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# Abstracts

*Section 1 - Bioorganic and Pharmaceutical Chemistry*

EFSA-CDN

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**BPC1**

# INFLUENCE OF LIPOPHILIC SUBSTITUTION ON THE PYRAZINE CORE OF PYRAZINAMIDE ON ANTIMYCOBACTERIAL ACTIVITY

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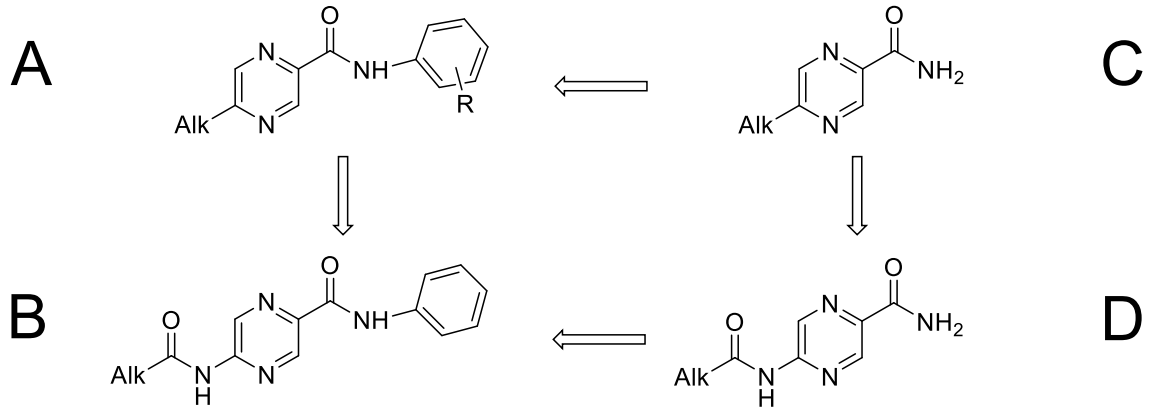
As part of our long-term investigation in preparing potentially active antimycobacterials, we report the design, synthesis, and antimicrobial evaluation of a series of 5-alkyl and 5-alkylamido derivatives of the first-line agent, pyrazinamide. The series is divided into four main general structures, refer to the figure below. For general structure C, we also prepared the 5-alkylpyrazine-2-carboxylic acid derivatives. All prepared compounds were evaluated for their *in vitro* antimycobacterial activity against *Mtb* H37Rv, and the most active compounds were evaluated for their cytotoxicity in HepG2 cell line. Enoyl reductase (InhA) was explored as a possible target for prepared compounds. Furthermore, obtained activities were compared to previously published alkyl derivatives at positions 3 and 6 of the pyrazine core. Further results are discussed in the presentation.




*The study was supported by EFSA-CDN, grant number CZ.02.1.01/0.0/0.0/16_019/0000841, co-funded by ERDF; and by the Ministry of Health of the Czech Republic, grant nr. NU21-05-00482.*

# SYNTHESIS OF PURINE DERIVATIVES WITH ANTIMYCOBACTERIAL ACTIVITY

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Tuberculosis (TB) is one of the top 10 causes of death worldwide from a single infectious agent. The World Health Organization estimated 10 million new cases and 1.2 million deaths from TB in 2020.[1] Some strains of mycobacteria causing TB show numerous resistances to first-line drugs (isoniazid /INH/ and rifampicin) and to second-line drugs (fluoroquinolones, amikacin, bedaquilin, etc.). The development of new anti-TB drugs with new mechanism of action is necessary to improve TB therapy and to fight against resistant TB as well. We identified compound K1297 with good anti-TB activity with minimum inhibitory concentration $MIC_{99}$ = 4 μM against $H_{37}R_v$ strain (for comparison, $MIC_{99}$ /INH/ = 0.5 μM). The main structural motif of this molecule is purine scaffold, which was modified and functionalized to elucidate the structure-activity relationships (SAR) and to identify derivatives with low toxicity and higher efficiency than the initial hit K1297. The effects of individual structural fragments on *in vitro* antimycobacterial activity, toxicity and selectivity of action have been evaluated. Finally, derivatives with optimized activity/toxicity ratio have been found and their pharmacokinetic profile and *in vivo* efficacy will be evaluated. In the cooperation with foreign collaborators, we have also determined the mechanism of action of these compounds using genome sequencing of the resistant mutants and metabolic radiolabelling experiments.

*The study was supported by the project "Grant Schemes at CU" (reg. no. CZ.02.2.69/0.0/0.0/19_073/00169359)*

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# 4-AMINOSALICYLIC ACID HYBRID COMPOUNDS AS POTENTIAL ANTITUBERCULOTICS

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Tuberculosis, caused by Mycobacterium tuberculosis, is the leading cause of death worldwide from a single infectious organism and a major threat to public health due to growing antimicrobial resistance.[1] Tuberculosis was effectively treated with first-line anti-TB drugs; however, due to the raising antimicrobial resistance, newer approaches to eradicate the disease are needed.

