Ph.D. Thesis

Transformations of electron-rich aromatic compounds toward bioactive scaffolds

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A. INTRODUCTION AND AIMS

The Mannich reaction is one of the most important basic reaction types in organic chemistry for C–C and C–N bond formation. A special variation of this latter reaction (modified Mannich reaction: *m*MR) uses benzaldehyde rather than formaldehyde, ammonia instead of secondary amine and replacing the C–H acid by an electron-rich aromatic compound, such as 1- or 2-naphthol or nitrogencontaining naphthol analogues leading to chelating compounds with improved antiproliferative activity.

Based on previous studies, an unexpected transformation between 1-*α*-aminobenzyl-2-naphthol and 3,4-dihydroisoquinoline enabled the synthesis of naphth[1,2-e][1,3]oxazino[2,3-*a*]isoquinolines under microwave (MW) irradiation. As a next step, the reaction was extended to 2-aminoalkyl-1-naphthols and other C=N dienophiles (cyclic imines) allowing the synthesis of new naphthoxazino-isoquinoline, -benzazepine and -thienopyridine derivatives. The synthesis of new nonracemic naphth[1,3]oxazino[3,2-*a*]quinoxalinones starting from enantiomeric (4*aS*,8*aS*)-4*a*,5,6,7,8,8*a*-hexahydro-2-quinoxalinone and 1-aminoalkyl-2-naphthols or 2-aminoalkyl-1-naphthols has already been achieved. As a result of a recent work, the reactivity of highly functionalised aminonaphthol or aminophenanthrol derivatives with different cyclic imines was also tested in [4+2] cycloaddition. According to literature data the formation and transformation of aromatic *ortho*-quinone methides (*o*-QM) via [4+2] cycloaddition has been examined. To the best of our knowledge, the stabilisation of partially aromatic *o*-QMs with cyclic imines has not been studied. Consequently, our first aim was to prepare bifunctional glycine-type precursors substituted with 2- and 1-naphthol. The stabilization of precursors via partially aromatic *ortho*-quinone methide intermediate was tested with different cyclic imines in [4+2] cycloaddition. Regarding literature data, 8-hydroxyquinoline is a biologically active moiety considered as a formal 1-naphthol analogue. 8-Hydroxyquinoline can be interpreted as possible substrate of the Mannich reaction, which is a privileged structure in many biologically active compounds and several marketed drugs used for the treatment of infectious diseases, neuropathies, and cancers. Taking into consideration that the diverse biological activities of 8- hydroxyquinoline derivatives can be finetuned by modification of the substitution pattern of the scaffold, we proposed to examine the behavior of 5-chloro-8-hydroxyquinoline in Mannich reaction.

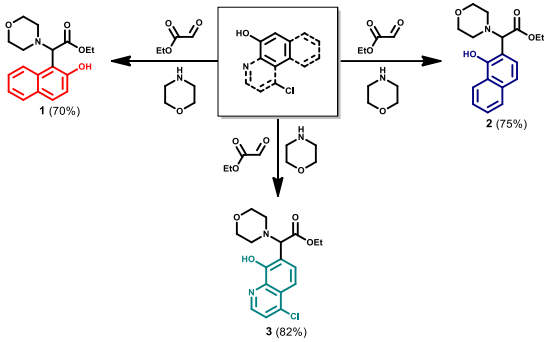
In recent studies the formation of triarylmethane derivatives consisting of indole known as potent biological moiety and 2-naphthol or the ethyl ester of kynurenic acid has been described. As a further aim of my PhD work, testing the reactivity of glycine derivatives substituted with 2- and 1-naphthol with indole and 7-azaindole was also planned. We targeted additionally to study the possibilitiy of transformation of the formed Mannich base consisting 5-chloro-8-hydroxyquinoline skeleton to give diarylmethane derivatives with indole and 7-azaindole as well as the effect of the aldehyde component and the amine part of the Mannich base on the synthetic pathway.

As a special *m*MR, the modified *aza*-Friedel–Crafts reaction by direct coupling of different cyclic imines and indole derivatives as electron-rich aromatic compounds have been studied. To investigate the scope and limitations of the reaction and examine the effect of structural modifications were part of our experiments, which can offer possibilities for having a preliminary overview about the structure–activity relationship for C-3-coupled indole and azaindole derivatives. Therefore, we focused our efforts on resynthesizing selected compounds (3-isoquinolyl-, 3-thieno[3,2-*c*]pyridyl-, 3-*β*-carbolinyl- and 3-benz[*c*]azepinyl-indole and –azaindole derivatives) as well as preparing new derivatives starting from 6,7-dimethoxy-3,4-dihydroisoquinoline, (4*aR*,8*aR*)-4*a*,5,6,7,8,8*a*-hexahydroquinoxalin-2(1*H*)-one and 7-azaindole.

