptors $5HT_{1a}$ and $5HT_{2a}$ (102 and 97 sets, respectively), which proves the important role of fluorine in these receptors in a broad range of chemotypes. The receptors with the lowest or no fluorinated ligands are the histamine H_2 receptor (6 compounds), the alpha-2b adrenergic receptor (5 compounds), and the serotonin 5-HT_4 and 5-HT_{1e} receptors (4, 0 compounds, respectively).

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P54

N-Benzyl-3-aryl-4-(diethoxyphosphoryl)azetidin-2-ones as antibiotic enhancers

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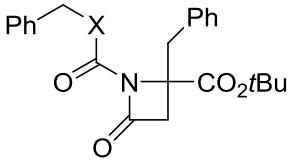
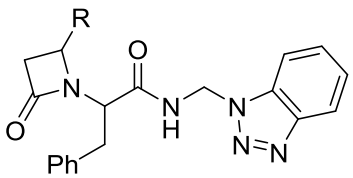
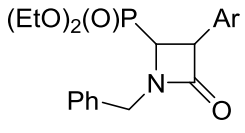
The compounds containing azetidinone are of special importance both in chemistry and medicine. Since the discovery of penicillin, the application of azetidinone derivatives has been mainly associated with their antibacterial activity. Azetidinone ring is also a common structural motif of a vast number of compounds possessing a wide range of other biological properties, including antiviral activity. For example, non-nucleoside analogues of azetidinone **1** exhibited activity towards HCMV [1]. On the other hand, compounds **2** have been recognized as inhibitors of HIV-1 protease [2]. The search for effective antiviral drugs, among newly designed compounds as well as already known ones, became even more challenging in the eyes of the coronavirus pandemic. In fact, symptomatic therapy is appropriate in the treatment of milder illnesses, and antimicrobial drugs are often necessary when bacterial complications occur. Especially, the methicillin resistant *Staphylococcus aureus* (MRSA) is a Gram-positive member of the most problematic bacteria in clinical treatment. Various clinical isolates of MRSA are multidrug resistant (MDR), *i.e.* resistant to antibiotics representing different classes, including β-lactams. Taking into account the structural analogy between β-lactam antibiotics and functionalized derivatives of azetidinones, the second ones provide some hope in search for effective agents against MRSA, either as new antibacterials less susceptible to MDR mechanisms or as antibiotic "adjuvants" that, being bioisosters of antibiotics, may be mistakenly recognized as substrates of various bacterial MDR proteins. We designed a new series of compounds of general formula **3** and their antimicrobial properties were assayed. Studies on docking and molecular dynamic simulations for the most active compound were also undertaken.



1 X = O, NH



2 R = (CO)NHCH(Bn)(COOMe), COOBn



3

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P55

Synthesis of new Cu(II) complexes of thiosemicarbazide derivatives with potential biological activity

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Copper ions are able to form complex bonds with various ligands, including organic ones. Currently, a number of studies are carried out on coordination connections of ligands with the copper atom, which result in innovative results and an increase in biological activity [1]. Many copper complexes have been found to exhibit potent antitumor activity, especially combinations with the S, N, O-donor system [2]. The antiproliferative activity of Cu(II) complex compounds is sometimes higher than that of ligands [3].

In our experiment, copper (II) complexes with thiosemicarbazide derivatives were synthesized as potential anticancer agents. 1-(pyridin-2-,3-,4-yl)acetyl-4-substituted thiosemicarbazide were used as substrates for the planned syntheses. The obtained connections were characterized by chemical and elemental analysis, FTIR spectroscopy and the TGA method. All obtained complexes are stable in air at room temperature. The nature of metal-ligand coordination has been studied. The FTIR spectra of the obtained coordination compounds show several absorption bands. Analysis of these spectra suggests that in all cases the organic ligands coordinate with the copper (II) ion. The thermal behavior of all complexes was tested using the TG, DTG and DTA methods under dynamic flow conditions in the air atmosphere. The shape of the TG curves, solid intermediates and end products clearly indicate the preparation of coordination compounds. The decomposition of the synthesized complexes is a multistage process.

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P56

Does polymorphism of the β_2-adrenergic receptor influence its interactions with lipid bilayer and G_s protein?

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The gene encoding the β_2-adrenergic receptor (β_2-AR) is extremely polymorphic. It has been shown that to certain types of polymorphisms are correlated with some clinical features of asthma, including airways reactivity, whereas the influence of other is not yet understood. Among polymorphisms affecting amino acids at positions 16, 27, 34, 164 and 220, the latter three are present in crystal structure of β_2-AR, which facilitate studying them by means of molecular dynamics simulations. The current study was focused on investigating to what extent the three polymorphisms of β_2-AR (i.e. Val34Met, Thr164Ile and Ser220Cys) affect the interaction of β_2-AR with its natural molecular environment which includes: lipid bilayer (in the case of all three cases) and G_s protein (which participates in β_2-AR-mediated signaling; in the case of Ser220Cys). We have designed and carried out a series of molecular dynamics simulations at different level of resolution (i.e. either coarse-grained or atomistic simulations), accompanied by thermodynamic integration protocol, in order to identify potential polymorphism-induced alterations in structural, conformational or energetic features of β_2-AR features. The results indicate the lack of significant differences in the case of magnitude of the β_2-AR-lipid bilayer interactions but also some non-negligible differences when considering the strength of β_2-AR-G_s protein binding. The observed alterations are discussed in the context of their significance for β_2-AR-mediated signaling.

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P57

New 3-(3-benzenesulfonylguanidinyl)thiourea derivatives with activity against methicillin-resistant *Staphylococcus spp.*

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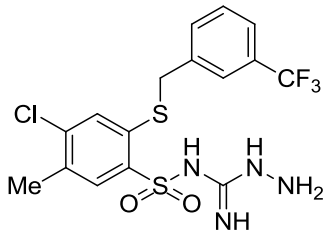
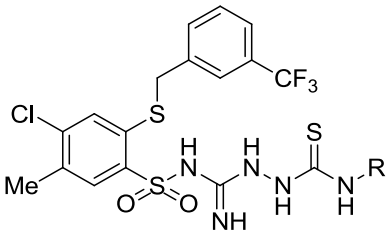
Methicilin-resistant staph, especially MRSA, has become major problems of modern epidemiology and chemotherapy. One of the methods of combating the growing resistance of bacterial strains is the search for new antibacterial agents. The numerous studies prove that the novel class of promising compounds with activity against *Staphylococcus spp.*, including MRSA, comprises derivatives with thiosemicarbazide fragment [1-2]. This encourages us to the incorporation of the mentioned structural element to the benzensulfonamide skeleton. As a result, a series of new 3-(3-benzenesulfonylguanidinyl)thiourea derivatives were synthesized in the nucleophilic addition of *N*-amino-*N'*-{4-chloro-5-methyl-2-[(3-trifluoromethylphenyl)methylthio]benzenesulfonyl}guanidine with appropriate isothiocyanates with variable R substituents.



1



2-6

Compd	R
2	Ph
3	4-Cl-Ph
4	4-F-Ph
5	3,4-diCl-Ph
6	iPr

The obtained derivatives **2-6** were tested for antimicrobial activity against *S. aureus* and *S. epidermidis*, including MRSA and MRSE strains, using microdilution assay. All compounds displayed significant both bacteriostatic and bactericidal activity toward non-resistant strains (MIC in the range of 1.56 – 12.5 µg/ml; MBC in the range of 3.125 – 25 µg/ml for *S. aureus* and S*. epidermidis*). With regard to the methicillin-resistant strains, the effect was observed only for compound **3** (MIC = 3.125 µg/ml against MRSA and MRSE, MBC = 3.125 and 6.25 µg/ml for MRSA and MRSE, respectively), however derivative **5** was bacteriostatic against MRSA (MIC = 3.125 µg/ml).

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P58

Synthesis and antimicrobial potential of new compounds from hydrazide-hydrazone group

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Searching for novel effective and non-toxic antimicrobial agents is nowadays especially important due to the increase of bacterial strains resistant to antibiotics and chemotherapeutics [1, 2].

Among various classes of organic compounds hydrazide-hydrazones play important role as potential antimicrobial agents [3]. Many scientific reports focus their attention also on 5-nitrofuran derivatives due to their promising biological properties [4]. In this study we decided to synthesize and perform *in vitro* antimicrobial activity assays of novel hydrazide-hydrazones of 5-nitrofuran-2-carboxylic acid [5].

New compounds were synthesized in two step reaction. In the first stage of the synthesis methyl ester of 5-nitrofuran-2-carboxylic acid was transformed into hydrazide in the reaction with hydrazine hydrate. In the second step obtained hydrazide was subjected to condensation reaction with diverse aldehydes to obtain novel hydrazide-hydrazones of 5-nitrofuran-2-carboxylic acid. The chemical structure of all obtained compounds was established on the basis of spectral methods [5].

Newly synthesized substances were tested for *in vitro* antimicrobial activity against Gram-positive and Gram-negative bacterial strains and fungi belonging to *Candida* spp. Synthesized compounds displayed significant antibacterial activity especially against Gram-positive bacteria like *Staphylococcus* spp. and *Bacillus* spp. (MIC = 0.48–15.62 μg/ml and MBC = 0.98–62.5 μg/ml, MBC/MIC = 1–4), what confirmed that hydrazide-hydrazones of 5-nitrofuran-2-carboxylic acid may be regarded as potential antimicrobial agents [5].

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P59

Photostability studies of novel melanogenesis inhibitors from the group of cinnamic acid derivatives

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One of the most common dermatological problems constitute skin hyperpigmentation disorders. The methods of their treatment include among others the use of topical formulations containing ingredients with melanogenesis inhibitory activity such as kojic acid or arbutin. These compounds have many limitations such as low efficacy and adverse effects like hyperactivity reactions. Moreover, kojic acid similarly to benzophenone derivatives, parabens and triclosan was identified as a very high-risk cosmetic ingredient with potential endocrine disrupting properties [1]. This prompted us to search for novel melanogenesis inhibitors. Cinnamic acid derivatives are widely used in cosmetic products as UV filters, UV absorbers, skin and hair conditioners, antioxidants, perfuming, masking, and antimicrobial ingredients [2] thus we focused on this group of compounds.

The aim of the study was to evaluate photostability of six *trans*-cinnamic acid derivatives demonstrating promising melanogenesis inhibitory activity. It is a very important study, because tested compounds are intended for potential application to the skin, especially facial skin, which is the most exposed to the ultraviolet radiation. The photostability of the tested compounds was investigated in ethanol solutions by the measure of the changes in the shape of ultraviolet absorption curves. Additionally LC-MS analysis was performed. Irradiation of samples was conducted with solar light simulator (Suntest CPS+, Atlas) at 500 W/m^2 for 5 to 120 minutes (cumulative dose of ultraviolet radiation from 18 to 436 kJ/m^2). The absorption spectra and the absorption value at the peak were recorded at eight time points of irradiation.

Upon solar light exposure the changes in the ultraviolet absorption curves such as hypsochromic shift and hypochromic effect were observed. The analysis of chromatograms and mass spectra of compounds obtained after irradiation suggests that the changes in absorption curves result from *trans-cis* photoisomerisation processes taking place in solution. There were not observed any products of photodegradation. The photoreaction was fast during the first 20 minutes of irradiation, but it slowed down as the exposure time increased. It indicates that the ratio of *trans* and *cis* isomers in solution achieved the photostationary state [3]. Regardnig that photoisomerisation is the main photochemical reaction of *trans*-cinnamate derivatives and is considered to be a very efficient way of dispersing the absorbed energy, all tested compounds may be identified as photostable. Considering that both isomers may have different activity and toxicity it is worth to extend the activity and safety assessment by testing the mixture of isomers.

