Journal of Labelled Compounds and Radiopharmaceuticals
J Label Compd Radiopharm 2007; **50**: 189–196.
Published online in Wiley InterScience
(www.interscience.wiley.com). DOI: 10.1002/jlcr.1254

JLCR

Research Article

Expedient synthesis of deuterium-labelled amides within micro-reactors

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Received 12 December 2006; Revised 8 January 2007; Accepted 9 January 2007

Abstract: The pharmaceutical industry relies heavily on the synthesis of small quantities (10–500 mg) of stable, isotopically labelled compounds in the evaluation of new drug candidates for metabolism studies. As a result of the phenomenal cost of labelled materials even the preparation of small quantities can be extremely expensive.

In this paper, for the first time, we report that micro-reactor technology may be used to prepare stable deuterium-labelled compounds by conducting all optimization experiments using unlabelled precursors and simply substituting the labelled derivatives once the optimization is complete. Here, we wish to present a simple, general procedure for the synthesis of amides containing isotopic labels demonstrated using [*C*-$^{2}H_3$]acetyl chloride **1**. The reaction is carried out within a micro-reactor set-up which we believe offers superiority over other reported methods viz requiring stoichiometric quantities of reagents, high containment of the system and generality of the technique, obtaining products in high yields. Copyright © 2007 John Wiley & Sons, Ltd.

Keywords: micro-reactor; amide; deuterium; label; synthesis

Introduction

Micro-reactors are best described as a network of interconnecting micron sized (10–500 μm) channels etched into a solid substrate and a number of typical designs are illustrated schematically in Figure 1. Fabrication of such devices in many materials such as ceramics, metal and polymers is possible but for this study glass micro-reactors manufactured in house by photolithography and wet etching have been used.[1] Made from this material micro-reactor 'chips' are economical to produce but more importantly they are inert to all the chemicals and solvents used in synthesis. One of the simplest micro-reactor designs is a 'T' whereby two separate reagents are moved through the reactor, either hydrodynamically (using syringe pumps) or electroosmotically (by applying a voltage), towards the outlet.[2] When the two streams are brought together within the micron-sized channels they flow in a laminar fashion, parallel to one another and are free of turbulence.[3] Consequently, mixing transpires exclusively by diffusion which, at these small dimensions, occurs very rapidly. Additionally the small channel size gives rise to high surface to volume ratios leading to rapid heat dispersion and efficient temperature control.[4] The overall result is a highly controlled regime that, when optimized, provides a continual flow of desired product from the device. The optimization process for a reaction is commonly focused on minimizing undesirable by-products and maximizing productivity, typically achieved by varying concentrations and flow rates. The end product is an efficient system which, when coupled with the high containment and low volumes of chemicals is both safer and produces less waste. Transferring from laboratory to bulk-scale production is then a simple process. Since scaling the apparatus up would alter the unique dimensions of the reactor, it is far simpler to run many micro-reactors in parallel. Scaling out in this way retains the original methodology and reduces lab to plant development time and cost significantly. Many traditional synthetic reactions have been successfully performed using micro-reactors within our laboratories and across the globe[5] and of late potential in the area of PET chemistry has been demonstrated in the synthesis of nanogram quantities of ^{11}C and ^{18}F labelled compounds.[6] For example a multi-step preparation of the popular

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Contract/grant sponsor: The University of Hull

tumour imaging agent ^{18}F-fluroxydeoxyglucose in a micro-flow cell has recently been described.[7] The approach of using micro-reactors allows for rapid radiosynthesis which is a driving factor in the development of new methodologies.[8] Additionally, for both labelling with radioactive or stable isotopes, the cost of specifically labelled precursors is also of importance and the inherent, well-established effects of a miniaturized reactor to deliver high yields using only stoichiometric quantities of reagents make them ideal candidates for isotopic labelling.

The most well-known pharmaceutical to contain the amide unit is undoubtedly paracetamol, however, it is also found in many other naturally occurring or biologically active molecules. For example the basic core of all penicillin's contain the amide linkage as does oseltamivir (Tamiflu) which is currently being stockpiled due to its inhibitory effects towards certain strains of influenza. The amide functionality is also found in paclitaxel (Taxol) which is used to treat cancerous tumours and in the famously misused but highly active lysergic acid diethylamide (LSD). Although diverse in their actions it is clear that the ability to effectively label amides with isotopes is paramount in the development and testing of both lead compounds and known drugs so that their effects and metabolic fate *in situ* may be fully understood. Specific examples of preparing amides containing isotopic labels may be found in the literature, for example, the labelling of melatonin via 1.5 equivalents of [*C*-$^{2}H_3$]acetyl chloride **1** has been reported with a quoted yield of 80% after 1.5 h but the need for excess labelled reagent is clearly a disadvantage.[9] Additionally Courtyn *et al.*[10] published a synthesis for [^{11}C]acetylhomotaurine using [^{11}C]acetyl chloride with the focus primarily on biological studies using the radiolabelled compound. These are rare examples though, since, as highlighted by Shevchenko *et al.*[11] preparing labelled amides using reagents such as acetic anhydride or acetyl chloride **2** is difficult due to their high cost and volatility. This problematic issue may be addressed through the use of micro-reactors which are, for the most part, closed units thus providing the ability to accurately control reagent consumption.