Generally, in favor of having a preliminary overview about the structure–activity relationship, we aimed to test the antibacterial and anticancer activity of synthesized compounds by preliminary biological screening systems.

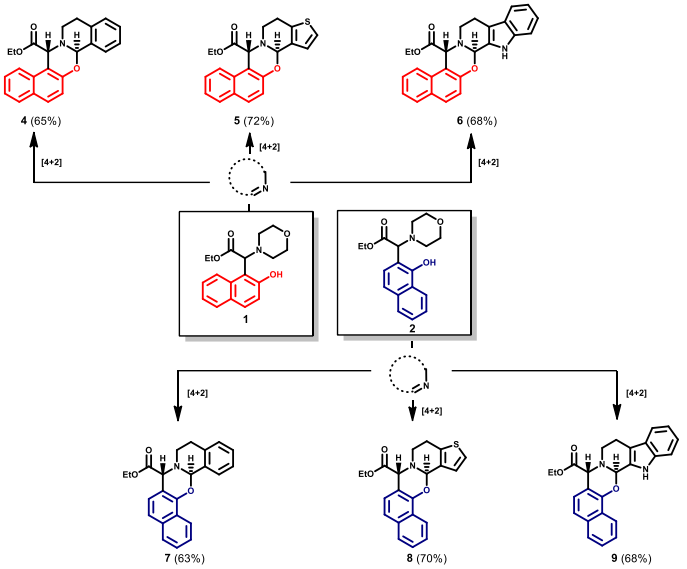
B. RESULTS AND DISCUSSION

1. In the frame of my PhD work, new bifunctional precursors substituted with 2- and 1-naphthol (**1**, **2)** or 5-chloro-8-hydroxyquinoline (**3)** were synthesised by the reaction of 2- and 1-naphthol or 5-chloro-8-hydroxyquinoline, morpholine and ethyl glyoxylate as the aldehyde component by using a modified Mannich-type synthetic pathway (Scheme 1). As a general procedure, 80–100 °C and reaction times of 30–150 min under microwave irradiation were found to be the optimal reaction conditions.



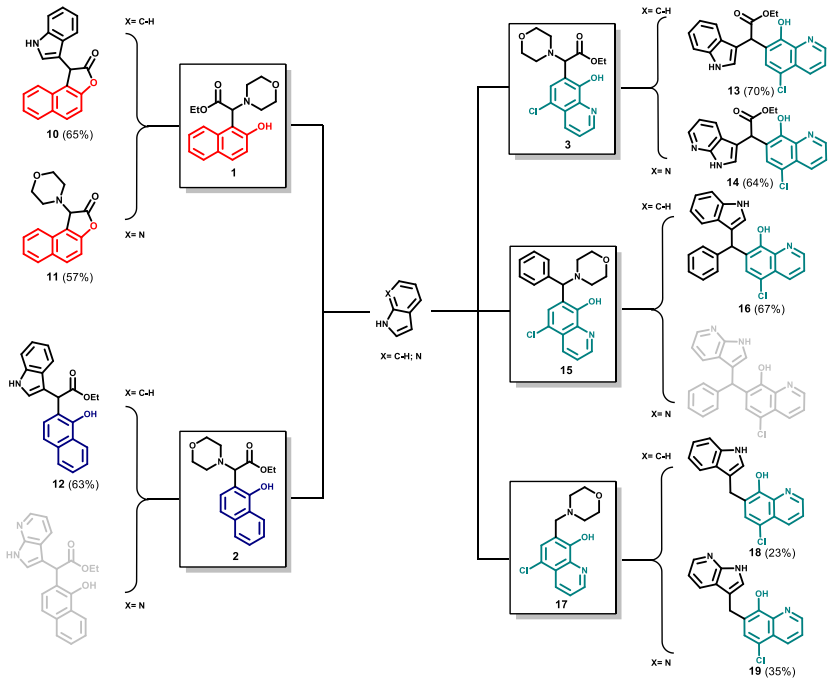
Scheme 1

2. The stabilization of Mannich bases **1, 2** via partially aromatic *ortho*-quinone methide intermediate was tested with different cyclic imines (3,4-dihydroisoquinoline, 6,7-dihydrothieno[3,2-*c*]pyridine, 3,4-dihydro-*β*-carboline) in [4+2] cycloaddition (Scheme 2). Based on ^{1}H-NMR analysis, in the case of new *α*-amino acid esters **4-9** the formation of a single product has been assumed. The NOE spectrum proved that the relative configuration of the newly formed stereogenic centres was *trans*.



