Summary of PhD Thesis

EXPLOITING THE BENEFITS OF CONTINUOUS-FLOW PROCESSING FOR THE DEVELOPMENT OF NOVEL SUSTAINABLE CATALYTIC PROCEDURES

Sándor B. Ötvös

Supervisor:
Prof. Dr. Ferenc Fülöp

University of Szeged
Institute of Pharmaceutical Chemistry

2013

University of Szeged
Doctoral School of Pharmaceutical Sciences

Programme of Pharmaceutical Chemistry and Drug Research
Programme director: Prof. Dr. Ferenc Fülöp

Institute of Pharmaceutical Chemistry

Supervisor: Prof. Dr. Ferenc Fülöp

Sándor B. Ötvös

Exploiting the benefits of continuous-flow processing for the development of novel sustainable catalytic procedures

Final Exam Committee:

Head:	Dr. Árpád Molnár
Members:	Dr. György Dombi
	Dr. György Dormán

Reviewer Committee:

Head:	Dr. György Dombi
Reviewers:	Dr. György Dormán
	Dr. István Pálinkó
Members:	Dr. Pál Perjési
	Dr. Csaba Tömböly

1. INTRODUCTION AND AIMS

The progress toward increased sustainability in chemical industry requires developments and novel approaches that imply improved performance and value in chemical production in association with a reduction of the environmental impact. The intense need for novel industrial methodologies has opened up new routes in compact-scale chemical engineering, and continuous-flow (CF) chemistry has emerged as a novel sustainable alternative for the conventional batch-based techniques in the synthetic chemistry of fine chemicals.

CF technologies offer numerous advantages over classical segmented unit operations. The well-regulated flow reactor concept renders an increased parameter space for chemical synthesis and enables reactions to be performed with an unprecedented level of control due to the greatly enhanced heat and mass transfer and improved mixing properties. This implies higher reaction rates, outstanding selectivity, and safer and greener chemistry as compared with mechanically stirred reaction vessels. These features offer a plausible opportunity for automation and excellent transferability between laboratory-based investigations and industrial-scale production and have pushed CF technology to the frontiers of modern sustainable chemistries.

Our major goal was to exploit the benefits of flow processing for reaction optimization and synthesis, and to develop novel sustainable synthetic methodologies with possible usefulness for the pharmaceutical industry. We set out to broaden the chemical space and practical applicability of transition metal-catalyzed and organocatalytic procedures to obtain pharmaceutically relevant intermediates and potentially bioactive compounds in a safe, simple and efficient manner. To achieve our goals, we intended to focus on the following areas in CF: (*i*) heterogeneous catalytic deuterations, (*ii*) Cu(I)-catalyzed azide–alkyne cycloadditions (CuAAC) and (*iii*) organocatalytic asymmetric aldol and conjugate addition reactions.

Enantiomerically pure compounds are crucially important in pharmaceutical research, and organocatalysis has contributed appreciably to the recent advances in asymmetric syntheses. However, long reaction times, difficulties in product isolation and selectivity issues often limit the practical applicability of organocatalytic reactions. Accordingly, we aimed to develop a novel sustainable methodology which relies on immobilized peptidic catalysts in a packed-bed CF reactor and eliminates most of the above drawbacks. Utilization of a peptide is highly beneficial, as such a modular catalyst offers an unprecedented level of structural diversity, and facilitates the creation of optimized organocatalysts.

A large number of 1,2,3-triazole-containing compounds have been described with various biological activities, such as antibacterial, antiviral or antifungal effects. CuAAC is a convenient

way to obtain 1,2,3-triazoles. However, most CF examples of CuAAC reactions rely on either costly apparatus or special catalyst types, thereby hampering their sustainable applications. We therefore planned to develop an easily available and inexpensive CF technique for CuAAC reactions, which is at the same time safe, efficient and productive and additionally conveniently adaptable for the preparation of potentially bioactive compounds for the drug discovery.

There are a broad array of pharmacologically interesting molecules among heterocyclic structures, and deuterium-labeled heterocycles are of considerable importance in drug discovery. However, conventional deuteration methodologies suffer from several drawbacks. For this reason, it was our aim to employ a convenient and safe CF technique with on-demand electrolytic D_2 gas production for the highly selective deuteration of nitrogen-containing heterocycles which are precursors for a series of bioactive compounds.

