ICCBIKG 2021

Conference Programme

BBMC.2A.9– Effect of plant extracts on bacterial growth and potential mechanism of action– *Stanković M, Pribojac J, Terzić J, Stefanović O.*

BBMC.2A.10– DNA and BSA interactions of copper(II) and zinc(II) complexes with antifungal agent fluconazole– *Stevanović N, Stanković M, Andrejević T, Ašanin D, Stanojević I, Glišić B.*

BBMC.2A.11– DNA/BSA binding study of dinuclear gold(III) complexes with aromatic nitrogen-containing heterocycles as bridging ligands–*Andrejević T, Ašanin D, Savić N, Stevanović N, Djuran M, Glišić B.*

BBMC.2A.12– *In silico* prediction of pharmacokinetic properties and druglikeness of novel thiourea derivatives of naproxen– *Nedeljković N, Dobričić V, Mijajlović M, Radić G, Nikolić M, Stanković A, Vujić Z.*

Session AST.2A: 09:30-10:15 (Room 2)

Applied sciences and technologies (Poster session)

Chair: Gorica Cvijanović

AST.2A.1– Quantification of Essential Elements in *Papaver Rhoeas* L. and *Papaver Somniferum*– *Uđilanović M, Vasiljević S, Šimšić Z.*

AST.2A.2– Effect of humic and fulvic acids on microbial growth– *Radulović M, Mitrovski S.*

AST.2A.3– HPLC analysis of phenols of Slovenian red wines: Cabernet Sauvignon and Merlot– *Šaćirović S, Ćirić A, Antić M, Marković Z.*

AST.2A.4– Possibility of application pulsating electromagnetic field in a safer soybean production– *Cvijanović G, Bajagić M, Đukić V, Đurić N.*

AST.2A.5- Essential oil analysis of the *Micromeria juliana* (L.) Benth., from Luštica, Montenegro– *Filipović S, Radulović N.*

AST.2A.6- Potentially toxic elements in lowland Great Morava River – bioindication with bleak (*Alburnus alburnus*)– *Milošković A, Kojadinović N, Radenković M, Đuretanović S, Veličković T, Nikolić M, Simić V.*

AST.2A.7- Improving production efficiency in the food production sector– *Veselinović N, Nikolić J.*

11:00 - 11:30	Coffee Break

Session BBMC.2B: 11:30-13:00 (Room 1)

Bioorganic, bioinorganic, and medicinal chemistry (Poster session)- Part II

Chair: Miroslav Živić

11:30 - 11:55	Section lecturer: **Ralph Puchta**, Assistant Professor, Department of Chemistry and Pharmacy, Friedrich-Alexander University, Erlangen, Nuremberg. Topic: **Computational Supramolecular Chemistry investigated by pupils and undergraduate students**

BBMC.2B.1– Phenolic content and potential Application of *Lysmachia vulgaris* L. aerial part and root extracts– *Srećković N, Katanić Stanković J, Mihailović V.*

BBMC.2B.2– Exploring the potential of α-arbutin as the inhibitor of neurodegenerative disorder– *Ristivojević P, Cvijetić I, Krstić-Ristivojević M, Milojković-Opsenica D.*

BBMC.2B.3– Nucleophilic substitution reactions of caffeine-derived Pt(II) and Pd(II) complexes with important bio-molecules– *Jovanović Stević S, Bogojeski J, Caković A, Petrović B.*

BBMC.2B.4– Influence of vanadium on the growth and metabolism of coprinellus truncorum fungal mycelium– *Žižić M, Živić M, Atlagić K, Karaman M, Zakrzewska J.*

BBMC.2B.5– 17-substituted steroidal tetrazoles - novel ligands for human steroid-converting cyp enzymes– *Jovanović Šanta S, Oklješa A, Sachanka A, Dzichenka Y, Usanov S.*

BBMC.2B.6– Binding of 17-substituted 16-nitrile 16,17 secoestrane compounds to estrogen receptors - in vitro and in silico study– *Jovanović Šanta S, Isenović E, Petrović J, Dzichenka Y.*

BBMC.2B.7– The Effects of Selenite on Filamentous Fungi Lipid Droplets Monitored in Vivo Label Free Using Advanced Nonlinear Microscopy Technique– *Pajić T, Todorović N, Stefanović D, Rabasović M, Krmpot A, Živić M.*

BBMC.2B.8– The Influence of Selenite on Fillamentous Fungi Hypha Morphometry Parameters– *Pajić T, Todorović N, Stefanović D, Rabasović M, Krmpot A, Živić M.*

