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Fully Automated Continuous Flow Synthesis of Highly Functionalized Imidazo[1,2-a] Heterocycles

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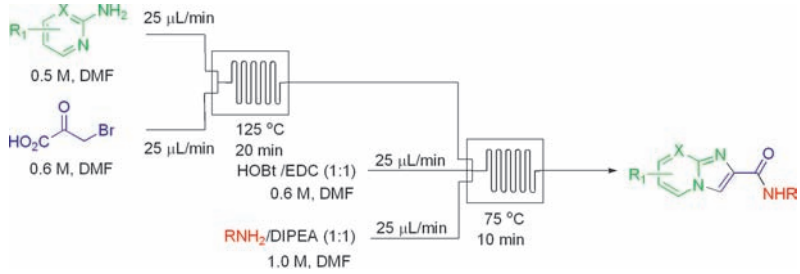
ABSTRACT



The first continuous flow synthesis of imidazo[1,2-a]pyridine-2-carboxylic acids directly from 2-aminopyridines and bromopyruvic acid has been developed, representing a significant advance over the corresponding in-flask method. The process was applied to the multistep synthesis of imidazo[1,2-a]pyridine-2-carboxamides, including a Mur ligase inhibitor, using a two microreactor, multistep continuous flow process without isolation of intermediates.

Over the past several years, high-throughput chemical synthesis has become increasingly important because of its potential to positively impact the drug discovery process.[1−3] Two of the most common hurdles encountered in the development of high-throughput syntheses of libraries of complex molecules have been slow reaction times and reliance on harsh or inconvenient reaction conditions. Moreover, the optimization and purification of these reactions are often time-consuming and inefficient. Automated microreactor-based (microfluidic chip) continuous flow systems have the potential to greatly accelerate the production of small molecule libraries.[4,5] Advantages include efficient heat transfer, enhanced reagent mixing, small reaction volumes, and the potential to run multistep reactions in a single, uninterrupted microreactor sequence using continuous flow conditions.[6,7] We are developing flow chemistry methods

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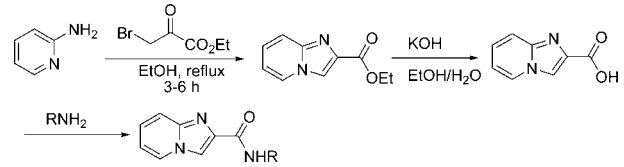
for transformations that are either not possible or highly inefficient using in-flask chemistry. This will enable the rapid synthesis of highly functionalized, drug-like small molecules in high purity and on a scale sufficient for evaluation in multiple biological assays. We previously reported the preparation of 1,2,4-oxadiazoles from arylnitriles and activated carbonyls in a single continuous flow sequence.[7a] We now report the efficient, multistep, continuous flow synthesis of diverse, highly functionalized imidazo[1,2-a] heterocycles directly from commercially available starting materials.

The imidazo[1,2-a] heterocyclic scaffold is found in compounds with anticancer, antiviral, and antimicrobial activities and in modulators of the nervous system.[8,9] Typically, the synthesis of imidazo[1,2-a] heterocycles requires long reaction times and high temperatures, limiting the accessibility of these biologically important structures.[10] Direct condensation of 2-bromopyruvic acid with 2-aminopyridines to generate imidazo[1,2-a]pyridine-2-carboxylic acids is inefficient due to competing decarboxylation of the product at high temperatures. Therefore, stepwise protocols are generally used for the formation of imidazopyridine carboxylic acid derivatives. For example, in a typical in-flask procedure, ethyl bromopyruvate is condensed with 2-aminopyridine (Scheme 1).

Scheme 1. In-Flask Preparation of Imidazo[1,2-a]pyridine-2-carboxylic Acids and Imidazo[1,2-a]pyridine-2-carboxamides



The product ester is isolated and subjected to saponification to provide the product.[11] Naturally, each in-flask step involves workup and purification of the intermediates. Herein we report the first continuous flow synthesis of imidazo[1,2-a]pyridine-2-carboxylic acids from 2-aminopyridines and bromopyruvic acid using a single microreactor to enable the rapid synthesis of these compounds. We further demonstrate the incorporation of this procedure into a continuous, two microreactor method for the highly efficient preparation of a diverse library of amide derivatives.