2. EXPERIMENTAL SECTION

CF experiments were carried out in an H-Cube® mesoreactor system containing a stainless steel cartridge as catalyst bed (Figure 1). The filled cartridge was embedded in a heating unit which included a coiled stainless steel reaction line for preheating of the liquid phase before entering the catalyst bed. Constant pressure was ensured by a built-in backpressure regulator, and the CF of the reaction medium was provided by a conventional HPLC pump. The system contained an electrolytic gas-generation unit for heterogeneous hydrogenations. The *in situ* generated gas was combined via a gas–liquid mixer with the solution of the substrate, and the mixture was then transported to the catalyst bed, where the triphasic reaction took place. CF deuterations were carried out by simply changing the hydrogen source from deionized water to D_2O. In the case of CF organocatalysis and CF CuAAC reactions, the gas generation unit was turned off. To determine optimal conditions, most important reaction parameters (such as temperature, pressure, flow rate and substrate concentration) were systematically fine-tuned.

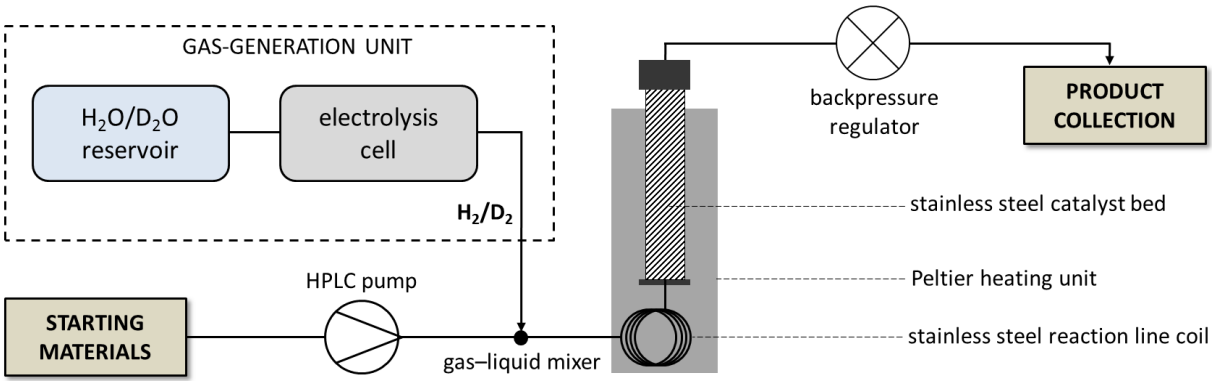

Figure 1. *Experimental setup for the CF reactions.*

The peptidic catalysts were prepared by means of solid-phase peptide synthesis (SPPS), utilizing Fmoc/*t*Bu protecting group chemistry. Amino-functionalized non-trifluoroacetic acid (TFA)-labile resins were utilized as supports for the SPPS: polyethylene glycol–polystyrene (PS) copolymer without any linker (TentaGel), and PS resin with a 4-methylbenzhydrylamine linker (PS-MBHA). TentaGel-bound catalysts were characterized by suspension-phase ^{13}C NMR measurements, and the structure of MBHA-immobilized peptides was verified by means of MS and RP-HPLC investigations after cleaving from the resin.

The products of the CF reactions were characterized by means of NMR spectroscopy (^{1}H, ^{13}C, ^{13}C HMBC and ^{13}C HSQC), MS and elemental analysis. In cases of chiral compounds, *ee* was assigned with chiral NP-HPLC, and *dr* was determined from the ^{1}H NMR spectra of the crude material. In CF CuAAC reactions, the copper contents were determined by ICP-MS.

3. RESULTS AND DISCUSSION*

Novel, sustainable heterogeneous catalytic methodologies were developed for the CF synthesis of pharmaceutically relevant intermediates and potentially bioactive compounds.