4-Aminosalicylic acid is a second line agent for tuberculosis. In presented series we attempted hybrid compounds bearing this moiety. Title compounds are based on positional derivatives of picolinic acid linked to 4-aminosalicylic acid or 4-aminobenzoic acid by amidic bond. Compounds were tested for biological activity against selected strains of Mycobacterium (M. tuberculosis H37Rv, M. tuberculosis H37Ra, M. kansasii, M. avium, M. smegmatis, M. aurum). The minimum inhibitory concentration (MIC) for tested mycobacterial strains was determined for all tested compounds beside isoniazid, ciprofloxacin and rifampicin as a reference. Results of the biological testing and structure-activity relationships are discussed in the presentation.

*The study was supported by the Ministry of Education, Youth and Sports of the Czech Republic (SVV 260 547) and EFSA-CDN (Grant CZ.02.1.01/0.0/0.0/16_019/0000841) cofunded by ERDF (Dr. Ghada Bouz).*

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# DESIGN, SYNTHESIS, AND SAR OF 3-AMIDOPYRAZINE-2-CARBOXAMIDES AS ANTIMICROBIALS

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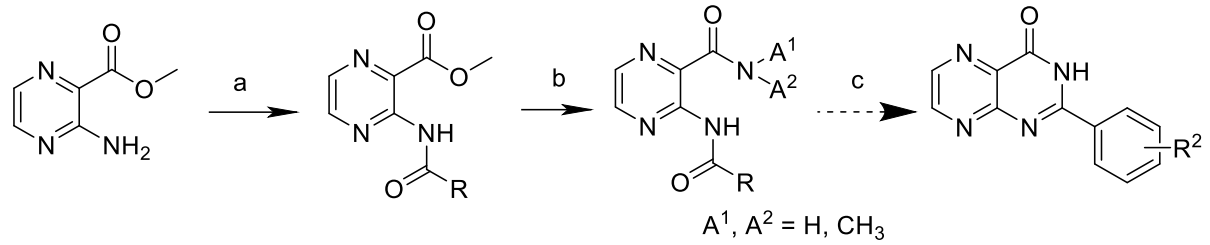
We present the design and synthesis of novel 3-amidopyrazine-2-carboxamides and their respective cyclic derivatives, along with their biological evaluation. The synthesized compounds were prepared according to Scheme 1 and evaluated for their *in vitro* activity against various mycobacterial strains and other strains of pathogenic bacteria and fungi. The active compounds 3-amidopyrazine-2-carboxamides (**3**) (where R is a substituted phenyl) with unsubstituted carboxamide at C-2. The most active compounds exerted MIC (Minimum Inhibitory Concentration) ranging from 1.98 to 7.81 μg.mL$^{-1}$ and are highly selective towards *Mycobacterium tuberculosis* (both H37Rv and H37Ra strain) inhibition (over other bacterial strains). The synthesized compounds were non-toxic on HepG2, in contrast to confirmed human prolyl-tRNA synthetase inhibitor, previously described in the literature.[1] No significant activity was observed for cyclic derivatives (**4**).




Scheme 1. Synthesis of final compounds

a) Acyl chlorides/ pyridine in ACN/ argon; b) 2M Ammonia in EtOH; or $CH_3NH_2$ in EtOH; or $(CH_3)_2NH$ in EtOH
c) KOH, $H_2O$, DMSO

*The study was supported by the Charles University, project GA UK No. 349721 and project SVV 260 547.*

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**BPC5**

# SYNTHESIS AND EVALUATION OF 2,5-DISUBSTITUTED 1,3,4-OXADIAZOLES AS POTENTIAL ANTITUBERCULOTICS

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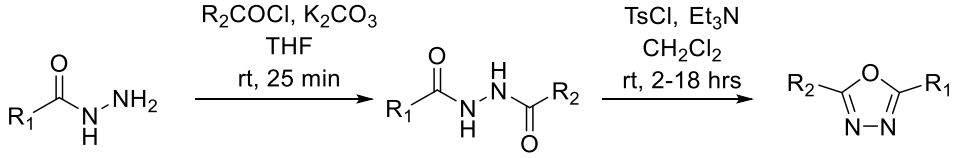
Many new structures that contain the 1,3,4-oxadiazole ring have been studied as potential bioactive agents. Compounds with this structural motif also target *M. tuberculosis* (*Mtb*.), including resistant strains.[1] We have prepared a series of 2,5-disubstituted 1,3,4-oxadiazoles, many without structural similarity to any drug developed so far. Asymmetric 1,2-diacylhydrazides were prepared by the reaction of monosubstituted (acyl) hydrazine ($R_1$; commercially available or in-house prepared) with the appropriate acyl chloride ($R_2$) in the presence of a base using anhydrous tetrahydrofuran (THF) as the solvent. 2,5-Disubstituted oxadiazoles were obtained directly by dehydrative cyclization of 1,2-diacylhydrazides (Scheme 1). The compounds were evaluated for their *in vitro* antimycobacterial activity against drug-susceptible *Mtb*. H37Rv, non-tuberculous mycobacteria (NTM; *M. avium*, *M. kansasii*) and panel of eight *Mtb*. strains with various resistance profiles. The mechanism of action of the most potent compounds was investigated. No activity against Gram-positive and Gram-negative bacteria, as well as fungal pathogens, was identified.