Focusing initially on the flow synthesis of imidazo[1,2,a] heterocyclic acids, we screened a variety of substrates and reaction conditions using a single microreactor and found that the use of a catalytic amount of *p*-toluenesulfonic acid (PTSA) (0.25 equiv) efficiently provided the desired products from 2-aminopyridines (1.0 equiv) and bromopyruvic acid (1.2 equiv) in dimethylformamide (DMF) at 125 °C. Although previous in-flask studies reported the use of protic solvents for the synthesis of this class of heterocycles, DMF was selected due to its high boiling point and excellent solubilizing properties. The 2-aminopyridine (0.5 M) and bromopyruvic acid (0.5 M) were introduced into the microreactor, and PTSA was either flowed in via a separate channel or premixed with the bromopyruvic acid. Reaction monitoring by LCMS analysis showed that the conversion of bromopyruvic acid to the corresponding imidazo[1,2-a]-pyridine-2-carboxylic acid derivative was complete within 10 min in a preheated microreactor at 125 °C and 4.0 bar. Under these conditions, a series of imidazo[1,2-a]pyridine-2-carboxylic acids were efficiently synthesized in moderate to high yields (Table 1).

Using the optimized reaction conditions, different imidazo[1,2-a]heteroaryl-2-carboxylic acids were synthe-

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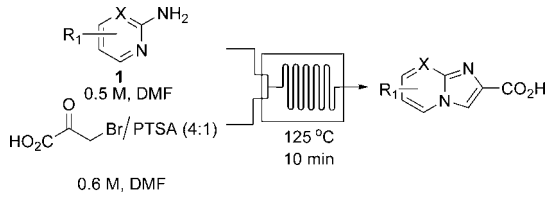
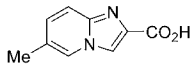
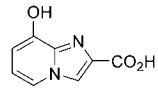
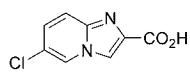
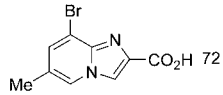
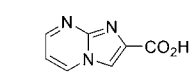
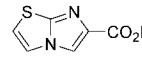
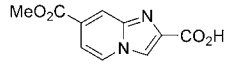
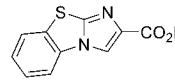
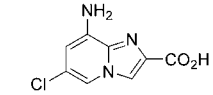
Table 1. Flow Synthesis of Imidazo[1,2-a]heteroaryl-2-carboxylic Acids



entry	product	yield (%)[a]	entry	product	yield (%)[a]
1		72	6		52
2		71	7		72
3		60	8		68
4		68	9		63
5		65			

[a] Isolated yield based on **1** after purification of a crude reaction mixture using preparative HPLC. Reactions were performed on 0.45 mmol scale.

sized (Table 1). Reactions of 2-aminopyridines having electron-donating (Table 1, entry 1) or electron-withdrawing groups (Table 1, entries 2 and 4) proceeded efficiently with moderate to excellent yields. Notably, functionalized 2-aminopyridines underwent cyclization leading to chloro- (Table 1, entries 2, 5), bromo- (Table 1, entry 7), hydroxy- (Table 1, entry 6), and amino- (Table 1, entry 5) substituted imidazo[1,2-a]pyridine-2-carboxylic acids. One intriguing observation was that this new method can be used to generate free amine or hydroxyl group-containing imidazopyridine carboxylic acids in an efficient manner, without the need to use protecting groups. The product in Table 1, entry 4 is notable since it is a dicarboxylic acid that is monoprotected as the ester, allowing selective functionalization of the free acid in subsequent synthetic transformations. Additionally, the new flow method can be applied to the synthesis of other imidazoheterocyclic scaffolds such as pyrimidines (Table 1, entry 3), thiazoles (Table 1, entry 8), and benzothiazoles (Table 1, entry 9). Significantly, the product in Table 1, entry 9 is a key intermediate for the preparation of AB530, a recently disclosed FLT3 kinase inhibitor under evaluation for the treatment of cancer.[12] Our single-step synthesis that provides the product in 63% yield constitutes a major improvement over the reported two-step, in-flask procedure to afford the product in 22% overall yield.

