Supporting Information

Segmented Flow Sampling with Theta Push-Pull Pipettes

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Fabrication of theta pipette push-pull microfluidic device:

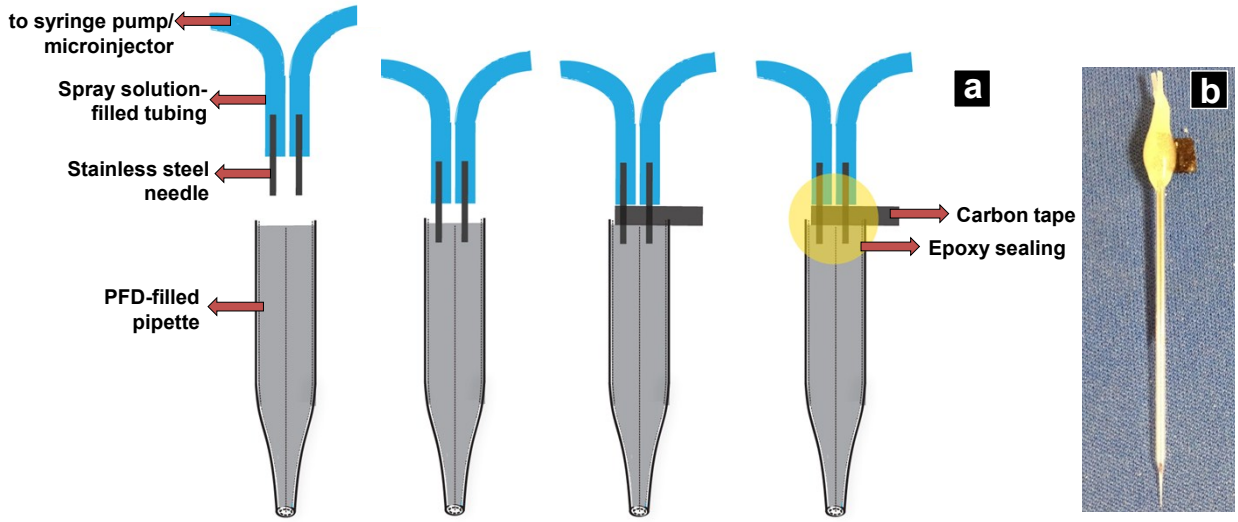

Figure S1. (a) Schematic showing the steps involved in fabrication of the push-pull microfluidic device from theta pipette. (b) Picture of a theta pipette push-pull microfluidic device

Segmented flow sampling:

Number of segments collected depends on volume of each segment and the geometry (shank length, width etc) of the pipette. In our lab, as many as 17 segments were collected through manual control (**Figure S2a**). If the goal of the experiment is to collect large number of segments then segments can be formed inside the stem of the pipette and correspondingly the segment volumes need to be increased to prevent coalescing inside the stem. In this study, our goal was to demonstrate formation and MS analysis of smallest segment reported to date and hence the latter strategy was not explored.

