Microwave-Assisted Transformation of Esters into Hydroxamic Acids

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Abstract: A general, mild and efficient procedure with which to access hydroxamic acids, in good yields and purity, is reported. Esters are used as substrates and reacted with hydroxylamine, in the presence of a base, under microwave activation. The method has been successfully applied to enantiomerically pure esters without loss of stereochemical integrity.

Key words: esters, chemoselectivity, hydroxamic acid, microwave reactions, metalloproteinases

Hydroxamic acids are bidentate chelating agents that interact with several zinc(II)-containing proteins. Molecules containing this functional group behave as powerful inhibitors of metalloproteinases1–4 and histone deacetylase,5–6 which are two important biological targets for anti-cancer therapy. Moreover, the hydroxamic acid motif occurs in other biomolecules such as naturally occurring siderophores7,8 like pseudobactines, desferrioxamines, ferrichromes and the fosmidomycin antibiotic9 and its analogues10 which have been recovered and found to be potent antimalarial agents.10–11

Due to its low stability, this hydroxamate moiety is usually introduced in the last step of the synthetic sequence and, recently, several methods have been developed for this purpose. So far, the most commonly used approaches can be summarized as: reaction of O/N-protected hydroxylamine with activated carboxylic acids,12–15 reaction of esters with hydroxylamine,16,17 treatment of N-aclyoxazolidines with hydroxylamine in the presence of samarium triflate,18 and coupling of carboxylic acids with hydroxylamine in the presence of 2,4,6-trichloro[1,3,5]triazine19 or cyclic phosphonic anhydride (PPAA).20 However, a general, mild and efficient procedure with which to access hydroxamic acids, in good yields and purities, is still required.

Stemming from our interest in the development of new metalloproteasase inhibitors,21,22 we have recently faced the problem of transforming esters into the corresponding hydroxamic acids at the end of multistep sequences without loss of stereoechemical integrity in the final product. As a convenient methodology, we envisaged the use of microwave (MW) activation in this simple transformation, using hydroxylamine as a reagent, in the presence of bases such as potassium hydroxide or sodium ethoxide.

Since it is known that MW irradiation generally speeds up slow reactions by simple, very efficient heating of the system,23 we hoped to overcome the problems associated with the long reaction times that are normally required for the above transformation, in this way. To test our hypothesis, we submitted an array of methyl esters to MW irradiation, in the presence of hydroxylamine and potassium hydroxide as base; the results are collected in Table 1.

The data reported in Table 1 clearly show that the formation of hydroxamic acids from esters under MW irradiation, occurs smoothly and is quite general; indeed it can be performed on simple alkylaryl esters (entry 1), on both N-protected (entries 2, 3, 5, 6) and unprotected (entry 4) amino esters, on dipetides (entries 7, 8), on protected hydroxysters (entry 9) and on sulfonamido esters (entry 10), though in the latter case a partial decomposition of the starting material was observed. Furthermore, it is worthwhile noting that protective groups are also well tolerated under these reaction conditions. The tert-butoxy-carbonyl (Boc) group for example, which can be lost under MW irradiation in the presence of silica gel24 or Lewis acids,25 remains intact under our reaction conditions, though a partial deprotection was observed in the reaction with Boc-proline ester (entry 5). Amidic bonds (entries 7, 8) and ketals (entry 9) also survive without any detectable decomposition. All the reactions went to completion in about six minutes, except in the case of the conversion of Boc-protected phenylalanine methyl ester, which required longer reaction times (12 min).

Of particular interest is the dipetide in entry 8, which has been designed to be a new matrix metallo-protease (MMP) inhibitor. Whilst substrate 1h, which has previously been obtained through a multistep synthesis,26 could not be transformed into the desired hydroxamic acid through usual methods (as mainly starting ester was recovered), MW irradiation led to the desired product 2h, albeit in moderate yield.

