Electronic Supplementary Material (ESI) for Analytical Methods.

Supplementary Information: Understanding partial saturation in paper microfluidics enables alternative device architectures

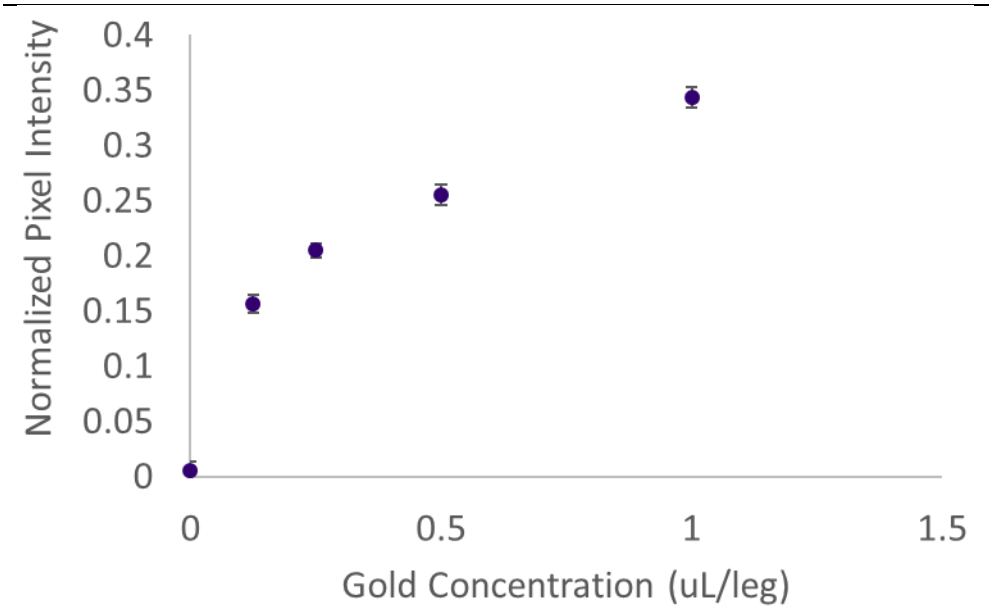


Figure S1. T20 biotin-streptavidin gold calibration curve for the automated dilution series device.

$$Concentration\ Factor = \frac{Input\ Volume}{Elution\ Volume} * \%\ Recovery \qquad \textbf{(Equation 1)}$$

Table S1. Summary of patient urine samples used in testing. Samples in red have pH values that are above the ideal value for chitosan purification and are expected to show reduced recovery of target DNA. Samples in green have pH values at or below the value required for chitosan purification.

Sample ID	pH	Salinity (mM)	Total NA in Sample (ng/µL)	Total non-target NA added to chitosan from 1 mL sample (ng)
01	7.3	181	0.5	500
02	6.2	108	9.8	9,800
03	6.0	11	0.3	300
04	7.8	132	1.0	1,000
05	6.8	37	3.0	3,000
08	5.8	214	2.4	2,400
09	7.0	78	2.1	2,100
10	5.8	236	15.8	15,800