Scheme 1




*The study was supported by the Czech Science Foundation (reg. No. 20-19638Y), Charles University (SVV 260 547) and "Grant Schemes at CU" (reg. no. CZ.02.2.69/0.0/0.0/19_073/00169359).*

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**BPC6**

# DESIGN, SYNTHESIS, AND PRELIMINARY BIOLOGICAL EVALUATION OF NEW BORONIC ACIDS DERIVATIVES

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The presented compounds follow up the research of a previously studied series of hybrid compounds, that exerted high *in vitro* antimycobacterial activity. They were designed as hybrids combining pyrazinamide and *para*-aminosalicylic acid.[1] The current study is focused on the combination of pyrazinamide with 4-aminophenyl boronic acid, bioisoster of *para*-aminobenzoic acid or with 6-aminobenzoxaborole, crucial pharmacophore of several antimicrobials. The use of free boronic acid or benzoxaborole moiety could afford reversible covalent bonds towards the potential targets in microorganisms.

The compounds were synthesized by condensation of 4-aminophenyl boronic acid pinacol ester or 6-aminobenzoxaborole with variously substituted heteroaromatic acids that underwent the previous activation. The subsequent deprotection of boronic acid pinacol ester proceeded smoothly to afford novel compounds.[2]

The synthetic products and the isolated condensation intermediates were subjected to biological *in vitro* screening against fungi and bacteria, including mycobacteria. Some of the compounds showed moderate antimycobacterial activity.

*The study was supported by SVV 260 547.*

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# DIENE TRANSMISSIVE DIELS-ALDER REACTION OF ELECTRON-POOR DENDRALENES

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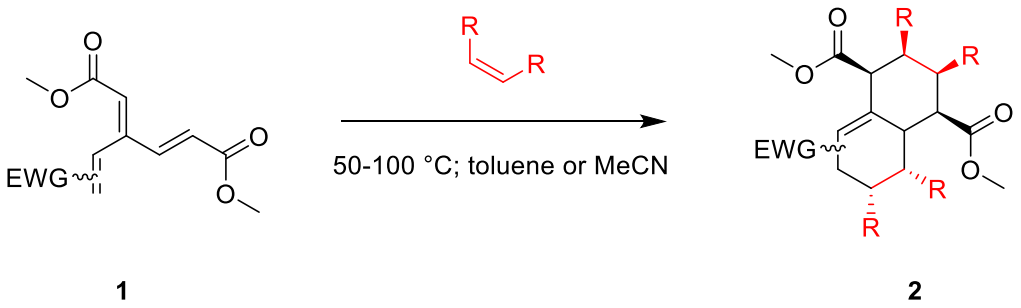
Dendralenes[1] **1** (Scheme 1) are group of cross-conjugated, acyclic polyenes. Unlike other polyenes, their structure allows them to undergo not only standard Diels-Alder (DA) reaction but more importantly Diene Transmissive Diels-Alder Reaction (DTDA). In this fashion the simplest [3]dendralene (number define number of cross-conjugated double bonds) can easily form a complex bicyclic products 2 with up to 8 new chiral centers. With the increasing size of the dendralene the possible complexity of the products rises as well.

Our work includes preparation of [3]dendrales with electron-withdrawing groups, and investigation of their reactivity in DA/DTDA as well as other reactions. In terms of DTDA, our dendralenes exhibit high stereoselectivity accompanied with generally high, easily isolable yields in reactions with selected dienophiles.




**1** **2**

Scheme 1. General structures

*This work was supported by the project EFSA-CDN (Reg. No. CZ.02.1.01/0.0/0.0/16_019/0000841) co-funded by the European Union.*

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# IN SITU GENERATION AND REACTIONS OF HETEROCYCLIC DENDRALENES

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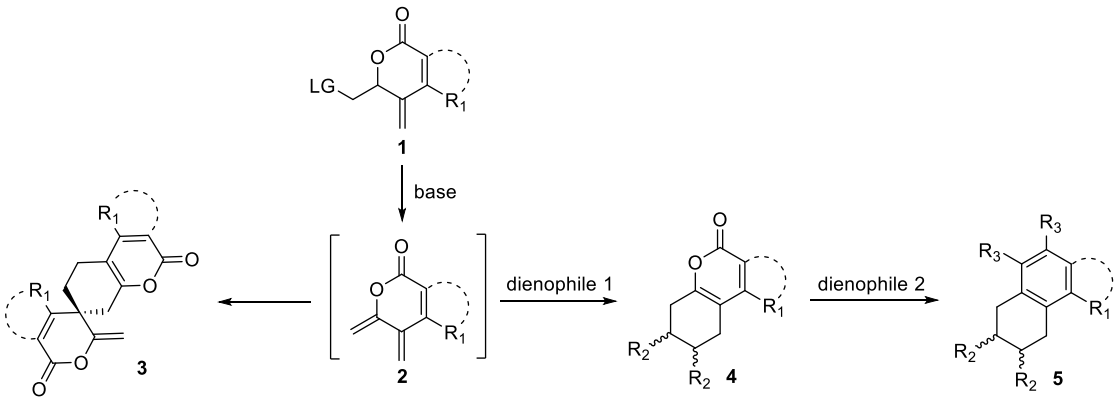
General structure of lactone **1**[1] (Scheme 1), binding an appropriate leaving group (e.g. iodide or mesylate), serves under basic conditions as a precursor of non-isolable and highly reactive exocyclic dienes or [3] and [4] dendralenes **2** (depending on side cyclic or acyclic chain $R_1$). Such intermediates act as substrates of diene-transmissive Diels-Alder cycloadditions. Depending on the presence of a dienophile in the reaction mixture, spirocyclic bislactone **3** or substituted lactone **4 (**sharing structural similarity with a number of biologically active natural products)[2,3] can be isolated. Formation of a new diene moiety in the molecule **4** allows another Diels-Alder cycloaddition to get a more complex structure **5**.

