Supplementary Material for Chemical Communications

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General methods. All chemicals were reagent grade and used as supplied. Trimethylsilyl trifluoromethanesulfonate (TMSOTf) was purchased from Acros Chemicals. Dichloromethane (CH_2Cl_2) was refluxed and distilled over fresh calcium hydride, diethyl ether (Et_2O), and tetrahydrofuran (THF) were refluxed and distilled over sodium metal and benzophenone, and Toluene was refluxed and distilled over sodium metal prior to use. Analytical thin-layer chromatography was performed on E. Merck silica column 60 F254 places (0.25 mm). Compounds were visualized by dipping the plates in a cerium sulfate-ammonium molybdate solution followed by heating. High-Performance liquid chromatography was performed using a Waters Nova-pak© silica column (3.9 x 150 mm) with EtOAc/hexanes as the mobile phase, monitoring at 257.9 nm. 1H NMR spectra were obtained on a Bruker (400 MHz) or a Varian VXR-500 (500 MHz) and 13C NMR spectra were obtained on a Bruker (100 MHz) or a Varian VXR-500 (125 MHz).

HPLC reference standards for all compounds were prepared according to established literature. Batch reactions were performed with 1.2 equivalents glycosylating agent (donor), 1.0 equivalents nucleophile (acceptor), 0.2 equivalents TMSOTf in dichloromethane. Compounds were purified by liquid column chromatography using forced flow of the indicated solvent on Silicycle 230-400 mesh (6 nm pore diameter) silica gel. Prior to HPLC analysis, compounds were stored neat, at -20°C.

Reactor design and fabricaton. The five-port silicon microreactor contains three primary inlets, one secondary inlet and one outlet. The intent of this design is to mix and react the contents of the three primary inlets and then quench with a late-entering secondary stream before exiting the device. The microfluidic channels were etched into a single crystal silicon wafer and capped by a Pyrex wafer via an anodic bond (Figure 1a [in the main paper]). This construction was chosen for its excellent compatibility with a wide range of chemical reagents, as well as the high thermal conductivity of silicon - facilitating rapid thermal equilibration and temperature control. Moreover, the silicon can be oxidized to create a glass surface throughout the resulting microchannels. Deep reactive ion etching techniques (DRIE) make it easier to realize deep aspect ration structures in silicon than glass. Thus, the use of DRIE and subsequent oxidation and anodic bonding to pyrex facilitates making microreactors with glass surface properties. Additionally, the anodic bond, performed in a cleanroom environment, provides a hermetic seal at all points of contact between the silicon and pryex, preventing cross-channel contamination. All ports were directly connected to stainless steel tubing, sealed by brass ferrules which were attached to the tubing and subsequently soldered to copper pads deposited onto the silicon surface (Figure 1c).

The device flow channels and inlet ports were patterned onto 650μm thick, double side polished (DSP), oxidixed, silicon wafers using standard photolithographic techniques. The oxide layer was etched by buffered oxide etch (BOE) to produce a hard mask of the device pattern. The inlet ports on the back-side of the silicon wafer were then etched to a depth of 300μm using deep reactive ion etch (DRIE). Following the back-side etch, the flow channels were patterned and etched to a depth of 400μm as previously described. In order to ensure adequate mixing and long residence times the reactor is split into two main zones. The first zone is the mixing zone where the primary inlet steams are combined. Once mixed, the reactants enter the reaction zone, which is a high residence time zone terminated by the secondary inlet which is used as a quench inlet. The quenched reaction stream then exits the reactor.

The small dimensions of the microreactor imply that the flow is laminar, each fluid stream forms a lamina layer that mixes with the adjacent layers by diffusion. The time required to mix laminae can be estimated by $t = w/D$ where w is the width to diffuse and w is the diffusivity of the component of interest. In this system the first two inlets are split into two streams and the resulting streams are stacked to form five lamellae such that the component introduced in the first inlet must diffuse through the lamina resulting from the second inlet before reaching the lamina of the third inlet. The splitting of these streams also makes the concentration profile across the channel symmetric, reducing the diffusion width to half that of the channel (Figure 2). The mixing zone for this device is 119 mm in length, and 200μm wide, and 400μm deep. The reaction zone is a 400μm wide channel 430 mm in length, 400μm deep. This section has a much larger volume than the mixing zone and is intended to increase the residence time of the device. The residence time for this zone is $\tau = V/Q$, where V is the volume of the section and Q is the total volumetric flow rate through that section. The output of the reaction zone combines with the secondary inlet before exiting the reactor. The volumes of the mixing and reaction zones are 9.5 μl and 68.8 μl, respectively, for a total pre-quench reactor volume of 78.3 μl.