3.1. CF deuteration of nitrogen-containing heterocycles[I]

- For the CF deuteration of nitrogen-containing heterocyclic structures, on-demand electrolytic D_2 production was applied, which afforded much higher deuterium efficiency and improved safety as compared with the conventional batch techniques.
- EtOAc was chosen as aprotic solvent to prevent D–H exchange and to maximize deuterium incorporation. 5% $Pd/BaSO_4$ proved insufficiently active even at high temperatures, and 5% Pt/Al_2O_3 was therefore chosen as optimal heterogeneous catalyst. It was observed that the utilization of pressures as high as 50 bar was necessary to obtain high reaction rates.
- Besides heterocyclic model compounds and substituted derivatives, a spasmolytic drug, drotaverine, was also deuterated (**13d**). The deuterium-labeled products were obtained in excellent conversions (96–99%) and deuterium contents (95–98%, Figure 2). It was found that deuterium incorporation was highly selective, as the benzene ring remained intact in all cases.

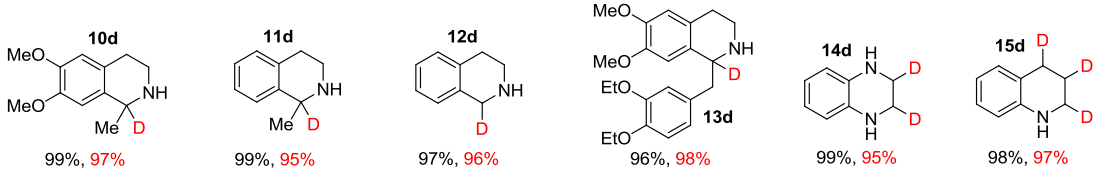

Figure 2. *Deuterated compounds with the representation of the conversions and deuterium contents (in red).*

* Compound numbering is identical to that applied in the thesis.

- The described CF deuteration procedure means a sustainable alternative for the conventional approaches, as it is safe, simple, rapid and cost efficient, whilst it allows pharmaceutically relevant deuterated products without ponderous purification steps.

3.2. Copper-catalyzed azide–alkyne cycloadditions in CF[V,VI]

- A simple, inexpensive and rapid CF technique was developed for 1,3-dipolar cycloaddition reactions between organic azides and acetylenes, which eliminated the need for costly special apparatus and applied Cu powder as a readily available Cu(I) source.
- Initially, the merits of flow processing were exploited by increasing the reaction rates with high-pressure/high-temperature conditions. Full conversion was reached in the test reaction between benzyl azide and phenylacetylene at 100 bar, 100 °C and a flow rate of 0.5 mL min^{-1}.
- Subsequently, it was demonstrated that the harsh reaction conditions can be relieved through the joint use of *N*,*N*-diisopropylethylamine (DIEA) and AcOH as basic and acidic additives, the CF CuAAC thereby being accomplished with maximum operational safety at RT (Figure 3).

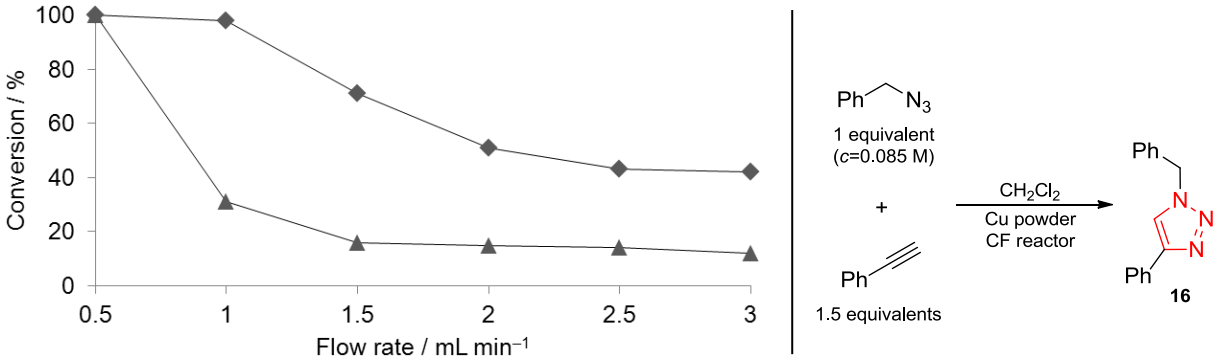

Figure 3. *The flow rate dependence of the test CF CuAAC reaction between benzyl azide and phenylacetylene clearly demonstrates that the joint use of basic and acidic additives relieves the harsh reaction conditions. (▲: 100 bar, 100 °C, no additives; ♦: 100 bar, RT, 0.04 equivalents of DIEA + 0.04 equivalents of AcOH.)*