The research was financed by National Centre for Research and Development within LIDER XI program (contract number *LIDER/26/0094/L-11/19/NCBR/2020*).

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P60

Quantification of gasotransmitter, hydrogen sulfide, in brain and liver tissue using LC-MS/MS technique

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Hydrogen sulfide (H_2S) is a small molecule that is, together with NO and CO, termed a gas transmitter. It exists widely in mammalian tissues, where plays an important role in numerous physiological and pathological processes. H_2S acts as an intracellular messenger, neuromodulator, blood vessel relaxant as well as regulator of inflammation and apoptosis. It is an important compound in cellular energy metabolism in the peripheral tissues and in the brain. Recent studies indicate that the regulation of its level in tissues may have a therapeutic potential. The precise determination of H_2S level in biological matrices is therefore an important analytical challenge.

The aim of this study was to develop the liquid chromatography-mass spectrometry (LC-MS/MS) method for the determination of free and dithiothreitol DTT-released forms of H_2S in brain and liver homogenates by quantification of a derivative of hydrogen sulfide and monobromobimane named sulfide dibimane (SDB) in aerobic conditions. The derivatization process was carried out in the dark at 20 °C in TRIS-HCl buffer (pH=8.5) for 60 min. The chromatographic separation of SDB was performed on the Hypersil GoldTM C18 analytical column (2.1 × 50 mm, 3 µm; Thermo Scientific, USA) with a gradient elution using a mobile phase containing 0.1% (v/v) of formic acid in acetonitrile and in water. Exion LC AC HPLC was coupled with a Sciex QTRAP 4500 triple-quadrupole mass spectrometer (both from Danaher Corporation, Washington, DC, USA). Electrospray ionization (ESI) in the positive ion mode was used for ion production. The mass spectrometer was operated at unit resolution in the selected reaction monitoring mode, monitoring the transition of the protonated molecular ions m/z 415–193 (CE = 25 eV) and m/z 415–223 (CE = 31 eV) for SDB and m/z 436–235 (CE = 42 eV) for the valsartan (internal standard;IS). The calibration curve was prepared by spiking tissue homogenate supernatant with the standard working solution of SDB (synthetized in-house) and was linear in the range from 50 to 3200 nM. The method was selective, sensitive and had satisfactory precision and accuracy.

This method has been successfully used to measure free and DTT-released forms of H_2S levels in the selected biological matrices derived from two animal models of diseases, namely brain ischemia model (MCAO, middle cerebral artery occlusion) and concanavalin A induced hepatitis.

P61

Evaluation of biological activity of novel selenoesters in MCF-7 human breast cancer cells

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There is a continuous increase in the number of breast cancer cases that is one of the most popular neoplasms worldwide. For this reason, there is a need to develop compounds that are more efficient and selective against tumor cells than those commonly used. Recently, seleno-organic compounds are an interesting group of substances in this field. Many studies have shown that selenium compounds exhibit anticancer activity that, among other things, includes induction of apoptosis and oxidative stress, DNA fragmentation, inhibition of angiogenesis and cell proliferation, decrease in protein kinase C (PKC) and NF-κB expression, or arrest cells in the G_1 and G_2/M phase of the cell cycle [1,2]. It was also found that some of the seleno-organic compounds can sensitize cancer cells to commonly used therapeutic methods (chemotherapy, radiotherapy) and increase their effectiveness while decreasing their side effects [1].

Therefore, we attempted to evaluate the biological activity of novel selenoesters (EDAG-1, -7, -8, -10, EDA-71, and E-NS-4) against MCF-7 breast cancer cells and MCF-10A normal breast epithelial cells. In the first step, the cytotoxicity of these compounds was tested by MTT assay according to Carmichael's method using tetrazolium salt. All tested compounds had cytotoxic activity against breast cancer cells, but EDAG-1 and EDAG-8 exhibited the strongest properties (0.47 ± 0.06 and 0.61 ± 0.05 μM, respectively) and these compounds were used in further studies. The next steps included cytometric analysis of the effects of selected selenoesters on apoptosis induction and cell cycle arrest in breast cancer cells using annexin V/propidium iodide and propidium iodide, respectively.

We observed that the high cytotoxic activity of the tested compounds against breast cancer cells was related to the strong apoptotic induction. Furthermore, it was found that EDAG-1 arrested cells in the G_1 phase of the cell cycle, while EDAG-8 arrested cells in the G_2/M phase. These results suggest that the high biological activity of these selenoesters can be related to cell cycle arrest in breast cancer cells.

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P62

Investigation of the mechanism of selective functionality of 5-HT$_7$ receptor ligands by in silico methods

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The GPCRs proteins may exist in many conformational states which are stabilized by their ligands. Many ligands of GPCR proteins stabilize one of many conformational states of these receptors which may selectively activate the G protein pathway of signaling or other different from G protein pathways e.g. connected with β-arrestin. That phenomenon is called functional selectivity. What is important, the activation of different signaling pathways causes different biological responses. The occurrence of various signaling proteins dependent on GPCR receptors allows designing ligands to stabilize the receptor conformations that preferentially activate one of the signaling proteins, which opens the possibility of designing drugs activating selected signaling pathways.

We have created a hybrid model of the 5-HT$_7$R with G protein using the GPCRdb service and the structures present in the PDB databank. The system was immersed in the cholesterol-containing lipid bilayer to best reflect the real conditions. The system was relaxed by performing 100 ns molecular dynamics simulations. The created system allows for observation of the interaction between the Gα subunit and the intracellular part of the receptor.

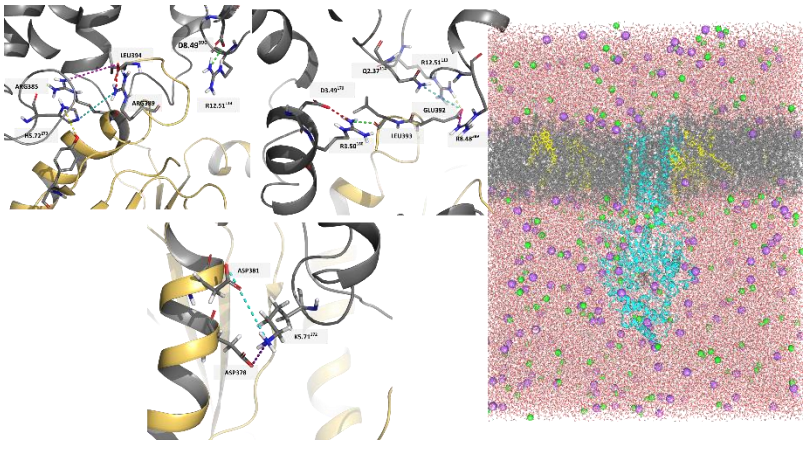

Figure 2. Visualization of the system and key receptor-modulator interactions

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P63

Hydroxycobalamin(*c*-lactam) as an inducer of cobalamin deficiency in melanoma cells

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Cobalamin (vitamin B12) has an important role in cellular functioning, especially in DNA synthesis, methylation, and mitochondrial metabolism. Its depletion may result in genome instability and an accumulation of methylmalonic acid and homocysteine (Hcy) [1]. Much is now known about the biochemistry of vitamin B12, however, the effect of its deficiency on homeostasis of particular cell types is still unclear. This is due to the difficulty of obtaining a suitable experimental model.

Cobalamin analogues with a modification of the amide group present at the *c*-position of B pyrrolic ring were demonstrated to be an efficient antagonists of the vitamin because the use of these agents in various biological systems (experimental animals, cell lines) resulted in an inhibition of cobalamin-dependent enzymes [2]. Previously we demonstrated that the use of the vitamin B12 antagonist hydroxycobalamin(*c*-lactam), abbreviated as HCCL, may be an effective method of inducing hypocobalaminemia *in cellulo* [3,4]. Nonetheless, to obtain the powerful experimental model, the conditions of incubation with the agent must be optimized and adapted to a specific cell type.

The aim of this study was to develop an *in vitro* model of hypocobalaminemia in melanoma cells. The experiments were performed on human melanoma cell line C32 treated with HCCL. The study included evaluation of cell proliferation and morphology, determination of the hypocobalaminemia marker – Hcy, and cell cycle assay. Analysis was performed using an immunoenzymatic test and the fluorescence image cytometer NuceloCounter NC-3000.

Based on the obtained results, we revealed that HCCL at a concentration of 100 µg/ml induced cobalamin deficiency in tested melanoma cells after 14 days of incubation, as indicated by a significant increase in the level of homocysteine - a marker of cobalamin deficiency. A combined treatment of melanoma cells with HCCL and vitamin B12 resulted in a complete antagonization of the HCCL-induced HCY accumulation. We also observed that the addition of vitamin B12 to the culture medium contributed to an increase of the number of cells as compared to the control, which proves its proproliferative properties toward tested melanoma cells. However, it was shown that HCCL did not affect melanoma cells proliferation and cell cycle progression in the presented experimental model.

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P64

Synthesis of novel oxazolo[5,4-*d*]pyrimidines and study of their *in vitro* cytotoxicity against several cancer cell lines

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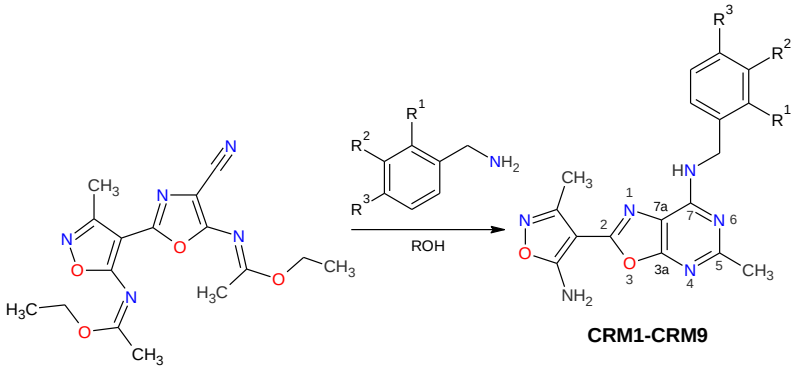
Cancer is a disease characterized by uncontrolled cell growth with loss of differentiation, which becomes a major cause of human death throughout the world [1]. In anticancer treatment, the oxazolo[5,4-*d*]pyrimidine system represents an important pharmacophore due to its structural similarity to purine bases. Oxazolo[5,4-*d*]pyrimidine derivatives in the biological evaluation were found to inhibit the growth of selected cancer cell lines [2] and angiogenesis, which is a process that actively supports cancer growth and spread [3].



CRM1-CRM9

Previously, we reported the synthesis and *in vitro* studies of a series of 7-aminooxazolo[5,4-*d*]pyrimidine derivatives showing immunosuppressive, antiviral and anticancer activity [4]. To explore the structure/activity relationship, novel 7-aminooxazolo[5,4-*d*]pyrimidines with differently substituted 7-*N*-benzyl groups (**CRM1**-**CRM9**) were designed and synthesized. The structures of all new compounds were confirmed using elemental analysis and various spectroscopic technics such as HR-MS, IR, ^{1}H and ^{13}C NMR spectroscopy. Nine derivatives (**CRM1**-**CRM9**) were evaluated for their cytotoxicity against three human cancer cell lines: A549 (adenocarcinoma of the lung), HT-29 (adenocarcinoma of the colorectal), A375 (melanoma cell line), and the normal cell line: NHDF (Normal Human Dermal Fibroblasts). Two compounds, i.e. **CRM6** and **CRM8** appeared to have the most potent cytotoxic activity against the tested cancer lines.

