Highly-customizable 3D-printed peristaltic pump kit

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Commercially available peristaltic pumps for microfluidics are usually bulky, expensive, and not customizable. Herein, we developed a cost-effective kit to build a micro-peristaltic pump (~50 USD) consisting of 3D-printed and off-the-shelf components. We demonstrated fabricating two variants of pumps with different sizes and operating flow-rates using the developed kit. The assembled pumps offered a flowrate of 0.02 \( \sim \) 727.3 lL/min, and the smallest pump assembled with this kit was 20 \( \times \) 50 \( \times \) 28 mm. This kit was designed with modular components (i.e., each component followed a standardized unit) to achieve (1) customizability (users can easily reconfigure various components to comply with their experiments), (2) forward compatibility (new parts with the standardized unit can be designed and easily interfaced to the current kit), and (3) easy replacement of the parts experiencing wear and tear. To demonstrate the forward compatibility, we developed a flowrate calibration tool that was readily interfaced with the developed pump system. The pumps exhibited good repeatability in flowrates and functioned inside a cell incubator (at 37 \( ^\circ \)C and 95 % humidity) for seven days without noticeable issues in the performance. This cost-effective, highly customizable pump kit should find use in lab-on-a-chip, organs-on-a-chip, and point-of-care microfluidic applications.

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1. Hardware in context

This paper describes the fabrication and operation of a do-it-yourself (DIY) peristaltic pump kit. Flow control is paramount in the development of microfluidic systems for lab-on-a-chip (LoC) and organs-on-a-chip (OoC) applications [1,2]. On-chip flows are typically controlled by integrating a microfluidic chip with external pumps to deliver fluidic samples at microscale through the microchannels (usually in the order of μL/min) [3–5]. To this end, commercially available flow devices such as extrusion syringe pumps, peristaltic pumps, and pneumatic pumps have been widely used [6–13]. However, existing pumps suitable for microfluidic applications are relatively bulky and costly [14–16]. Pressure-controlled flow systems cost approximately ten thousand US dollars, while syringe pumps and peristaltic pumps cost hundreds to thousands of US dollars. For LoC and OoC applications, pumps with a small footprint are preferable [1,17]. While miniaturized pumps are available [4,18,19], such pumps include proprietary and costly control systems (> 1000 USD). Importantly, these commercially available pumps are not amenable to customization. Since each experiment has its own requirements (i.e., flowrate, working environment, space constraint), rapid customization of the instrument would be advantageous to users; the users should be able to tailor the flow control system specific to their experiments rather than to tailor their experiments around the flow control system.

With the introduction of open-source electronic prototyping platforms such as Arduino, precise control of motors is becoming accessible to non-experts. Arduino also empowers scientists with little background in electronics and programming to build complex scientific instruments [20]. With the advance in 3D printing technologies, scientists no longer need to rely on manufacturers to produce parts; they can design and print them at an affordable cost. 3D printers have become commodities found in most scientific laboratories today. 3D printers based on stereolithography (SLA) can cost as little as 300 USD for liquid-crystal display (LCD) variants. SLA 3D printers produce structural parts with good repeatability and dimensional tolerance (± 0.15 %, with a limit of ± 0.01 mm) [21,22]. With the low-cost SLA printers and the open-source electronics, miniaturized micropumps comparable to commercial options can be easily designed, fabricated, and assembled in-house. 3D printing and open-source electronics have enabled end-users to build a flow control system in microfluidics and relevant fields. Despite successful demonstrations, however, existing DIY pumps have yet focused on developing a finished product rather than a platform that can be extended, configured, and hacked [23–25]. We believe that a kit has the intrinsic ability to evoke the culture of hacking and tinkering. In light of that, we proposed to develop a kit to assemble peristaltic pumps, which we believe promotes the design, fabrication, and customization of open-source scientific infrastructure with forward compatibility by the scientific community.

To this end, our current work has addressed two challenges pertinent to micro-peristaltic pumps: (1) the lack of affordable pumps suitable for microfluidics and (2) the lack of pumps suited for customization. To overcome these challenges, we first designed and built a micro-peristaltic pump kit with 3D-printed parts and the open-source Arduino platform. Instead of building a finished product, we designed this kit with modular and standardized components. Modularity offers advantages in reconfiguring and customizing the setup to better tailor to each experiment. Furthermore, modularity encourages the scientific community to expand the kit by designing new modules continually to add functionality. We demonstrated the expansion of the kit by developing a flow calibration tool that was readily interfaced with the other pump systems. Our work enhanced the availability of the flow control systems with improved customizability and expandability, benefitting the researchers in microfluidics, chemical engineering, and biomedical engineering.