- The applicability of the CF methodology was found extremely wide, for different azides and alkynes. Excellent yields (72–99%) were obtained with either aliphatic or aromatic starting materials. In some of the cases, the joint use of basic and acidic additives (each in 0.04 equivalents) at RT resulted in higher yields than at 100 bar and 100 °C.
- The scope of the CF CuAAC process was extended to the preparation of 1,2,3-triazole-substituted alicyclic β-amino acid derivatives, as novel potentially bioactive compounds (Figure 4). Phenylacetylene, diethyl acetylenedicarboxylate and ethynyl ferrocene were reacted as dipolarophiles. Yields were as high as 99% in some of the cases, and the copper impurities detected in our systems compared well with CF or batch results from the literature. No epimerization of the triazole products were observed even upon applying basic conditions which is due the very short residence time (1.5 min) applied on the catalyst bed.

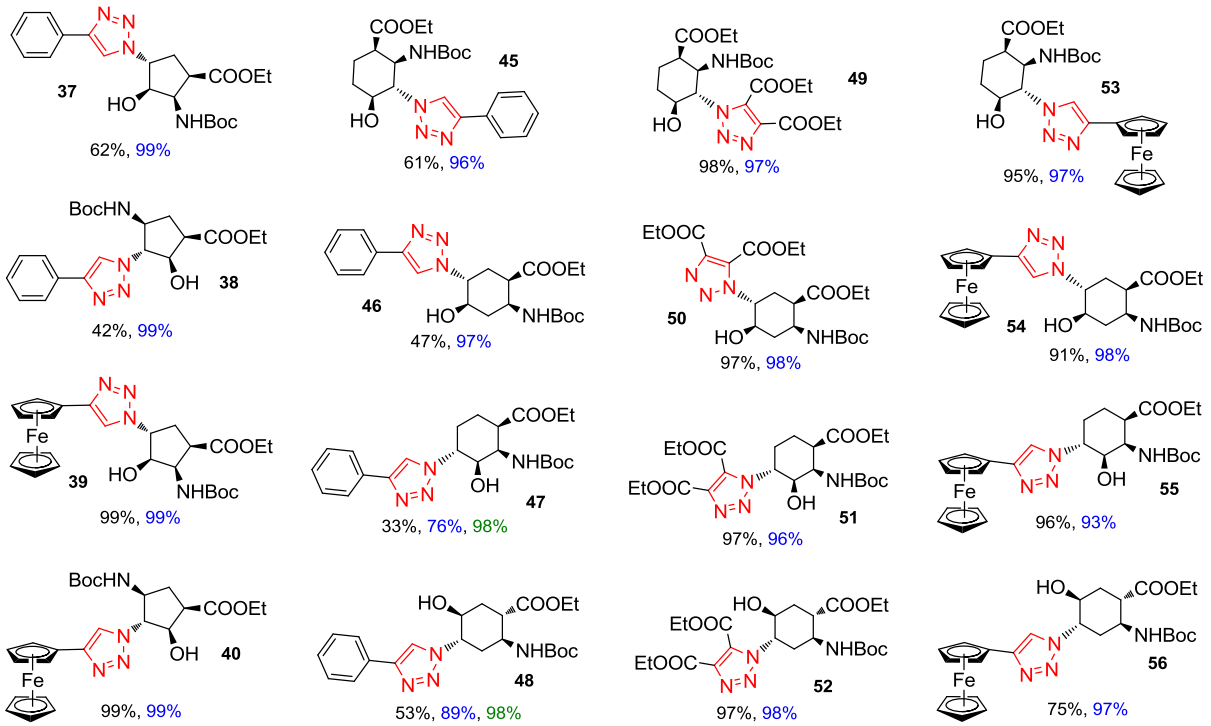

Figure 4. *1,2,3-Triazole-modified alicyclic β-amino acid derivatives. Yields are represented with colored numbers. (Black: 100 bar, 100 °C, without any additives; blue: 100 bar, RT, with 0.04 equivalents of DIEA + 0.04 equivalents of AcOH; green: 100 bar, 100 °C, with 0.04 equivalents of DIEA + 0.04 equivalents of AcOH.)*

- Large-scale synthesis was implemented simply and safely as a function of process time. 3 g of triazole **16** was isolated in 3 h, and 2.06 g of triazole **52** was prepared in 100 min.
- In all reactions with terminal alkynes, the 1,4-disubstituted regioisomers were formed selectively due to the efficient Cu(I) catalysis.