Scheme 1




*The study was supported by Charles University (SVV reg. No. 260 547 and GAUK reg. No. 205007).*

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**BPC9**

# NOVEL SYNTHETIC APPROACHES TOWARD AMARYLLIDACEAE ALKALOIDS AND DERIVATIVES

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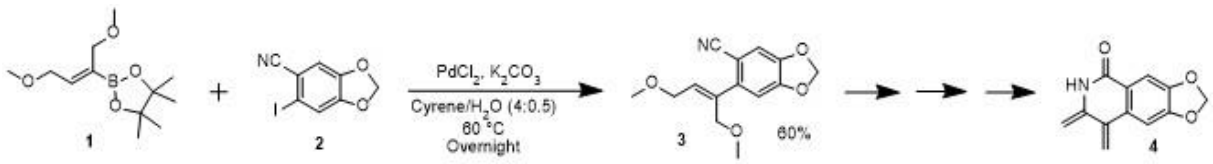
*Amaryllidaceae* alkaloids (AAs) are one of the most diverse groups of secondary metabolites that can be found in living organisms and possess a wide array of structure types, biosynthetic pathways, and pharmacological activities.[1]

Innovative approaches towards the synthesis of these alkaloids (AAs) and other biologically active isoquinoline derivatives were designed. The first strategy was based on the development of a unique Pd-catalyzed cross-coupling/allylic isomerization sequence leading to the precursor of a highly reactive isoquinoline radialene **4**. Due to disappointing results, we attempted to develop a new, green chemistry approach. The reaction starts with Suzuki coupling, where a novel combination of reagents and recyclable solvent has been employed with good results. After that, a cascade of reactions was performed to synthesize the AAs precursor **4** (Scheme 1).

Scheme 1




*The study was supported by Charles University (SVV 260 547, GAUK 362421) and the Czech Science Foundation (Project No. 18-17868S).*

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# TRIPLE LABELED ODN PROBES: ADVANTAGE OR NOT?

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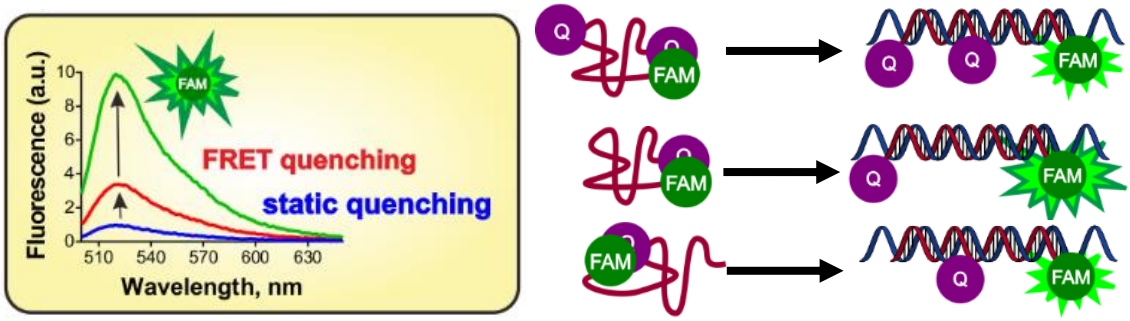
Long oligodeoxynucleotide (ODN) probes are used for the detection of viral genetic information. The advantage of long ODN probes lies in the possibility of recognizing viruses even though they have several point mutations. ODN probes for Taq-man assays are usually consist of ODN strand with one fluorophore and one quencher. However, several studies indicate that the presence of one fluorophore and two quenchers is beneficial for quenching efficiency.[1] In our study, we compare the presence of one or two quenchers to quenching efficiency. Two different structures of quenchers were chosen for comparison – the first one, commercially available BlackBerry® Quencher 650, and the second one was quencher synthesized in our group – tetrapyrazinoporphyrazine.[2] We tested different positions of quencher - *e.g.*, two quenchers (intrastrand and 3′-end), only intrastrand quencher, or only quencher at 3′-end. After analysis of quenching efficiency in random coil and after hybridization, we observed results that were in contrast with observation other groups because presence of the second has not a significant role in quenching efficiency.




