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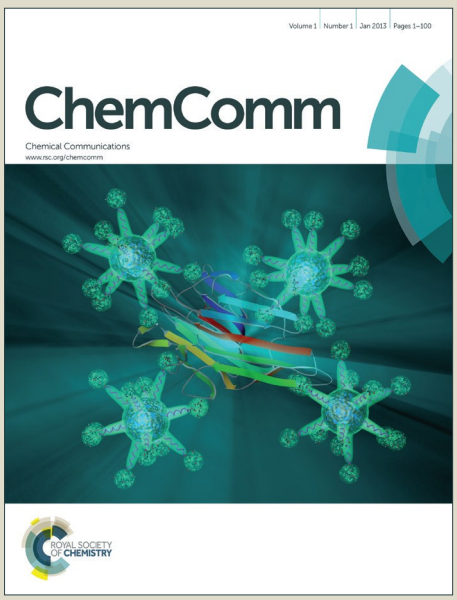

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Borinic acid Catalysed Peptide Synthesis

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The catalytic synthesis of peptides is a major challenge of modern organic chemistry hindered by the well established use of stoichiometric coupling reagents. Herein, we describe the first borinic acid able to catalyse this reaction under mild conditions with an improved activity compared to our recently developed thiophene-based boronic acid. This catalyst is particularly efficient for peptide bond synthesis affording dipeptides in good yields without detectable racemization.

The synthesis of peptides *via* the assembly of chiral *N*-protected α-amino acids and α-aminoesters is a well-known methodology.[1] This essential transformation could be easily realized with high level of predictability owing to the progress made with the use of stoichiometric peptide coupling reagents (Figure 1).[2]

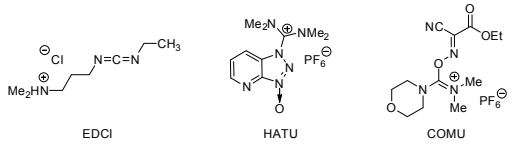

Figure 1. Representative coupling reagents in amide and peptide synthesis.

Despite those great accomplishments, a catalyst that would be able to achieve similar efficiency is still to be discovered. Albeit the catalytic synthesis of amides from simple carboxylic acids and amines has been reviewed several times[3], almost no information concerning the assembly of two α-amino acids derivatives is available. Compared to simple carboxylic acids and amines, protected α-amino acids are challenging substrates, due to their weaker nucleophilic character of the amine function and increased steric hindrance caused by the side chain.

Inspired by Yamamoto's pioneering work[4], Hall[5] (**3a-b**, Figure 2), Tam[6] (**4**, Figure 2) and our laboratory (**5**, Figure 2)[7] have recently reported boronic acids able to promote room temperature coupling of non-activated carboxylic acids and amines. Regarding peptide synthesis, 25 mol% of boronic acid **5** was found to promote the formation of Boc-Phe-Val-OMe in 50% yield with no detectable racemization, thus achieving the first synthesis of a dipeptide using a simple chemical catalyst.

Previously, the coupling of protected α-amino acids has been explored by Whiting[8] using stoichiometric quantities of a combination of aryl boronic acids **1**-**2** (Figure 2). However, low to moderate yields of the corresponding dipeptides were obtained. Interestingly, a cooperative effect of combined catalyst systems was suggested to be beneficial when 50 mol% of **1a** and 50 mol% of **1b** were used together for the coupling of hindered valine derivatives.[9]

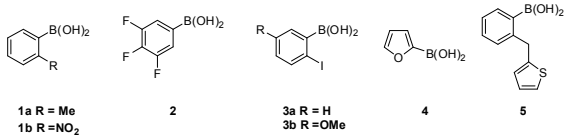

Figure 2. Aryl boronic acids investigated by Whiting, Hall, Tam and Blanchet.

Although peptides are of great value to pharmaceutical industry, their catalysed synthesis is still unknown. Therefore, the next challenge lies in identifying an efficient catalyst that would directly couple *N*-protected and *C*-protected α-amino acids to provide the desired dipeptides in reasonable yields without racemization.

Since boronic acid **5** has shown moderate activity in peptide coupling, we decided to shift to a potentially more reactive boron derivative. We hypothesized that the use of borinic acids, which possess an amplified Lewis acidity at the boron centre[10], can provide an improved catalytic system. Additionally, they offer an improved structural flexibility for further tuning of the electronic and steric properties with two aromatic moieties being attached to the same boron atom. In this context, we report the use of borinic acids for catalysed peptide synthesis.