3.3. CF organocatalysis with solid-supported peptidic catalysts[II-IV]

- We introduced the first CF organocatalytic methodology for asymmetric 1,4-addition of aldehydes to nitroolefins, and extended the scope of the sustainable procedure to aldol reactions.
- Solid supported peptidic catalysts containing a proline unit at the N-terminus and an acidic side-chain at the C-terminus were applied (Figure 5).

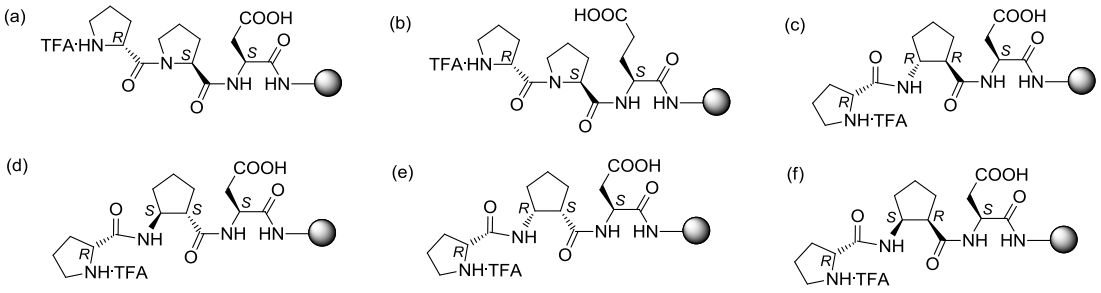

Figure 5. *Structures of the immobilized peptidic catalysts utilized in this study. H-D-Pro-Pro-Asp-NH-resin (a), H-D-Pro-Pro-Glu-NH-resin (b), H-D-Pro-(1R,2R)-ACPC-Asp-NH-resin (c), H-D-Pro-(1S,2S)-ACPC-Asp-NH-resin (d), H-D-Pro-(1S,2R)-ACPC-Asp-NH-resin (e), H-D-Pro-(1R,2S)-ACPC-Asp-NH-resin (f). (ACPC=2-aminocyclopentanecarboxylic acid, resin=TentaGel or PS-MBHA with different loadings.)*

- The catalysts were readily synthetized and immobilized in the same step by SPPS without the cleavage of the peptide from the resin. The heterogeneous support of the SPPS served also as catalyst carrier. The experimental setup was simple, time- and cost-efficient, as it eliminated the need for the peptide work-up and purification steps, and there was no product loss.
- For both organocatalytic transformations, appropriate test reactions were chosen and the most important reaction conditions were thoroughly fine-tuned.
- In the case of conjugate additions, it was observed that the diastereoselectivity decreased continuously when the flow rate was reduced. It was verified that the catalyst itself epimerized the product and consequently, the shorter the residence time on the catalyst bed, the higher the *dr* (Figure 6). The enantioselectivity was not dependent on the flow rate.