To demonstrate the utility of the newly developed methodology, our next goal was to develop a continuous flow method to access imidazo[1,2-a]pyridine-2-carboxamides in a single, uninterrupted process directly from readily available starting materials. Optimization studies showed that a combination of EDC/HOBt/DIPEA (1:1:2) for 10 min at 75 °C were the best conditions for complete conversion of the imidazoheterocyclic acids to the corresponding amide derivatives. Initial efforts to directly combine these two reactions using two microreactors were unsuccessful. Experimentation with a variety of reaction conditions demonstrated that removal of PTSA from the first reaction and the separate additions of solutions of EDC/HOBt (1:1) and amines/DIPEA (1:1) were necessary to achieve conversion. The major reaction competing with the amidation is the nucleophilic addition of the amine to EDC. Thus, to compensate for this side reaction the amidation reactions were performed using an excess of base and amine. The final optimized conditions employed were: (i) 1.2 equiv of bromopyruvic acid, (ii) a residence time on the first microreactor of 20 min and a temperature 100 °C, and (iii) the stream containing the imidazo[1,2-a]pyridine-2-carboxylic acid exiting the first microreactor was combined with a solution of EDC/HOBt and amines/DIPEA in a second microreactor at 75 °C for 10 min (Figure 1).

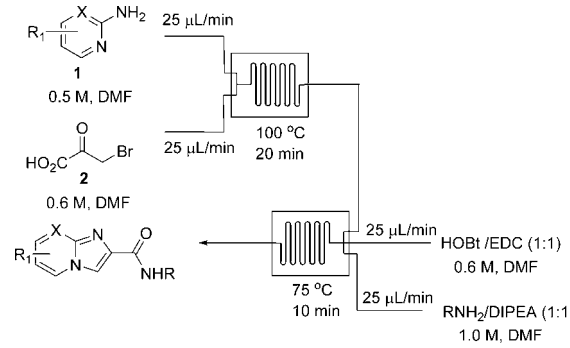


Figure 1. Continuous flow sequence for the synthesis of imidazo[1,2-a]pyridine-2-carboxamides.

Several aspects of this new continuous flow process are noteworthy. First, this is the first example of the synthesis of imidazo[1,2-a]heteroaryl-2-carboxamides in a single continuous process, directly from readily available starting materials (i.e., 2-aminopyridines and bromopyruvic acid). As noted previously, the in-flask synthesis of these compounds involves multiple reaction steps involving workup and purification of multiple intermediates,[13] often requiring the

(12) Patel, H. K.; Grotzfeld, R. M.; Lai, A. G.; Mehta, S. A.; Milanov, Z. V.; Chao, Q.; Sprankle, K. G.; Carter, T. A.; Velasco, A. M.; Fabian, M. A.; James, J.; Treiber, D. K.; Lockhart, D. J.; Zarrinkar, P. P.; Bhagwat, S. S. *Bioorg. Med. Chem. Lett.* **2009**, *19*, 5182–5185.

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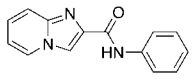
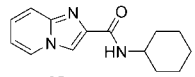
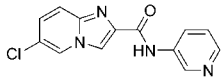
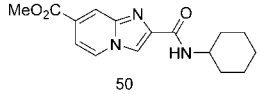
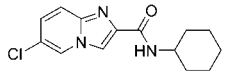
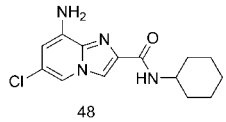
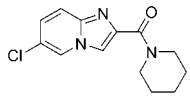
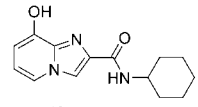
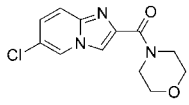
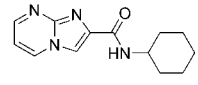
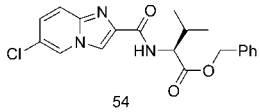
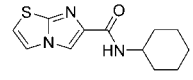
use of strong Lewis acids.[14] Second, until now, the accessibility of these amides was limited due to the lack of applicable methodologies to generate various amides using commercially available starting materials. Finally, since this method involves the introduction of amines in the final step, it provides a convenient handle for further derivitization of the products, and a wide variety of amides are accessible simply by changing the amines.