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P65

Analytical estimation of cefepime photodegradation process

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The primary source of drug contamination in water is the pharmaceutical industry, hospitals, veterinary clinics and households. Animal husbandry poses a high risk of contamination with medicinal substances, especially antibiotics and their metabolites. The use of prophylactic doses of antibiotics in poultry, cattle and pig farming is much greater than for therapeutic purposes. Fertilizing fields with animal excrements, which contain a very high concentration of antibiotics in their original form, leads to their presence in the natural environment. Landfills and poorly disposed of overdue drugs may be an additional source of antibiotics in surface and groundwater.

Pharmaceuticals that are more difficult to decompose in water may accumulate in aquatic ecosystems, which leads to increased toxicity to fish and microorganisms. They can also penetrate into groundwater and are presented in drinking water, which poses a risk to human health. The highest their concentrations are recorded in post-production sewage from pharmaceutical companies. The most commonly found drugs in surface waters are non-steroidal anti-inflammatory drugs (NSAIDs), estrogens, lipid-regulating drugs and antibiotics. In these environments, biologically active substances undergo biodegradation at different rates (from several minutes to several dozen days).

The process of photodegradation under the influence of UV rays is of great importance in the biodegradation of antibiotics in surface waters. Based on the above information, extensive analyzes are needed to comprehensively assess the risks associated with the potential toxicity and ecotoxicity of drugs. The activities undertaken as part of the work will complement the panel of research on the fourth generation cephalosporin - cefepime, including issues related to remediation.

In the first stage of the research, a qualitative and quantitative analysis (using the validated UPLC-MS/MS method) of the cefepime phototransformation process in aqueous solutions with the use of simulated solar radiation was carried out. The irradiation time, necessary to decompose half of the initial concentration of the tested active substance, was determined, as well as potential photodegradation products. Based on the obtained results, further analyzes will be undertaken to assess the environmental toxicity and potential mutagenicity of the obtained products.

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P66

Analysis of retention parameters of selected NSAIDs by TLC method using phases with different properties

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The distribution of a compound in a living system can be viewed as a series of division steps, coupled with diffusion across several regions. The affinity of a molecule for biological membranes and its biological activity can be represented by its lipophilicity. The lipophilicity of a compound is one of the essential physicochemical properties affecting drug absorption, distribution, metabolism and elimination (ADME). According to the International Union of Pure and Applied Chemistry (IUPAC), this property represents affinity molecules or moieties for a lipophilic environment. It is commonly measured by assessing the distribution behavior of compounds in two-phase systems. In most cases, this feature regulates the ability of active substances to pass through biological membranes and thus assessing the effectiveness of a drug in biological environment. Knowledge of important pharmacokinetic properties, which largely determine bioavailability, contributes to the optimization of the chemical synthesis of the target molecule and its biological activity, with significant financial savings and acceleration of the development of a potential drug.

The interpretation and predictability of chromatographic retention-biological activity models is supported by the fact that the separation of substances in the chromatographic system may correspond to that of the solute on lipid bilayer biological membranes. Various parameters (e.g. steric, polarity) significantly affect the retention of a chemical, so it is not possible to rely only on the calculated lipophilicity values. As is known, many factors other than just hydrophobic forces are involved in this mechanism. The choice of the appropriate stationary and mobile phase for the determination of lipophilicity can significantly influence the obtained values of the chromatographic parameters.

Retention properties of selected drugs from the NSAIDs group were examined by TLC chromatography. As stationary phases, adsorbents with different properties were used, both RP-silica gel and diol or amine phases. The influence of the chemical structure of substances and used adsorbent on their retention was examined. The established chromatographic retention parameter, RM0, for each compound was correlated with software-calculated partition coefficients, as a frequently used measure of lipophilicity (especially in the analysis of new compounds, drug candidates). The obtained results indicate that the applied conditions and the nature of compounds influence their retention behavior.

P67

Synthesis of novel 1,2,4-triazole-3-thione-derived *N*-Mannich bases with potential pharmacological activity

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1,2,4-triazole-3-thione and their derivatives belong to a class of exceptionally active compounds possessing a wide spectrum of biological properties including antioxidant, anti-inflammatory, analgesic, antibacterial and cytotoxic activity [1]. Moreover, a large number of Mannich bases have been synthesized and evaluated as anticancer and antimicrobials agents as well as enzyme inhibitors, e.g. COX inhibitors [2]. Milošev et al. prepared 1,2,4-triazole-3-thione *N*-Mannich bases to study their cytotoxic activity. The results clearly showed that the new compounds exhibited good cytotoxic activity against cancer cell lines and low toxic effects toward normal cells [3].

Taking into account the above information and continuing our studies on the identification of novel biologically active molecules, we decided to obtain new 1,2,4-triazole-3-thione derivatives containing dimethylpyridine and phenylpiperazine moieties, presented by the general structure in the Figure 1.

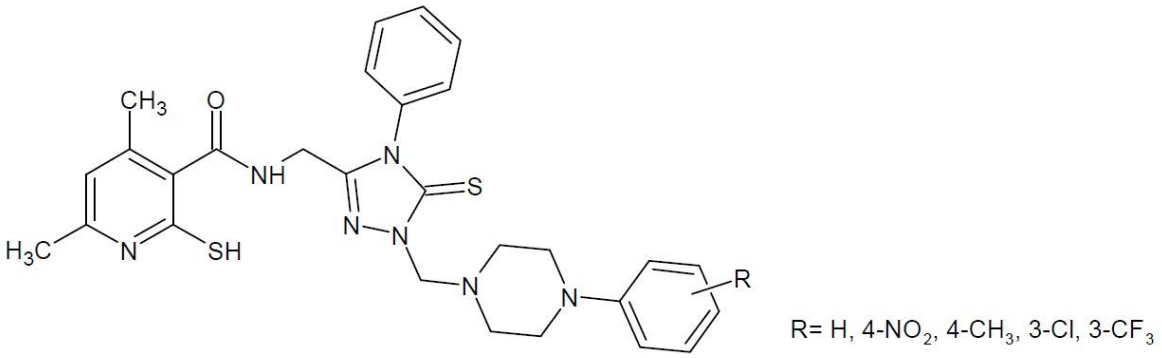

Fig. 1 General structure of new *N*-Mannich bases derived from 1,2,4-triazole-3-thione

The starting material for the synthesis was (4,6-dimethyl-2-sulfanylpyridine-3-carboxamide)acetic acid hydrazide which was obtained by the general procedure developed in Department of Medicinal Chemistry in Wroclaw Medical University [4]. Hydrazide was condensed with phenyl isothiocyanate, and then cyclized to 1,2,4-triazole-3-thione. The final compounds were prepared in a Mannich reaction with formalin and corresponding different substituted phenylpiperazines.

Structures of new compounds were confirmed by FTIR, ^{1}H NMR and ^{13}C NMR. New *N*-Mannich bases were passed on pharmacological tests to investigated their activity.

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P68

Analysis of the cytotoxic activity mechanisms of 1,3-disubstituted thiourea derivatives

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Cancer, after cardiovascular diseases, is the most common cause of death in the world [1]. The currently used and most effective method of cancer treatment is chemotherapy, however its administration is associated with serious side effects. A great challenge for the pharmaceutical industry is to find new, more effective and selective anticancer compounds, that will be concomitantly safe for normal cells.

Thioureas have a great medicinal applications among other as an anti-cancer therapeutics. They can act by different cytotoxic mechanisms e.g. protein tyrosine kinases (PTK), SIRT1 (sirtuins) and SIRT2 proteins or type II topoisomerases inhibition [2]. Additionally, some terminal fragments of the thiourea moiety, enriched with electronegative substituents, exhibited antibacterial [3,4], antiviral, antituberculotic [5,6] or anti-inflammatory [7] potential, as well as the ability to activate the central nervous system [8]. It is known that a large number of 1,3-disubstituted urea and thiourea derivatives have an antiproliferative effect against various tumor cell lines, in particular solid tumors and leukemias, while giving slight side effects. The 3-(trifluoromethyl)phenylthiourea moiety is commonly used as a scaffold in medical chemistry to design new compounds with more promising pharmacological effects [6, 9].

The aim of this study was to evaluate the cytotoxic effect of 1,3-disubstituted thiourea derivatives on selected cell lines and to understand the mechanisms of cytotoxic activity of selected compounds. A series of 3-(trifluoromethyl) phenylthiourea analogues with different terminal systems was tested. In present study we evaluated the anti-proliferative effects of 1,3-disubstituted thiourea derivatives on human primary (SW480) and metastatic (SW620) colon cancers, metastatic prostate cancer (PC3), chronic myelogenous leukemia (K-562) and normal immortalized keratinocytes (HaCaT) cell lines.

Cytotoxicity of studied compounds was measured by MTT assay while cellular apoptosis will be evaluated using annexin assay.

The results of this study clearly indicate that SW480, SW620, PC3 and K-562 cells were highly sensitive to our compounds (1-3, 8, 9), compared to human normal keratinocytes (HaCaT). The most promising compounds belong to the group of dihalogenophenyl (1-3) and para-substituted phenyltioureas (8,9). Their effectiveness was even better than that of the leading cytostatic – cisplatin.

It seems that selected compounds can exhibited cytotoxic effect on cancer cells at lower toxicity against normal cells. Further studies are needed to gain more insight into the mechanism of action of tested derivatives.

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P69

Synthesis and both pharmacological and ADMET profile *in vitro* of 1,3,5-triazine-derived 5-HT$_6$ receptor ligands with ethyl-branched linker

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Serotonin receptors are a very important protein target in Central Nervous System diseases [1]. Lately, a new chemical class of strong 5-HT$_6$R agents – phenyl-1,3,5-triazine-piperazine derivatives - has been identified in our studies [2]. The subject of the presented research is selected representative very potent ($K_i \leqslant 55$ nM) triazine-based derivatives (**1-11**) containing ethyl linker, varied in a substitution: substituted with two chlorine atoms (**2**,**3**), two fluorine atoms (**4**,**5**), fluorine and chlorine atoms (**6**-**9**) and CF_3 containing (**10**,**11**) as well as unsubstituted (**1**) in the aromatic ring (R^1) (**1-11**, Fig. 1).

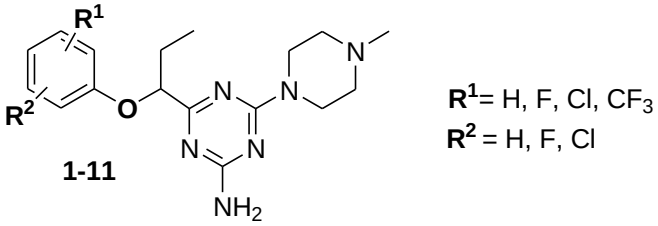

Fig. 1. General structure of investigated compounds.

The compounds were obtained within 2-step synthesis, including *O*-alkylation and cyclic condensation. The affinities towards 5-HT$_6$R and its off-targets were tested in the radioligand binding assays. For selected active compounds (**1**-**3**), a broader binding profile and comprehensive *in vitro* evaluation of their drug-like parameters have been examined. Two compounds, the 2,3-dichlorophenyl- and the phenyl-unsubstituted ones, showed particularly promising properties of drug-like, strong and selective 5-HT$_6$R ligands.

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P70

Overexpression of prolidase modulate p53-dependent apoptosis in MCF-7 cells.

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Background: Prolidase [E.C. 3.4.13.9] is a multifunctional protein of ability to bind and inactivate p53 function. This study was undertaken to establish the role of prolidase in the regulation of p53-dependent apoptosis in a model of prolidase overexpressed breast cancer MCF-7 cells and zebrafish model. Adriamycin was used to induce apoptosis through p53 protein in both models. The effectiveness of apoptosis was confirmed by analysis of phosphatidylserine, caspases 3 and caspases 9 expression and localization in MCF-7 cells and cell proliferation on the zebrafish embryo xenograft model.

