Supplementary Figures and Videos for

Motorized actuation system to perform droplet operations on printed plastic sheets

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**Figure S1** Screenshot of the Graphical User Interface (GUI) to remotely control the droplet actuation system. The connect button automatically searches for available COM port numbers. The circle button at the center resets the stage’s horizontal position. The single arrow buttons in four directions tilt the stage and return it to its initial position according to the speed (i.e. revolutions per minute, r.p.m.) and distance (i.e. finite steps). The double arrow buttons tilt and hold the platform in the four directions. In addition, GUI offers the option to preview a live video, take a snapshot, or record a video file for future analysis.
Figure S2 Determination of the optimal line width and inter-symbol spacing for plus symbols. In all cases, the droplet volume is 10 µL, tilting speed is 100 r.p.m., and number of steps is 14. (a) Sectional images of the printed plus symbols having different line widths and inter-symbol spacing. A droplet is placed on a row of plus symbols and its movement is tracked upon tilting the stage to the right side. (b) The error in droplet displacement is plotted for different line widths; each line width with four inter-symbol spacing. The optimal design occurs when the displacement error is negligible. (c) Images of a droplet over plus symbols demonstrating our definition of positive error, zero error, and negative error when the stage is tilted to the right side. A positive error occurs when the droplet displacement is more than one symbol, a zero error occurs when the droplet displacement is exactly one symbol, and a negative error is noted when the droplet stays in its original position.
**Figure S3** Physical model for the droplet detachment from a hydrophilic plus symbol. (a) Side-view of the droplet ready to detach from the hydrophilic symbol when the stage is tilted to a critical angle $\alpha$. (b) Top-view of the droplet showing its contact with a plus symbol and the superhydrophobic surface.
**Figure S4** Illustration of the procedure for measuring the volume of smaller droplet left or dispensed on a dot symbol. (a) 10 µL of water droplet initially pipetted (pipet-lite SL 10™) on the first dot symbol moves to the second dot symbol by tilting the stage. (b) A residual volume is dispensed on the first symbol and the remaining volume moves to the second dot symbol. (c)-(d) The remaining droplet on the second symbol is extracted into the pipette tip. (e) Using a 10 µL micro-syringe (Hamilton Microliter™ Syringe), the volume from the pipette tip detached from the pipette is transferred into the syringe. (f) The reading from the syringe shows the volume of the droplet after transport over one symbol (e.g. 9.4 µL as shown in the illustration).
**Table S1** Data displaying the remaining volume (or volume left) on second symbol and volume dispensed (or volume lost) on different symbols for an initial water droplet volume of 10 µL. The symbols used in our experiment are plus symbols (line width = 0.02 cm, line length = 0.24 cm) and two different-sized, solid circular dot symbols (diameter = 0.109 cm and 0.148 cm, respectively). For each symbol, the number of repeats (n) for every experimental and control tests is 10.

<table>
<thead>
<tr>
<th>Initial droplet volume = 10µL</th>
<th>Volume loss on different symbols (µL)</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>Volume left</td>
<td>9.71 ± 0.05  9.41 ± 0.05  9.12 ± 0.06  9.81 ± 0.07</td>
<td></td>
</tr>
<tr>
<td>Volume lost</td>
<td>0.29 ± 0.05  0.59 ± 0.05  0.88 ± 0.06  0.19 ± 0.07</td>
<td></td>
</tr>
</tbody>
</table>

**Table S2** Data displaying the remaining volume (or volume left) on the final symbol and volume dispensed (or volume lost) on multiple dot symbols for an initial water droplet volume of 10 µL. Each solid circular dot symbol has a diameter = 0.148 cm. For each symbol, the number of repeats (n) for every experimental and control tests is 10.

<table>
<thead>
<tr>
<th>Initial droplet volume = 10µL</th>
<th>Volume loss on different symbols (µL)</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>Volume left</td>
<td>9.12 ± 0.06  8.39 ± 0.09  7.69 ± 0.1  9.81 ± 0.07</td>
<td></td>
</tr>
<tr>
<td>Volume dispensed</td>
<td>0.88 ± 0.06  1.61 ± 0.09  2.31 ± 0.1  0.19 ± 0.07</td>
<td></td>
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</tbody>
</table>
Figure S5 Images of merged droplets (from 5 µL of a red droplet and 20 µL of a yellow droplet) as a function of time show the effect of stage agitation on enhancing molecular diffusion in the droplet. The blend ratio is 1:4 to produce an orange droplet, as per the datasheet of the dyes (Tone’s Food Color Kit™). A webcam (Logitech C920) is used to record the droplet mixing as a video file. The video file (resolution: 1280 x 720 pixels) is analyzed to extract the color intensities of four detection zones (15 x 15 pixels) within the droplet. A detection zone outside the droplet is used as the control zone. (a) Side-view of the merged droplet with no stage agitation. (b) Side-view of the merged droplet with stage agitation. Scale bar = 2 mm.
Figure S6 Characterizing the mixing profile in a merged droplet with stage agitation. (a) Side-view of a red droplet (5 µL) and yellow droplet (20 µL) placed on individual symbols. (b) Side-view of the merged droplet. (c) Plot of the averaged RGB color intensities of the four detection zones, along with RGB color intensities of the control zone. (d) Plot of the averaged grayscale color intensities of the four detection zones, along with grayscale color intensities of the control zone.
Figure S7 Characterizing the mixing profile in a merged droplet by passive diffusion (i.e. without stage agitation). (a) Side-view of a red droplet (5 µL) and yellow droplet (20 µL) placed on individual symbols. (b) Side-view of the merged droplet. (c) Plot of the averaged RGB color intensities of the four detection zones, along with RGB color intensities of the control zone. (d) Plot of the averaged grayscale color intensities of the four detection zones, along with grayscale color intensities of the control zone.

Figure S8 Glucose testing in 24-well plates. (a) In each well, different concentrations of 250 µL of glucose standard solutions are pipetted. After that, 500 µL of the assay reagent is added in each well. A webcam is used to record the color intensity changes within each well plate for 30 minutes. Each glucose concentration is tested three times (n = 3). (b) A Matlab script estimates the color intensity in each well at different time points. (c) The maximum slope of each color intensity graph is plotted to obtain the standard curve equation and to estimate the concentration of glucose in sheep serum.

Additional video files:
Supplemental video 1. Transport of single and multiple droplets (10 µL), transport of larger droplets (80 µL and 300 µL), and merging of three droplets.
Supplemental video 2. One-directional transport on single greater-than symbol and three converging greater-than symbols, dispensing small droplets on symbols (dot, rectangular, and diamond-shaped), and glucose detection test.

Supplemental video 3. Tests showing the volume range of three fluids (water, milk, and ethylene glycol) that can be transported using fixed operating conditions (speed =100 rpm, number of steps = 14).