The use of electroosmotic flow as a pumping mechanism for semi-preparative scale continuous flow synthesis

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By employing a series of reactions we demonstrate the use of electroosmotic flow as a continuous pumping mechanism suitable for semi-preparative scale synthesis, affording an array of small organic compounds, of analytical purity, with yields ranging from 0.57–1.71 g h^{-1}.

One of the slowest steps associated with lead compound generation is the efficient synthesis and purification of small organic compounds, in particular those used to introduce diversity into combinatorial libraries. At present, the majority of synthetic transformations performed in research laboratories employ techniques, and glassware, that has remained largely unchanged for decades. Furthermore, the use of long standing protocols can mean that such reactions are performed using un-optimised reaction conditions, the outcome of which is largely operator dependent. Consequently, when target compounds are identified and prepared for transfer to production, the synthetic route frequently requires re-optimization, not only to address changes in scale, but also to enable the process to be operated under a more efficient continuous flow regime.

Owing to the pharmaceutical importance of α,β-unsaturated compounds, we were interested in developing a simple synthetic technique that would enable the rapid production of gram quantities of analytically pure material, using continuous flow methodology. Whilst a range of green approaches have been investigated for the synthesis of such analogues, the use of water[1] or ionic liquids[2] as reaction media demands that a formal work-up be performed in order to isolate, and where possible recycle, the catalyst. To circumvent this problem, numerous authors have reported the use of solid-supported bases, which can simply be filtered from the reaction mixture.[3] However, difficulties with the technique arise due to mechanical degradation of the support upon prolonged stirring, or shaking, of the reaction mixture in a batch mode.

In the late 1980s, Venturello and co-workers[4] reported the Knoevenagel condensation under continuous flow, using a vertical, double jacketed glass column packed with aminopropyl functionalised silica gel. Reactions were performed by simply placing a solution of aldehyde and activated methylene at the top of the column, where it passed through the catalyst under gravity (as in a liquid chromatographic process). Collection of the reaction mixture at the outlet, followed by evaporation of the solvent system, afforded the desired product and any unreacted starting materials. Although this technique illustrated advantages associated with continuous flow syntheses, namely the ease of product isolation and catalyst recycle, the technique employed large volumes of solvent (30 ml $mmol^{-1}$) and provided no control over flow rate. This inability to readily alter a reagent's residence time led to incomplete product conversions when less reactive analogues were employed, resulting in the need for additional off-line purification. To address this shortfall, continuous flow techniques have evolved to employ pressure-driven flow (in the form of HPLC or displacement pumps), enabling the rate at which reagents pass through the packed beds to be controlled.[5] Using this approach, an array of synthetic transformations have been demonstrated including hydrogenations,[6] oxidations,[7] reductions,[8] Diels–Alder reactions,[9] Suzuki couplings[10] and Michael additions,[11] culminating in the ability to perform automated multi-step syntheses, as illustrated by Ley and co-workers[12] for the synthesis of (±)-oxomaritidine. Whilst the aforementioned examples serve to illustrate the versatility of continuous flow systems, in order for the technique to become more widely adopted problems such as irreproducible flow (especially at low flow rates), back-pressure generation and cumbersome operating systems, need to be addressed.

Having recently demonstrated the ability to synthesise milligram quantities of analytically pure compounds in a series of miniaturised electroosmotic flow (EOF) based reactors,[13] we were interested in exploring methodologies capable of increasing the throughput of the system. One option was to scale-out the technique, *i.e.*, increase the number of optimised micro-reactor units employed, while an alternative approach was to simply increase the size of the catalyst bed.

To date, EOF has largely found use as a pumping mechanism for the manipulation of nl to μl quantities of material within micron-sized capillaries and channel networks.[14] However, as the volumetric flow rate is largely dependent upon the cross-sectional area of a channel, increasing the size of the channel enables the flow rate to be readily increased. As such reactions are diffusion limited, increasing the size of a reaction channel can be detrimental to reaction efficiency as it leads to inefficient mixing. This is, however, not the case when employing packed catalyst beds, as the diffusion distance between the liquid phase and the solid-supported catalyst, or reagent, remains the same irrespective of the overall bed size. Unlike simple pressure-driven systems where packed-bed size is limited by a reactor's tolerance to pressure, electroosmotic systems generate minimal back-pressure and therefore have the potential to be scaled-up with ease. In addition, the absence of mechanical pump drivers greatly reduces the footprint of the reaction set-up, which simply consists of a power supply (16 cm (w) × 6 cm (d) × 27 cm (l)) and a flow reactor

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(6 cm (w) × 1 cm (d) × 6 cm (l)). Automation of the system also enables the reactors to be operated remotely from a safe working distance, reducing the amount of valuable fume cupboard space required. Using this approach, we investigated the ability to scale-up our previously optimised reaction set-up which employed a 0.5 mm (id) capillary to a 3.0 mm (id) capillary, the results of which are reported herein.