Material and Methods: MCF-7, MCF-7 ^{PL} (with a prolidase cDNA expression plasmid) and the zebrafish embryo (inoculated with labelled cancer cells) were incubated with different concentrations of Adriamycin for 24h. Cell viability was performed using the method described in the NucleoCounter kit. Expression and translocation of p53, caspase 9 and 3 were assessed by fluorescent microscopy.

Results: Adriamycin decreased survival and induced apoptosis in MCF-7 cell line and zebrafish embryo xenograft model in a dose-dependent manner. Increased expression of p53, caspase 9 and 3 and their translocation to the nucleus we observed. However, the transfection of MCF-7 cells with prolidase vector (MCF-7^{PL}) protects cells against the pro-apoptotic effects of Adriamycin.

Conclusions: Overexpression of prolidase in MCF-7 cells (MCF-7^{PL}) counteracts Adriamycin – induced, p53-dependent apoptosis in these cells. It suggests, that p53 can be suppressed by forming a complex with prolidase. In fact, we found that in MCF-7^{PL} cells, prolidase expression was elevated providing conditions for sequestration of p53 and the creation of pro-survival pathways.

P71

Oligopeptides as *Escherichia coli* type I signal peptidase inhibitors

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Due to the constantly growing threat of antimicrobial resistance, there is a need for new therapeutics that will treat bacterial infections. To overcome bacterial strains that have become resistant to the currently available antibiotics, new antibiotics should display novel mechanisms of action. Bacterial type I signal peptidase, an essential part of the bacterial secretion system, plays a key role in bacterial viability and virulence which makes it an attractive drug target for new class of antibacterials.

In our research, we focused on the development of a new series of oligopeptides with potential activity against *Escherichia coli* type I signal peptidase. The compounds were designed based on the structure of previously published *Escherichia coli* type I signal peptidase inhibitors: compound 1 with a boronic acid warhead [1] and arylomycin derivative GO775 [2] (Figure 1). A series of new oligopeptides were synthesized by a multistep solid phase peptide synthesis. The results of *in vitro* inhibitory activity towards signal peptidase revealed their weak inhibitory activity, thus obtained compounds need further development and optimization.

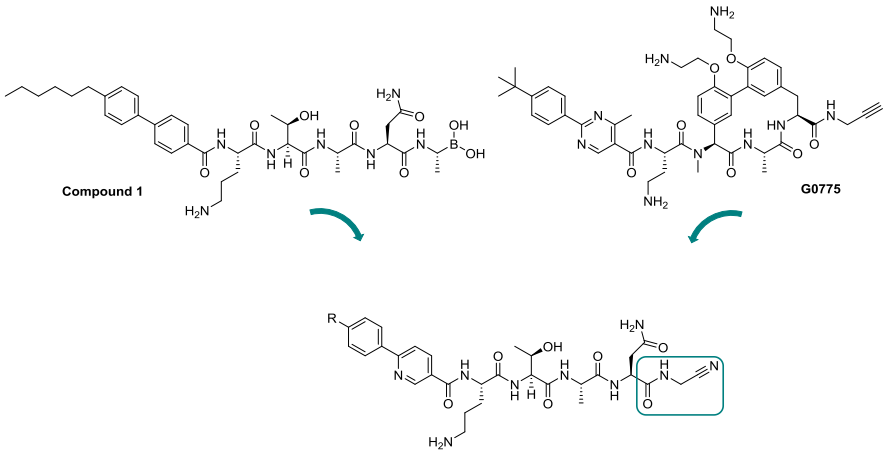

Figure 1. Design of oligopeptides as *Escherichia coli* type I signal peptidase inhibitors: combination of an oligopeptide core from compound 1 and a nitrile warhead from arylomycin derivative G00775.

Financial suport: *N42/DBS/000022.*

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P72

Comprehensive studies on new, potent COX-2 inhibitors based on 1,2,4-triazole derivatives of pyrrolo[3,4-*d*]pyridazinone

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Most of drugs commonly used in treatment of pain and inflammation act as, mainly non-selective, inhibitors of both isoforms of cyclooxygenase (COX) and belong to varied group of medicines named as Non-Steroidal and Anti-Inflammatory Drugs (NSAIDs). The analgesic and anti-inflammatory effects associated with administration of these medicaments relies on the reduction of COX-dependent prostaglandins (PGs) production. Unfortunately, in patients receiving NSAIDs adverse effects related to gastroduodenal tract, such as heartburn, dyspepsia, stomach ache or even ulceration, may occur. Selective COX-2 inhibitors – COXIBs, were supposed to spare gastric mucosa, but the therapy with those drugs carry a serious risk of hazardous cardiovascular incidents which can even lead to patient death [1, 2].

Therefore there is still a current need to search and develop new, safe and efficient analgesic and anti-inflammatory compounds, because a long-term usage of already known and available NSAIDs is often restricted by their severe side effects [1, 2]. We have reported formerly the synthesis and biological evaluation of new 1,3,4-oxadiazole based derivatives of pyrrolo[3,4-*d*]pyridazinone, which revealed remarkable *in vitro* cyclooxygenase inhibitory activity and acted as specific or selective COX-2 inhibitors [3].

Encouraged by those results we have decided to modify the structure of above mentioned derivatives, by replacing the 1,3,4-oxadiazole ring with 1,2,4-triazole pharmacophore, which is an important moiety present in numerous potent bioactive molecules with wide range of therapeutic applications. Obviously, it also acts as a valuable building block used in the design and synthesis of promising analgesic and anti-inflammatory agents, especially those with good affinity towards an inducible COX-2 isoform [4, 5].

Summing up, here we report the design, synthesis and complex *in vitro* and *in silico* investigations of new series of *N*-substituted-1,2,4-triazole based derivatives of pyrrolo[3,4-*d*]pyridazinone.

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P73

Novel classes of TRPA1 channel modulators with amide and hydrazide moieties – design, synthesis and biological activity

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The results of recent studies indicate that the dysfunction of the Transient Receptor Potential Cation Channel Subfamily A Member 1 (TRPA1) is associated with various disease states, including neuropathic pain. The searching for TRPA1 channel antagonists is one of the most important strategies for new analgesics in the treatment of inflammatory and neuropathic pain [1].

The previous studies in the group of purine-2,6-dione and benzimidazole derivatives possessing amide and hydrazide groups, allowed to select several of TRPA1 antagonist with analgetic activity [2,3]. The aim of the present study was the computer-aided design and synthesis of several new series of purine-2,6-dione, benzimidazole and flufenamic acid derivatives as TRPA1 channel modulators with potential application in the treatment of chronic pain. Design supported by molecular modeling methods included docking the proposed structures to a specially constructed model of a biological target and analysis of interactions at the binding site and then were synthesized in the multistep procedure. In order to synthesize the final amide derivatives, the previously obtained esters were hydrolysis to appropriate acids which than were used for synthesis amides in the presence of coupling reagents. For the synthesis of hydrazide derivatives, the starting esters were converted into the corresponding hydrazides and then their *N*-substituted derivatives were obtained by a nucleophilic substitution reaction using the appropriate aldehyde. For the new series of compounds, their ago/antagonistic activity towards the human TRPA1 (hTRPA1) channel were performed using the kinetic fluorescent determination of calcium influx in human TRPA1 transfected HEK-293 cells using Fluo-4. Among the group of purine-2,6-dione derivatives some potent TRPA1 antagonist were selected while the benzimidazole analogues showed agonistic properties. The replacement of the aromatic heterocyclic system (purine-2,6-dione or benzimidazole) by the aromatic core (antranilic acid) due to the loss of TRPA1 channel activity.

Acknowledgements: The study was supported by the Jagiellonian University, grants *No N42/DBS/000062* and *N42/DBS/000019*.

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P74

Substituted pyridine-2-yloxyethyl derivatives of 1-(1-benzoylpiperidin-4-yl)methanamine as novel 5-HT$_{1A}$ receptor biased agonists.

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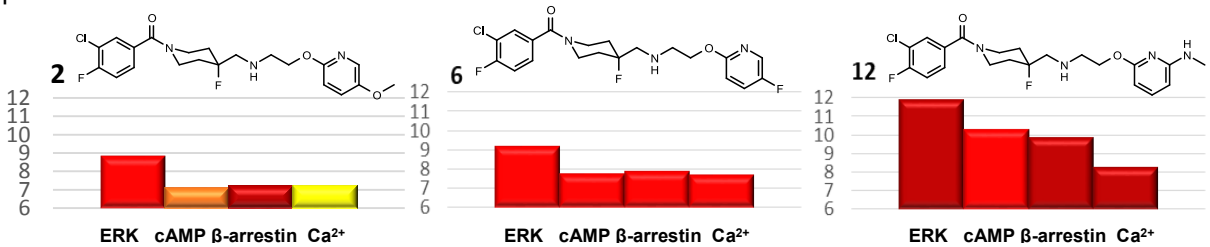
Here we present the discovery of pyridine-2-yloxyethyl derivatives of 1-(1-benzoylpiperidin-4-yl)methanamine (NLX-204 analogs) as a novel serotonin 5-HT$_{1A}$R biased agonists, with prominent druglike properties (CNS-MPO, Fsp3, LELP). Those compounds may be of significant importance in the search for the therapy of psychiatric and neurodegenerative disorders, in particular depression, negative and cognitive symptoms of schizophrenia, Parkinson's disease and Rett syndrome [1,2].

Based on the structure of NLX-204 previously described by our group, we designed, with the support of molecular modeling and obtained by chemical synthesis, a small library of 17 variously substituted pyridine-2-yloxyethyl derivatives of 1-(1-benzoylpiperidin-4-yl)methanamine. The obtained compounds displayed high affinity for the 5-HT$_{1A}$R (pK_i 8.20–10.35), high selectivity versus key biological "anti-targets" – adrenergic α_1 and dopaminergic D_2 receptors (K_i ratio over 1000-fold for most compounds), and very high values of parameters predicting developability of the tested compounds (CNS MPO, Fsp3 or LELP).

The selected 15 compounds were tested in 4 functional assays associated with the 5-HT$_{1A}$ receptor activation, i.e: ERK1/2 phosphorylation (pERK1/2), adenylyl cyclase inhibition (cAMP), calcium mobilization (Ca^{2+}) and β-arrestin recruitment. These studies revealed, that similarly to the lead structure NLX-204, the whole series of derivatives showed high preference to stimulate ERK1/2 phosphorylation, which is particularly important for antidepressant activity and treatment of negative and cognitive symptoms of schizophrenia. We identified 6 novel ERK1/2 phosphorylation-preferring biased agonists, and three of them (**2**, **6** and **12**) elicited even higher preference than NLX-204.



Compounds displaying the most interesting functional profiles were selected for in vivo studies. In the rat forced swim test (FST, Porsolt test), compounds **2** and **6** showed dose-dependent antidepressant-like activity with minimal effective dose of 2.5 and 1.25 mg/kg p.o., respectively. Further characterization of this promising series is ongoing.

This work was supported by The National Science Centre (NCN) grant no. 2015/19/B/NZ7/03543.