*The study was supported by Dean's fund (PROGRES Q42).*

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# STUDYING PROPERTIES OF SUPRAMOLECULAR COMPLEXES OF PHTAHLOCYNINES WITH CUCURBITURIL

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Phthalocyanines (Pcs) are macrocyclic compounds structurally related to porphyrins, which are used also as photosensitizers in photodynamic therapy. Pcs have planar structure and because of π-π interactions they form aggregates in water environment and therefore lose their photodynamic activity. Based on formation of a supramolecular complex with cucurbiturils (CBs), we can potentially improve solubility and decrease aggregation of Pcs in water. CBs are pumpkin-shaped macromolecules composed of methylene bridged glycoluril oligomers[1] with hydrophobic cavity and hydrophilic portals where they can bind appropriate guest molecules.

In this project, we used one of the strongest reported supramolecular interactions between CB[7] and 1-aminoadamantane[2] as substituent on the Pc ring.

Seven zinc Pcs, peripherally or non-peripherally substituted with aminoadamantane, were prepared. Their absorption spectra showed substantially improved (but not complete) monomerization in water after addition of four equivalents of CB[7]. Biological tests on HeLa cells did not show higher photodynamic activity of Pc-CB[7] complex even though higher monomerization should also result in higher activity. Lower activity of Pc-CB[7] complex was due to lower cell uptake of this complex.

*The study was supported by SVV 260 547 and Czech Science Foundation (20-09212S).*

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# OLIGODEOXYNUCLEOTIDE PROBES LABELING: INFLUENCES ON A CLICK REACTION EFFICIENCY

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Labelling of oligodeoxynucleotides (ODN) chains is a crucial process of preparation of diagnostic or theranostic probes. Unfortunately, this procedure is nowadays the most problematic step in otherwise highly efficient process carried out on solid phase in oligo synthesizers. The wide diversity of labels together with large ODN chain demands highly specific yet mild conditions during the labelling process. One of the most efficient approach is copper-free click reaction through Huisgen's cycloaddition.[1] Though its exceptional reaction kinetics, there is significant yield fluctuation.

We decided to investigate the influence of different conditions on the efficiency of the click reaction in ODN probes preparation. More than 350 probes were labeled, deprotected, and purified. Four types of molecules (azaphtalocyanine, BODIPY, acridine, and cyanine dye derivative) were used for the labelling in five diferent concentrations (10 uM to 100 mM). Three positions in the strand of a 24-base identical sequence were tested (2, 13, and 24, counted from 3´-end of oligonucleotide). An influence of solid phase support was observed using two commercially most frequently used types (controlled pore glass, polystyrene) and their various porosity. Further more the time efficiency of most promising condition combination was deeply investigated. And special attention was paid to deprotection method of highly base sensitive dyes.

Experiments prooved that hydrophilic/-phobic compatibility of solid phase support and the label has the crutial impact on the concentration of label needed for fully labelled ODN probe preparation decreasing the commertialy recommended concentration even 15 times.

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# SYNTHESIS OF NEW UNSYMMETRICAL PHTHALOCYANINES FOR PHOTOINDUCED DRUG RELEASE

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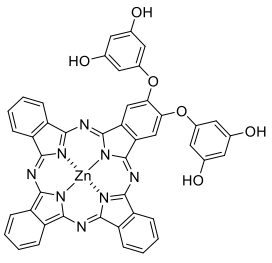
Phthalocyanines (Pcs) are group of organic dyes with interesting photophysical properties (strong absorption in area over 600 nm and strong singlet oxygen production). Thus, they can be used as photosensitizers (PSs) in photodynamic therapy.

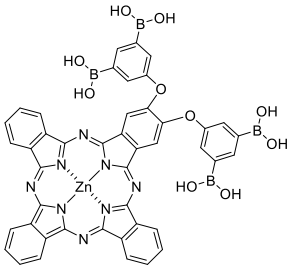
The aim of this project was the synthesis of novel unsymmetrical Pcs. Due to their amphiphilic character, they can be incorporated into bilayer of liposomes (which can already have some hydrophilic cargo inside). Upon activation of the Pc by light, the reactive singlet oxygen is produced. This destroys liposomal membrane and releases cargo (hydrophylic anticancer drug) only in the irradiated area. This process is called photoinduced drug release.

Precursor for Pc 1 was obtained by nucleophilic substitution of 4,5-dichlorophthalonitrile with 3,5-dimethoxyphenol. Unsymmetrical magnesium complex of Pc 1 was prepared by statistical condensation of phthalonitrile with 4,5-disubstituted precursor and magnesium butoxide as initiator. The reaction led to the mixture of six different congeners. The required congener of ABBB type was easily isolated from the mixture by chromatographic methods. Magnesium complex will be converted to metal-free ligand and then to zinc complex. Final step will be deprotection of protective groups. Precursors for Pcs 2 and 3 were also prepared by nucleophilic substitution and synthesis of final Pcs will be optimized.