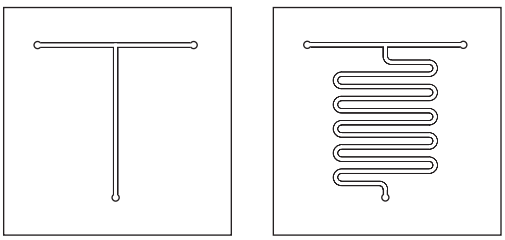
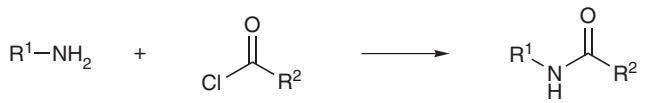


Figure 1 Typical 'T' (left), serpentine (right) micro-reactor designs.

Results and discussion

As Figure 2 illustrates, the micro-reactions were (see Table 1) carried out in a first-stage chip (T1 = 201 μm × 75 μm × 2.0 cm) to react the amine and organic base. The outlet of this chip was connected via

Table 1 General reaction for the acylation of amines



Amine (R^1)	Acylating agent (R^2)	Amide product
Ph **3**	CH_3 **2**	*N*-Phenyl-acetamide **4**
4′-MeOPh **5**	CH_3 **2**	*N*-(4-Methoxy-phenyl)-acetamide **9**
3′-AcOPh **6**	CH_3 **2**	*N*-(3-Acetyl-phenyl)-acetamide **10**
$PhCH_2$ **7**	CH_3 **2**	*N*-Benzyl-acetamide **11**
$PhCH_2CH_2$ **8**	CH_3 **2**	*N*-Phenethyl-acetamide **12**
Ph **3**	Ph **13**	*N*-Phenyl-benzamide **14**
4′-MeOPh **5**	Ph **13**	*N*-(4-Methoxy-phenyl)-benzamide **15**
3′-AcOPh **6**	Ph **13**	*N*-(3-Acetyl-phenyl)-benzamide **16**
$PhCH_2$ **7**	Ph **13**	*N*-Benzyl-benzamide **17**
$PhCH_2CH_2$ **8**	Ph **13**	*N*-Phenethyl-benzamide **18**
Ph **3**	CD_3 **1**	*N*-Phenyl-[*C*-$^{2}H_3$]acetamide **19**
4′-MeOPh **5**	CD_3 **1**	*N*-(4-Methoxy-phenyl)- [*C*-$^{2}H_3$]acetamide **20**
3′-AcOPh **6**	CD_3 **1**	*N*-(3-Acetyl-phenyl)- [*C*-$^{2}H_3$]acetamide **21**
$PhCH_2$ **7**	CD_3 **1**	*N*-Benzyl-[*C*-$^{2}H_3$]acetamide **22**
$PhCH_2CH_2$ **8**	CD_3 **1**	*N*-Phenethyl-[*C*-$^{2}H_3$]acetamide **23**

DOI: 10.1002.jlcr

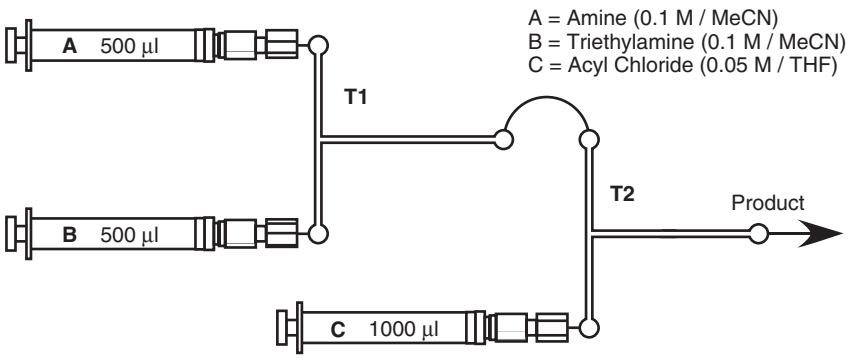


Figure 2 Schematic of the micro-reactor set-up used in initial experimentation. Note the differing syringe sizes and reagent concentrations; the overall effect gives rise to stoichiometric mixing quantities when all syringes are driven at an equal flow rate.

a 2.5 cm length of PEEK™ tubing (360 µm o.d., 150 µm i.d.) to one inlet of a second-stage reactor (T2 = 158 µm × 54 µm × 1.5 cm) which allowed the intermediate to then react with the acyl chloride. Employing a modular-type set-up, comprising of two micro-reactors, allowed for the rapid optimization of reactions by simple interchange of the second-stage reactor for one of longer channel length (S1 = 170 µm × 60 µm × 23.5 cm) when required. PEEK™ tubing of length 5 cm was used to connect the micro-reactor to the syringe pump via PEEK™ HPLC connectors. Glass luer lock syringes were automatically operated by a syringe pump capable of simultaneously delivering a maximum of three solutions. Reactions were operated at a concentration of 0.1 M (amine and base, respectively) in acetonitrile and 0.05 M (acyl chloride) in THF, which provided a final concentration of 0.025 M on collection and correlates to a 1:1:1 ratio of reagents. A mixed solvent system was selected as the polarity of acetonitrile was necessary to avoid precipitation of ammonium salts whilst THF was found to be a stable solvent for the daily storage of acyl chloride solutions.