2. Hardware description

We designed a DIY peristaltic pump kit as a low-cost alternative to existing peristaltic pumps that are either expensive, large, or not amenable to hacking and tinkering. The kit was designed to achieve easy variations in size, flowrate, and the number of channels. We also developed a tool to calibrate flowrates. Only open-source and readily available components (e.g., Arduino, stepper motors) were used to keep the cost of the materials low (~ 50 USD). Currently, low-cost peristaltic pumps (< 10 USD) are commercially available in the market, although they are designed for aquarium applications with high flowrate (~ mL/min) (Table 1). These aquarium pumps are not suitable for applications in microfluidics that require low flowrate (~ μL/min) and precise control over the flowrate (Table 1). Commercially available micro-peristaltic pumps (e.g., Takasago 6-channel pump, Takasago RP-TX series) for microfluidics typically cost greater than 1000 USD (Table 1) [18,19]. The development of FAST pump was recently reported by Jönsson et al. (Table 1) [23]. FAST pump is a DIY, SLA-printed peristaltic
which helped reduce the overall footprint of the pump. The motor required a lower current (150 mA) than conventional step-

we further reduced the footprint of the pump. Overall, we introduced a low-cost (85 USD) stepper motor than the one demonstrated in the FAST pump (42 USD). FAST pump also included ball bearings while our pumps operated well without using the ball bearings. By negating the use of ball bearings, we further reduced the footprint of the pump. Overall, we introduced a low-cost (~ 50 USD), DIY peristaltic pump kit with a customizable footprint (30 × 85 × 17 mm and 20 × 50 × 28 mm), a wide range of flowrates (0.02 ~ 727.3 μL/min), and the number of channels. Lastly, the developed kit was designed with modularity, envisaging end-users to design and incorporate additional modules to improve the functionality of this kit in the future.

2.1. Peristaltic Pump: Working principle

A peristaltic pump delivers fluids by alternating compression and relaxation of a flexible fluidic tube. The dynamics of the tube permit drawing and propelling the fluid through the pump [26–28]. We developed the pump consisting of three rollers. The three-roller assembly was mechanically simple, where the rotational torque from a single motor directly translated to the propulsion of fluid. The rotation of the rollers compressed and relaxed the flexible tubing, creating negative and positive pressures that drew and delivered the fluid through the tube (Fig. 1).

2.2. Kit: pump, calibration tool, fluid reservoir

The current kit contains the parts to assemble two variants of the peristaltic pump and a tool to calibrate the flowrates (Fig. 2). We demonstrated assembling two variants of the pump using the parts developed for the kit (Fig. 3A). Variant A was a four-channel peristaltic pump with a footprint of 30 × 85 × 17 mm with flowrates of 0.05 ~ 727.30 μL/min. It was also possible to change the configuration of Variant A, from a bottom-loaded configuration to a top-loaded configuration, by swapping Parts #1–2 with Parts #7–8 (Fig. 3B-C). The bottom-loaded configuration required the removal of multiple parts to access the tubing. The top-loaded configuration allowed for access to individual tubing without the hassle of removing the multiple parts. Variant A had a circumferential diameter, \( D = 15 \text{ mm} \), and rollers diameter, \( d_r = 4 \text{ mm} \). \( D \) and \( d_r \) determined the volume of fluid displaced per cycle. For Variant A, the displaced volume per cycle was ~ 6.3 μL. Variant B was a 3-channel peristaltic pump with an overall footprint of 20 × 50 × 28 mm with flowrates of 0.02 ~ 143.00 μL/min. Variant B was with \( D = 6.8 \text{ mm} \) and \( d_r = 1.8 \text{ mm} \), which resulted in a volume displacement of ~ 2.3 μL per cycle. The same motor operating at the same revolutions per minute (rpm) yielded a lower flowrate in Variant B than Variant A.

To power the pump, we selected a two-phase four-wire micro-stepper motor coupled to a gearbox with a gear ratio of 300:1. While this stepper motor was not in the class of National Electrical Manufacturers Association (NEMA) stepper motor, we selected this motor because of the small size and the low required current. The volume of the motor was ~ 10 × 10 × 10 mm, which helped reduce the overall footprint of the pump. The motor required a lower current (150 mA) than conventional step-