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P75

Synthesis and antimicrobial activity of novel hybrids of 2-thiohydantoin and 2-quinolone derivatives

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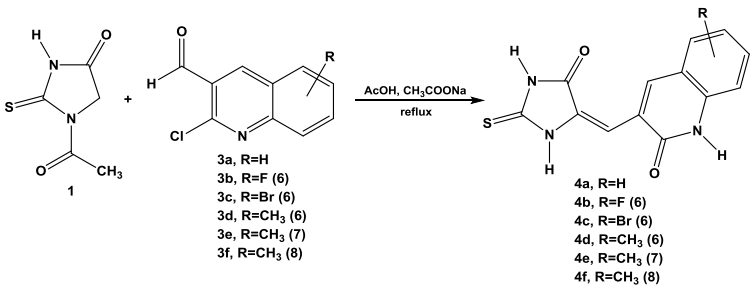
Observed increase in drug resistance of pathogenic microorganisms to the drugs used so far makes it necessary to look for new substances with a therapeutic effect. One of the strategies used, involves research into the use of hybrids of two different compounds with antimicrobial activity. Such hybrids may be more active than either component alone. In our research, we paid attention to 2-thiohydantoin and 2-quinolone derivatives. Compounds containing the 2-thiohydantoin core show antibacterial, antifungal, antitumor, antiparasitic and antiviral activity.[1] Compounds containing the 2-quinolone core exhibit a similar type of biological activity. They show, inter alia, anti-cancer, anti-tuberculosis, antibacterial [2] and antifungal properties. Based on literature reports, we decided to synthesize hybrids of 2-thiohydantoin and 2-quinolone derivatives and then to test the biological activity of the compounds obtained.

Knoevenagel condensation in between was used to obtain hybrids 1-acetyl-2-thiohydantoin with 2-chloroquinoline-3-carbaldehyde derivatives according to a modified procedure described in earlier works. [3] Under the conditions used, the acyl group was detached from 1-acetyl-2-thiohydantoin and the chlorine atom was replaced with an oxygen atom. Scheme 1.



Scheme 1.

The antibacterial activity of the novel hybrid compounds was investigated against reference bacterial strains: *Staphylococcus aureus* ATCC 292313*, Bacillus subtilis* ATCC 6633 *Enterococcus faecalis* ATCC 29212*, Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853 and *Klebsiella pneumoniae* ATCC 700603. The antimicrobial assays were carried out using a serial dilution method to obtain the MIC (Minimal Inhibitory Concentration). Antimicrobial properties were investigated both in the dark and after irradiation of bacterial colonies placed in microtitrate plates with blue light (420 nm).

Most of the hybrid compounds showed bacteriostatic activity to the reference gram-positive bacterial strains, but three of them were also bacteriostatic towards gram negative bacteria. Blue light activation enhanced bacteriostatic effect of the tested compounds.

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P76

New rhodanine-hydrazide and rhodanine-thiosemicarbazide hybrids: Synthesis and their antibacterial properties *in vitro*

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There are more and more cases of drug-resistant bacteria which have become a serious health problem in the world. The World Health Organization (WHO) has declared antimicrobial resistance as one of the top 10 global public health threats against humanity. An analysis of innovative drugs approved by the FDA in the last decade for specific pharmacological groups shows that antimicrobials are at a second place (~ 16% of all approved drugs) after anticancer drugs (~ 27%).

The development of antimicrobial resistance is a serious problem and at the same time it is a major driving force for new research into the discovery of new antimicrobials. The thiazolidin-4-one derivatives, including rhodanine (2-thioxothiazolidin-4-one), represent considerable interest for design of antimicrobial agents. There are reports from the literature that the rhodanine derivatives have been identified as potential antimicrobial ligands as inhibitors of Mur B, C, D, E, F ligases (selective and non-selective), penicillin-binding proteins (PBPs), mannosyl transferase 1 (PMT1) [1-4].

In our study we synthesized new series of rhodanine-hydrazide and rhodanine-thiosemicarbazide hybrids and evaluated their antibacterial properties *in vitro*. The rhodanine was starting material and its reaction with triethyl orthoformate led to obtaining of 5-ethoxymethylidenerhodanine. Next, modification of 5-ethoxymethylidenerhodanine in the reaction with corresponding aroylhydrazides and thiosemicarbazides led to obtaining of target hybrids.

The *in vitro* antibacterial activity of the target compounds was primarily screened using agar dilution method (with concentration of 1000 μg/mL). Next, MIC (minimum inhibitory concentration) was determined using broth dilution method (with concentration range from 7.81 to 1000 μg/mL) for potentially active compounds. Both Gram-positive and Gram-negative reference strains were used.

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P77

Crosslinking of chitosan with dialdehyde chitosan for biomedical applications

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Polysaccharides are an important class of biological polymers universally found in all living organisms. They are biocompatible, non-toxic, and wide availability in nature [1]. Chitosan is a pseudo-natural polysaccharide composed of glucosamine and N-acetylglucosamine derived from chitin shells of crustaceans [2]. For medical applications, it is more eligible to utilize cross-linking agents that have a potential to become a low-toxicity alternative to common toxic low-molecular weight cross-linking agents. One of the most acceptable biopolymers is chitosan and its derivatives as dialdehyde chitosan (DACS) [3]. DACS was obtained by controlled oxidation with sodium periodate (Fig. 1).

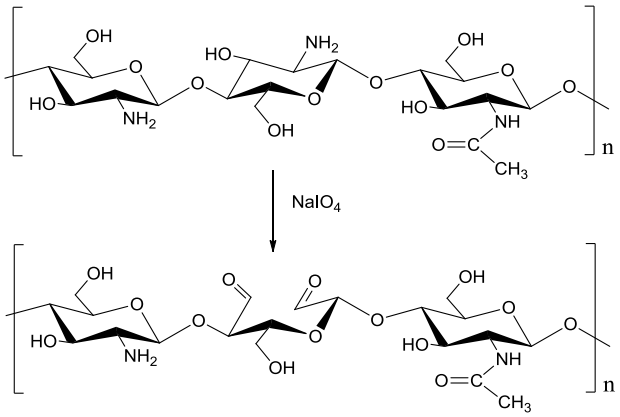

Figure 1. Oxidation of chitosan to dialdehyde chitosan (DACS) with sodium periodate.

In this study, chitosan was oxidized by sodium periodate to prepare a dialdehyde functionalized material. The obtained modified chitosan have been characterized with scanning electron microscopy, contact angle measurement, thermal analysis, and ATR-FTIR spectroscopy. Dialdehyde chitosan was used for the cross-linking of chitosan films. Obtained materials were characterized in order to define the structure, morphology, thermal stability, mechanical properties, degree of swell, and toxicity. The results were compared with the materials obtained by cross-linking chitosan with low molecular weight glutaraldehyde. It has been shown that dialdehyde chitosan is a very promising crosslinking agent.

Acknowledgements *The project was supported by research grant: National Science Centre 2016/23/N/ST8/00211.*

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P78

Synthesis and cytostatic activity of new derivatives of quinazolin-2-one

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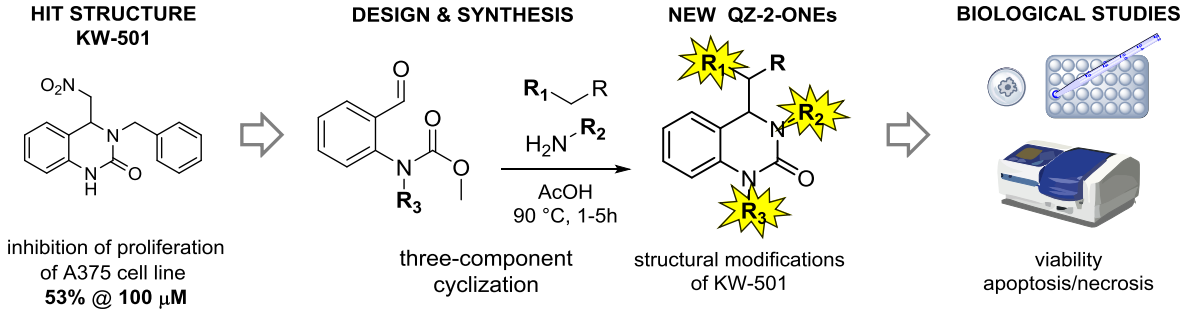
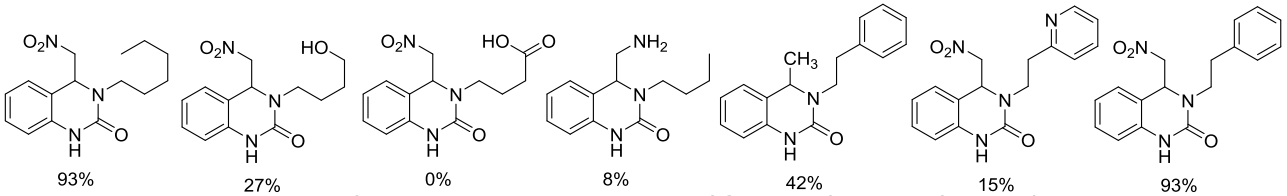
This project is part of chemical and pharmacological studies in the group of quinazolin-2-one (QZ-2-one) derivatives. [1] These studies were inspired by KW-501, a quinazoline derivative that showed minor cytotoxic activity in a screening on skin cancer cell line. The aim of this project was to design and synthesize a series of new QZ-2-ones, analogs of KW-501, and to assess their cytotoxic and cytostatic activity.



As the result of these studies a series of new analogs of KW-501 was obtained. Some of the structural modifications resulted in a significant increase cytostatic activity. A number of these derivatives showed antiproliferative effect against various neoplastic cell lines in the range of 10-25 μM (MTT test). Preliminary studies of the mechanism of action indicate a pro-apoptotic effect, suggesting potential antineoplastic activity.



Examples of active and inactive derivatives of QZ-2-one (analogs of KW-501)
(activity given as % inhibition of A375 cell line proliferation at 25 mM)

The obtained results allowed for a better understanding of the structure-activity relationship in this group of compounds. These results will help in choosing the right direction for further development of this project.

Financial suport: Narodowe Centrum Nauki: 2019/03/X/NZ7/01116 (K/MNT/000120)
Uniwersytet Jagielloński – Collegium Medicum: N42/DBS/000185

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P79

Influence of selected substances on the activity of tyrosinase

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Melanins belong to the group of compounds responsible for photoprotection. They protect against the harmful effects of UV radiation. Unfortunately, in some cases, this dye can accumulate, causing discoloration. The developing industry is looking for more and more effective and safer solutions in the fight against hyperpigmentation. Many brightening substances are of natural origin, but their derivatives are increasingly used [1].

Melanins are formed as a result of reactions taking place in the melanogenesis pathway. The key enzyme and regulator in this process is tyrosinase (TYR). Many of the brightening agents used affect the activity of tyrosinase. Tyrosinase is a glycoprotein produced only by specialized cells, melanocytes. Substances with bleaching potential can influence the TYR at various stages. Melanogenesis can be regulated in the steps of TYR expression, maturation, degradation, or direct inhibition of activity [2].

Melanogenesis is a complex and multi-step process. Many brightening compounds, despite their proven action, do not affect the activity of TYR, inhibiting the process of melanin formation at a different stage.

The use of antioxidants may affect the process of melanogenesis by reducing oxidative stress. The resulting free radicals can stimulate the activity of tyrosinase. Additionally, antioxidants may limit the photo-oxidation process of melanin deposited in melanosomes [3].

The strongest whitening agents include hydroquinone, azelaic acid, arbutin and kojic acid. Unfortunately, long-term use of hydroquinone causes many side effects, which is why it has been restricted or banned for use in cosmetic products by many countries [4].

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P80

Phototoxic effect of meloxicam on normal human melanocytes

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One of the most frequently occurring side effects of pharmacotherapy is cutaneous adverse drug reactions [1]. They often lead to deterioration of patient's health by limiting the possibility of drug usage and require hospitalization with additional treatment [2,3]. Phototoxicity is the most common type of skin adverse-drug reaction resulting from simultaneously internal or external usage of photosensitizing agents and exposure to sunlight [4]. The phototoxic reaction causes damages to a cellular structure by induction and generation of free radicals which are activated by UV radiation. The symptoms of drug-induced photosensitivity are similar to sunburn [5].