As Fig. 1 illustrates, the reaction set-up employed consists of a borosilicate glass capillary (3.0 mm (id), 3.0 cm (length)), fritted at both ends to retain the solid-supported reagent, attached to two borosilicate glass reagent reservoirs *via* rubber stoppers (No. 9, Suba Seal). To perform a reaction, the packed-bed is primed with anhydrous acetonitrile (MeCN) (to form a complete electrical circuit) and platinum electrodes (0.25 mm (od) × 2.5 cm (length)) are then placed in reservoirs A and B, respectively. A solution of starting material is then placed in reservoir A and an aliquot of solvent placed in reservoir B. The reaction mixture is subsequently passed through the packed-bed by application of a positive voltage to reservoir A (167 to 300 V cm^{-1}) and the reaction products collected in reservoir B (0 V cm^{-1}). Unless otherwise stated, reactions are carried out for 10 min, after which the contents of reservoir B are removed and analysed, off-line, by GC-MS. Once optimised, the reactors are operated continuously for a period of 1 h (2.5 h in some instances), the reaction products collected, concentrated *in vacuo* and the crude product dissolved in $CDCl_3$ prior to purity evaluation by NMR spectroscopy.

To assess the reactor's performance, we firstly investigated the incorporation of silica-supported piperazine (0.100 g, 1.70 mmol g^{-1}) into the packed-bed, demonstrating the Knoevenagel condensation of two activated methylenes with a series of aldehydes (Table 1). Employing an applied field of 200 V cm^{-1}, a pre-mixed solution of aldehyde and activated methylene (both 1.0 M in anhydrous MeCN) was placed in reservoir A and passed through the packed-bed at flow rates in the range from 48.3 to 62.1 μl min^{-1}, depending on the aldehyde employed. In all cases, optimised reaction conditions afforded the desired α,β-unsaturated product (*trans*-isomer only) in excellent product purity (>99.9%) and yield (>98.8%), demonstrating reactor stability over ⩾15 runs. To further demonstrate the feasibility of operating such reactors continuously, the syntheses of 3-(4-bromophenyl)-2-cyanoacrylic acid ethyl ester and 2-(4-brombenzylidene)malononitrile were investigated over an extended 2.5 h period, affording 2.30 g (99.4%) and 1.70 g (99.8%) of analytically pure material, respectively.

Based on the results presented in Table 1, it can be seen that the use of continuous flow reactors not only leads to enhanced product purity but also to a dramatic reduction in reaction time, affording near quantitative yields with residence times in the range of 0.15–0.19 min.† The generality of the technique was subsequently evaluated by incorporating polymer supported diazabicyclo[2.2.2]octane (1.45 mmol N g^{-1}), 3-(dimethylamino)-propyl (1.50 mmol N g^{-1}), 3-aminopropyl (1.70 mmol N g^{-1}) and 3-(1,3,4,6,7,8-hexahydro-2*H*-pyrimido[1.2.1]-pyrimidino)propyl (2.40 mmol N g^{-1}) functionalised silica gels into the flow reactor, whereby 3-(4-bromophenyl)-2-cyano-3-phenylacrylic acid ethyl ester was obtained in excellent yield and purity (>99.0%). Compared with the gravity-based system reported by Venturello and co-workers,[4] the approach described here is also advantageous as it represents a significant reduction in solvent usage, requiring only 1 ml $mmol^{-1}$ of product, *cf.* 30 ml $mmol^{-1}$. In addition, as the products synthesised are obtained in excellent purity, no subsequent off-line chromatographic purification is needed, further reducing the environmental burden associated with the technique.

In addition to the base-catalysed syntheses discussed, the study also investigated the feasibility of performing acid catalysed reactions in such continuous flow systems. Based on our previous experience of solid-supported acid catalysts, the synthesis of dimethyl acetals was selected as a model reaction. In brief, reactions were performed by mobilising a pre-mixed solution of aldehyde and trimethylorthoformate (1.0 M and 2.5 M, respectively, in MeCN) through a packed bed, containing Amberlyst-15 (0.075 g, 0.315 mmol), upon application of 300 and 0 V cm^{-1}, to reservoirs A and B respectively. Again, the reaction products were

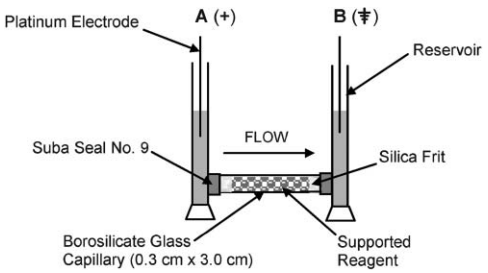
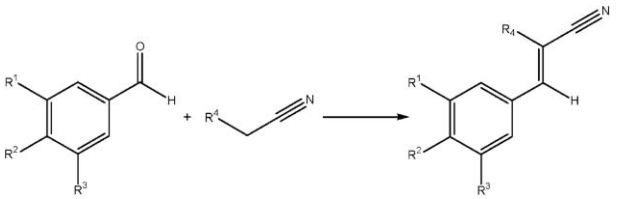


Fig. 1 Schematic illustrating the reaction set-up used for the continuous flow synthesis of small organic compounds by EOF.