Yields of the MW-induced transformations were generally good. All the final compounds gave a positive test for hydroxamic acid by treatment with an iron(III) chloride solution.27 The ester was consistently completely consumed to give the hydroxamic acid as the major product, sometimes accompanied by small amounts of the corre-
<table>
<thead>
<tr>
<th>Entry</th>
<th>Ester</th>
<th>Hydroxamic acid</th>
<th>Yield</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td><img src="image1.png" alt="Image of ester 1a" /></td>
<td><img src="image2.png" alt="Image of hydroxamic acid 2a" /></td>
<td>71%</td>
</tr>
<tr>
<td>2</td>
<td><img src="image3.png" alt="Image of ester 1b" /></td>
<td><img src="image4.png" alt="Image of hydroxamic acid 2b" /></td>
<td>100%</td>
</tr>
<tr>
<td>3</td>
<td><img src="image5.png" alt="Image of ester 1c" /></td>
<td><img src="image6.png" alt="Image of hydroxamic acid 2c" /></td>
<td>70%</td>
</tr>
<tr>
<td>4</td>
<td><img src="image7.png" alt="Image of ester 1d" /></td>
<td><img src="image8.png" alt="Image of hydroxamic acid 2d" /></td>
<td>88%</td>
</tr>
<tr>
<td>5</td>
<td><img src="image9.png" alt="Image of ester 1e" /></td>
<td><img src="image10.png" alt="Image of hydroxamic acid 2e" /></td>
<td>50%b</td>
</tr>
<tr>
<td>6</td>
<td><img src="image11.png" alt="Image of ester 1f" /></td>
<td><img src="image12.png" alt="Image of hydroxamic acid 2f" /></td>
<td>50%b</td>
</tr>
<tr>
<td>7</td>
<td><img src="image13.png" alt="Image of ester 1g" /></td>
<td><img src="image14.png" alt="Image of hydroxamic acid 2g" /></td>
<td>100%</td>
</tr>
<tr>
<td>8</td>
<td><img src="image15.png" alt="Image of ester 1h" /></td>
<td><img src="image16.png" alt="Image of hydroxamic acid 2h" /></td>
<td>40%</td>
</tr>
<tr>
<td>9</td>
<td><img src="image17.png" alt="Image of ester 1i" /></td>
<td><img src="image18.png" alt="Image of hydroxamic acid 2i" /></td>
<td>98%</td>
</tr>
<tr>
<td>10</td>
<td><img src="image19.png" alt="Image of ester 1k" /></td>
<td><img src="image20.png" alt="Image of hydroxamic acid 2k" /></td>
<td>33%c</td>
</tr>
</tbody>
</table>

Table 1 Conversion of Esters into Hydroxamic Acids under Microwave Irradiation

*a All the reactions were carried out by irradiating the ester and hydroxylamine in MeOH, in the presence of base, for 6 min at 80 °C and 150 W.

*b Protecting group was partially removed.

*c Partial decomposition to 4-methoxyphenylsulfonic acid was observed.
sponding carboxylic acid (entries 8, 9). Changing the base used (potassium hydroxide or sodium ethoxide) did not affect the outcome of the reaction; however, it must be pointed out that the use of methanol as solvent was crucial since alternative alcohols promoted initial transesterification, which led to a decrease in the rate of hydroxamic acid formation.

With compounds 1a, 1b and 1d, a comparison between classical heating (oil bath) and MW irradiation was carried out and the results obtained (Table 2), clearly show an improved performance of the MW-assisted process.

### Table 2: Comparison of Reactions with and without MW Irradiation

<table>
<thead>
<tr>
<th>Ester Without MW irradiation</th>
<th>With MW irradiation</th>
</tr>
</thead>
<tbody>
<tr>
<td>25 °C, 4 h</td>
<td>25 °C, 12 h</td>
</tr>
<tr>
<td>1a 49%</td>
<td>15%</td>
</tr>
<tr>
<td>1b 59%</td>
<td>55%</td>
</tr>
<tr>
<td>1d 64%</td>
<td>71%</td>
</tr>
</tbody>
</table>

Finally, we demonstrated that the use of MW irradiation did not affect the stereochimical integrity of the chiral compounds since the optical rotation of the hydroxamic acid derivative of Boc-alanine methyl ester was found to be in agreement with the literature value \[\{[\alpha]^{20}_{D}\} = -35 (c 0.8, MeOH); \text{Lit.} \]

In summary, we have shown that hydroxamic acids can be easily prepared from the corresponding esters, through a simple protocol, using hydroxylamine and a base under microwave irradiation. The method is quite general, and can be applied to enantiomerically pure amino esters without any loss of stereochimical integrity.