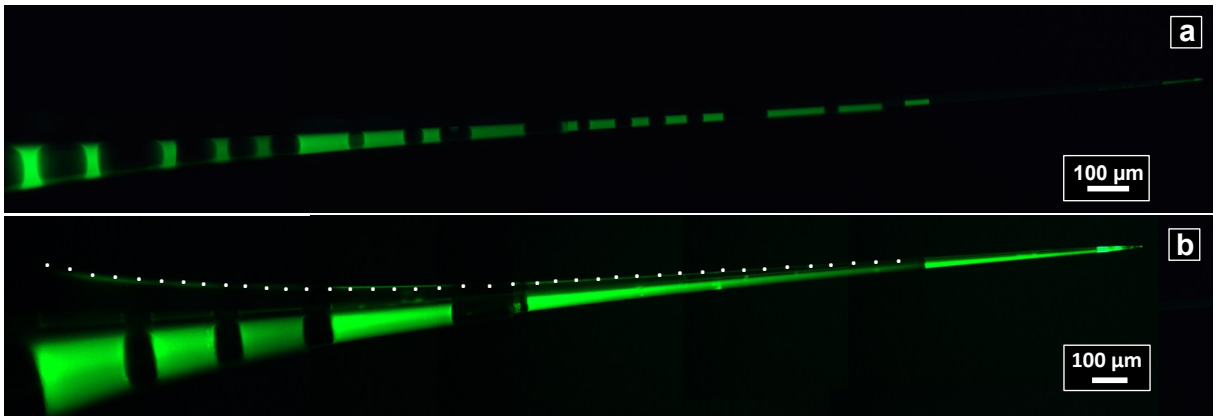

Figure S2 (a) Optical micrograph of a pipette showing 17 segments achieved through manually controlled segmented flow sampling. (b) Optical micrograph of a theta pipette showing segmented flow sampling in one barrel and PFD filled second barrel outlined by white dotted line

Carry-over studies:

Figure S3 shows the total ion chromatogram of a theta pipette with three aqueous segments. First segment delivered to the mass spectrometer contained fluorescein (m/z= 333.03) and dopamine (m/z=154.08). The following two segments contained only dopamine. Shown below are the total ion chromatogram of three aqueous segments delivered to the mass spectrometer and extracted ion chromatogram (EIC) at m/z= 333.03 and 154.08. The intensity of second and third peak in (b) is very small, indicating a very small amount of fluorescein is delivered to the mass spectrometer with the second and third aqueous segment, and thus a very small amount of carry-over is observed.