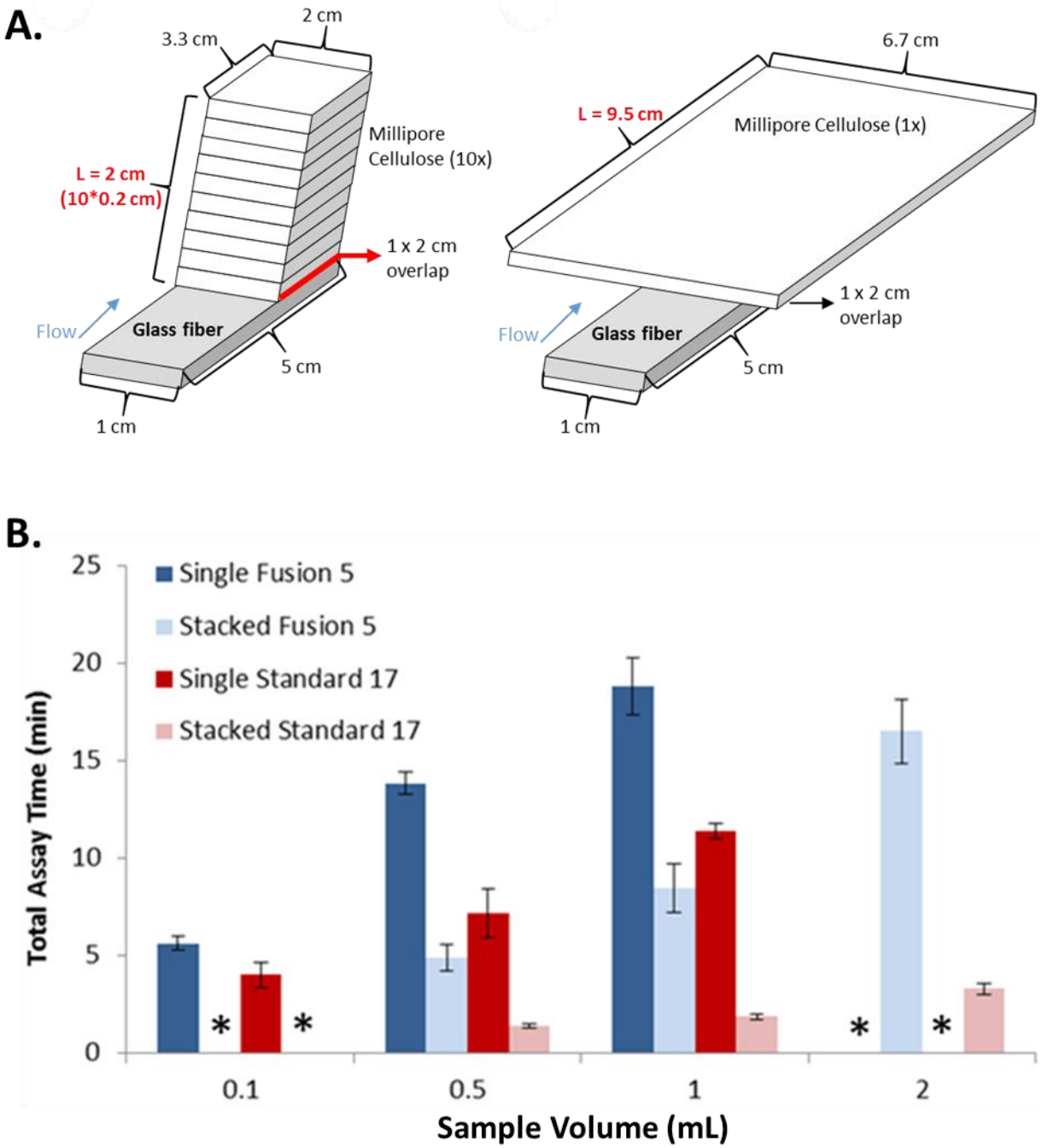
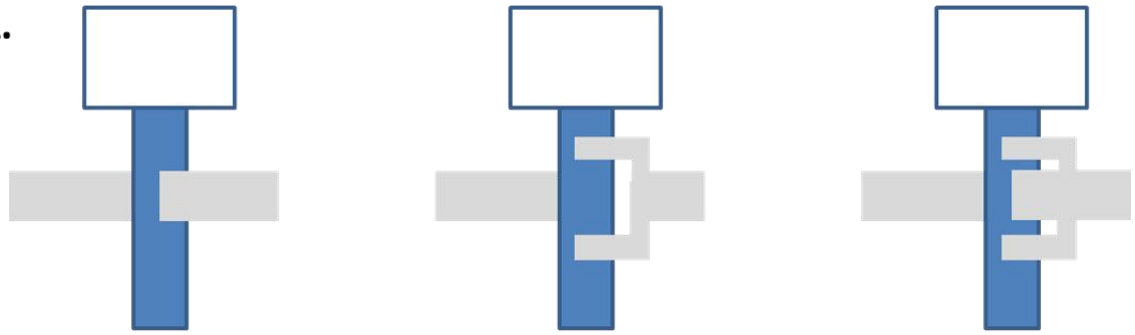
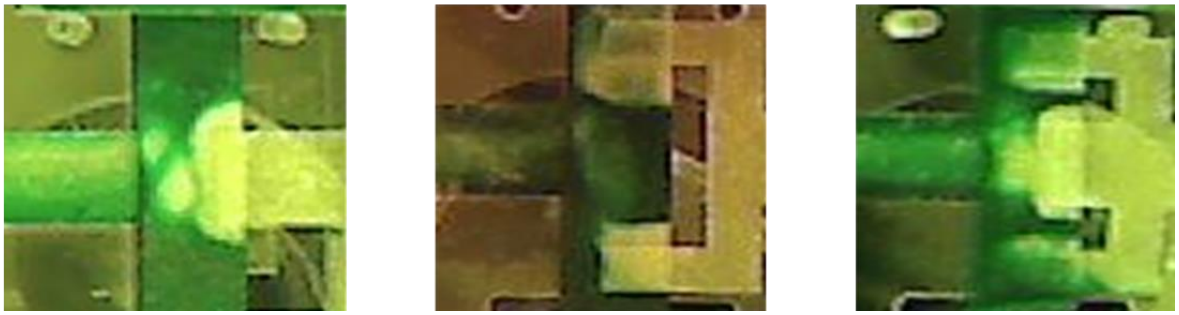