We started our investigations by synthesizing a library of biaryl borinic acids according to known procedures (See Supporting Information). The reactivity of these borinic acids was compared to a set of commercially available boronic acids in the model reaction carried out between phenylacetic acid **6** and benzylamine **7** (Table 1). Interestingly, higher yields were systematically obtained with

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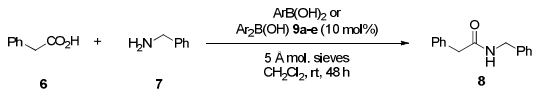
the borinic acids. With electron donating group substituted borinic acids **9c** (4-t-Bu), **9d** (4-MeO) and **9e** (3-MeO) 39-59% isolated yields were obtained when the corresponding boronic acids were found to be essentially unreactive (Table 1, Entries 3 and 4).

Table 1. Borinic acids vs boronic acids in the coupling of acid **6** and amine **7**.[a]



6 7 8

Entry	Ar	$Ar_2B(OH)$	Yield with $ArB(OH)_2$ (%)	Yield with $Ar_2B(OH)$ (%)
1		**9a**	13	19
2		**9b**	5	15
3		**9c**	0	59
4		**9d**	4	45
5		**9e**	5	39

[a] Reaction conditions: Phenylacetic acid **6** (0.55 mmol), benzylamine **7** (0.50 mmol) and molecular sieves (1g of 5 Å activated powder) were stirred in dry CH_2Cl_2 (7 mL) with borinic acids **9a-e** or boronic acid (10 mol%) under inert atmosphere for 48h at room temperature.

Encouraged by these results, the screening of various aryl borinic acids was undertaken. Based on Hall's previous work,[5] we focused our attention on halogen substituted borinic acids.

The presence of a fluorine atom at the *ortho* position in **9f** decreased the yield to 63% compared to the 81% yield obtained with the fluorine at the *para* position **9g** (Table 2, Entries 1 and 2). A different picture was observed with the introduction of a chlorine atom. Indeed, the 2-chloro substituted **9h** was found to be very effective delivering a quantitative yield of the amide **8** (Table 2, Entry 3) and suggesting that increased electronegativity at the *ortho* position in **9f** lowered the reactivity of the catalyst. The importance of the *ortho*-chloro substituent was shown by the lower yields (76% and 46% respectively) obtained with the *meta* **9i** and para **9j** substituents (Table 2, Entries 4 and 5). 2-Bromo substituted **9k** led to a similar reactivity as **9h** (Table 2, Entry 6), however, with a much less pronounced availability.[11] These results suggest the need for a specific electronic requirement in the *ortho* position for an optimal catalytic performance.

Hall and co-workers have reported a dramatic improvement of the performance of boronic acid **3a** (Figure 2) when an electron donating group was introduced *para* to the halogen atom. In their study, the 5-methoxy substituted **3b** was identified to provide faster reactions thus supporting the role of the *ortho* iodo substituent as a hydrogen-bond acceptor in the proposed transition state.[5a] Based on these results, we prepared the 5-methoxy substituted borinic acid **9l**.

Table 2. Aryl borinic acid screening.[a]

6 7 8

Entry	$Ar_2B(OH)$	Ar	Isolated Yields (%)
1	**9f**		63
2	**9g**		81
3	**9h**		99 (99[b])[d]
4	**9i**		76
5	**9j**		46
6	**9k**		98 (99[b])[d]
7	**9l**		80
8	**9m**		37
9	**9n**		18
10	**9o**		99 (97[c])[d]

[a] Reaction conditions: Phenyl acetic acid **6** (0.55 mmol), benzylamine **7** (0.50 mmol) and molecular sieves (1 g of 5 Å activated powder) were stirred in dry CH_2Cl_2 (7 mL) with borinic acids **9f-o** (10 mol%) under inert atmosphere for 48h at room temperature. [b] ^{1}H NMR conversion after 6 hours. [c] ^{1}H NMR conversion after 4 hours. [d] Conversions calculated using 1,3,5-trimethoxybenzene (10 mol%) as an internal standard.

However, in our hands, catalyst **9l** was less effective affording amide **8** with 80% yield (Table 2, Entry 7). Substituting the methoxy group with a chlorine or fluorine atom significantly decreased the catalyst performance (**9m** and **9n**, 37% and 18% yields respectively, Table 2, Entries 8 and 9). Interestingly, switching the fluorine position *para* to the boron atom in **9o** enhanced the catalytic activity with 97% conversion being attained within 4 hours (99% yield, Table 2, Entry 10) compared to 99% conversion attained within 6 h with catalyst **9h** (99% yield, Table 2, Entry 3). These results illustrate the importance of fine-tuning the electronic properties of the catalyst in terms of electron-density of the *ortho*-halogen and boron Lewis acid property.