Working within the given concentration range was found to effectively avoid precipitation of solids, a problem which can often hamper any attempts at optimization and reproducibility. To monitor a particular reaction 10 µl of product solution was collected from the reactor outlet into a small vial containing 50 µl of quench agent (1% water in acetonitrile) and analysed immediately by HPLC; this enabled us to confirm that the reaction occurred on chip rather than in the vial. Percentage conversions were calculated based on the amount of product (amide) present with respect to the amine remaining. Retention times and relative responses of all components were determined using synthetic standards.

Initially the reaction between phenylamine **3** and unlabelled acetyl chloride **2** to afford *N*-phenyl-acetamide **4** was investigated using a set-up consisting of two micro-reactors, T1 and T2, joined in a series arrangement as shown in Figure 2. Since it is necessary to carry out the reaction in the presence of a base, to sequester any hydrogen chloride formed, we decided to investigate the reaction in conjunction with triethylamine due to its ready availability, low toxicity and known reactivity. The syringe driver was set to the following flow rates; 20, 10, 5, 2 and 1 µl min^{-1} and each reaction repeated 10 times (at each flow rate) to determine reproducibility. In all cases the relative standard deviation (RSD) was observed to be less than 5% and the relationship between flow rate and conversion can be clearly seen in Figure 3. At the lowest investigated flow rate of 1 µl min^{-1}, the complete conversion of phenylamine **3** into *N*-phenyl-acetamide **4** was observed. The acetylation of four other amines namely 4-methoxy-phenylamine **5**, 1-(3-amino-phenyl)-ethanone **6**, benzylamine **7** and phenylethylamine **8**, to afford *N*-(4-methoxy-phenyl)-acetamide **9**, *N*-(3-acetyl-phenyl)-acetamide **10**, *N*-benzyl-acetamide **11** and *N*-phenethyl-acetamide **12**, respectively, was then investigated and the results summarized in Table 2.

In all cases near quantitative conversions were achieved, however, for the acetylation of phenylamine **3** and 1-(3-amino-phenyl)-ethanone **6** an increased residence time (achieved by employing a slower flow rate) was necessary to allow the reactions enough time to reach completion. Resonance stabilization of the former and electron withdrawing effects of the latter's acetyl group affect the reactivity of the molecules. In an effort to improve the productivity of the system for these particular amines the second stage micro-reactor, T2 which had a channel length of 1.5 cm, was substituted for one of longer length (23.5 cm). Based on the flow rates and the channel volume of T2 (0.11 µl) the relationship between conversions and residence time was plotted (Figure 4). In this way an appropriate serpentine micro-reactor, S1, with a channel length of 23.5 cm, was chosen so as to provide an adequate

DOI: 10.1002.jlcr

residence time ($\geqslant$2.6 s) when the reagents were infused at a faster flow rate of 20 µl min^{-1}, thus providing a higher overall throughput. In this way comparable results to those seen in the first system when running at lower flow rates were achieved and conversions of 100.0 and 97.7% were obtained for phenylamine **3** and 1-(3-amino-phenyl)-ethanone **6**, respectively.

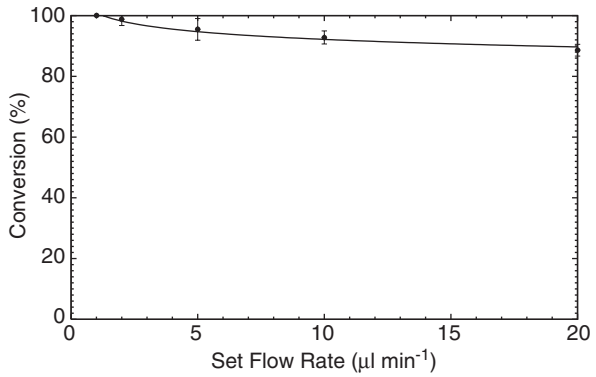


Figure 3 Graph illustrating the effect of varying flow rate on the conversion of aniline **3** to *N*-phenyl-acetamide **4**.