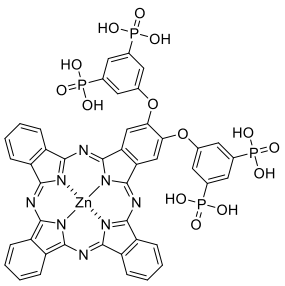
*The study was supported by Czech Science Foundation, grant. No. 19-14758Y and SVV 260 547.*



**Pc 1**



**Pc 2**



**Pc 3**

**BPC14**

# SYNTHESIS OF NEW ACRIDINE-4-CARBOXAMIDES

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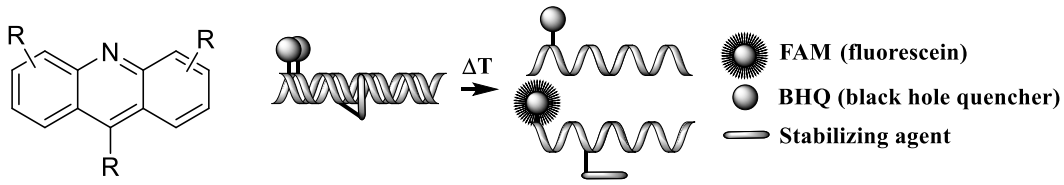
Melting temperature difference ($\Delta T_m$) between complementary and mismatched duplex plays a crucial role in discrimination of point mutations or single nucleotide polymorphisms. The $\Delta T_m$ is decreasing with the length of oligodeoxynucleotide. From this point of view, shorter oligodeoxynucleotide probes are advantageous due to higher $\Delta T_m$ compared with longer probes. On the other hand, their low melting temperature is the main disadvantage. Stabilizing agents are used for elimination of the disadvantage. There are three types of stabilizing agents that can be used for thermal stabilisation of oligodeoxynucleotide duplexes: polyamines, minor groove binders, and intercalators. In our work we focused on preparation of new acridine derivatives. New type of acridine derivative bearing azido group within the sidechain connected through C-C bond was proposed due to known instability of the C-N bond in position 9 of acridine core[1], during oligonucleotide deprotection by $NH_4OH$. A synthetic procedure for preparation of precursor and acridine derivatives was developed and the new acridine derivatives were prepared. Acridine derivative FK-106 was tested for activity in solution and compared to the best acridine derivatives from previous series. FK-106 was comparable or better than the best derivative from previous series (FK-8). FK-106 was than covalently attached to the oligodeoxynucleotide probes by click chemistry. The stability of FK-106 in presence of $NH_4OH$ was proved. Probes were tested at melting system, and at PCR conditions.




*The study was supported by The Grant Agency of Charles University (reg. No. 994218) and The Technology Agency of the Czech Republic (reg. No. TH03010251).*

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**BPC15**

# SYNTHESIS AND CHARACTERIZATION OF THIOL-TERMINATED CERAMIDES FOR GOLD NANOPARTICLES COATING

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The main skin permeability barrier is situated in the uppermost layer, the stratum corneum, which is composed of flattened cells, corneocytes, embedded in a hydrophobic lipid matrix. This lipid matrix is composed of a mixture of ceramides, free fatty acids and cholesterol. Certain ceramides are covalently anchored to crosslinked proteins on the corneocyte surface[1]. The role of this "first lamella" is not entirely explained yet, but it is considered to provide a scaffold for the orientation of free extracellular lipids. In this project, we aim at the construction of model membranes containing lipid-coated gold nanoparticles surrounded by isolated human skin lipids to study the mutual interactions between the anchored and free lipids. To achieve this goal, first, modified ceramides containing a terminal thiol group for the attachment to the gold nanoparticles were prepared. Ceramide precursors were constructed from 16-hexadecanolide by multiple reaction steps up to the stage of succinimid-1-yl 32-hydroxydotriacontanoate[2]. This product was then converted to the thiol derivative by a subsequent halogenation/tosilation and a reaction with thiourea/hexamethyldisilathiane. The final modified ceramide molecule will be prepared by an *N*-acylation reaction with a suitable sphingoid base and it will be attached to gold nanoparticles to imitate the corneocytes behavior in physiological conditions. The gold nanoparticles will be characterized by $^{1}$H-NMR, $^{13}$C-NMR, scattering techniques and UV-vis spectroscopy. Model lipid membranes to probe the interactions between free and bound lipids will be constructed in later stages of the project.