Starting materials were purchased from commercial sources and used without further purification. Sulfonamide 1k was synthesized according to the literature.\[^{26}\]

Dipeptide N-Boc-Val-Phe-OMe (1g) was prepared following typical peptide synthesis coupling procedures.\[^{29}\]

Dipeptide ester 1h was synthesized via a multistep sequence.\[^{26}\]

Ethereal extracts were dried with Na2SO4. Reactions were monitored by TLC using SiO2-coated plates; detection was accomplished through staining with a KMnO4 basic solution. Flash column chromatography was performed using glass columns (10–50 mm wide) and SiO2 (230–400 mesh).\[^{1}\]

\[^{1}\] H NMR spectra were recorded at 200 or 400 MHz. For those compounds that are present as slowly interconverting rotamers, \[^{1}\]H NMR experiments were performed at 50 °C and signals of the averaged spectrum are reported when possible.\[^{1}\]C NMR spectra were recorded at 50 MHz. Chemical shifts were determined relative to the residual solvent peak (MeOH, \(\delta = 3.31\) ppm for \[^{1}\]H NMR; MeOH, \(\delta = 49.0\) ppm for \[^{13}\]C NMR). Polarimetric measurements were performed at \(\lambda = 589\) nm at the temperature specified.

### Hydroxamic Acid Preparation; Typical Procedure

Methyl ester (0.2 mmol) was dissolved in anhydrous MeOH (0.5 mL) under nitrogen. The solution was charged with a preformed slurry of hydroxylamine hydrochloride (44 mg, 0.6 mmol) and KOH (71 mg, 1.2 mmol) in MeOH and the sealed flask was inserted into the cavity of a Discovery Microwave System apparatus (CEM) and heated for 6 min at 80 °C by microwave irradiation at 150 W (the power level was previously allowed to settle).

The reaction mixture was acidified with 3 M HCl (or aq 25% AcOH in the case of acid-sensitive compounds) to apparent pH 4. The resulting solution was concentrated and the residue was dissolved in a small amount of H2O and extracted several times with EtOAc until TLC analysis of the aqueous phase revealed that no ferric chloride-positive component remained.

The organic layer was dried over anhydrous Na2SO4 and the filtrate was concentrated to give the desired hydroxamic acid, which was purified by reverse phase HPLC (semipreparative column, Ultrasphere C-8, \(5 \mu m\), 250 × 10 mm, flow rate 3.5 mL/min, MeCN–H2O, gradient containing 0.05% TFA).

\[^{1}\]H NMR (CD3OD, 200 MHz): \(\delta = 7.26–7.31\) (m, 5 H), 3.44 (s, 2 H).

\[^{13}\]C NMR (CD3OD, 50 MHz): \(\delta = 166.2, 136.2, 130.1, 129.2, 127.6, 35.9\).

(5)-\[^{N}\text{-tert-Butoxy carbonyl}\]alanine Hydroxamate (2b)\[^{17}\]

Yield: 200 mg (100%); oil; \(\{[\alpha]_{D}^{20}\} = 29 \text{ (c 1, MeOH)}\)

\[^{1}\]H NMR (CD3OD, 400 MHz): \(\delta = 4.01\) (dd, \(J = 7.0, 1.1\), 1 H), 1.43 (s, 9 H), 1.28 (dd, \(J = 7.2, 3\), 1 H).

\[^{13}\]C NMR (CD3OD, 50 MHz): \(\delta = 169.9, 155.6, 78.1, 49.4, 28.5, 17.9\).