The concentration of a component in the reactor is determined by both the concentration inside the syringe and the flow rate of each stream. The flow rate of a given material stream is calculated according to Equation (1) where Q is the volumetric flow rate, r is the syringe internal radius and $\dot{h}$ is the linear speed of the syringe plungers.

$$Q = \pi r^2 \dot{h} \tag{1}$$

As the plunger speed ($\dot{h}$) is the same for all of the syringes, it is possible to calculated the concentration of reagents within the reactor. The total flow rate through the reaction zone is given by Equation (2) and the total flow rate after the quench has been added is given by Equation (3) where r_i^2 is the radius of syringe i (i.e. r_{p1}^2 is the radius of the syringe attached to primary flow port 1 and r_q^2 is the radius of the syringe attached to the quench, secondary inlet).

$$Q_{react} = \pi\left(r_{p1}^2 + r_{p2}^2 + r_{p3}^2\right)\dot{h} \tag{2}$$

$$Q_{react} = \pi\left(r_{p1}^2 + r_{p2}^2 + r_{p3}^2 + r_q^2\right)\dot{h} \tag{3}$$

Thus the concentration inside the reaction zone of any component can be determined by:

$$C_i = C_{i,syringe} \frac{r_i^2}{r_{p1}^2 + r_{p2}^2 + r_{p3}^2} \quad (4)$$

An inert species was used as an HPLC standard to normalize the data. The use of an HPLC standard compensates for collection errors and solvent evaporation. This inert species was added to the quenching syringe. To calculate the outlet concentration of the inert species the dilution due to the addition of the quench stream must be included, thus the inert concentration is determined by:

$$C_{i,out} = C_{i,syringe} \frac{r_i^2}{r_{p1}^2 + r_{p2}^2 + r_{p3}^2 + r_q^2} \quad (5)$$

Prior to data collection, syringes with freshly distilled CH_2Cl_2 were attached to the four fill ports to flush the device. Finally, the reactor was silylated by rinsing with TMSOTf to deactivate the surfaces of the reactor.

General Procedure for Data Collection. Prior to the introduction of reagents, the microchemical device was rinsed with 20 to 50 reactor volumes of anhydrous dichloromethane. Immediately before priming the device with reagent, 5 to 10 reactor volumes of 0.025 M TMSOTf in CH_2Cl_2 are flushed through the activator port and throughout the device. This procedure ensures that the activator line is fully primed and free from air bubbles, and deactivates the surface of the reactor by silylation. Following the installation of the reagent containing syringes, the device is flushed with 10 to 20 reactor volumes to displace gas bubbles, and prime the remaining reagent lines.

Samples were prepared by azeotroping the glycosylating agent and acceptor separately, and drying overnight on vacuum. Samples were diluted with freshly distilled dichloromethane to the desired concentrations: 0.03 M for mannosyl donor, 0.025 M for the nucleophile (acceptor), 0.025 M TMSOTf, and 0.05 M standard in Et_3N with 25% by volume dichloromethane. The following gas-tight syringes were employed: 5.0 ml syringes for both glycosyl donor and nucleophile, a 2.5 ml syringe for the quench/standard, and a 1.0 ml syringe for the activator.