![Fig. 1. Illustration of the working principle of a peristaltic pump. As the rollers rotate, they compress and relax the flexible tubing, creating a positive and negative pressure that draws fluid through the tube. \( D \) is a circumferential diameter and \( d_r \) is a roller diameter.](image-url)
per motors (1A), and it was possible to drive the motor by the output pins of Arduino (40 mA, 5 V). The standalone microstepper motor (without the gearbox) has a step angle of $18^\circ$ ($\pi/10$ rad). With the gear ratio of 300:1, each angular step was decreased to $\pi/3000$ rad. The gear ratio increased the output stall torque from 5 g cm to 1.6 kg cm. This magnitude of torque...
was sufficient to overcome mechanical resistance (such as frictional forces) to the rotation. This motor offered a wide range of operating rpm (0.01 – 9 rpm). We note that the stepper motor does not have a lower limit in rotation rate (rpm); the rotation rate can be decreased by increasing the duration of the pause between the steps. In practice, we set the lowest rotational speed to be 0.01 rpm (corresponding to 0.02 rotation rate can be decreased by increasing the duration of the pause between the steps. In practice, we set the lowest rotational speed to be 0.01 rpm (corresponding to 0.02 rotation). In our experiments, we found that both clear water and water with blue dye were suitable for calibration. Using this configuration, we measured the time taken for the liquid to travel from the first sensor to the second sensor to get the linear speed (mm/s). Accounting for the tube diameter, we then calculated the volumetric flowrate. We note that this calibration tool is not for the real-time monitoring of the flowrate. Rather, it is meant to perform rapid flowrate calibration prior to the experiment. This capability is beneficial for the applications requiring precision in the flowrate of the fluids (such as perfusion cell culture).

Lastly, a fluidic reservoir was an essential component in a microfluidic system. Intending to use common lab supplies to build the system, we modified a standard 2-ml Eppendorf tube to create a fluid reservoir (Fig. 3G). To secure this Eppendorf tube in place, we designed and 3D printed a tube holder (Part #9). Overall, we demonstrated the versatility of the current kit to assemble (1) two variants of the pump with varying specifications, (2) a flowrate calibration tool, and (3) a fluid reservoir holder. Our demonstration suggested both customization and extension of the pump were feasible using the kit we developed.

### 2.3. Modularity and forward compatibility

Modularity is one of the key features of the developed kit. Here, we define modularity as deconstructing the peristaltic pump into small subunits called modules, which can be independently printed, modified, replaced, or exchanged with other modules in the same system or between different systems. Identifying each module was essential to understand the function of the pump. Crucially, each module is 3D printable or commercially available. Each module was standardized to ensure (1) customizability (users can easily reconfigure the various components to comply with their experiments), and (2) forward compatibility (additional standardized parts can be interfaced to the current kit). To standardize the design of the device and promote the assembly of the modules, we used a breadboard consisting of M2 through-holes spaced 10 mm apart. The M2 through-holes allowed mounting various modules to the breadboard, analogous to studs on Lego® bricks. Modules were designed with through-holes spacing in a multiple of 10 mm.

We identified three major advantages to deconstruct the pump into 3D printable modules. Firstly, it permits the module-by-module replacement of the subparts. For example, the use of tubes of different sizes was possible without replacing the entire parts. We assembled two types of tubes with (1) ID = 1 mm and OD = 2 mm and (2) ID = 2 mm and OD = 3 mm (ID and OD are inner and outer diameters, respectively) with Variant A by changing Parts #1 and #2 (Fig. 4A). Similarly, we assembled the tubes with different sizes to Variant B by changing Parts #10 and #12 (Fig. 4B). It is also possible to replace the module that might wear off faster than others. For example, the end supports, being exposed to constant frictional forces with the driveshaft, may wear out after a prolonged operation, which can be easily replaced. Secondly, it allowed designing the flow system by uniquely positioning the parts (e.g., pumps, fluid reservoirs) for the intended microfluidic experiments. This capability was advantageous in reconfiguring the whole experimental setup (i.e., number of channels, orientations, and proximity). For example, we illustrated the setup with a single petri-dish and two fluidic channels (Fig. 5A), individual Petri-dishes with four fluidic channels (Fig. 5B), and a standalone pump for point-of-care applications (Fig. 5C).

Thirdly, it ensured the fidelity of the fabrication of the parts during 3D printing. 3D features requiring accuracy and alignment (e.g., through-holes) were oriented parallel to the Z-axis during 3D printing. We found that the direction of through-holes that were aligned perpendicularly to the Z-axis (i.e., along XZ-plane or YZ-plane) did not result in prints with good fidelity. In such cases, 3D-printed parts did not match the 3D CAD files in terms of dimensional tolerance; the through-holes often appeared deformed due to the inadvertent curing of the polymer. The deformed holes would affect the alignment of shafts and compromised the rotation of the shafts. In particular, the holes in the end support (Parts #3 and #16) and the end brackets (Parts #5, #13, and #14) required good fidelity of printing. To this end, the direction of the through-holes was aligned parallel to the Z-axis (see also Fig. 7).