*The study was supported by the Czech Science Foundation (19-09135J).*

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**BPC16**

# LOSS OF LINOLEATE TAIL IN OMEGA-O-ACYLCERAMIDES CHANGES THE MOLECULAR ARCHITECTURE OF SKIN BARRIER LIPIDS

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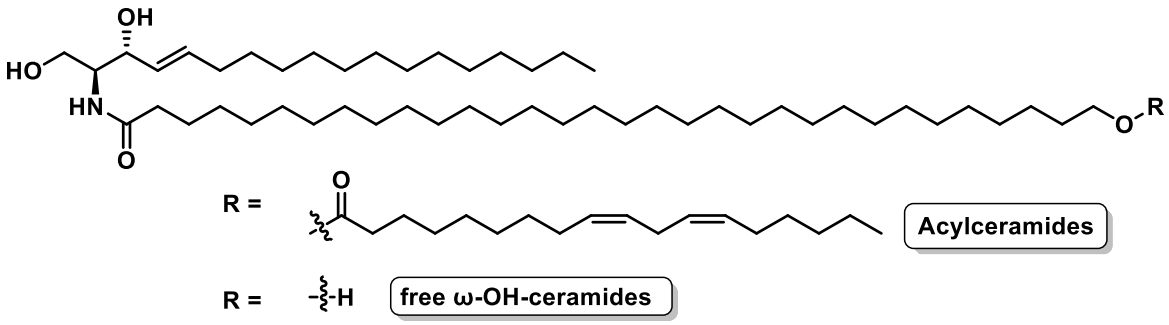
Acylceramides are essential for the correct multilamellar lipid organization and proper skin barrier formation in the outermost skin layer, *stratum corneum*. The complete depletion of acylceramides is lethal whereas their decreased levels were found in several skin disorders like atopic dermatitis or ichthyoses. The linoleate tail is attached to ω-OH group of ceramides via PNPLA1 enzyme. Dysfunctional *Pnpla1* leads to significantly decreased levels of acylceramides and accumulation of free ω-OH-ceramides.




In this study we successfully modified and optimized the synthesis of ω-OH-ceramides. Furthermore, the effect of linoleate tail and its absence on lipid organization was studied. Electron microscopy revealed a loss of lamella pairing in lamellar bodies (lipid transport vesicles) and disrupted lamellar organization in *Pnpla1*$^{-/-}$ mice epidermis. X-ray diffraction showed different lamellar arrangement in model membranes upon the linoleate tail loss, such membranes were unable to provide the physiological long periodicity phase and were less thermally stable. Models lacking the linoleate tail also proved to be in general more permeable for model drugs and water, especially under stress conditions.

*The study was supported by the Czech Science Foundation (19-09135J) and by Charles University (GAUK 1194119)*

# CERAMIDE-DECORATED DENDRIMERS AS CORNEOCYTE LIPID ENVELOPE MIMICKING MODEL

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Skin is the biggest organ in human body and protects it from excessive water loss while hampers the entrance of undesired substances, as allergens and microbes. Skin's outermost layer, stratum corneum (SC), holds the principal skin barrier. Human SC consists of flattened dead cells known as corneocytes, that are embedded in a hydrophobic lipid matrix. Corneocytes are having ceramides attached to their surface which are forming the so called "corneocyte lipid envelope" (CLE).

Most research strategies to mimic the SC function, are taking only the lipidic matrix into consideration. In our approach, we are focusing on the development of CLE mimicking compounds. The further goal is to create a better SC model which would incorporate CLE mimicking entities together with the lipidic matrix.

In our first approach, we plan to immobilize ultralong-chain ceramides on PAMAM dendrimers of fourth generation. For the development of an effective synthetic protocol, long aliphatic chain alcohols were used as ceramide mimicking compounds, in order to undergo different modifications and be coupled with PAMAM dendrimers.

We attempt to immobilize ultralong chain ceramides on 4th generation PAMAM dendrimers to study the role of the corneocyte lipid envelope, a structure vital for mammalian survival on dry land. We started coupling PAMAM with long-chain alcohols to probe the conditions for the lipid anchoring. Copper-catalyzed alkyne azide cycloadditions were not successful as well as various other protein-lipid coupling approaches. We succeeded with activating the alcohol with N,N-disuccinimidyl carbonate and coupling it with the PAMAM peripheral amino groups. This strategy will be exploited to prepare the PAMAM-ceramide conjugates and their deuterated or fluorescent counterparts to study the possible scaffolding/signaling roles of this peculiar skin barrier structure.

*The study was supported by GAUK 262821 and Czech Science Foundation (GACR 19-09135J)*

**BPC18**

# 2-AMINOOXAZOLES AS A VIABLE ALTERNATIVE TO 2-AMINOTHIAZOLES IN ANTIMICROBIAL AGENTS

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Antimicrobial drug resistance is currently one of the most critical health issues. Pathogens resistant to even last-resort antibiotics are increasing, and very few effective antibacterial compounds have been introduced in recent years. The promising drug candidates are often discontinued in the primary stages of the drug discovery pipeline due to their unspecific reactivity (PAINS), toxicity, insufficient stability, or low water solubility. In this work, we investigated a series of 2-aminothiazoles and their 2-aminooxazole isosteres to potentially overcome the above-mentioned problems.

Synthesis (see **Figure 1**) followed conventional methods and led to 30 final compounds. All derivatives were tested against several microbial strains, including clinical drug-resistant (myco)bacterial isolates. Several compounds showed high activity against mycobacteria (MIC < 3.91 μg/mL), and some also showed promising results against various bacterial strains. None of the compounds was significantly cytotoxic against Hep G2 cell lines. The lipophilicity of the derivatives was experimentally measured and correlated with the observed activity.