Meloxicam belongs to the group of non-steroidal, anti-inflammatory drugs (NSAIDs). NSAIDs are commonly and extensively used for their analgesic, antipyretic and anti-inflammatory properties. The mechanism of action is associated with blocking the cyclooxygenase – enzyme which is responsible for the synthesis of proinflammatory prostaglandins [6].

The aim of the study was to examine the influence of meloxicam and UV radiation on human normal light pigmented melanocytes (HEMn-LP). Melanocytes are cells responsible for synthesizing melanin, during in process of melanogenesis. The study included evaluation of cell proliferation and morphology, cell vitality and, cell cycle analysis. Analysis was performed by using the fluorescence image cytometer NuceloCounter NC-3000.

Based on the obtained analysis, it was found that meloxicam in combination with UVA radiation results in a reduction in survival of melanocytes, increases the number of cells with low vitality, and affects cycle progression. In the photos taken, it can be observed that meloxicam causes a significant reduction in the number of HEMn-LP cells compared to control. In addition, cell morphology changes can be observed – loss of the original shape and contact between neighboring cells, as well as a reduction in their size. The obtained results suggest that meloxicam has phototoxic properties, following the influence of UVA radiation on human normal light pigmented melanocytes.

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P81

Antimalarial activity of novel 6,7-dimethoxy-quinazoline derivatives

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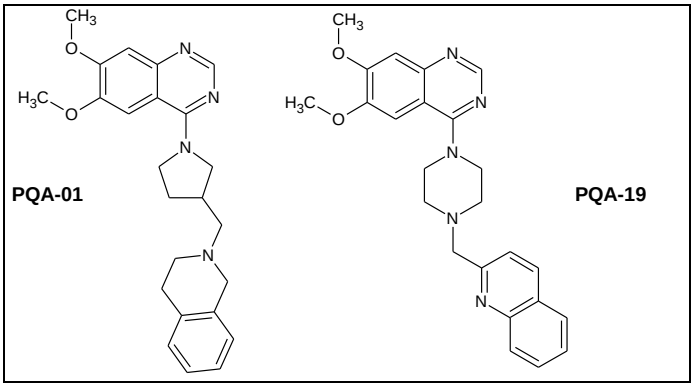
Malaria is a life-threatening disease, affecting mainly children and pregnant women from tropical countries. Current antimalarial drugs are rapidly losing their effectiveness due to rising parasite resistance. Therefore there is an urgent need to discover and develop next-generation therapeutics and novel compounds characterized by high selectivity and effectiveness [1, 2]. The aim of the study was the evaluation of antiplasmodial activity and cytotoxicity of novel 6,7-dimethoxy-quinazoline derivatives compounds PQA-01 and PQA-19 as promising antimalarial compounds.



These compounds were evaluated against the asexual stages of two strains of *Plasmodium falciparum*, namely, the chloroquine-sensitive (D10) and chloroquine-resistant (W2) strains. The antiplasmodial effect of PQA-19 against the W2 strain (IC_{50} 5.0 µg/ml) was comparable to the D10 strain (IC_{50} 4.0 µg/ml), while the IC_{50} values of PQA-1 on both strains were significantly higher (> 8.8 µg/ml). Moreover, both compounds were not cytotoxic against human keratinocyte cells, at concentrations they were active against parasite strains. Our research proved that a novel 6,7-dimethoxy-quinazoline derivative (PQA-19) could act as good lead compounds for future effective antimalarial agents.

The project is co-financed by the Polish National Agency for Academic Exchange (PPN/BIL/2018/2/00108) and the Italian Ministry of Foreign Affairs and International Cooperation “Executive Programme for Scientific and Technological Cooperation between the Italian Republic and the Republic of Poland” (PO19MO10) and supported by JUMC grant no *N42/DBS/000178*.

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P82

FPPQ, a dual-acting 5-HT_3/5-HT_6R antagonist with antipsychotic and pro-cognitive properties

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Schizophrenia is a severe neuropsychiatric disorder, characterized by positive symptoms such as hallucinations and delusions and negative symptoms including apathy and anhedonia. Third group of symptoms is represented by cognitive impairment, which involves disturbances in memory and thinking. Despite over 60 years of experience in the pharmacological treatment of schizophrenia, there is still a need of safe and more efficacious agents, especially for cognitive decline. Taking into account the results from the clinical studies, indicating that 5-HT_3 receptor (5-HT_3R) antagonist, ondansetron, ameliorates cognitive deficits of schizophrenia[1] as well as the pro-cognitive effects of 5-HT_6 receptor (5-HT_6R) antagonists,[2] we applied a hybrid approach to design dual-acting 5-HT_3/5-HT_6R antagonists. The study allowed for the identification of the first-in-class compound FPPQ, which behaves as 5-HT_3R antagonist and neutral antagonist of 5-HT_6R in the Gs pathway. FPPQ displays selectivity over 87 targets and decent brain penetration. Likewise, **FPPQ** inhibits phencyclidine (PCP)-induced hyperactivity, and displays pro-cognitive properties in the novel object recognition task. In contrast to **FPPQ**, neither 5-HT_6R inverse agonist SB399885 nor neutral 5-HT_6R antagonist CPPQ reversed (PCP)-induced hyperactivity. Thus, combination of 5-HT_3R antagonism and 5-HT_6R antagonism, exemplified by **FPPQ**, contributes to alleviating the positive-like symptoms.

The study was financially supported by the Polish Ministry of Science and Higher Education Grant No. N N405 671540; the project “Prokog”, UDA-POIG.01.03.01-12-063/09-00, co-financed by the European Union from the European Fund of Regional Development (EFRD); statutory funds from the Institute of Pharmacology, Polish Academy of Sciences, University of Montpellier, CNRS, INSERM and Agence Nationale de la Recherche (no. ANR-17-CE16-0013-01 and ANR-17-CE16-0010-01).

P83

A novel method for the synthesis and preliminary evaluation of the antitumor activity of sulfonamide derivatives of cyclic arylguanidines for the treatment of glioblastoma multiforme

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Gliomas are cancers of the central nervous system (CNS) that morphologically resemble glial cells. Glioblastoma (glioblastoma multiforme; GBM), is the most aggressive and most common primary brain tumor in adults and children,[1,2] containing 16% of all primary brain and cancers.[3] The average incidence rate corrected for age is 3.2 per 100000 population.[4]

We proposed a new type of compounds, showing antitumor activity against GBM, from the group of arylsulfonamide derivatives of cyclic arylguanidines. The selected compounds were subjected to MTS cytotoxicity preliminary tests against astrocytoma (GBM) line. The tested compound, *N*-(3,4-dihydroquinazolin-2-yl)naphthalene-1-sulfonamide, showed high cytotoxic activity against mentioned line.

In order to enable the further development of this group, we have developed a fast and universal method of synthesis, which is an alternative to the reaction between dimethyl(arylsulfonyl)-carbonodithioimidates and aryl diamines described in the literature.[5] The new method is based on the reaction between arylsulfonamides and 2-(methylsulfanyl)-1,4dihydroquinazoline. This reaction can be carried out in the field of microwave radiation, under solvent-free conditions. It allows to obtain a product with high efficiency in a short time, improving the atomic economy of the previously described reaction.[5]

Acknowledgements

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P84

New furanochromone derivatives – synthesis and preliminary studies

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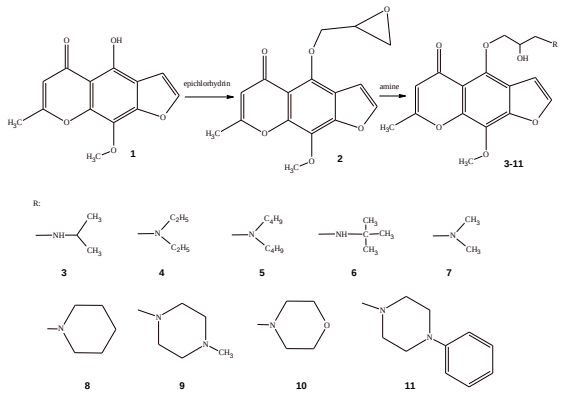
Despite the new registrations of novel therapies for cancer treatment, the efficient and safe drugs of this disease are still an unmet medical need [1]. The research to find a new anticancer agent is still going on. According to the literature data, the furanochromone analogs seem to be a good candidate as potential therapeutic molecules [2,3].

The preliminary, unpublished researches in the Department of the Biochemistry Medical University of Warsaw show that some furanochromones have strong cytotoxic activity against cancer cells. Unfortunately, these newly synthesized molecules also revealed a strong toxic effect against normal cells.

Based on that, we have decided to synthesize a group of furanochromone derivatives with potential biological activity. The first step of our research was to synthesize a series of furanochromone analogs which were linked with the selected amines in two steps reaction (Scheme **1**). The structure of all obtained compounds was confirmed by ^{1}HNMR, ^{13}CNMR, and MS spectra. Next, we tested cytotoxic properties of all derivatives against both cancer (SW 480, SW 620) and normal cells (V79) line to compare the toxicity of compounds in cancer and non-cancer cells. The synthesized furanochromone analogs did not show a strong cytotoxic effect against tested cell lines. We are going to synthesize next derivatives and after preliminary screening, the most promising compounds will be used in further biological tests.



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P85

Docking-based 3D-QSAR studies for 1,3,4-oxadiazol-2-one derivatives as FAAH inhibitors

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The Endocannabinoid System and its activity recently became a hot topic widely examined by medicinal chemists. Several studies suggest that medication affecting endocannabinoid signaling can be used in the treatment of several complex, currently incurable diseases e.g. chronic pain or schizophrenia[1], [2]. One of the enzymes responsible for the degradation of endogenous cannabinoids is termed fatty acid-amide hydrolase. It is suggested that inhibition of this enzyme enhances the signaling within the endocannabinoid system and comprises a potential therapeutic strategy in treating mentioned above diseases [3]. One way to design selective and potent FAAH inhibitors is to examine the structure-activity relationship among a series of compounds with experimentally determined inhibitory activity. Quantitative-structure activity relationship techniques (QSAR) can be easily implemented for such process. Therefore, we decided to take advantage of the currently available X-ray structure of fatty acid-amide hydrolase and construct 3D-QSAR (the CoMFa and CoMSIA) models for a series of previously published compounds (1,3,4-oxadiazol-2-one derivatives)[4], [5]. The obtained models were characterized by good statistical parameters: CoMFA Q^2= 0.61, R^2=0.98; CoMSIA Q^2=0.64, R^2=0.93. The CoMFA model field contributions were 54% and 46% for steric and electrostatic fields, respectively. In the CoMSIA model, steric, electrostatic, hydrogen bond donor, and hydrogen acceptor properties were equal to 24%, 35%, 18%, and 23%, respectively. These models were validated applying the leave-one-out technique, the 7-element test set (CoMFA $r^2_{test\text{-}set}$= 0.91; CoMSIA $r^2_{test\text{-}set}$= 0.91), a progressive scrambling test, and an external validation criteria developed by Golbraikh and Tropsha (CoMFA r^2_0=0.98, k=0.95; CoMSIA r^2_0=0.98, k=0.89). As the statistical significance of the obtained models was confirmed, the CoMFA and CoMSIA field calculation results were mapped onto the enzyme binding site.

We believe that promising results obtained in this study will contribute to a better understanding of the structure-activity relationship among FAAH inhibitors and assist in designing novel, more potent compounds.