Having identified **9o** as a useful catalyst, we monitored the reaction using ^{19}F-NMR to collect some insights regarding the mechanism of the reaction. Indeed, it was important to rule out any boronic acid catalysis consecutive to a side protodeboronation reaction facilitated by the increased Lewis acidity of borinic acids.[12]

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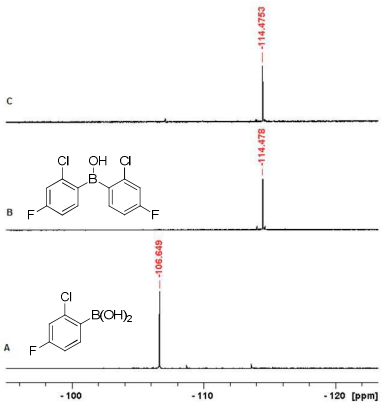
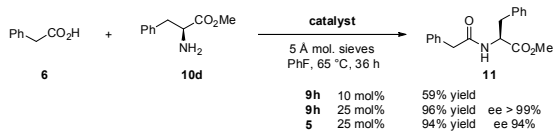


Figure 3. Monitoring the catalyst **9o** behaviour using ^{19}F probe (375 MHz; $CDCl_3$; 25 °C). **A**: isolated 2-Cl-4-F-C_6H_4-$B(OH)_2$. **B**: isolated **9o**. **C**: NMR monitoring of the reaction corresponding to entry 10, table 2, after 6h (97% conversion).

During the whole course of the reaction, a single signal at -114.5 ppm in ^{19}F NMR (Figure 3) was observed, with no intermediates being detected. This signal matched the isolated **9o**, while the corresponding boronic acid was measured at -106.7 ppm, suggesting the stability of the catalyst **9o** during the reaction.

While **9o** was found to be slightly more reactive than **9h**, the combined stability and higher availability of **9h** led us to select it as the optimal catalyst for the further investigations.

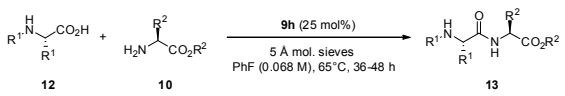
Before exploring dipeptide synthesis, a control reaction was carried out between phenyl acetic acid **6** and phenylalanine methyl ester **10d** to ascertain the absence of racemization (Scheme 1). The use of less reactive α-amino ester **10d** required the use of 25 mol% of catalyst **9h** and a switch to a different low polarity solvent and a higher temperature (fluorobenzene, 65 °C).



Scheme 1. Coupling between phenyl acetic acid **6** and phenylalanine methyl ester **10**.

Amide **11** was previously reported by Whiting for its propensity to provoke racemization, providing an enantiomeric excess of 68% under similar reaction conditions with catalysts **1a** and **1b**.[8] This result was previously improved using our catalyst **5** (ee: 94%).[7] Interestingly, **9h** afforded the desired amide **11** in 96% yield with the complete retention of enantiopurity (ee > 99%, Scheme 1). First attempts of coupling two protected α-amino acids at room temperature resulted in the total absence of conversion. However using previously optimized conditions (Scheme 1), Boc-protected phenylalanine (Boc-Phe) was efficiently coupled with various methyl α-amino esters derived from hindered Valine, Alanine, Glycine and phenylalanine in average 47-61% yields (Table 3, Entries 1-4). Similarly, Z-proline (Z-Pro) was coupled with various α-amino esters (Table 3, Entries 5-11) in 47-73% yields. While the modification of the nature of the ester group led to marginal change when valine α-amino ester was used (Table 3, Entries 7-8), a higher yield was obtained when an ethyl glycine ester (H-Gly-OEt) was used compared to benzyl or methyl counterparts (Table 3, Entries 9-11). Highest 80-72% yields were also obtained with the less hindered Boc-glycine (Boc-Gly-H) as the nucleophilic component of the reaction (Table 3, Entries 12-13) while Z-methionine (Z-Met) led to a non-optimized 40% yield (Table 3, Entry 14). Careful examination of the ^1H NMR data of all the synthesized dipeptides incorporating two chiral centres showed that a single diastereomer was obtained.