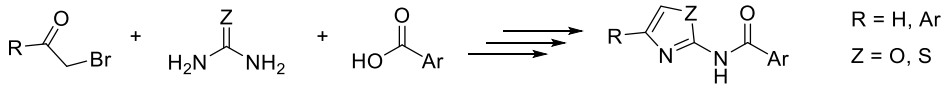


**Figure 1.** Synthetic diagram and general structure of the investigated derivatives

*This study was supported by the Grant Schemes at CU (reg. no. CZ.02.2.69/0.0/0.0/19_073/0016935).*

# ANTIBACTERIAL EFFECTS OF OXAZOLE DERIVATIVES

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Oxazoles are essential intermediates for the synthesis of new chemical entities with a wide spectrum of biological activities. [1] In our project, a collection of 14 freshly synthesized oxazole derivatives were subjected to the basic antibacterial screening. 8 reference bacterial strains were employed to determine the minimal inhibitory concentrations (MIC) of these derivatives. 2 compounds have shown moderate activity against reference bacterial strains. One of them, designated AB15, was selected for advanced *in vitro* studies contributing to an anti-infective drug discovery research. Then, 18 clinical isolate strains (kindly provided from the Faculty Hospital in Hradec Králové) have been employed in the confirmation and evaluation of MIC. The most promising activity was revealed predominantly against specific strains of *Acinetobacter baumannii* (MIC = 31.25 μmol/L) and *Pseudomonas aeruginosa* (MIC = 62.5 μmol/L). To reveal a possible synergic effect of AB15 with commercially available antibiotics, the checkerboard assays have been included, as well. In summary, 6 antibiotic agents with various mechanisms of action and commonly used in the treatment of infections caused by Gram-negative pathogens have been selected. For the comprehensive insight into biological activities of AB15, anti-biofilm activity evaluation, *in vivo* toxicity, or evaluation of *in vivo* efficacy will also be included.

*The study was supported by the "Grant Schemes at CU" (reg. no. CZ.02.2.69/0.0/0.0/19_073/0016935) and Ministry of Health of the Czech Republic, grant nr. NU21-05-00482.*

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# SYNTHESIS OF NOVEL INHIBITORS OF PROLYL-TRNA SYNTHETASE

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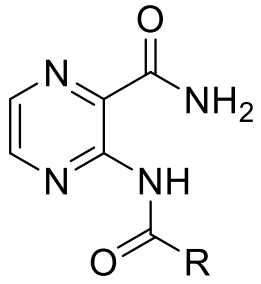
This work relates to the effort to synthesize novel inhibitors of prolyl-tRNA synthetase (PRS). Such inhibitors could have a wide scale of use, including antimicrobials. The design started from a confirmed inhibitor of human prolyl-tRNA synthetase (hPRS) based on the 3-aminopyrazinamide fragment.[1] This inhibitor binds to the ATP site of the hPRS. Our design of compounds focuses more on performing similar interactions as the ATP cofactor. To achieve that, we need some polar substitutions in the R-chain of our compounds (see the structure). We supported this idea by *in silico* simulations. For successful synthesis, it is necessary to keep anhydrous conditions and protect hydroxy or amino substituents with protective groups before the acylation step. Synthesized compounds will be tested for antimicrobial activity and cytotoxicity. We will try to confirm the binding of our compounds in the ATP site of PRS.




R = chains with hydroxy or amino substitutions

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# AN INTEGRATED COMPUTATIONAL APPROACH TO THE DISCOVERY OF A NEW TGR5 AGONIST

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TGR5 is a promising breakthrough in type 2 diabetes therapy [1,2]. Most attempts to develop novel TGR5 agonists were focused on the modification of bile acids which are endogenous ligands of TGR5. The cryo-electron microscopy structures of TGR5 are expected to facilitate the finding of TGR5 agonists through a computational approach [3]. This study aims to find completely new TGR5 agonists employing a structure-based pharmacophore modeling approach and docking-based virtual screening against CHEMBL and ZINC libraries. Candidate molecules obtained in previous stages underwent interaction analysis using molecular docking and molecular dynamics simulations. Three candidate molecules emerged as showing the best docking score and interaction similar to that of the original TGR5 ligand. In silico examination of ADMET showed that these hits could potentially serve as TGR5 agonists.

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**BPC22**

# CALCULATION OF NMR SPIN-SPIN COUPLINGS FOR INTRINSICALLY DISORDER PROTEINS: A PROSPECTIVE TOOL TO FACILITATE EXPERIMENTAL NMR STUDIES

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Intrinsically disorder proteins (IDPs) are post-translationally modified polypeptide chains that fail to shape single well defined three dimensional (3D) structures. The phosphorylation and de-phosphorylation of IDPs adjust a great variety of molecular processes that are linked to the development of neurodegenerative disorders such as Alzheimer's and Parkinson's. The theoretical modeling of NMR spectroscopy parameters such as the indirect spin-spin coupling constants assists the interpretation of experimental NMR data and thus also the structure characterization of the proteins.

In this work, we perform multiscale calculations of the 3JHN-Ha spin-spin couplings for the protein fragment Tau(210-240) that belongs to the IDP family. The multiscale calculations build on the sampling of IDP configurations by molecular dynamics (MD) in order to account for the flexibility of the system. The MD simulation is used to construct a structural ensemble for density functional NMR calculations. The computation of J couplings employ molecular clusters that we construct through a fragmentation by the adjustable density matrix assembler (ADMA) technique. In our contribution, we will demonstrate the performance of the computational procedure through a comparison against experimental NMR data as well as against predictions made based on empirically parametrized equations.

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