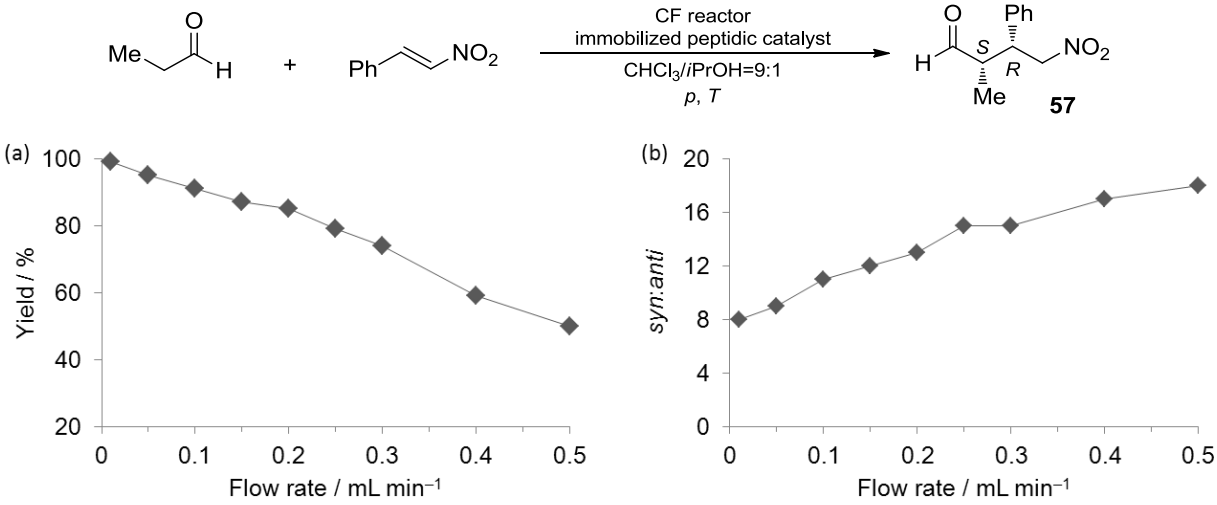

Figure 6. *The adjustment of the flow rate in the test CF reaction between propanal and E-β-nitrostyrene influenced not only the yield, but also the diastereoselectivity as the catalyst epimerized the γ-nitroaldehyde product. (Conditions: 1 equivalent of BNS (c=8 mg mL^{-1}) and 5 equivalents of propanal, 60 bar, RT.)*

- Pressurizing improved reaction rates in both transformations up to an optimal value of 60 bar. To probe the diffusion dependence, the Koros-Nowak test was performed. It was established that the CF reactions are diffusion-controlled and the role of pressure is to promote the transport of the reactants into the matrix of the catalyst carrier.
- It was found that heating also enhanced the reaction rates, but it proved best to employ RT, as higher temperatures dramatically lowered stereoselectivities.
- The effect of *N*-methylmorpholine as a basic additive was examined to improve the catalytic activity in cases of 1,4-additions. However, the best result was obtained without the base, as it epimerized the product and did not affect reaction rates.
- It emerged that for 1,4-additions the catalyst H-D-Pro-Pro-Asp-NH-*resin* on PS-MBHA was the most favorable. On investigation of the bioinspired substitution of the central proline unit with various β-amino acids, it was found that the insertion of (1*R*,2*R*)-ACPC into the sequence

gave comparable results with the parent catalyst. However, the exchange to other ACPC isomers generated significant distortion in peptide conformation, and allowed lower yields.

- For aldol reactions, H-Pro-Pro-Asp-NH-*resin* on TentaGel resin performed best. The D-proline N-terminal catalysts showed significantly lower reactivity and selectivity and inverted the absolute configuration of the corresponding β-hydroxyketone product.
- In aldol reactions, the reusability of the peptidic catalyst was extremely high, as it did not lose any of its activity or selectivity during 17 h of continuous use in the flow reactor. The developed CF methodology is therefore prominently robust.
- In cases of conjugate additions, the heterogenous catalyst was reusable, but its activity decreased slowly in the course of the consecutive reaction cycles.
- With the CF method, chiral γ-nitroaldehydes were obtained in yields up to 91% with high diastereoselectivites (up to a *dr* of 36:1) and excellent enantioselectivites (up to an *ee* of 93%). β-Hydroxyketones, as aldol adducts, were furnished with excellent yields (up to 91%) and high enantioselectivities (up to an *ee* of 85%). The CF results were nicely comparable with those of the literature batch procedures. The batch reactions required hours of reaction time, whereas the flow method relied on short residence times (7 or 6 min) on the catalyst bed, and thus, prominently short process times.