Table 1 Summary of the results obtained for the synthesis of α,β-unsaturated compounds in an EOF-based continuous flow reactor (unless stated otherwise, all reactions were conducted for 1 h)



R^1	R^2	R^3	R^4	Flow rate^{-1} /μl min	Conv. (%)[a]	Yield /g
H	H	H	CO_2Et	62.0	99.9	0.75 (99.7)[b]
H	CO_2Me	H	CO_2Et	56.1	100.0	0.87 (99.8)
OMe	H	OMe	CO_2Et	50.1	99.9	0.78 (99.9)
H	OBn	H	CO_2Et	51.1	99.9	0.94 (99.7)
H	Br	H	CO_2Et	55.1	99.9	2.30[c] (99.4)
H	H	H	CN	62.1	99.9	0.57 (99.4)
H	CO_2Me	H	CN	60.4	99.9	0.76 (98.8)
OMe	H	OMe	CN	55.7	100.0	0.71 (99.2)
H	OBn	H	CN	48.4	99.9	0.75 (99.4)
H	Br	H	CN	48.3	100.0	1.70[c] (99.8)

[a] n = 15. [b] Number in parentheses represents % yield. [c] Reaction conducted for 2.5 h.

Table 2 Synthesis of dimethylacetals in a wide bore, EOF-based, continuous flow reactor (unless stated otherwise, all reactions were conducted for 1 h)

Starting material	Flow rate/ μl min^{-1}	Conv. (%)[a]	Yield/g
Benzaldehyde	139.0	100.0	1.26 (99.2)[b]
4-Cyanobenzaldehyde	124.0	99.8	1.31 (99.2)
4-Chlorobenzaldehyde	120.0	99.9	1.33 (99.5)
4-Benzyloxybenzaldehyde	111.0	99.9	1.71 (99.7)
3,5-Dimethoxybenzaldehyde	112.0	99.9	1.42 (99.4)
2-Naphthaldehyde	126.0	99.9	1.52 (99.8)
Methyl-4-formylbenzoate	121.0	99.9	1.52 (99.4)
5-Nitro-2-thiophenecarboxaldehyde	123.0	99.9	1.50 (99.8)
trans-Cinnamaldehyde	113.0	99.9	1.20 (99.4)
4-Bromobenzaldehyde	105.0	99.6	3.64 (99.7)[c]

[a] n = 15. [b] Number in parentheses represents % yield. [c] Reaction conducted for 2.5 h.

Table 3 Evaluation of the catalytic activity of an acid and a base catalyst employed within the EOF-based flow reactor

	Catalyst/ mmol	Product/ mmol	Turnover number
Silica-supported piperazine	0.17	42.00	247
Amberlyst-15	0.32	80.71	256

[a] Based on the data presented herein (catalysts remain active).

collected at 10 min intervals and analysed off-line by GC-MS. Once optimised, reactions were operated continuously for 1 h and the products isolated by evaporation of the reaction solvent: the purity of the 'crude' material was subsequently evaluated by NMR spectroscopy. Employing flow rates in the range of 111.0–139.0 μl min^{-1} resulted in optimal conversion of an array of aldehydes to their respective dimethylacetal (Table 2), obtaining greater product purity compared with analogous batch reactions. This observation is attributed to the unique reaction conditions attained within continuous flow reactors, which enable reaction products to be removed from the reactor prior to, in this case, competing acid-catalysed deprotection occurring. Again, extended operation was demonstrated for the synthesis of 1-bromo-4-dimethoxymethylbenzene, affording a space time yield of 1.46 g h^{-1}. Furthermore, system generality was demonstrated *via* the incorporation of additional solid-supported Lewis acid catalysts, including silica-supported sulfonic acid (1.50 mmol g^{-1}), polymer supported *para*-toluene sulfonic acid (2.00 to 3.50 mmol g^{-1}), ytterbium polystyryl sulfonate(III) (0.80 mmol g^{-1}), whereby excellent yields and purities were obtained in all cases.

Based on the data presented herein it can be concluded that the catalysts remain active over prolonged periods of time, enabling catalytic turnovers in excess of 247 times to be attained (Table 3).

