Supplemental information for:

## Fully 3D printed Fluidic Devices with Integrated Valves and Pumps for Flow Injection Analysis

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<u>Overview</u>

- 1. Six STL files: 2 STL files per layer (with the 3 layers being a Flow Layer, Valve Layer, and Cover Layer), with one STL for the Agilus30 component and one for the VeroClear material.
- 2. Video (AVI file) of the peristaltic pumps in action
- 3. Video (AVI file) of the loading step (of the injection process)
- 4. 2 supplemental figures (Fig. S1 and S2)



**Figure S1**. A) Bright field micrograph showing cross sectional area of analysis channel used in the devices. The micrograph was taken with a Keyence VHX-500K digital microscope (Japan) and the cross-section area measurement made with their VHX Measurement Software. The cross-sectional area was found to be 8.2 x 10<sup>-4</sup> cm<sup>2</sup>. Considering the length of the injection plug shown in B) (3660 μm), the injection plug volume was calculated to be 300 nL.



**Figure S2**. Plot of peak elution (using apex of peak) vs pump frequency as well as peak width at half-height vs pump frequency. As can be seen on the figure, as pump frequency increased from 0.5 to 2 Hz, the peaks eluted more quickly and were narrower. It was found in separate studies that at frequencies greater than 5 Hz, the plugs did not migrate down the channel (frequency was too high for pumping action). These studies utilized a pump frequency of 2 Hz but the range of 2-4 Hz are clearly suitable for pumping in devices of these dimensions.