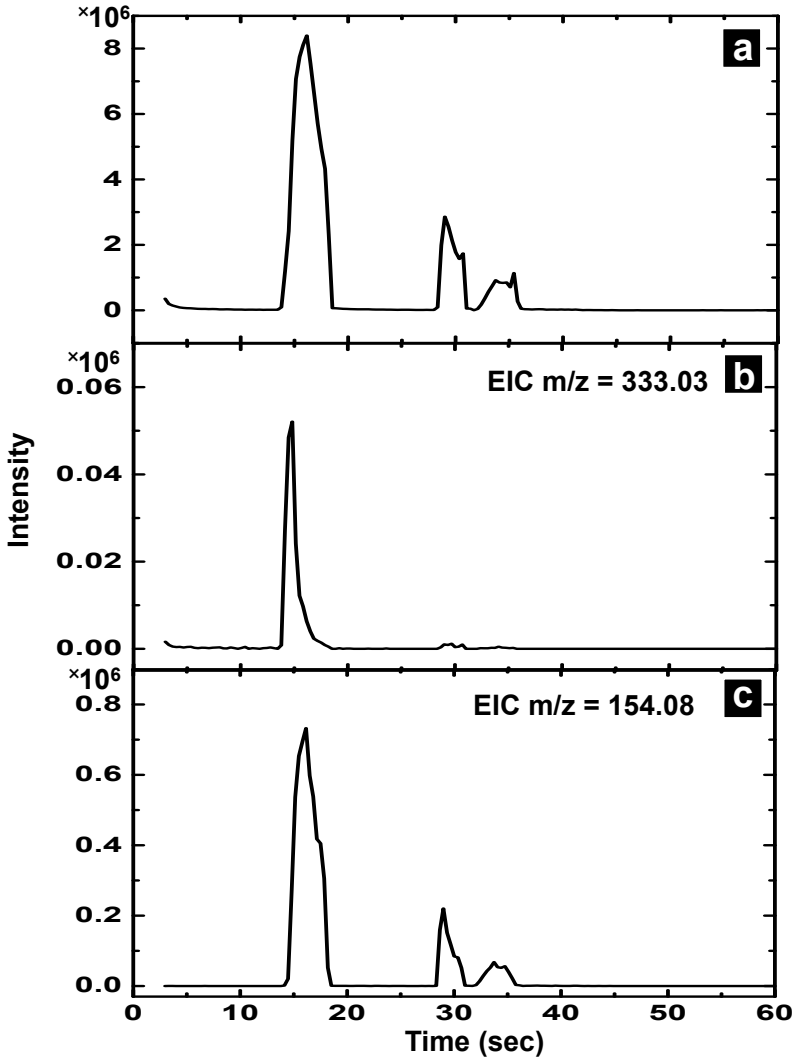

Figure S3 (a) Total ion chromatogram of the theta pipette for carry-over studies with three aqueous segments. (b) EIC of m/z = 333.03 and (c) EIC of m/z = 154.08

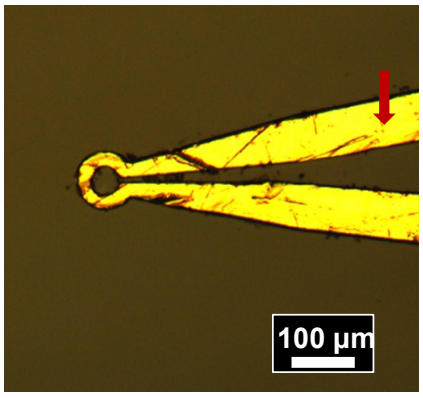

Figure S4. Optical micrograph of the gold electrode (bright yellow) used to perform electrochemical oxidation of dopamine to dopamine hydroquione (Dop HQ) in **Figure 3 and 4**. Arrow indicated the spot of sample collection.

Study of intercellular heterogeneity among *Allium cepa* cells

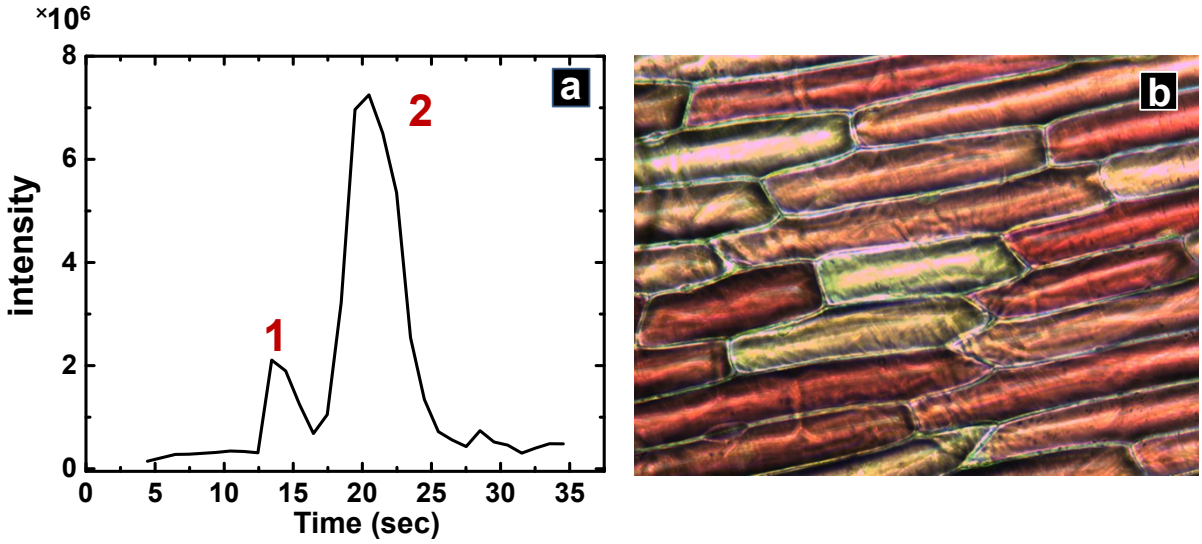

Figure S5. (a) Total ion chromatogram showing the delivery of two segments containing cytoplasm from colorless (1) and red (2) *Allium cepa* cells, respectively, sampled with a theta push-pull pipette (b) Optical micrograph of *Allium cepa* cells showing the intercellular heterogeneity in color.