The scope of the technique was subsequently extended to evaluate the feasibility of performing continuous flow, multi-step syntheses. As Fig. 2 illustrates, the model reaction selected involved an acid-catalysed acetal deprotection, followed by a base-catalysed condensation, of the *in situ* generated aldehyde with

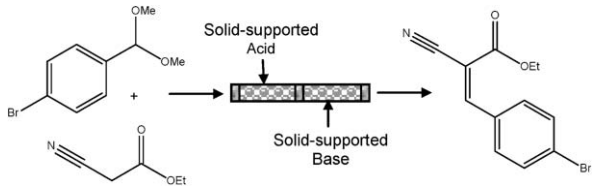


Fig. 2 Schematic illustrating the multi-step synthesis of 3-(4-bromophenyl)-2-cyanoacrylic acid ethyl ester in an EOF based, continuous flow reactor.

ethylcyanoacetate, to afford 3-(4-bromophenyl)-2-cyanoacrylic acid ethyl ester. To perform a reaction, a pre-mixed solution of 1-bromo-4-dimethoxymethylbenzene and ethylcyano acetate (1.00 M in MeCN) was placed in reservoir A and pumped through a packed-bed containing Amberlyst-15 (0.036 g, 0.151 mmol) and silica-supported piperazine (0.050 g, 0.085 mmol). By ensuring that each step of the reaction proceeds to completion, multiple reaction steps can be performed in series, without the need to purify the reaction intermediates. Consequently, operation of the reactor at an optimised flow rate of 54.9 μl min^{-1} afforded 3-(4-bromophenyl)-2-cyanoacrylic acid ethyl ester in excellent purity (100.0% by GC-MS) with a system throughput of 0.926 g h^{-1}.

From the examples presented, it can be seen that EOF is a versatile pumping technique that affords accurate, pulse-free reagent delivery, enabling reactions to be readily optimised. Furthermore, the ease with which the supported reagents are recycled provides reaction reproducibility and catalyst lifetimes unobtainable in traditional agitated reaction systems.

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Notes and references

† The total volume of the flow reactor was found to be 9.0 μl, therefore when operating at 50.0 μl min^{-1} a residence time of 0.18 min is obtained.

1 A. Pande, K. Ganesan, A. K. Jain, P. K. Gupta and R. M. Malhotra, *Org. Process Res. Dev.*, 2005, **9**, 133.
2 D. W. Morrison, D. C. Forbes and J. H. Davis, *Tetrahedron Lett.*, 2001, **42**, 6053.
3 (*a*) J. Simpson, D. L. Rathbone and D. C. Billington, *Tetrahedron Lett.*, 1999, **40**, 7031; (*b*) F. Texier-Boulett and A. Foucod, *Tetrahedron Lett.*, 1982, **23**, 4927.
4 (*a*) E. Angelletti, C. Canepa, G. Martinetti and P. Venturello, *Tetrahedron Lett.*, 1988, **29**, 2261; (*b*) E. Angelletti, C. Canepa, G. Martinetti and P. Venturello, *J. Chem. Soc., Perkin Trans. 1*, 1989, 105.
5 A. Kirschning, W. Solodenko and K. Mennecke, *Chem.–Eur. J.*, 2006, **12**, 5972.
6 (*a*) R. V. Jones, L. Godorhazy, N. Varga, D. Szalay, L. Urge and F. Darvas, *J. Comb. Chem.*, 2006, **8**(1), 110; (*b*) N. Yoswathananont, K. Nitta, Y. Nishiuchi and M. Sato, *Chem. Commun.*, 2005, 40.
7 C. Wiles, P. Watts and S. J. Haswell, *Tetrahedron Lett.*, 2006, **47**, 5261.
8 S. Itsuno, K. Ito, T. Maruyama, N. Kanda, A. Hirao and S. Nakahama, *Bull. Chem. Soc. Jpn.*, 1986, **59**, 3329.
9 K. Kamahori, K. Ito and S. Itsuno, *J. Org. Chem.*, 1996, **61**, 8321.
10 I. R. Baxendale, C. M. Griffiths-Jones, S. V. Ley and G. K. Tranmer, *Chem.–Eur. J.*, 2006, **72**, 4407.
11 F. Bonfils, I. Cazaux, P. Hodge and C. Caze, *Org. Biomol. Chem.*, 2006, **4**, 493.
12 I. R. Baxendale, J. Deeley, C. M. Griffiths-Jones, S. V. Ley, S. Saaby and G. K. Tranmer, *Chem. Commun.*, 2005, 2566.
13 (*a*) C. Wiles, P. Watts and S. J. Haswell, *Tetrahedron*, 2005, **61**, 5209; (*b*) C. Wiles, P. Watts and S. J. Haswell, *Tetrahedron*, 2004, **60**, 8421.
14 S. J. Haswell, *Analyst*, 1997, **112**, 1R.