Figure S2. Optimizing waste pad configuration for large volume sample processing. **A.** Schematic of waste pad configurations. Stacked waste pads where L = 2 cm (10*0.2 cm, thickness of the cellulose membranes) and large single waste pad where L = 9.5 cm. **B.** Comparing total flow time of stacked waste reservoirs and single large reservoirs for flow through fusion 5 and standard 17. Averages of at least N=3 +/- one standard deviation are reported. *These conditions have not yet been tested within this system

A.



Post flow:

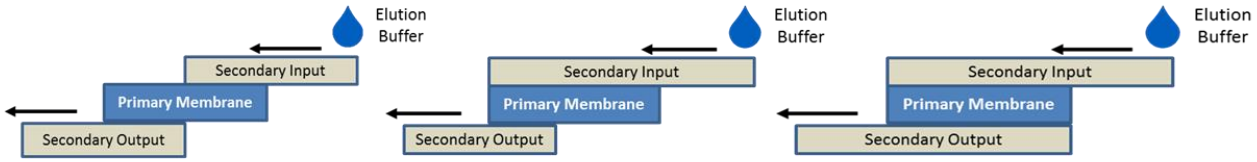


B.

Double Partial

Top Full, Bottom Partial

Double Full



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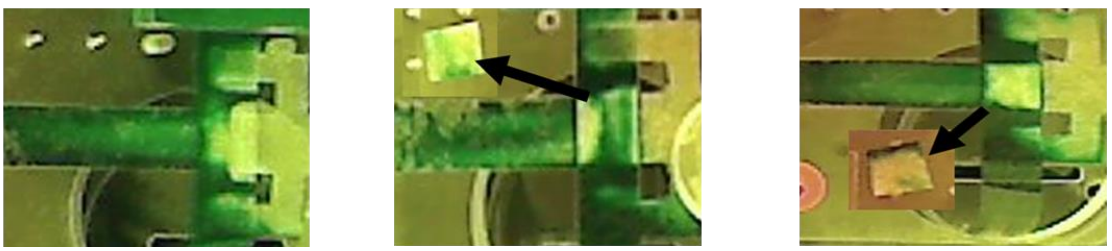

C.