BBMC.2B.9– Antimicrobial activity of 3- (1- (3- hydroxyphenyl) amino) ethylidene) chroman-2,4-dione and its corresponding palladium(II) complex– *Avdović E, Milanović Ž, Radulović M, Dimić D.*

BBMC.2B.10– Design, Synthesis and pharmacological evaluation of novel N-{4-[2-(4-Aryl-piperazin-1-yl)-ethyl]-phenyl}-arylamides–*Andrić D, Dukić-Stefanović S, Penjišević J, Jevtić I, Šukalović V, Suručić R, Kostić-Rajačić S.*

13:00 - 14:00	Buffet Lunch

14:00 - 14:30 (Room 1)	Invited speaker: **Dragoslav Nikezić,** Full Professor, Faculty of Science, University of Kragujevac and State University of Novi Pazar, Serbia Topic: **Computational dosimetry- international comparison of different simulational software within EURADOS organization** Chair: Tatjana Miladinović

Session CCMD.2A: 14:30-15:30 (Room 1)

Chemoinformatics, chemogenomics and molecular design (Poster session)

Chair: Dejan Milenković

CCMD.2A.1– The exploration of CYP17A1 ligand space by the qsar model–*Boboriko N, Liying H, Dzichenka Y.*

CCMD.2A.2– Direct scavenging activity of 4,7-dihydroxycoumarin derivative towards series of chloromethylperoxy radicals– *Milanović Ž, Avdović E, Antonijević M, Marković Z.*

CCMD.2A.3– Cytotoxic activity and molecular docking study of 4-substituted flavylium salt – *Milenković D, Živanović M, Dekić M, Stanojević Pirković M, Đorović Jovanović J.*

CCMD.2A.4– Molecular docking of some cyclohexadiene derivatives– *Đorović Jovanović J, Dimić D, Stanojević Pirković M, Jeremić S, Milenković D.*

CCMD.2A.5– Inhibitory effect of coumarin benzoylhydrazones on MCL-1 protein– *Simijonović D, Antonijević M, Avdović E, Petrović Z, Marković Z.*

CCMD.2A.6– Spectroscopic and quantum-chemical investigation of testosterone Propionate, a commonly misused anabolic steroid– *Ristivojević N, Dimić D, Đošić M, Mišić S, Gavran A, Đorović Jovanović J, Dimitrić Marković J.*

CCMD.2A.7– Understanding effect of laser speed and formulation factors on printability and characteristics of SLS Irbesartan tablets- application of decision tree model– *Madžarević M, Ibrić S.*

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DESIGN, SYNTHESIS AND PHARMACOLOGICAL EVALUATION OF NOVEL *N*-{4-[2-(4-ARYL-PIPERAZIN-1-YL)-ETHYL]-PHENYL}-ARYLAMIDES

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Abstract:

$5HT_{1A}$ receptor targeting drugs have been used as the treatment for the many neuropsychiatric disorders, such as schizophrenia and depression. As a part of ongoing research, we designed series of new compounds that share arylpiperazine common structural motif with the $5HT_{1A}$ receptor ligand aripiprazole. Receptor-ligand interactions were determined by the molecular docking simulations, revealing the positive impact of the phenyl substitution in the arylpiperazine part of the molecules. Nine selected compounds were synthesized in four reaction steps in high overall yields (59-73%). *In vitro* pharmacological evaluation of the synthesized compounds revealed three compounds (**5b**, **6b** and **6c**) with high $5HT_{1A}$ binding affinity, comparable with aripiprazole (Ki 12.0, 4.8, 12.8, 5.6 nM, respectively). Compounds from **b** series, **5b** and **6b**, possess 2-methoxyphenyl substituents, while **6c** possess 2,3-dichlorophenyl substituent in the arylpiperazine part of the molecule. The pharmacological results are therefore in accordance with the molecular docking simulations thus proving the rational design. Compounds **5c**, **6b** and **6c** can be considered as the candidates for further evaluation as new, potential antidepressants.

Keywords: $5HT_{1A}$, aripiprazole, arylpiperazines, molecular docking, binding assay

1. Introduction

Depression is one of the most abundant neuropsychiatric disorders, present in more than 300 million people worldwide. Followed by serious symptoms affecting person's behaviours and feelings which often lead to suicidal intentions, represents a global health issue.[1] Having that in mind, the development of new therapeutics for the treatment of depression symptoms has been scientificaly imortant form many years because of the limited efficiancy of the curent aproved drugs.[2] One of the targets for the development of new antidepressants is serotonine $5HT_{1A}$ repceptor. Some antidepressants known to bind to $5HT_{1A}$ receptor possess *N*-arylpiperazine structural motifs. Aripiprazole represents one of such molecules, recently approved for the treatment of the major depressive disorder.[3]