Table S1. Tentative Peak assignments from mass spectra of *Allium cepa* epidermal cells.

Name	***Exact mass***	***Obs mass***
Monosaccharide +K (not in spectra range shown)	***219.0285***	***219.0142***
Cyanidin	***287.0515***	***287.0545***
Delphinidin/ quercetin +H	***303.0505***	***303.0478***

Disaccharide + K	***381.0799***	***381.0798***
Cyanidin glucoside	***449.1006***	***449.1072***
Delphinidin glucoside/ quercetin glucoside+H	***465.1033***	***465.1034***
Cyanidin glucoside-pyruvic acid	***517.0982***	***517.0914***
Cyanidin malonyl glucoside	***535.1087***	***535.1089***
Trisaccharide +K	***543.1328***	***543.1330***
Tetrasaccharide	***705.1875***	***705.1856***

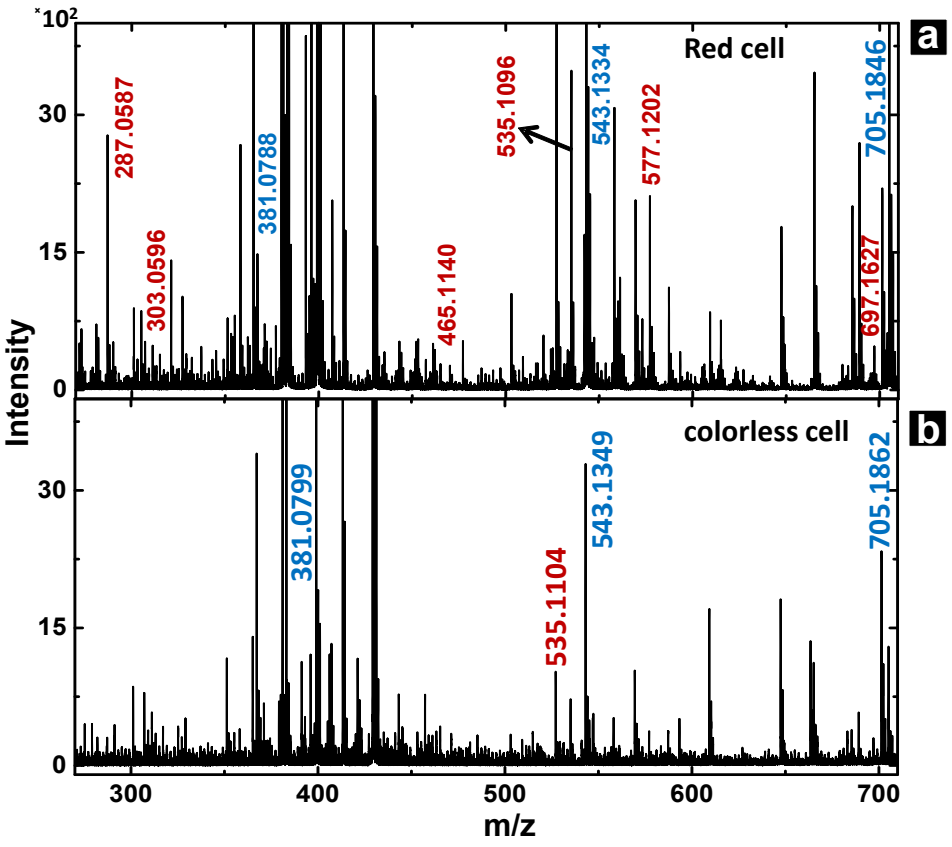

Figure S6. Mass spectra of a red (a) and a colorless (b) *Allium cepa* cell sampled using push-pull probe. The peaks labeled in red are anthocyanins and peaks in blue are oligosaccharides. Peak at 535 in (b) could be a result of carry-over or the cell had a faint pink color not

discernible to human eye. If carry-over is the major contributing factor here then other anthocyanin peaks should also be observed.