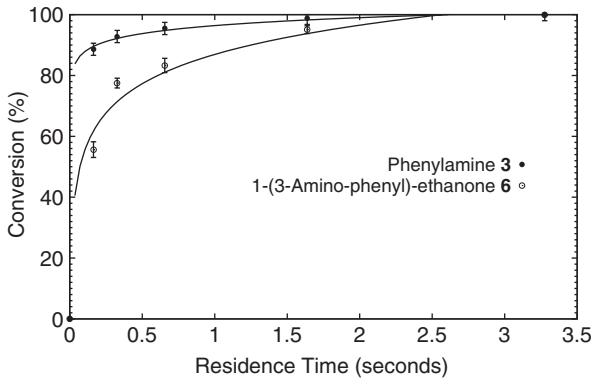


Figure 4 The relationship between residence time and conversion for phenylamine **3** and 1-(3-amino-phenyl)-ethanone **6** in T2.

Using the optimized micro-reactor configuration and conditions the reaction between the same five amines, **3** and **5**–**8**, with benzoyl chloride **13**, to form *N*-phenyl-benzamide **14**, *N*-(4-methoxy-phenyl)-benzamide **15**, *N*-(3-acetyl-phenyl)-benzamide **16**, *N*-benzyl-benzamide **17** and *N*-phenethyl-benzamide **18** was investigated. Although benzoyl chloride **13** is less reactive than the acetyl analogue **2**, similar results were obtained (Table 3). The poor conversions observed in the case of *N*-(3-acetly-phenyl)-benzamide **16** can be attributed to a combination of the decreased reactivity of benzoyl chloride **13** and delocalization through the ring between the amino and acetyl group which stabilizes the molecule. The net result significantly slows the reaction and consequently a reduction in conversion is observed.

In order to demonstrate the concept of employing such a system for deuterium labelling, acetyl chloride **2** was substituted for its deuterated equivalent [*C*-^{2}H$_3$]acetyl chloride **1**. To confirm the synthesis of labelled amides *N*-phenyl-[*C*-^{2}H$_3$]acetamide **19**, *N*-(4-methoxy-phenyl)-[*C*-^{2}H$_3$]acetamide **20**, *N*-(3-acetyl-phenyl)-[*C*-^{2}H$_3$]acetamide **21**, *N*-benzyl-[*C*-^{2}H$_3$]acetamide **22** and *N*-phenethyl-[*C*-^{2}H$_3$]acetamide **23**, the micro-reactor system was typically run for 2.5–3.0 h to prepare sufficient compound (20 mg) for analysis, following work-up and recrystallization to remove unwanted reaction components. As expected, the system produced analogous results with the labelled reagent and the reaction efficiency was comparable to the pre-optimized conditions (Table 2). As can be seen from

Table 2 Acetylation of amines in a double T micro-reactor (n = 10)

Amide product	Set flow rate (µl min^{-1})	Conversion (%)	RSD (%)
N-Phenyl-acetamide **4**	1	100.0	0.00
N-(4-Methoxy-phenyl)-acetamide **9**	20	99.3	0.83
N-(3-Acetyl-phenyl)-acetamide **10**	1	99.9	0.83
N-Benzyl-acetamide **11**	20	95.3	1.44
N-Phenethyl-acetamide **12**	20	96.4	1.45

Table 3 Conversions obtained at optimal conditions for the benzoylation of amines (n = 10)

Amide product	Reactor	Conversion (%)	RSD (%)
N-Phenyl-benzamide **14**	T1, S1	90.5	4.91
N-(4-Methoxy-phenyl)-benzamide **15**	T1, T2	94.0	5.50
N-(3-Acetyl-phenyl)-benzamide **16**	T1, S1	65.5	4.70
N-Benzyl-benzamide **17**	T1, T2	97.4	0.94
N-Phenethyl-benzamide **18**	T1, T2	91.9	2.46

DOI: 10.1002.jlcr

Table 4 Summary of the results obtained for the isotopic labelling of primary amines within micro-reactors

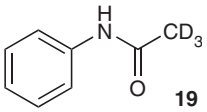
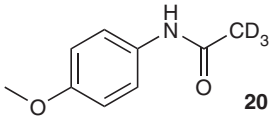
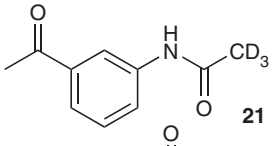
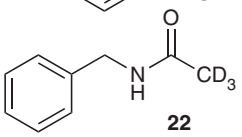
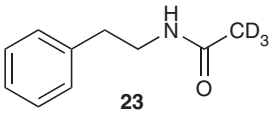
Entry	Amide product	Reactor	Residence time (s)	Throughput (mg h^{-1})	Conversion (%)	Yield (%)
1	 **19**	T1, S1	5.54	7.63	95.0	92
2	 **20**	T1, T2	2.62	9.89	98.0	98
3	 **21**	T1, S1	5.54	9.95	95.8	92
4	 **22**	T1, T2	2.62	8.95	98.2	98
5	 **23**	T1, T2	2.62	9.58	96.4	96

Table 4 the deuterium label was incorporated into the final product and in all cases excellent isolated yields were obtained after recrystallization. It should be emphasized that the ability to obtain similar yields when substituting for the labelled reagent is a significant result, as this is often difficult to obtain in batch.

