Supporting Information for:

## Droplet-based µChopper device with a 3D-printed pneumatic valving layer and a simple photometer for absorbance based fructosamine quantification in human serum

Yvette Kayirangwa, Md Mohibullah, and Christopher J. Easley\*

Department of Chemistry and Biochemistry, Auburn University, Auburn, AL, 36849

Email: chris.easley@auburn.edu

Supporting Information (SI) Contents:

- Page S-2: Photodiode slit characterization and optimization and path length measurement, Figure S-1.
- Page S-3: LED intensity characterization and optimization, Figure S-2.
- **Page S-4:** Analysis time comparisons between  $\mu$ C hopper and plate reader.



**Figure S-1. (A)** The photodiode seat was shaded with black oil-based paint marker, leaving only the slit area for light from the LED through the microdevice to pass through to the detector. Four layers were applied (labeled #1 - #4). **(B)** A sliced channel cross-section of the PDMS fluidic channel in the region of interest above the optical slit confirms the path length to be about 300 µm.



**Figure S-2.** LED intensities were varied and characterized to decide the optimum LED intensity for the experiments. In series with a 220  $\Omega$  resistor, a 10 k $\Omega$  potentiometer (variable resistor) was used to limit the current through the LED, and four intensities were evaluated (low, medium, high, and maximum) using quarter turns of the potentiometer. At the maximum light intensity (zero added resistance on the potentiometer), the PD was oversaturated and thus gave poor readings for both low and high BPB concentrations. The best results were observed with about one quarter turn on the variable resistor, labeled as "high intensity" in the figure. As shown in the figure, we also observed that consistent absorbance values for four different concentrations of BPB were measured at varying light intensities (low, medium, or high), as long as signal saturation was avoided, indicating that the photometer was functioning appropriately at any of these three light intensities.

## Analysis Times: µChopper versus Plate Reader

While plate readers are mature as a commercial analytical technology, and improved versions of the droplet-based  $\mu$ Chopper (with normally open valves, on-chip mergers, and more complex optics) have already been shown capable of rapid measurement and combinatorial capabilities,<sup>45</sup> here we have compared the run times needed with the devices in the current state.

The time required for each standard to be run in triplicate on our  $\mu$ Chopper device was determined to be 132 s. If a typical 5-point calibration curve is run (15 total samples), the device requires 660 s = 11 min to run all the samples in triplicate. In comparison, the plate reader required approximately 5 minutes to run the equivalent number of samples (15 samples). Thus, the  $\mu$ Chopper device run time was relatively short, even with sequential additions, channel priming, and rinses included. For either technique, these analysis times are small compared to the 2-hour incubation time needed with the fructosamine assay kit.