List of publications and lectures

Papers related to the thesis

I. *Sándor B. Ötvös*, István M. Mándity, Ferenc Fülöp:
Highly selective deuteration of pharmaceutically relevant nitrogen-containing heterocycles: a flow chemistry approach
Mol. Diversity **2011**, *15*, 605-611. IF.: 3.153

II. *Sándor B. Ötvös*, István M. Mándity, Ferenc Fülöp:
Highly efficient 1,4-addition of aldehydes to nitroolefins: organocatalysis in continuous flow by solid supported peptidic catalysts
ChemSusChem **2012**, *5*, 266-269. IF.: 7.475

III. *Ötvös Sándor*, Mándity István, Fülöp Ferenc:
Organokatalízis folyamatos áramú reaktorban
Magy. Kém. Lapja **2012**, *67*, 143-146. IF.: 0.000

IV. *Sándor B. Ötvös*, István M. Mándity, Ferenc Fülöp:
Asymmetric aldol reaction in a continuous-flow reactor catalyzed by a highly reusable heterogeneous peptide
J. Catal. **2012**, *295*, 179-185. IF.: 5.787

V. *Sándor B. Ötvös*, István M. Mándity, Lóránd Kiss, Ferenc Fülöp:
Alkyne-azide cycloadditions with copper powder in a high-pressure continuous-flow reactor: high-temperature conditions vs. the role of additives
Chem. Asian. J. **2013**, *8*, 800-808. IF.: 4.572*

VI. *Sándor B. Ötvös*, Ádám Georgiádes, István M. Mándity, Lóránd Kiss, Ferenc Fülöp:
Efficient continuous-flow synthesis of novel 1,2,3-triazole-substituted β-aminocyclohexanecarboxylic acid derivatives with gram-scale production
Beilstein J. Org. Chem. **2013**, *9*, 1508-1516. IF.: 2.801*

Other paper

VII. *Sándor B. Ötvös*, Ottó Berkesi, Tamás Körtvélyesi, István Pálinkó:
Synthesis and spectroscopic and computational characterization of Zn_4O(alicyclic or aromatic carboxylate)$_6$ complexes as potential MOF precursors
Inorg. Chem. **2010**, *49*, 4620-4625. IF.:4.325

Cumulative impact factor: 28.113

*The impact factors for the year 2012 are presented.

Scientific lectures related to the thesis

1. *Sándor B. Ötvös*, István M. Mándity, Ferenc Fülöp:
   Highly selective deuteration of some heteroaromatic compounds, a flow chemistry approach
   Advances in Synthetic Chemistry (ASC), Munich, Germany, 8-9 April 2010. P105.

2. *Ötvös Sándor*:
   Gyógyszerkémiailag jelentős nitrogén tartalmú heterociklusos vegyületek deuterálása folyamatos áramban
   A Szegedi Ifjú Szerves Kémikusok Támogatásáért Alapítvány 10. Tudományos Előadóülése, Szeged, Hungary, 5 May 2010. (II. Prize lecture)

3. *Ötvös Sándor*, Mándity István, Fülöp Ferenc:
   Deuterált heterociklusok előállítása áramlásos kémiával
   MTA Heterociklusos Kémiai Munkabizottság Ülése, Balatonszemes, Hungary, 19-21 May 2010.

4. *Sándor B. Ötvös*, István M. Mándity, Ferenc Fülöp:
   Organocatalysis in flow: solid supported peptide catalyzed enantioselective synthesis of γ-nitroaldehydes
   Organocatalysis, New Methodologies for Sustainable Chemistry, CATAFLU.OR Symposium, Bologna, Italy, 24-25 March 2011. Abstr.: p. 26, P14.

5. István M. Mándity, *Sándor B. Ötvös*, Ferenc Fülöp:
   Bioinspired organocatalyst design harnessing flow chemistry technique
   Organocatalysis, New Methodologies for Sustainable Chemistry, CATAFLU.OR Symposium, Bologna, Italy, 24-25 March 2011. Abstr.: p. 25, P13.

6. *Ötvös Sándor*:
   Organokatalízis folyamatos áramú reaktorban
   A Szegedi Ifjú Szerves Kémikusok Támogatásáért Alapítvány 11. Tudományos Előadóülése, Szeged, Hungary, 18 April 2011. (I. Prize lecture)

7. *Sándor B. Ötvös*, István M. Mándity, Ferenc Fülöp:
   Effective continuous flow synthesis of chiral γ-nitroaldehydes utilizing solid supported peptide as organocatalyst
   The 17th European Symposium on Organic Chemistry (ESOC), Hersonissos, Crete, Greece, 10-15 July 2011. Abstr.: p. 251, P1.249.