Experimental

Materials

All solvents used in micro-reactions were puriss grade ($\geqslant$99.5%) over molecular sieves ($H_2O \leqslant 0.005\%$) purchased from Fluka. All other reagents were purchased from Sigma-Aldrich. 4-Methoxy-phenylamine **5** and 1-(3-amino-phenyl)-ethanone **6** were recrystallized (DCM/hexane) before use. All other reagents were used as received without further purification.

Instruments

Nuclear magnetic resonance (NMR) spectra were recorded on a Jeol (GX400) spectrometer operating at room temperature in solutions of deuteriochloroform ($CDCl_3$) doped with trimethylsilane (TMS, 0.03%) as an internal standard. Chemical shifts are quoted in parts per million (ppm) and coupling constants in Hertz (Hz). The following abbreviations are used when reporting NMR data; s = singlet, d = doublet, t = triplet, *br* s = broad singlet, m = multiplet, sep = septet and C_0 = quaternary carbon. Infra-red spectra were recorded on a Perkin Elmer Paragon 1000 FT-IR spectrometer in the range 4000–600 cm^{-1} and peaks (ν_{max}) reported in wavenumbers (cm^{-1}). Mass spectra were recorded on a Varian GC (CP-3800) coupled to a Varian MS (Saturn 2000) with a CP-Sil 8 (30 m) column manufactured by Phenomonex (Zebron ZB-5). Ultra-high-purity helium (99.999% Energas) was used as the carrier gas. Analysis was carried out using the following method; injector temperate 200°C, helium flow rate 1 ml min^{-1}, oven temperature 50°C for 4 min, and then increased to 250°C at a rate of 30°C min^{-1} with a 3.0 min filament delay. The HPLC set-up used for experimental analysis was a Shimadzu (LC-10AD) pump coupled to an in-line Shimadzu (DGU-14A) degasser and a Prodigy ODS-2 (5 μm, 4.6 × 250 mm) column manufactured by Phenomonex. Analyte detection was carried out at $\lambda = 254$ nm using a Polymer Laboratories (LC 1210) UV/Vis detector. All analyses were carried out at room temperature in isocratic mode using acetonitrile containing 0.1% trifluoroacetic acid (TFA) and water in a 9:1 ratio as the mobile phase. The borosilicate glass used to fabricate micro-reactors was crown white glass (TELIC, USA) pre-coated with layers of chrome and photoresist. PEEK™ tubing was purchased from Upchurch Scientific, glass luer lock syringes (Hamilton), HPLC fittings (1/16 in) and female–female luers interfacing the

DOI: 10.1002.jlcr

tubing and syringes were all purchased from Supelco. The syringe driver (MD-1001) was manufactured by Bioanalytical Systems Inc. and could operate at set flow rates ranging between 0.1 and 100 $\mu l\,min^{-1}$.

General batch preparation methodology

To a stirred solution of pre-weighed amine dissolved in DCM (150 ml) was added an equimolar amount of acylating agent (acetyl chloride **2** or benzoyl chloride **13**). After 2 h continued stirring at room temperature any solids were removed by filtration and the reaction mixture was washed with *sat.* NH_4Cl solution (3×150 ml). The organic layer was dried over anhydrous $MgSO_4$, filtered and concentrated *in vacuo*. Subsequent recrystallization (DCM or ether/hexane) yielded the corresponding amide, confirmed by comparison to data in the literature.

N-Phenyl-acetamide (**4**).[12] (0.27 g, 27%) as fine white needles; δ_H (400 MHz, $CDCl_3$/TMS) 2.18 (3 H, s, CH_3), 7.10 (1 H, m, Ar), 7.31 (2 H, m, $2 \times$ Ar), 7.36 (1 H, *br* s, NH) and 7.50 (2 H, m, $2 \times$ Ar); δ_C (100 MHz, $CDCl_3$/TMS) 24.6 (CH_3), 119.9 (CH), 124.3 ($2 \times$ CH), 129.0 ($2 \times$ CH), 137.9 (C_0) and 168.3 (CO); *m/z* (EI) 136 ($M^+ + 1$, 30%), 135 (84), 93 (100) and 65 (6).

N-(4-Methoxy-phenyl)-acetamide (**9**).[12] (0.21 g, 21%) as white crystals; δ_H (400 MHz, $CDCl_3$/TMS) 2.15 (3 H, s, CH_3), 3.78 (3 H, s, CH_3), 6.85 (2 H, d, *J* 9.0, $2 \times$ Ar), 7.19 (1 H, *br* s, NH) and 7.38 (2 H, d, *J* 9.0, $2 \times$ Ar); δ_C (100 MHz, $CDCl_3$/TMS) 24.3 (CH_3), 55.5 (CH_3), 114.1 ($2 \times$ CH), 121.9 ($2 \times$ CH), 130.9 (C_0), 156.4 (C_0) and 168.1 (CO); *m/z* (EI) 166 ($M^+ + 1$, 100%) and 108 (20).