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P86

Towards property modeling: Synthesis and similarity-based SAR probing of new tetracyclic diazaphenothiazine analogues

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A series of new tertacyclic phenothiazine derivatives containing a quinoline and a pyridine fragment were obtained. The compounds were prepared by the reaction of 1-methyl-3-benzoylthio-4- (buthylthio) quinolinium chloride with 3-aminopyridine derivatives bearing various substituents on the pyridine ring. The direction and mechanism of the cyclization reaction of intermediates with the structure of 1-methyl-4- (3-pyridyl)aminoquinolinium-3-thiolate was dependent on the substituents in the 2- and 4-pyridine positions. The structure of the compounds obtained was analyzed by ^{1}H, ^{13}C NMR (COSY, HSQC, HMBC) and X-ray methods. The biological activity of the obtained derivatives was tested. Antiproliferative activity was tested against A549, T47D, SNB-19 neoplastic cells, NHDF normal cell lines and antibacterial activity against gram-positive and gram-negative strains. Both the antitumor and antimicrobial activity depend on the presence of substituents in the pyridoquinothiazine system. *In silico* computation of the intermolecular similarity was performed using the principal component analysis (PCA) and hierarchical clustering analysis (HCA) on the pool of structure/property-related descriptors calculated for novel tetracyclic diazaphenothiazine derivatives. The distance-oriented property evaluation for the congeneric series of compounds was correlated with the experimental anticancer activities and empirical lipophilicity profile, respectively. A range of various software logP predictors for estimation of the numerical lipophilic values was employed and subsequently cross-compared with the experimental parameters. Moreover, the newly synthesized adducts were subjected to the quantitative shape comparison (CoMSA) with the generation of an averaged pharmacophore pattern to illustrate the key 3D steric/electronic/lipophilic features. Finally, the quantitative sampling of similarity-related activity landscape (SALI) provided a subtle picture of favorable and disallowed structural modifications that are valid for determining the activity cliffs.

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P87

The *in silico* prediction of human-specific metabolites from selected organic UV filters

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Chemical UV filters are widely used components of sunscreens and other cosmetic and personal care products to protect the skin against the acute and chronic consequences of ultraviolet radiation. Previous studies indicated that some UV filters can be absorbed into and across the skin, further metabolized in the body and excreted [1-5]. That is the reason why studies focused on the evaluation and determination of these agents and their metabolites in human body fluids are important area of research.

Nowadays different *in silico* approaches to predict compounds metabolism based on the interactions of agents with cytochrome P450 (CYP450) enzymes and their metabolic endpoints are available [6]. MetaSite (Molecular Discovery Ltd.) is a computational model created to simulate metabolic conversion reactions in phase I metabolism by CYP450 and flavin-containing monooxygenase 3 (FMO3). The software ensures the prediction as a result of its unique algorithm that is independent of the training dataset [7, 8].

The main objective of this study was to evaluate human liver metabolism of selected chemical UV filters most *frequently used* in sunscreen formulations using MetaSite software. The following compounds were studied: Benzophenone-3 (BP-3), 4-Methylbenzylidene camphor (4-MBC), Homosalate (HMS), Octinoxate (OTN) and Octocrylene (OTC).

MetaSite suggested the following major metabolites of the analyzed compounds: 2,4-dihydroxybenzophenone for BP-3, both 3-(4-carboxybenzylidene)camphor and 3-(4-hydroxymethylbenzylidene)camphor for 4-MBC, 3,3,5-trimethylcyclohexyl-dihydroxybenzoate for HMS, demethylated parent ethylhexyl methoxycinnamate for OTN, and 2-ethyl-5-hydroxyhexyl 2-cyano-3,3-diphenyl acrylate for OTC. All of the major metabolites given by the software were previously reported in the literature as the major *in vivo* or *in vitro* metabolites.

In general MetaSite offers a large number of predictions, which should be always complied with experimental investigations. It represents a valuable tool in predicting human metabolites.

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P88

Effect of the position of a chlorine substituent on the antimicrobial activity and crystal structure of 4-methyl-1,6-diphenylpyrimidine-2(1*H*)-selenone derivatives

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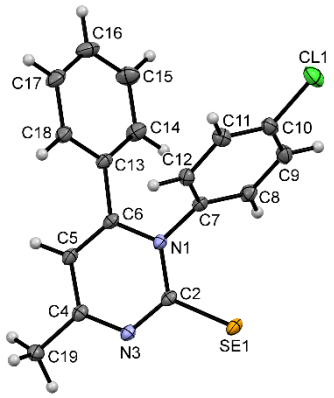
Antimicrobial resistance is still the reason to be a public threat worldwide. The continuous need to develop new antimicrobial drugs that are effective against multidrug-resistant pathogens has forced researchers to invest in various drug discovery strategies. We cannot avoid resistance nor predict directly the emergence of new infectious agents, but we can try to mitigate the problem with new agents development presenting novel mechanisms of action and chemical classes. Nowadays, it is important to develop new classes of compounds with more effective mechanisms due to drug resistance development which can reduce effective disease treatment [1,2].

In the group of compounds showing antimicrobial activities there are Se-containing compounds. The results of our previous studies showed very good antimicrobial activities of 4-methyl-1,6-diphenyl-2[1*H*]-pyrimidineselenone and its methoxy derivatives [3,4]. Now, the previously active 4-methyl-1,6-diphenyl-2[1*H*]-pyrimidineselenone was modified by introduction of chlorine substituent to phenyl ring at the N1 atom and these derivatives were evaluated against antimicrobial activities. For compound containing chlorine substituent in position *para* we have determined crystal structure.



The derivative with *p*-Cl substituent shows the best activity against bacteria *S. aureus* ATCC 25923 and ATCC 43300, *S. epidermidis* ATCC 12228, *M. luteus* ATCC 1024 in comparison to other two derivatives. Furthermore, this compound possesses very good antifungal activity, wherein its activity against *C. parapsilosis* ATCC 22019 is better than activity of the reference compound, namely fluconazole. The molecular geometry in crystal of investigated compound mainly differs in arrangement of the aromatic rings with respect to the heterocyclic ring in comparison to other derivatives. The intermolecular interactions are dominated by C-H···Cl, C-H···N and C-H···Se contacts.

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P89

2-(2-Arylmethylthio-4-chloro-5-methylbenzenesulfonyl)-1-phenylguanidines – synthesis and anticancer activity

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Sulfonamides are a classic group of chemotherapeutic drugs with a broad spectrum of pharmacological action. Historically, this class of compounds derives from the simple sulfanilamide which was the leading structure for development of important drugs such as antibacterial sulfathiazole [1], antiglaucoma acetazolamide [2], diuretic furosemide [3], hypoglycemic agent glibenclamide [4] or antiviral amprenavir [5]. Scientific reports of recent years indicate that sulfonamide derivatives show in vitro and in vivo antitumor activity with various mechanisms of action, including carbonic anhydrase inhibition, cell cycle perturbation in G1 phase, inhibition of tubulin polymerization or angiogenesis inhibition (inhibition of extracellular matrix metalloproteinases) [6]. Cinnamic acid and its natural analogues have been used in the treatment of cancer for over centuries [7]. Natural products containing the cinnamon group attract a lot of attention due to their broad spectrum of biological activity and low toxicity. Thus designing of molecular hybrids, containing in their structure pharmacophore fragments of cinnamic acid and sulfonamide, may lead to compounds with increased biological activity as a result of synergistic effect of both fragments.

In search of innovative low-molecular chemotherapeutic agents, a series of new 2- (2-arylmethylthio-4-chloro-5-methylbenzenesulfonyl)-1-phenylguanidine derivatives with potential anticancer activity were designed. The new original compounds were molecular hybrids containing the pharmacophore core of 4-chloro-2-mercapto-5-methylbenzenesulfonamide and 4-acetylphenyl moiety or chalcone fragment. The designed compounds were obtained by organic synthesis. The structures of the compounds were confirmed by elemental analysis, spectroscopy (IR, ^{1}H NMR, ^{13}C NMR), X-Ray and mass spectrometry.

The obtained compounds were evaluated for their cytotoxic activity in the MTT test against three human tumor cell lines: breast cancer (MCF-7), colon cancer (HCT-116) and cervical cancer (HeLa). It has been shown that a number of 1-(4-acetylphenyl)-2-(benzenesulfonyl)guanidines (series 1) and 2-(benzenesulfonyl)-1-(4-cinnamoylphenyl)guanidines (series 2) are highly active against all cancer cell lines (IC_{50}: 10–21 μM, series one and 4.7–15 μM, series two). Additionally, in tests carried out on non-cancer human keratinocyte cell line (HaCaT), it was proved that the tested compounds were selective for cancer cells.

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P90

Schiff bases as antiparasitic agents

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Despite the great knowledge about parasites and the fight against them, as well as the improvement of hygiene conditions, parasitic diseases are currently still a global problem. New pathogens and diseases appear, which is mainly related to the popularization of tourism, human migration, and civilization changes. There was also a problem of resistance to existing drugs. Hence, it is necessary to search for new compounds with antiparasitic activity. One of the promising groups of compounds are Schiff bases, which are characterized by a broad spectrum of biological activity.

We decided to obtain new Schiff bases derived from 4-amino-5-(3-fluorophenyl)-2,4-dihydro-3*H*-1,2,4-triazole-3-thione and to study their activity against nematodes of *Rhabditis sp*. The highest activity was observed for 5-(3-fluorophenyl)-4-[(4-methylbenzylidene)amino]-2,4-dihydro-3*H*-1,2,4-triazole-3-thione, which at a concentration of 10 mg/ml showed activity comparable to albendazole. The weakest nematocidal properties were shown for 5-(3-fluorophenyl)-4-[(4-nitrobenzylidene)amino]-2,4-dihydro-3*H*-1,2,4-triazole-3-thione. On the basis of the obtained results, it was found that the presence of an electron donating substituent in the aromatic ring of the aldehyde was an element of the structure necessary to induce nematocidal activity in this group of compounds. The research is preliminary and may be the beginning of a new project aimed at obtaining effective nematicides.

P91

1,3,4-Thiadiazole as antibacterial agents

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The growing resistance of bacteria to known and therapeutic antibiotics is a serious health problem all over the world. Sepsis, a consequence of acute bacterial infection, is now one of the leading causes of death on a global scale. MRSA is a methicillin-resistant strain of *Staphylococcus aureus*, a common cause of difficult-to-treat nosocomial infections. It is resistant to all beta-lactam antibiotics, including penicillins, cephalosporins, monobactams and carbapenems. Despite the intense search for new antibiotics, there is still a lack of drugs that are effective against bacteria such as *Staphylococcus aureus.*

In response to this pressing problem, we have synthesized a series of 1,3,4-thiadiazole derivatives with potential antimicrobial properties. We tested all synthesized compounds for antibacterial activity. In the tests we used, among others, *Staphylococcus aureus* strains. Most of the thiadiazoles tested showed negligible to moderate antibacterial activity. The most promising compound turned out to be the thiadiazole having a substituted phenyl group with a bromo substituent in the *para* position (Fig. 1)

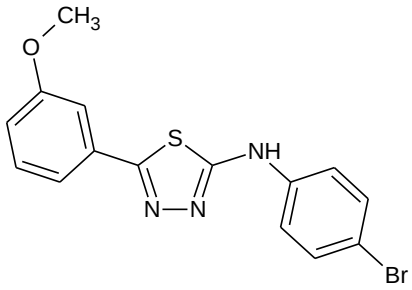

Fig. 3: The structure of the most active compound.