8. István M. Mándity, *Sándor B. Ötvös*, Ferenc Fülöp:
   Bioinspired organocatalyst design with a flow chemistry approach
   The 17th European Symposium on Organic Chemistry (ESOC), Hersonissos, Crete, Greece, 10-15 July 2011. Abstr.: p. 270, P1.268.

9. *Ötvös Sándor*, Mándity István, Fülöp Ferenc:
Enantioszelektív organokatalízis folyamatos áramban
MTA Heterociklusos Kémiai Munkabizottság Ülése, Balatonszemes, Hungary, 26-28 September 2011.

10. *Ötvös Sándor*:
Gyógyszerkémiailag jelentős vegyületek enantioszelektív szintézise hatékony folyamatos áramú technikával
XVIII. Szent-Györgyi Napok, Szeged, Hungary, 14-19 November 2011.

11. *Ötvös Sándor*:
Enantioszelektív organokatalízis folyamatos áramban
PhD témák az SZTE Gyógyszerkémiai Intézetben, Szeged, Hungary, 23 February 2012.

12. *Sándor B. Ötvös*, István M. Mándity, Ferenc Fülöp:
Organocatalysis in continuous flow via proline mimetic peptides
FloHet, Heterocyclic and Synthetic Conference, Gainesville, Florida, USA, 4-7 March 2012. Abstr.: p. 81, P3.

13. István M. Mándity, *Sándor B. Ötvös*, Ferenc Fülöp:
Highly efficient deuteration of heterocyclic compounds in continuous flow
FloHet, Heterocyclic and Synthetic Conference, Gainesville, Florida, USA, 4-7 March 2012. Abstr.: p. 80, P2.

14. *Sándor B. Ötvös*, István M. Mándity, Ferenc Fülöp:
Organocatalysis in continuous flow via proline mimetic peptides
Molekulától a Gyógyszerig: Tudomány – gyakorlat és hatósági elvárások a gyógyszerfejlesztésben, Szeged, Hungary, 24-25 May 2012. P4-GYKI.

15. *Ötvös Sándor*, Mándity István, Fülöp Ferenc:
Azid-alkin cikloaddíció áramlásos reaktorban
MTA Heterociklusos és Elemorganikus Kémiai Munkabizottság ülése, Balatonszemes, Hungary, 6-8 June 2012.

16. *Ötvös Sándor*, Georgiádes Ádám, Mándity István, Kiss Lóránd, Fülöp Ferenc:
Potenciálisan bioaktív 1,2,3-triazolok előállítása hatékony áramlásos technikával: az optimalizációtól a méretnövelésig
MTA Heterociklusos és Elemorganikus Kémiai Munkabizottság ülése, Balatonszemes, Hungary, 5-7 June 2013.

17. *Ötvös Sándor*, Georgiádes Ádám, Mándity István, Kiss Lóránd, Fülöp Ferenc:
Potenciálisan bioaktív 1,2,3-triazolok szintézise modern áramlásos technikával
Vegyészkonferencia, Hajdúszoboszló, Hungary, 26-28 June 2013. Abstr.: p. 28.

18. *Sándor B. Ötvös*, István M. Mándity, Ádám Georgiádes, Lóránd Kiss, Ferenc Fülöp: *Safe, efficient and scalable synthesis of novel potentially bioactive 1,2,3-triazoles in flow*
4th Conference on Frontiers in Organic Synthesis Technology, Budapest, Hungary, 16-18 October 2013. Abstr.: p. 26-27.

Other scientific lectures

19. *Sándor B. Ötvös*, Péter Berenji, Zoltán Németh, Ottó Berkesi:
IR and theoretical investigations of aromatic basic zinc carboxylates, precursors for the most popular MOF-s
The 13th International Symposium for Students in Chemistry, Timişoara, Romania, 21 November 2008. Abstr.: p. 18.

20. *Ötvös Sándor Balázs*, Berkesi Ottó, Körtvélyesi Tamás:
Aliciklusos és aromás karbonsavak bázikus négymagvú cink karboxilátjainak előállítása, szerkezetük elméleti kémiai és infravörös spektroszkópiai vizsgálata
XXXII. Kémiai Előadói Napok, Szeged, Hungary, 26-28 October 2009. Abstr.: p. 45-46.