Table 3. Scope of the reaction: peptide synthesis.[a]



Entry	α-Amino-acid **12**		α-Amino-ester **10**		Dipeptide **13**		Yield (%)
1	Boc-Phe-H	**12a**	H-Val-OMe	**10a**	Boc-Phe-Val-OMe	**13a**	51
2	Boc-Phe-H	**12a**	H-Ala-OMe	**10b**	Boc-Phe-Ala-OMe	**13b**	47
3	Boc-Phe-H	**12a**	H-Gly-OMe	**10c**	Boc-Phe-Gly-OMe	**13c**	55
4	Boc-Phe-H	**12a**	H-Phe-OMe	**10d**	Boc-Phe-Phe-OMe	**13d**	61
5	Z-Pro-H	**12b**	H-Phe-OMe	**10d**	Z-Pro-Phe-OMe	**13e**	60
6	Z-Pro-H	**12b**	H-Leu-OMe	**10e**	Z-Pro-Leu-OMe	**13f**	60
7	Z-Pro-H	**12b**	H-Val-OMe	**10a**	Z-Pro-Val-OMe	**13g**	58
8	Z-Pro-H	**12b**	H-Val-OBn	**10f**	Z-Pro-Val-OBn	**13h**	59
9	Z-Pro-H	**12b**	H-Gly-OEt	**10g**	Z-Pro-Gly-OEt	**13i**	73
10	Z-Pro-H	**12b**	H-Gly-OBn	**10h**	Z-Pro-Gly-OBn	**13j**	47
11	Z-Pro-H	**12b**	H-Gly-OMe	**10c**	Z-Pro-Gly-OMe	**13k**	51
12	Boc-Gly-H	**12c**	H-Gly-OBn	**10h**	Boc-Gly-Gly-OBn	**13l**	80
13	Boc-Gly-H	**12c**	H-Phe-OMe	**10d**	Boc-Gly-Phe-OMe	**13m**	72
14	Z-Met-H	**12d**	H-Phe-OMe	**10d**	Z-Met-Phe-OMe	**13n**	40

[a] Reaction conditions: *N*-protected α-amino acid **12** (0.46 mmol), α-aminoester **10** (0.46 mmol) and molecular sieves (1 g of 5 Å activated powder) were stirred in dry PhF (6.8 mL)with the borinic acid catalyst **9h** (25 mol%) under inert atmosphere for 32-48h at 65 °C.

Based on Hall's mechanism with boronic acids, we rationalized that the *N*-protected α-amino acid is activated in the form of the mixed boron anhydride **A** (Figure 4). This step is greatly facilitated by the increased Lewis acidity of the borinic acid **9h**. The next step involves

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the additions of the α-amino ester to generate a tetravalent intermediate **B**. The collapse of **B** is then assisted by water and the Lewis basicity of *ortho* chlorine substituents to yield eventually the desired dipeptide.

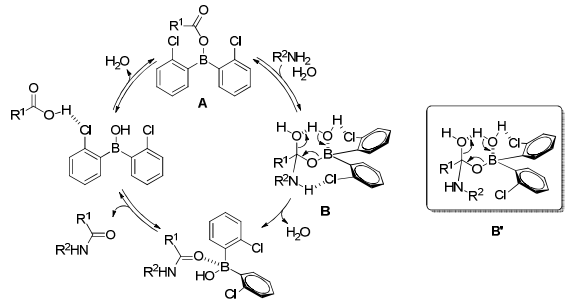
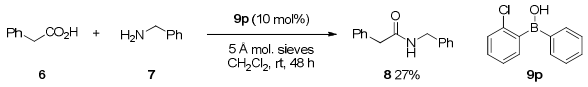


Figure 4. Proposed mechanism for the amide synthesis catalysed by the borinic acid 9h.

To clarify if both chlorine atoms are involved, the unsymmetrical borinic acid **9p** was prepared and tested (Scheme 2). With this catalyst bearing a single chlorine atom, the yield was reduced from 99% with **9h** (Table 2) to 27%.



Scheme 2. Reactivity of borinic acid **9p** in the model reaction between phenylacetic acid **6** and benzylamine **7**.

Accordingly, the specific reactivity of **9h** and **9k** tends to indicate that both halogen atoms are probably involved in the rate-determining step and that the intermediate **B** is formed rather than **B'** (Figure 4).

Indeed, mild hydrogen-halogen bondings might stabilize the transition state and favour the collapsing of **B**. Additionally we suggest that the formation of anhydride **A** is pre-organized by similar hydrogen-halogen bonding.

To summarize, we have uncovered the unique reactivity of borinic acids for amide and peptide syntheses. Fourteen dipeptides were prepared in synthetically useful yields without detectable racemization. Importantly, no stoichiometric peptide coupling reagents were used and thus minimal waste was generated with water being the sole side product of the reaction. However, there is still room for improvements; progress is necessary to match the requirements of solid phase peptide synthesis such as very high yields and short reaction times. We believe that those results will shed light on new strategies aiming at developing the catalysed peptide synthesis, which exhibits an utmost importance for the pharmaceutical industry.

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