P92

Development of novel thiazole derivatives as potential anticancer agents

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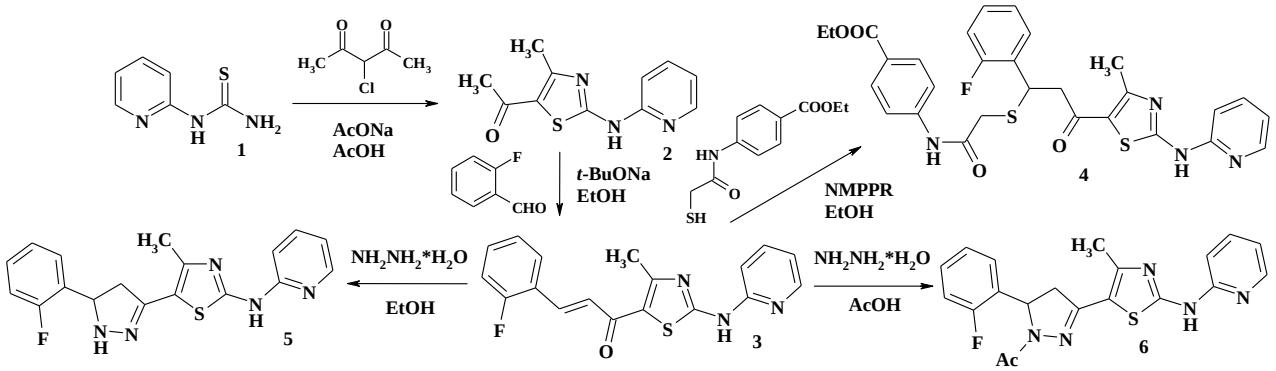
In modern medicinal chemistry, Michael acceptors (MA) possess the dualistic role. MA are considered as frequent hitters or pan assay interference compounds (PAINS) that are useless in the drug discovery. PAINS tend to react nonspecifically with numerous biological targets rather than specifically affecting one desired target. From another hand, low selectivity is linked to a polypharmacological approach where the affinity toward various targets is regarded as an advantage and is base-line for further optimization. In this context, it is worth mentioning that such MA are effective “covalent inhibitors” that opens new perspectives in the design of new anticancer agents. Currently, the MA are assigned as “old new tool” for drug design. To develop the theme of MA, we obtained a pyridine-thiazole **3** containing an "enone" (MA) moiety. Based on compound **3**, derivatives **4**-**6** were synthesized which can be interpreted as pro-Michael acceptors (pro-MA).



Synthesized compounds were studied for antitumor activity (MTT assay) on 16 cell lines with different origin. It was used colon, breast, glioblastoma, leukemia and lung cancer cell lines as well as pseudonormal cell lines (Hek293, HaCaT) and normal human lymphocytes. Cytotoxic effect of lead compound **3** was indicated in low μM range as IC50 value. For example, IC50 for HL-60 cell line (acute promyelocytic leukemia) was reached already by concentration of 0.57 μM of compound 3. Moreover, it shows a low toxic effect on pseudonormal cell lines and does not reach IC50 even by acting it in concentration of 50 μM. Starting compounds **1**,**2** had a lower cytotoxic effect, IC50 was higher than 50 μM and IC50 of pro-MA 5 and 6 was μM ranged from 30 to more than 50. Nevertheless, pro-MA derivative **4** has a similar effect to the compound MA derivative 3. Such selective action of MA/pro-MA derivatives **3** and **4** on cancer cell lines with low cytotoxicity toward pseudonormal cells is a promising feature of the mechanism of their action and can suggest them as perspective anticancer drug-like substances.

P93

Novel thiopyrano[2,3-d]thiazoles as anticancer agents

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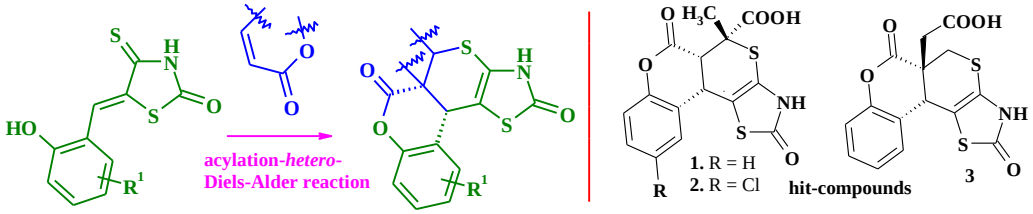
The thiopyranothiazole scaffold is characterized by "fixed" 4-thiazolidinone biophor in "rigid" fused system that allows to save the biological activity of their synthetic precursors 5-ene-4-thiazolidinones [1]. Considering mentioned arguments the design of new chemotherapeutic agents among thiopyrano[2,3-d]thiazoles is promising direction in medicinal chemistry. Recently, we reported a diastereoselective tandem acylation-*hetero*-Diels-Alder reaction of 5-(*o*-hydroxybenzylidene)-4-thioxo-2-thiazolidinones with α,β-unsaturated carboxylic acid derivatives for the synthesis of cromeno[4',3':4,5]thiopyrano[2,3-d]thiazoles [2]. Based on this reaction more than 200 novel derivatives were synthesized.



In vitro screening of anti-proliferative activity (MTT assay) of the more than 50 synthesized derivatives towards Jurkat (acute T-cell leukemia), HL-60, K562 (myeloid leukemia), MCF-7 (breast adenocarcinoma), U251 (glioblastoma) cell lines and non-tumor cells (HEK293 and HaCaT lines) allowed identification of three hit-compounds **1**-**3**. Leukemia Jurkat, K562 and HL-60 lines were the most sensitive to cytotoxic action of the tested compounds. Thus, compound **2** displayed cytotoxicity with IC_{50} values of 3.9-37.6 μM towards Jurkat, K562, HL-60, U251 and MCF-7 cells. Besides, mentioned derivative was proximally 10 times more active than temozolomide towards glioblastoma U251 cell line. At the same time, hits **1**-**3** possessed low toxicity towards non-tumor HaCaT and HEK293 lines. Compound **2** caused the cleavage (activation) of the pro-apoptotic caspase 3, and the cleavage of the DNA reparation enzyme PARP-1 in Jurkat cells (Western-blot analysis). Moreover, this derivative increased the content of the pro-apoptotic mitochondrial proteins endonuclease G (EndoG) and Bax in Jurkat cells. Thus, the anti-proliferation action of compound **2** in leukemia cells is associated with apoptosis induction *via* mitochondria-dependent pathway. Identified hit-compound **2** may serve as a lead for the in depth study as anticancer agent.

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P94

In vitro antileukemic activity of new xanthone derivatives

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Xanthones are often described as a privileged structures, as they have very diverse biological profiles, including antihypertensive, antiepileptic, antidepressant, antimicrobial and anticancer activity [1-6]. There are few structural characteristics which allow these compounds to exert such activity: a planar tricyclic core; the presence of a carbonyl functionality on the central ring and biaryl-ether modifying the electronics of this system and the possibility of its elaboration with a variety of substituents.

The aim of this study was to design and synthetize new structurally different xanthone derivatives and to determine their cytotoxic activity against human leukemic cells. One of the reason why we chose leukemic cells was that they tend to be resistant to chemotherapeutic agents and become refractory, resulting in failure of treatment [7]. The second reason was the results of literature review, which revealed that among xanthones – predominantly natural compounds (such as mangiferin, gambogic acid or psorospermin) leukemia are the subject of research in this field [8-10].

Six new xanthone derivatives were designed and synthesized. Xanthone core was modified with four amine substituents. Four compounds were additionally substituted with chlorine atom(s) at xanthone core. Next the drug like properties of the titled compounds were estimated by *in silico* methods. Finally, the cytotoxic effect of xanthone derivatives (at various concentration) on four different human leukemia cell lines was assessed. The changes in cell viability of the following leukemic cell lines were analyzed: HL-60 (acute promyelocytic leukemia), U937 (human promonocytic leukemia, isolated from histiocytic lymphoma), ML-1 (acute myeloblastic leukemia) and MOLT-4 (acute lymphoblastic leukemia).

All tested compounds decreased cell viability of each leukemic cell line at conc. below 25 μM. The IC_{50} values were in the range of 6.03-25.0 μM. The differences in cytotoxic activity of the agents against leukemic cell lines predominantly depended on the structural modification of xanthone core, less on the cell line type. Further experiments are planned for a better understanding of the new xanthone derivatives activity against human pathological cells .

Acknowledgements
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P95

The potential antifungal effect of newly synthesized derivatives containing the cyclopropane system

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Recently, the occurrence of fungal infections caused by *Candida* spp., has increased dramatically, especially in immunocompromised patients. Additionally, their treatment is often ineffective due to the resistance of yeasts to antimycotics. Moreover, the list of the commertially available antifungal agents, used for the treatment of candidiasis is limited to three major classes: polyenes, azoles and echinocandins. Therefore, there is a need to search for new antifungals [1, 2]. A series of nine newly synthesized thiazole derivatives containing the cyclopropane system, showing promising activity against *Candida* spp., has been further investigated [3]. We decided to verify their antifungal activity towards clinical *Candida albicans* isolated from the oral cavity of patients with hematological malignancies, investigate the mode of action on fungal cell, the effect of combination with the selected antimycotics (nystatin, chlorhexidine and thymol) and toxicity to erythrocytes. These studies were performed by the broth microdilution method, test with sorbitol and ergosterol, checkerboard technique and erythrocyte lysis assay, respectively [4, 5]. All derivatives showed very strong activity (similar and even higher than nystatin) against *C. albicans* isolates with minimal inhibitory concentration (MIC) = 0.008 – 7.81 μg/mL. Their mechanism of action may be related to action within the fungal cell wall structure and/or within the cell membrane. MICs of compounds alone, as well as MICs of combinations which exhibited inhibitory effects, were used to calculate fractional inhibitory concentrations (FICs) and ΣFIC (FICI – FIC index) values. The interactions between the derivatives and the selected antimycotics showed additive effect in the case of combination some of them and thymol (ΣFIC = 1) against reference yeast strains. In turn, the combinations of the studied compounds with nystatin (ΣFIC = 2 – 4) and chlorhexidine (ΣFIC = 2 – 3) were found to be noninterfering (FICI values between 1 and 4 were considered as indifferent). Moreover, the erythrocyte lysis assay confirmed the low cytotoxicity of these compounds as compared to nystatin. The present studies confirm that the studied thiazole derivatives containing the cyclopropane system appear to be a very promising group of compounds in treatment of infections caused by *C. albicans*. However, this requires further studies *in vivo*.

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P96

Molecular basis of antitumor activity of novel transition metal complexes with berenil and nitroimidazole

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Imidazole ring is a known structure in many natural or synthetic drug molecules and its metal complexes can interact with DNA and do the cleavage. The biological activity of berenil seems equally interesting. According to our studies, platinum(II) complexes with berenil have comparable or greater anti−tumor activity compared to cisplatin with less drug toxicity. Therefore, the goal of this study was synthesis and anticancer activities of novel transition metal (Pt, Pd, Au) complexes with berenil and nitroimidazole.

The cytotoxic activity of the novel complexes was examined using the MTT method of Carmichael. The results showed higher cytotoxicity of these compounds in comparison with cisplatin. The complexes in which the central atom was palladium or platinum appeared to be the compounds of highest cytotoxicity in relation to neoplastic breast cancer cells MCF-7. In relation to human normal breast epithelial cell MCF-10A the new complexes were characterized with lower cytotoxicity than in case of examined neoplastic cells. Additionally our experiments carried out with flow cytometry assessment of annexin V binding and propidium iodide revealed that these complexes inhibited the proliferation of breast cancer cells by increasing the number of apoptotic cells. In addition, we have observed that these complexes selectively concentrate in tumor cell mitochondria due to characteristic for these cells increased membrane potential which may explain the increased proapoptotic activity of these compounds. We have further confirmed the effectiveness of the synthesized compounds by our team on zebrafish embryo xenograft model. These results indicate that novel transition metal (Pt, Pd, Au) complexes with berenil and nitroimidazole may constitute a new class of compounds with potential antitumor activity.

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