Recent advances in serotonin receptors structure research gave us a number of crystal $5HT_{1A}$ receptor structures, most notably $5HT_{1A}$ bound to the aripiprazole.[4] This discovery prompted us to re-

evaluate a docking analysis of previously synthesized compounds,[5,6] since they share minimal common arylpiperazine structure with aripiprazole. Based on these findings we designed some new arylpiperazine derivatives which were subjected to the detailed molecular docking analysis using the crystal structure of $5HT_{1A}$ receptor with aripiprazole. Nine novel *N*-{4-[2-(4-aryl-piperazin-1-yl)-ethyl]-phenyl}-arylamides (**5a-c** - **7a-c**) were then selected for the synthesis and pharmacological evaluation.

2. Result and discussion

2.1. Molecular docking

Docking analysis was done using aripiprazole-bound serotonin $5HT_{1A}$ receptor-Gi protein complex receptor model (PDB: 7E2Z)[4] and selected ligands (Figure 1, Table 1.). All ligands were protonated at physiological pH (pH=7.4). Schrodinger Maestro software induced fit docking procedure was carried out using standard protocol and flexible ligand/flexible bind site. Receptor binding site was determined using bound aripiprazole position. The dimensions of the grid box and receptor setup were 10x10x10 (x,y,z) Å during docking study, respectively, with a grid space of 1 Å. Up to 20 docking poses within 30 kcal/mol of the best docked pose were generated and results were sorted based on the docking score and number of key receptor-ligand interactions.[7,8]

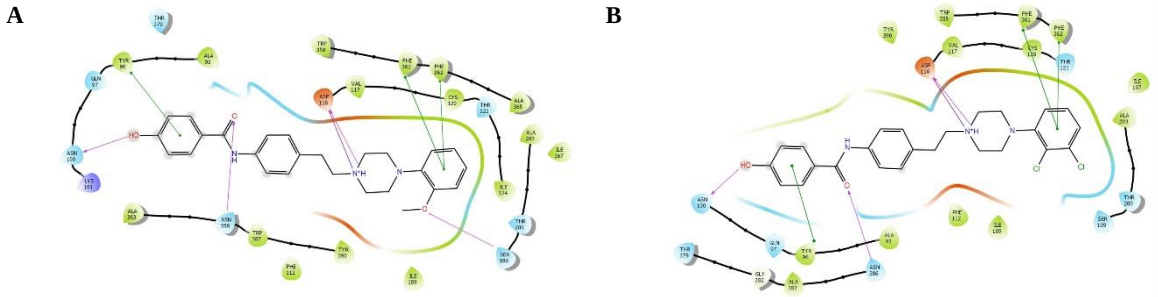


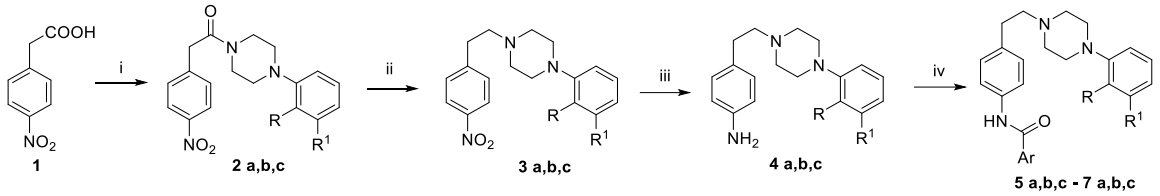
Fig. 1. 2D docking schematics of ligand and $5HT_{1A}$ receptor. A: Best docking pose of **6b** in the active site of the $5HT_{1A}$; B: Best docking pose of **6c** in the active site of the $5HT_{1A}$. The most relevant interacting residues are labeled. Key interactions are color coded as follows: purple – electrostatic interactions (salt bridge, hydrogen bonding), green – aromatic interactions (edge to face, C-H…Pi). Gray nodes represent solvent accessible ligand atoms.

$5HT_{1A}$ receptor binding site can be divided in two distinctive parts. Orthosteric binding site (OBS) that is located deep inside the binding cavity, between transmembrane helices 3 and 5 and extended binding pocket (EBP) bordering extracellular space, and formed in part by receptor extracellular loops. OBS is responsible for binding of arylpiperazine part of the ligand, and correct ligand orientation in the receptor binding site. Key interactions observed in OBS are with Asp116 (salt bridge), and number of aromatic interactions with Phe361 and 362 (edge-to-face, C-H…Pi) as well as hydrogen bond with Ser199. On the other side, EBP forms number of hydrogen bonds with ligand, that adds to binding affinity. Key interactions in the EBP are with Asn100 and Asn386, reinforced by aromatic interaction (edge-to-face) with Tyr96.