Scheme 2

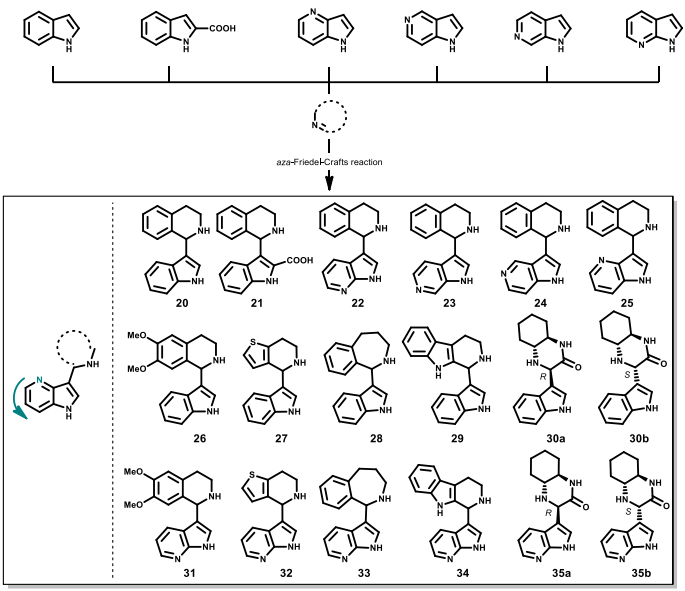
3. Glycine derivatives bearing 2- and 1-naphthol (**1, 2)** were reacted with indole and 7-azaindole. Reactions were found to depend on the naphthol skeleton. Starting from the 2-naphthol-substituted precursor **1**, lactam-type ring-closed products **10, 11** were isolated. The transformation of 1-naphthol, in turn, led to the formation of the desired biaryl ester **12**. The formed Mannich base substituted with 5-chloro-8-hydroxyquinoline **3** was subjected to give diarylmethane derivatives **13, 14** with indole and 7-azaindole. The effect of the aldehyde component and the amine part of the Mannich base on the synthetic pathway was also investigated. The series of 8-hydroxyquinoline skeleton containing derivatives was extended by reacting 7-aminobenzyl-8-hydroxyquinoline **15** and 7-aminomethyl-8-hydroxyquinoline **17** Mannich bases with indole and 7-azaindole. The synthesis of 7-((1*H*-indol-3-yl)methyl)-5-chloroquinolin-8-ol (**18**) from different precursors bearing the morpholine skeleton or L-proline as amines, was also achieved. These studies represent a systematic investigation on the effect of leaving groups on conversion (Scheme 3).



Scheme 3

4. A series of cyclic amines coupled with indole and azaindole derivatives has been systematically designed (Scheme 4). The investigated precursors were synthesized by the coupling of indole with cyclic imines such as 3,4-dihydroisoquinoline, 6,7-dimethoxy-3,4-dihydroisoquinoline, 6,7-dihydrothieno[3,2-*c*]pyridine, 3,4-dihydro-*β*-carboline, 4,5-dihydro-3*H*-benz[*c*]azepine and (4*aR*,8*aR*)-4*a*,5,6,7,8,8*a*-hexahydroquinoxalin-2(1*H*)-one. By fixing the isoquinoline part, the influence of the indole skeleton was also varied among indole-2-carboxylic acid, 4-, 5-, 6- and 7-azaindoles. To have a comprehensive analysis about the correlation between structure and biological activity, the 7-azaindole analogues of the previously mentioned C-3-substituted indoles were also resynthesized or synthesized by using the modified *aza*-Friedel–Crafts reaction. The preparation of new 7-azaindole derivatives **31, 35a**, **35b** starting from 6,7-dimethoxy-3,4-dihydroisoquinoline as cyclic imine and

(4*aR*,8*aR*)-4*a*,5,6,7,8,8*a*-hexahydroquinoxalin-2(1*H*)-one as chiral cyclic imine were also achieved by using microwave conditions.



Scheme 4

5. In favor of having a preliminary overview about the structure–activity relationships, our efforts were focused on having an overview about biological activity of synthesized compounds by preliminary biological screening systems. In the frame of biological evaluations of 2- and 1-naphthol-substituted glycine derivatives and α-amino acid esters, in the case of derivatives **1**, **7**, **8** and **9** efflux pump inhibitory (EPI) activity on *S. aureus* ATCC 25923 strain was observed. In addition, **7** and **8** were effective on the resistant *S. aureus* MRSA ATCC 43300. In point of correlation between structure and biological activity, referring to α-amino acid esters 1-naphthol as electron-rich aromatic compound could be identified as a significant moiety. Regarding the cytotoxic effect of indole and 8-hydroxyquinoline, 2- and 1-naphthol skeleton containing di- or triarylmethane derivatives, the biaryl structural element and 5-chloro-8-hydroxyquinoline skeleton could be identified as a significant moiety (compound **18**). For improved cytotoxic activity, the presence of