N-(3-Acetyl-phenyl)-acetamide (**10**).[13] (0.12 g, 12%) as white crystals; δ_H (400 MHz, $CDCl_3$/TMS) 2.22 (3 H, s, CH_3), 2.61 (3 H, s, CH_3), 7.43 (2 H, t and *br* s, *J* 7.8, Ar and NH), 7.70 (1 H, m, Ar), 7.90 (1 H, m, Ar) and 8.00 (1 H, m, Ar); δ_C (100 MHz, $CDCl_3$/TMS) 24.6 (CH_3), 26.7 (CH_3), 119.2 (CH), 124.2 (CH), 124.5 (CH), 129.4 (CH), 137.7 (C_0), 138.4 (C_0), 168.5 (CO) and 197.9 (CO); *m/z* (EI) 178 ($M^+ + 1$, 48%), 177 (100), 135 (55), 120 (70), 87 (15) and 63 (15).

N-Benzyl-acetamide (**11**).[14] (0.13 g, 13%) as a white crystalline solid; δ_H (400 MHz, $CDCl_3$/TMS) 2.02 (3 H, s, CH_3), 4.43 (2 H, d, *J* 5.6, CH_2), 5.85 (1 H, *br* s, NH) and 7.28 (3 H, m, $3 \times$ Ar) and 7.34 (2 H, m, $2 \times$ Ar); δ_C (100 MHz, $CDCl_3$/TMS) 23.3 (CH_3), 43.8 (CH_2), 127.5 (CH), 127.9 ($2 \times$ CH), 128.7 ($2 \times$ CH), 138.2 (C_0) and 169.8 (CO); *m/z* (EI) 150 ($M^+ + 1$, 100%), 106 (90) 91 (20), 77 (6) and 51 (5).

N-Phenethyl-acetamide (**12**).[15] (0.51 g, 51%) as a white crystalline solid; δ_H (400 MHz, $CDCl_3$/TMS) 1.93 (3 H, s, CH_3), 2.81 (2 H, t, *J* 7.0, CH_2), 3.52 (2 H, m, CH_2), 5.50 (1 H, *br* s, NH), 7.19 (2 H, m, $2 \times$ Ar), 7.24 (H, m, Ar) and 7.32 (2 H, m, $2 \times$ Ar); δ_C (100 MHz, $CDCl_3$/TMS) 23.3 (CH_3), 35.6 (CH_2), 40.6 (CH_2), 126.5 (CH), 128.6 ($2 \times$ CH), 128.7 ($2 \times$ CH), 138.8 (C_0) and 170.0 (CO); *m/z* (EI) 164 ($M^+ + 1$, 100%), 104 (37) and 65 (6).

N-Phenyl-benzamide (**15**).[16] (0.37 g, 37%) as fine white crystals; δ_H (400 MHz, $CDCl_3$/TMS) 7.16 (1 H, m, Ar), 7.38 (2 H, m, $2 \times$ Ar), 7.50 (2 H, m, $2 \times$ Ar), 7.56 (1 H, m, Ar), 7.65 (2 H, m, $2 \times$ Ar), 7.79 (1 H, *br* s, NH) and 7.88 (2 H, m, $2 \times$ Ar); δ_C (100 MHz, $CDCl_3$/TMS) 120.2 ($2 \times$ CH), 124.6 (CH), 127.0 ($2 \times$ CH), 128.9 ($2 \times$ CH), 129.1 ($2 \times$ CH), 131.9 (CH), 135.0 (C_0), 137.9 (C_0) and 165.7 (CO); *m/z* (EI) 198 ($M^+ + 1$, 46%), 197 (100), 105 (68) and 51 (4).

N-(4-Methoxy-phenyl)-benzamide (**16**).[17] (0.2235 g, 22%) as white crystals; δ_H (400 MHz, $CDCl_3$/TMS) 3.81 (3 H, s, CH_3), 6.91 (2 H, d, *J* 9.0, $2 \times$ Ar), 7.47 (2 H, m, $2 \times$ Ar), 7.54 (3 H, m, $3 \times$ Ar), 7.76 (1 H, *br* s, NH) and 7.86 (2 H, d, *J* 7.0, $2 \times$ Ar); δ_C (100 MHz, $CDCl_3$/TMS) 55.5 ($2 \times$ CH), 114.2 ($2 \times$ CH), 122.1 ($2 \times$ CH), 127.0 ($2 \times$ CH), 128.7 ($2 \times$ CH), 131.0 (C_0), 131.7 (C_0), 135.0 (CH), 156.6 (C_0) and 165.6 (CO); *m/z* (EI) 228 ($M^+ + 1$, 15%), 227 (93), 105 (100), 77 (57) and 51 (19).