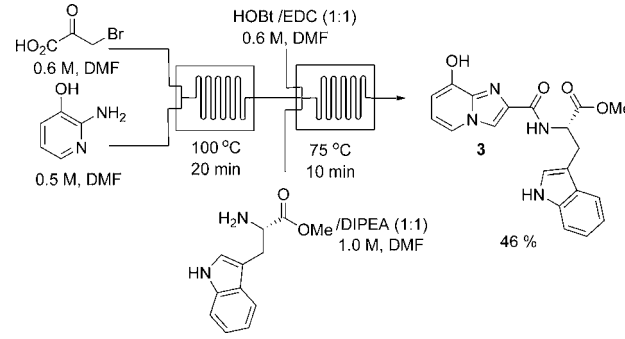
This novel continuous flow method tolerates a wide range of amines such as primary (Table 2, entries 3 and 7−12), secondary (Table 2, entries 4 and 5), aryl (Table 2, entry 1), heteroaryl (Table 2, entry 2), and heterocyclic (Table 2, entry 5). It is noteworthy that comparatively unreactive 3-aminopyridine couples efficiently (Table 2, entry 2). Additionally, amino acid derivatives can directly couple efficiently to introduce further complexity to the final products (Table 2, entry 6). Furthermore, different imidazo[1,2-a]heterocarboxamides such as thiazoles and pyrimidines can be accessed efficiently. Moreover, free amines (Table 2, entry 9) and free hydroxyl (Table 2, entry 10) containing amides can be synthesized efficiently. The tolerance of free amine and free hydroxyl groups is particularly noteworthy since previous syntheses of this type of scaffold were initially only achievable in a stepwise manner.[15] Finally, to demonstrate the utility of the new method, we synthesized compound **3**, a Mur ligase inhibitor with potential antibacterial activity.[16] The in-flask synthesis of this compound was reported to proceed in 16.4% overall yield over two steps. Using our continuous flow method, compound **3** was prepared in a single step in 46% yield (Scheme 2).

Table 2. Direct Multistep Synthesis of Imidazo[1,2-a]pyridine-2-carboxamides from **1** and **2**

entry	product	yield (%)[a]	entry	product	yield (%)[a]
1		58	7		65
2		55	8		50
3		62	9		48
4		46	10		42
5		44	11		44
6		54	12		48

[a] Isolated yield based on **1** after purification of the crude reaction mixture using preparative HPLC. Reactions were performed on a 0.38 mmol scale.

Scheme 2. Continuous Flow Preparation of Mur Ligase Inhibitor **3**



In summary, we have developed novel continuous flow methods for the synthesis of imidazo[1,2-a]pyridine-2-carboxylic acids and imidazo[1,2-a]pyridine-2-carboxamides using readily available starting materials and standard, commercially available reagents. This work documents the first highly efficient synthesis of imidazo[1,2-a]pyridine-2-carboxylic acids and amides directly from 2-aminopyridines and bromopyruvic acids. Furthermore, the continuous flow nature of our synthesis enables the facile scale-up of these compounds, which is highly advantageous for drug discovery. We anticipate that these advances will facilitate the rapid synthesis of these biologically important scaffolds. Further exploration of the reactivity features of the process and applications to focused library synthesis of complex biologically active molecules is in progress.

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Supporting Information Available: Experimental procedures and characterization data for all compounds including ^{1}H, ^{13}C NMR spectra and MS data. This material is available free of charge via the Internet at http://pubs.acs.org.

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