morpholine bearing cationic centre and 5-chloro-8-hydroxyquinoline is beneficial (compound **3**). In the case of cyclic amines coupled indole and azaindole derivatives, the most significant EPI activity was observed referring to 6,7-dihydrothieno[3,2-*c*]pyridine coupled indole derivative on *E. coli* AG100 strain. The reaction of 6,7-dimethoxy-3,4-dihydroisoquinoline with 7-azaindole resulted in the most potent biofilm formation inhibitor product. Applying indole and dihydro-β-carboline as well as dihydrothieno[3,2-*c*]pyridine led to the formation of C-3-coupled compound with the highest anticancer activity. 6,7-Dimethoxy-3,4-dihydroisoquinoline as electron-rich aromatic compound and indole skeleton have been found to be effective in the inhibition of ABCB1.

C. PUBLICATIONS

I. **Dóra Hegedűs**, Nikoletta Szemerédi, Gabriella and István Szatmári
Application of partially aromatic *ortho*-quionone-methides for the synthesis of novel naphthoxazines with improved antibacterial activity
Eur J Med Chem **2022**, *237*, 114391. DOI: 10.1016/j.ejmech.2022.114391 **IF: 6.7**

II. **Dóra Hegedűs**, Nikoletta Szemerédi, Maja Gábor, Judit Sas, Khadija Belasri, István Szatmári and Gabriella Spengler
Cyclic Amines Coupled to Indole Derivatives with Improved Efflux Pump Inhibiting Activity in Bacteria and Cancer Cells
Anticancer Res **2024**, *44*, 1149-1160. DOI:10.21873/anticanres.16910 **IF: 1.6**

III. **Dóra Hegedűs**, Nikoletta Szemerédi, Krisztina Petrinca, Róbert Berkecz, Gabriella Spengler and István Szatmári
Synthesis of tumor selective indole and 8-hydroxyquinoline skeleton containing di-, or triarylmethanes with improved cy-totoxic activity
Molecules **2024**, *29*, 4176. DOI:10.3390/ molecules29174176 **IF: 4.2**

D. CONFERENCE LECTURES

I. **Dóra Hegedűs** and István Szatmári
Részlegesen aromás *orto*-kinon metidek szintézise és továbbalakítása
Tudományos Diákköri Konferencia, Szeged, November 16, 2018.

II. **Dóra Hegedűs** and István Szatmári
Részlegesen aromás *orto*-kinon metidek szintézise és továbbalakítása
Szegedi Ifjú Kémikusok Támogatásáért Alapítvány előadóülése, Szeged, May 25, 2021.

III. **Dóra Hegedűs**, Gabriella Spengler, Nikoletta Szemerédi and István Szatmári
Bioaktív naftoxazin-származékok szintézise glioxilát-szubsztrát alkalmazásával
MTA Heterociklusos és Elemorganikus Kémiai Munkabizottság ülése, Balatonszemes, May 23-25, 2022.

IV. **Dóra Hegedűs**, Nikoletta Szemerédi, Gabriella Spengler and István Szatmári
A módosított Mannich-reakció alkalmazása bioaktív prekurzorok szintézisére és továbbalakításaira
Gyógyszerkémiai és Gyógyszertechnológiai Szimpózium, Herceghalom, September 19-20, 2022.

V. **Dóra Hegedűs**, Nikoletta Szemerédi, Gabriella Spengler and István Szatmári
Partially aromatic *ortho*-quionone-methides as precursors for the synthesis of novel naphthoxazines with improved antibacterial activity
22nd Tetrahedron Symposium, Lisbon/Portugal, June 28–July 1, 2022.

VI. **Dóra Hegedűs** and István Szatmári
Synthesis and transformations of bifunctional glycine-type precursors containing 8-hyroxyquinoline skeleton
23rd Tetrahedron Symposium, Göteborg/Sweden, June 27-30, 2023.

VII. **Dóra Hegedűs**, Nikoletta Szemerédi, Dorka Gubó, Gabriella Spengler and István Szatmári
Synthesis and transformations of bifunctional 8-hydroxyquinoline skeleton containing glycine-type precursors
Congressus Pharmaceuticus Hungaricus (CPH) XVII. and EUFEPS annual meeting, Debrecen/Hungary, May 23-25, 2024.

VIII. **Dóra Hegedűs**, Nikoletta Szemerédi, Krisztina Petrinca, Gabriella Spengler, István Szatmári
Synthesis and cytotoxic activity of bioconjugates consisting of indole and 8-hydroxyquinoline skeleton
24th Tetrahedron Symposium, Montpellier/France, June 18-21, 2024.