N-(3-Acetyl-phenyl)-benzamide (**14**).[18] (0.2770 g, 28%) as faint pink crystals; δ_H (400 MHz, $CDCl_3$/TMS) 2.61 (3 H, s, CH_3), 7.49 (3 H, m, $3 \times$ Ar), 7.57 (1 H, m, Ar), 7.73 (1 H, ddd, *J* 7.9, *J* 1.7 and *J* 1.1, Ar), 7.91 (2 H, m, $2 \times$ Ar), 8.07 (1 H, ddd, *J* 8.2, *J* 2.2 and *J* 1.1, Ar), 8.17 (1 H, t, *J* 2.0, Ar) and 8.19 (1 H, *br* s, NH); δ_C (100 MHz, $CDCl_3$/TMS) 26.7 (CH_3), 119.6 (CH), 124.4 (CH), 124.8 (CH), 127.1 ($2 \times$ CH), 128.8 ($2 \times$ CH), 129.4 (CH), 132.1 (CH), 134.6 (C_0), 137.8 (C_0), 138.5 (C_0), 165.9 (CO) and 197.9 (CO); *m/z* (EI) 240 ($M^+ + 1$, 10%), 239 (65), 105 (100), 77 (57) and 51 (15).

N-Benzyl-benzamide (**17**).[19] (0.1069 g, 10.7%) as fine white crystals; δ_H (400 MHz, $CDCl_3$/TMS) 4.64 (2 H, d, *J* 6.0, CH_2), 6.46 (1 H, *br* s, NH), 7.31 (1 H, m, $1 \times$ Ar), 7.35 (4 H, d, *J* 4.5, $4 \times$ Ar), 7.42 (2 H, m, $2 \times$ Ar), 7.50 (1 H, m, Ar) and 7.79 (2 H, m, $2 \times$ Ar); δ_C (100 MHz, $CDCl_3$/TMS) 44.1 (CH_2), 126.9 ($2 \times$ CH), 127.6 (CH), 127.9 ($2 \times$ CH), 128.6 ($2 \times$ CH), 128.8 ($2 \times$ CH), 131.5 (CH), 134.4 (C_0), 138.2 (C_0) and 167.3 (CO); *m/z* (EI) 212 ($M^+ + 1$, 26%), 211 (100), 105 (96), 77 (80) and 51 (28).

N-Phenethyl-benzamide (**18**).[20] (0.27 g, 27%) as fine white crystals; δ_H (400 MHz, $CDCl_3$/TMS) 2.94 (2 H, t, *J* 7.0, CH_2), 3.73 (2 H, m, CH_2), 6.15 (H, *br* s, NH), 7.25 (3 H, m, $3 \times$ Ar), 7.33 (2 H, m, $2 \times$ Ar), 7.40 (2 H, m, $2 \times$ Ar), 7.48 (H, m, Ar) and 7.68 (2 H, m, $2 \times$ Ar); δ_C (100 MHz, $CDCl_3$/TMS) 35.7 (CH_2), 41.1 (CH_2), 126.6 (CH), 126.8 ($2 \times$ CH), 128.5 ($2 \times$ CH), 128.7 ($2 \times$ CH), 128.8 ($2 \times$ CH), 131.4 (CH), 134.7 (C_0), 138.9 (C_0) and 167.4 (CO); *m/z* (EI) 226 ($M^+ + 1$, 10%), 134 (20), 105 (100), 77 (46) and 51 (18).

DOI: 10.1002.jlcr

Micro-reaction methodology

For preparing labelled compounds within the micro-reactor set-up it was necessary to run the system for a sufficient amount of time (2.5–3.0 h) in order to prepare 20 mg of product for analysis. Post-collection the reaction products were concentrated *in vacuo*, dissolved in DCM (25 ml) and washed with saturated NH_4Cl solution (3×30 ml). Drying over $MgSO_4$, filtration, concentration *in vacuo* and recrystallization (DCM/hexane) afforded the final product for characterization.

N-Phenyl-[C-$^2H_3$]acetamide (**19**). (18.30 mg, 92%) as fine white needles. ν_{max}/cm^{-1} 3300, 3196, 3137, 3080, 1667, 1599, 1434, 1320, 759 and 694; δ_H (400 MHz, $CDCl_3$/TMS) 7.10 (1H, m, Ar), 7.25 (1H, *br* s, NH), 7.32 (2H, m, $2 \times$ Ar) and 7.50 (2H, m, $2 \times$ Ar); δ_C (100 MHz, $CDCl_3$/TMS) 24.0 (CD_3, sep, *J* 19.5), 119.9 (CH), 124.4 ($2 \times$ CH), 129.1 ($2 \times$ CH), 137.9 (C_0) and 168.4 (CO); *m/z* (EI) 139 ($M^+ + 1$, 30%), 138 (63), 94 (100) and 65 (7).