(5)-\[^{N}\text{-tert-Butoxy carbonyl}\]phenylalanine Hydroxamate (2c)\[^{17}\]

Yield: 280 mg (70%); oil.

\[^{1}\]H NMR (CD3OD, 200 MHz): \(\delta = 7.34–7.23\) (m, 5 H), 4.20 (m, 1 H), 3.05 (dd, \(J = 8.4, 13.6\), 1 H), 2.85 (dd, \(J = 6.6, 13.6\), 1 H), 1.39 (s, 9 H).

\[^{13}\]C NMR (CD3OD, 50 MHz): \(\delta = 170.5, 157.0, 138.1, 130.1, 129.2, 127.4, 80.4, 55.1, 39.4, 28.6\).

(5)-Proline Hydroxamate (2d)\[^{16}\]

Yield: 166 mg (88%); oil.

\[^{1}\]H NMR (D2O, 200 MHz): \(\delta = 4.31–4.22\) (m, 1 H), 3.43–3.35 (m, 2 H), 2.45–2.35 (m, 2 H), 2.20–2.0 (m, 2 H).

\[^{13}\]C NMR (CD3OD, 50 MHz): \(\delta = 169.8, 59.4, 43.9, 26.3, 22.7\).

(5)-\[^{N}\text{-tert-Butoxy carbonyl}\]proline Hydroxamate (2e)\[^{19}\]

Yield: 230 mg (50%); oil.

\[^{1}\]H NMR (CD3OD, 200 MHz): \(\delta = 4.22–4.10\) (m, 1 H), 3.60–3.40 (m, 2 H), 2.0–1.80 (m, 4 H), 1.44 (s, 9 H).

\[^{13}\]C NMR (CD3OD, 50 MHz): \(\delta = 169.9, 158.1, 79.8, 56.8, 43.9, 28.8, 26.4, 21.8\).

(5)-\[^{N}\text{-tert-Butyl 4-(Hydroxycarbamoyl)-2,2-dimethyl oxazolidin-3-carboxylate (2f)}\[^{20}\]

Yield: 246 mg (71%); oil.

\[^{1}\]H NMR (CD3OD, 200 MHz): \(\delta = 3.31–3.13\) (m, 3 H), 3.43–3.35 (m, 2 H), 3.20–3.02 (m, 2 H), 2.50–2.35 (m, 2 H), 1.20–1.05 (m, 2 H), 1.00–0.85 (m, 2 H).

\[^{13}\]C NMR (CD3OD, 50 MHz): \(\delta = 166.5, 152.4, 80.3, 66.4, 65.8, 62.4, 27.2, 23.1\).
text-Butyl (S)-1-[(S)-1-(Hydroxycarbamoyl)-2-phenylethyl-carbamoyl]-2-methylpropylcarbamate (2g)
Yield: 80 mg (100%); colorless solid; mp 106–108 °C; [α]D25 + 3.5 (3.55, MeOH).
1H NMR (CD3OD, 400 MHz): δ = 7.27–7.26 (m, 5 H), 4.55 (t, J = 7.6 Hz, 1 H), 3.82 (d, J = 6.7 Hz, 1 H), 3.17–2.85 (m, 2 H), 1.98–1.83 (m, 1 H), 1.44 (s, 9 H), 0.83 (d, J = 6.8 Hz, 6 H).
13C NMR (CD3OD, 50 MHz): δ = 173.2, 169.6, 157.9, 137.9, 130.1, 129.2, 127.5, 80.6, 61.7, 53.5, 38.8, 31.9, 28.7, 19.6, 18.6.

(S)-1-[(S)-3-Benzylsulfanyl-2-(furan-2-ylmethylisobutylamino)-propionyl]piperidine-2-carboxylic Acid Hydroxyamide (2h)
Yield: 260 mg (33%); oil.

We thank Julie Colombel for her contribution in running some of the MW-induced reactions.

References