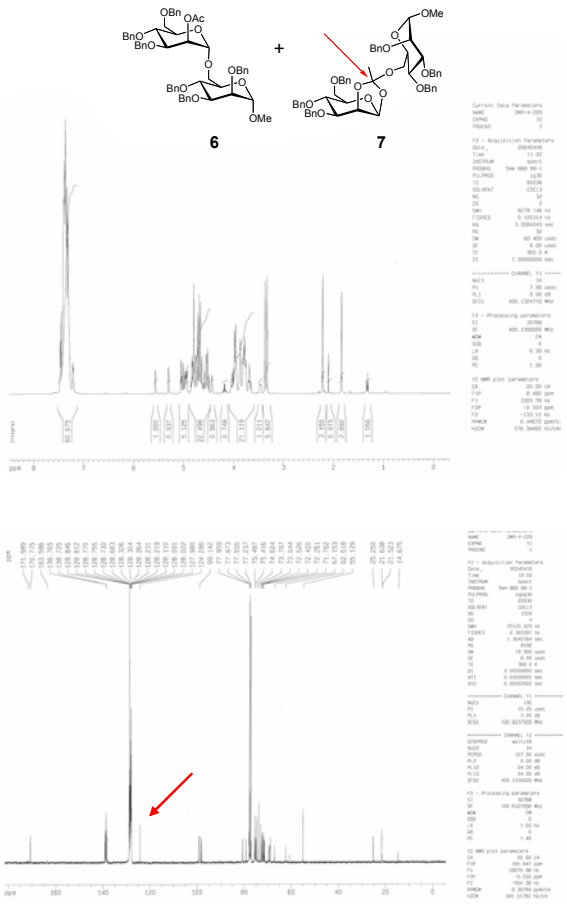
Sample collection was accomplished following equilibration of the temperature of the device (immersed in either a water or acetone bath, depending on temperature). 2.8 reactor volumes (220 µl) were delivered at the desired rate to flush the device, and equilibrate the reaction zone. Following the flush, 44 µl of material was collected for analysis, diluted with 20 µl hexanes, and the run was stopped. While maintaining the temperature, the device could be rerun at a different speed, or equilibrated to a different temperature.

Compound Characterization. All compounds were previously described in the literature.[1] 13C NMR was used to identify orthoester-containing mixtures by monitoring for peaks at ~120-125 ppm. Sample 1H and 13C NMR provided of product(**6**)/orthoester(**7**) mix (~1:1) (Supplementary Figure 1).

HPLC Analysis. Collected samples were analyzed using a Waters Nova-pak© silica column (3.9 x 150 mm) with EtOAc/hexanes as the mobile phase, monitoring at 257.9 nm. The data was normalized by dividing the area of a given peak by the area corresponding to the standard. See representative traces for chemistries depicted in supplementary Figure 3.

[1] For **2**, see J. Rademann, A. Geyer, R. R. Schmidt, *Angew. Chem. Int. Ed.* 1998, **37**, 1241. For **5**, see P. J. Garegg, S. Oscarson, A. –K. Tidén, *Carbohyd. Res.* **1990**, *200*, 475. For **6**, see X. L. Ding, F. Z. Kong, *J. Carbohyd. Chem.* **1998**, *17*, 915. Z. –J. Li, H. Li, M. –S. Cai, *Carbohyd. Res.* **1999**, *320*, 1.

Supplementary figures:



Supplementary Figure 1. Sample 1H and 13C NMR provided of product(**6**) /o rthoester(**7**) mix (~1:1)

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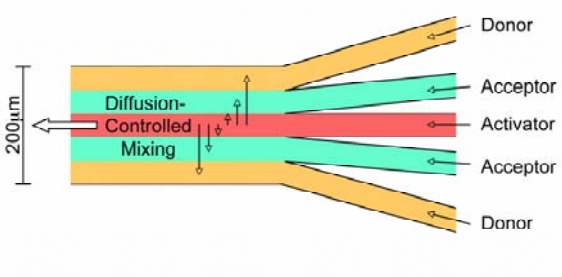
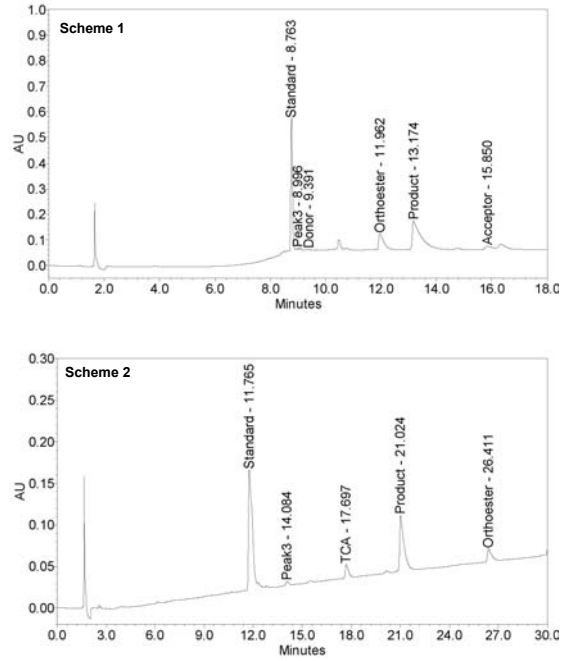


Supplementary Figure 2. Schematic of diffusion-controlled mixing of glycosyl donor, acceptor and activator laminae in the mixing zone.



Supplementary Figure 3. Representative HPLC scans from Schemes **1** and **2**.