N-(4-Methoxy-phenyl)-[C-$^2H_3$]acetamide (**20**). (19.58 mg, 98%) as white crystals. ν_{max}/cm^{-1} 3266, 2835, 1646, 1605, 1512, 1334, 1246, 1027 and 840; δ_H (400 MHz, $CDCl_3$/TMS) 3.78 (3H, s, CH_3), 6.83 (2H, d, *J* 9.0 $2 \times$ Ar), 7.39 (2H, d, *J* 9.0, $2 \times$ Ar) and 7.65 (H, *br* s, NH); δ_C (100 MHz, $CDCl_3$/TMS) 23.5 (CD_3, sep, *J* 19.5), 55.5 (CH_3), 114.1 ($2 \times$ CH), 122.0 ($2 \times$ CH), 131.0 (C_0), 156.4 (C_0) and 168.6 (CO); *m/z* (EI) 169 ($M^+ + 1$, 100%), 124 (33) and 108 (15).

N-(3-Acetyl-phenyl)-[C-$^2H_3$]acetamide (**21**). (18.42 mg, 92%) as white crystals. ν_{max}/cm^{-1} 3351, 2923, 1672, 1593, 1547, 1432, 1273, 792 and 687; δ_H (400 MHz, $CDCl_3$/TMS) 2.60 (3H, s, CH_3), 7.42 (1H, t, *J* 8.0, Ar), 7.68 (1H, m, Ar), 7.96 (1H, m, Ar), 7.99 (1H, *br* s, NH) and 8.02 (1H, m, Ar); δ_C (100 MHz, $CDCl_3$/TMS) 23.8 (CD_3, sep, *J* 19.3), 26.7 (CH_3), 119.2 (CH), 124.1 (CH), 124.6 (CH), 124.6 (CH), 129.3 (CH), 137.6 (C_0), 138.6 (C_0), 168.9 (CO) and 198.1 (CO); *m/z* (EI) 181 ($M^+ + 1$, 75%), 180 (100), 136 (30), 121 (57) and 64 (5).

N-Benzyl-[C-$^2H_3$]acetamide (**22**). (19.6 mg, 98%) as a white crystalline solid. ν_{max}/cm^{-1} 3293, 3087, 2928, 1627, 1552, 1454, 1320, 746 and 694; δ_H (400 MHz, $CDCl_3$/TMS) 4.44 (2H, d, *J* 5.6, CH_2), 5.74 (1H, *br* s, NH), 7.28 (3H, m, $3 \times$ Ar) and 7.34 (2H, m, $2 \times$ Ar); δ_C (100 MHz, $CDCl_3$/TMS) 22.6 (CD_3, sep, *J* 19.5), 43.8 (CH_2), 127.6 (CH), 127.9 ($2 \times$ CH), 128.7 ($2 \times$ CH), 138.2 (C_0) and 169.9 (CO); *m/z* (EI) 153 ($M^+ + 1$, 100%) and 107 (45).

N-Phenethyl-[C-$^2H_3$]acetamide (**23**). (19.1 mg, 96%) as white needles. ν_{max}/cm^{-1} 3291, 3086, 2930, 1646, 1558, 1451, 1314, 748 and 700; δ_H (400 MHz, $CDCl_3$/TMS) 2.82 (2H, t, *J* 7.0, CH_2), 3.52 (2H, m, CH_2), 5.50 (H, *br* s, NH), 7.20 (2H, m, $2 \times$ Ar), 7.24 (H, m, Ar) and 7.32 (2H, m, $2 \times$ Ar); δ_C (100 MHz, $CDCl_3$/TMS) 22.6 (CD_3, sep, *J* 19.4), 35.6 (CH_2), 40.6 (CH_2), 126.5 (CH), 128.7 ($2 \times$ CH), 128.8 ($2 \times$ CH), 138.9 (C_0) and 170.1 (CO); *m/z* (EI) 167 ($M^+ + 1$, 100%) and 103 (11).

Conclusion

We have effectively demonstrated the inherent benefits of combining a micro-reactor system with traditional synthetic isotope labelling and a number of deuterium-labelled amides were produced using the developed system at a minimal cost. In addition, close to quantitative isotope label incorporation into the desired products was obtained under stoichiometeric conditions. Although the reaction mixtures required purification, the use of a phase transfer micro-reactor[21] or coupling to a preparative HPLC system would incorporate this step into the on-line synthesis. Importantly, however, the developed system produces a continuous product stream which makes the rapid synthesis of isotopically labelled amides (reaction time ~3–6 s) a reality. For the system used in this article the throughput is approximately 10 mg h^{-1}. It is clear that in the realm of isotope chemistry there are significant cost advantages to pre-optimizing a reaction in such a system first with unlabelled products. Using micro-reactors to contain hazardous components of a reaction and control conventional chemistry more efficiently is another important advantage which will surely see the continued growth of such devices in this field in the future.

Acknowledgements

The authors would like to thank Charlotte Wiles for helpful discussion, Steve Clark for micro-reactor fabrication and The University of Hull for financial support (J.H.).

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