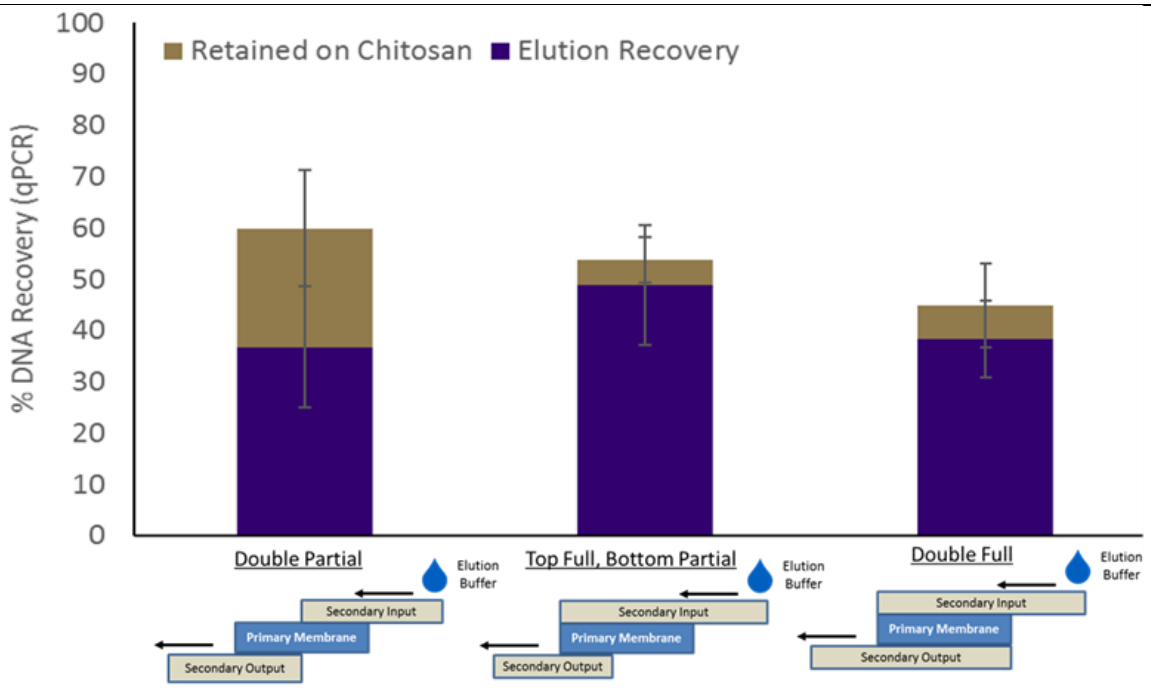

Figure S3. Optimizing junction geometry for automated p-switch devices. **A.** At the junction, the number of legs can help direct fluid into the elution region. The top and bottom legs of the three leg device cause a short-circuiting of the flow toward the waste region. **B.** The amount of overlap in the junction region can control wash-out into the elution region. **C.** DNA purification results from testing the different overlap geometries shown in part b. Devices were tested with DNA spiked into 1 mL of buffer and the elution was analysed by qPCR. The DNA remaining in the chitosan region was also quantified. Averages of N=3 are reported with error bars representing +/- one standard deviation.

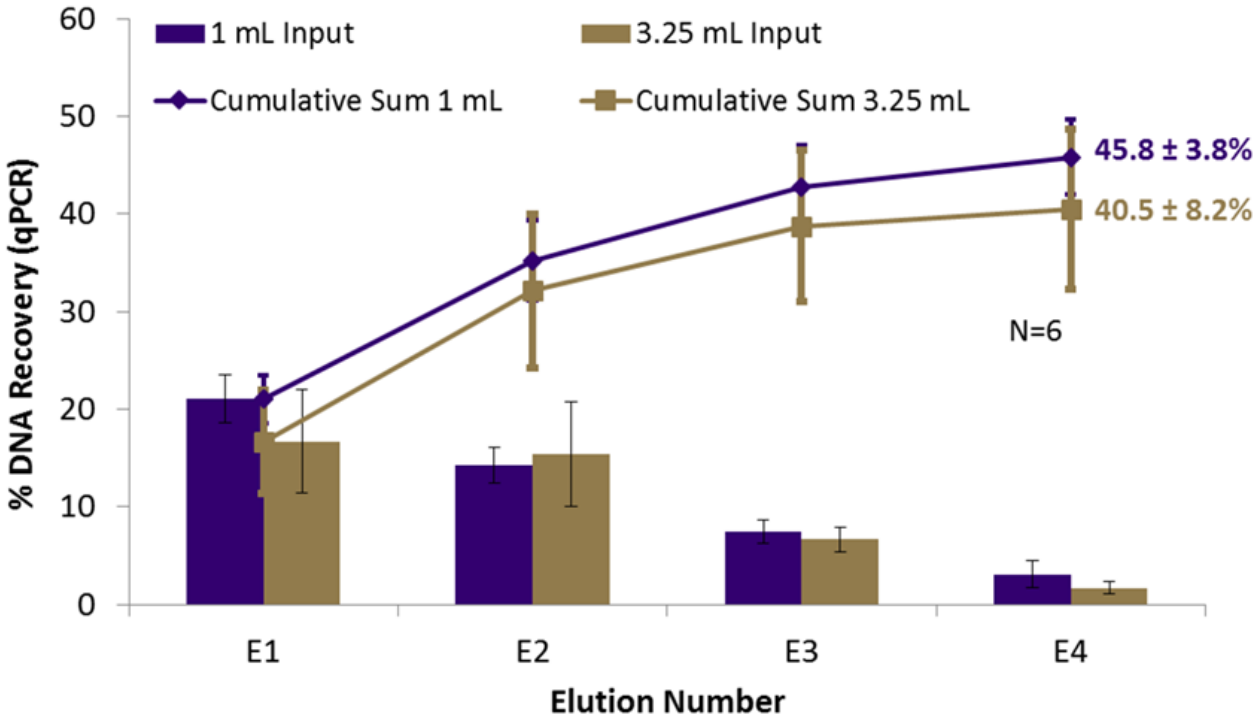

Figure S4. Automating the p-switch for the large volume sample concentration. Results from different elution regions from a DNA purification and concentration experiment using the automated p-switch device. Averages of N=6 are reported with error bars representing +/- standard error. The highest percentage of purified DNA was recovered in the first two elution volumes (each ~ 60 µL). The 1 mL samples were processed in 12-13 minutes while the 3.25 mL samples were processed in 32-33 minutes.