Obtained results showed that compounds **6b** and **6c** have most key interactions. The best docking pose of the compounds revealed the key interactions of the arylpiperazine part of the molecule in the OBS are the salt bridge with Asp116 and hydrogen bond with Ser199 (**6b**, Figure 1. A) and aromatic interactions with Phe361, Phe362 (**6c**, Figure 1. B). The most notable interactions of the carboxamide part of the molecule in the EBP included hydrogen bonds with Asn100, Asn386 and aromatic interactions with Tyr96 (Figure 1.).

2.2. Chemistry

The general synthetic route towards **5 a-c**, **6 a-c** and **7a-c** is presented in Scheme 1.



a: R=H, R^1=H; **b**: R=OCH_3, R^1=H; **c**: R=Cl, R^1=Cl.

Scheme1. Reagents and conditions for the synthesis of **5 a-c**, **6 a-c** and **7a-c**: (i) Arylpiperazine, propylphosphonic acid anhydride (PPAA), *N*,*N*-dimethylformamide (DMF), r.t.; (ii) B_2H_6, THF, 0°C for 6 h, r.t. for 1 h, then reflux for 2 h; (iii) NH_4CHO_2, 10% Pd/C, MeOH, (iv) $ArCO_2H$ (3-hydroxybenzoic acid for preparation of **5 a**,**b**,**c** or 4-hydroxybenzoic acid for preparation of **6 a**,**b**,**c** or 2-(4-hydroxyphenyl)acetic acid for preparation of **7 a**,**b**,**c**), PPAA, DMF, r.t.

Acylation of the appropriate arylpiperazine by 4-nitrophenylacetic acid **1** afforded amides **2 a-c**, which upon reduction by B_2H_6 provided corresponding amines **3 a-c**. Reduction of the nitro group in **3** with Pd/C/ammonium formate, followed by *N*-acylation of **4** with aryl acids, yielded final products **5 a-c**, **6 a-c** and **7a-c** in high overall yields (59-73%). All compounds were spectroscopically characterized.

2.3. $5HT_{1A}$ *binding assay*

Competitive binding assay at serotonin $5HT_{1A}$ receptors was performed using radio ligand [^{3}H] 8-OH-DPAT, as previously described.[5] The observed K_i values (Table 1.) indicated that introduction of 2-methoxy and 2,3-dichloro substituents on the phenyl ring of arylpiperazine part of the molecule has positive impact on the $5HT_{1A}$ receptor binding affinity.

Table 1**.** Summary of affinity activities (Ki) and observed key interactions for tested ligands and $5HT_{1A}$ receptor

Compound	K_i (nM)	Observed key interactions
5a	37.8	Asn100, Asp116, Phe361, Phe362
5b	12.0	Asn100, Asn386, Asp116, Phe361, Phe362, Ser199
5c	35.4	Asn100, Asp116, Phe361, Phe362
6a	52.2	Asn100, Asn386, Asp116, Phe361, Phe362
6b	4.8	Tyr96, Asn100, Asn386, Asp116, Phe361, Phe362, Ser199
6c	12.8	Tyr96, Asn100, Asn386, Asp116, Phe361, Phe362
7a	1887	Tyr96, Asn386, Asp116, Phe361, Phe362
7b	46.8	Asn100, Asn386, Asp116, Phe361, Phe362, Ser199
7c	25.7	Asn100, Asn386, Asp116, Phe361, Phe362
aripiprazole	5.6	Tyr96, Asp116, Phe361, Phe362

Biggest affinity gain was observed in the case of ligands **5b**, **6b** and **6c** compared to phenylpiperazine derivatives **5a** and **6a** respectively. The binding affinity of the most active compound from the series **6b** compared with aripiprazole [9] (4.8 and 5.6 nM, respectively), illustrates the positive impact of the 2-methoxy substituent on the phenyl ring in the arylpiperazine part of the molecule, which is in accordance with the molecular docking analysis.

3. Conclusions

In summary, molecular docking analysis was performed on aripiprazole-bound serotonin $5HT_{1A}$ receptor-Gi protein complex receptor model and newly designed ligands. Nine selected *N*-{4-[2-(4-aryl-piperazin-1-yl)-ethyl]-phenyl}-arylamides were synthesized and pharmacologically evaluated. Based on the observed interactions with the binding site, molecular docking analysis predicted the best binding affinities for the two compounds from the series, **6b** and **6c**. *In vitro* obtained $5HT_{1A}$ binding affinities are in accordance with molecular docking predictions. Three compounds (**5b**, **6b** and **6c**) are highlighted as the best candidates to be further investigated as potential antidepressants.

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