Features:

- Affordability; less expensive (~ 50 USD) than commercial alternatives (> 1000 USD)
- Wide range of flow control (0.02 ~ 727.30 μL/min)
- Modular design for reconfigurability
- Small footprint (20 × 50 × 28 mm)
Fig. 4. **A)** Photograph of Variant A and the additional modules (Parts #1 and #2) for varying tubing size. Parts #1-b and #2-b were designed for tubing with ID = 1 mm and OD = 2 mm. Parts #1-c and #2-c were designed for tubing with ID = 2 mm and OD = 3 mm. **B)** Photograph of Variant B and the additional modules (Parts #10 and #12) for varying tubing size. Parts #10-b and #12-b were designed for tubing with ID = 1 mm and OD = 2 mm. Parts #10-c and #12-c were designed for tubing with ID = 2 mm and OD = 3 mm.

Fig. 5. Schematic drawing of the setup consisting of **A)** one 100-mm Petri dish with two fluidic channels and two reservoirs, **B)** four 30-mm Petri dish with four fluidic channels and four reservoirs, and **C)** Variant B pump and three reservoirs.
3. Design files

Design files Summary

| Design file name | File type | Open source license | Location of the file |
|------------------|-----------|---------------------|----------------------|
| Part #1          | STL file; CAD file | CC | available with the article |
| Part #2          | STL file; CAD file | CC | available with the article |
| Part #3          | STL file; CAD file | CC | available with the article |
| Part #4          | STL file; CAD file | CC | available with the article |
| Part #5          | STL file; CAD file | CC | available with the article |
| Part #6          | STL file; CAD file | CC | available with the article |
| Part #7          | STL file; CAD file | CC | available with the article |
| Part #8          | STL file; CAD file | CC | available with the article |
| Part #9          | STL file; CAD file | CC | available with the article |
| Part #10         | STL file; CAD file | CC | available with the article |
| Part #11         | STL file; CAD file | CC | available with the article |
| Part #12         | Adobe Illustrator file for laser-cut | CC | available with the article |
| Part #13         | STL file; CAD file | CC | available with the article |
| Part #14         | STL file; CAD file | CC | available with the article |
| Part #15         | STL file; CAD file | CC | available with the article |
| Part #16         | STL file; CAD file | CC | available with the article |
| Part #28 – 20 mm | STL file; CAD file | CC | available with the article |
| Part #28 – 40 mm | STL file; CAD file | CC | available with the article |
| Part #19         | Laser cut file   | CC | available with the article |

**Part #1** is the pump bracket that functions as the boundary for the silicone tubing. As the roller rolls over the silicone tubing, the bracket and the roller ensure that the tubing is fully compressed, resulting in a negative pressure that draws the fluid.

**Part #2** is a platform consisting of the anchoring pins that function to secure silicone tubing and prevent the tubing from slipping during operation.

**Part #3** is the end support that secures the drive shaft (Part #20) in place.

**Part #4** is the roller that comes into direct contact with the silicone tubing.

**Part #5** is the end bracket that secures the rollers to the drive shaft.

**Part #6** is the frame that secures the motor in place.

**Part #7** is the bracket that functions as the boundary for the silicone tubing; an optional part for the *top-loaded* configuration (Fig. 3B-C).

**Part #8** is a platform consisting of the anchoring pins that function to secure silicone tubing and prevent the tubing from slipping during operation; an optional part for the *top-loaded* configuration (Fig. 3B-C).

**Part #9** is the Eppendorf tube holder that serves as a reservoir for the perfusion.

**Part #10** is the main frame for Variant B.

**Part #11** is the motor holder for Variant B.

**Part #12** is the tubing cover for Variant B.

**Part #13** is the end bracket that couples the rollers to the motor shaft.

**Part #14** is the end bracket to secure the drive shaft and roller shafts.

**Part #15** is the bracket to secure the motor in place.

**Part #16** is the end support to secure the drive shaft in place.

**Part #19** is the breadboard consisting of a PMMA sheet with M2 through-holes.
## 4. Bill of materials

### Bill of materials

| Designator | Component Description | Number | Cost/unit - SGD | Total cost - SGD | Source of materials | Material type |
|------------|-----------------------|--------|-----------------|-----------------|---------------------|--------------|
| Part #1    |                       | 1      | 486.00/L        | 0.63            | Formlabs            | Dental SG Resin |
| Part #2    |                       | 2      | 486.00/L        | 0.51            | Formlabs            | Dental SG Resin |
| Part #3    |                       | 2      | 486.00/L        | 0.45            | Formlabs            | Dental SG Resin |
| Part #4    |                       | 3      | 486.00/L        | 0.21            | Formlabs            | Dental SG Resin |
| Part #5    |                       | 2      | 486.00/L        | 0.33            | Formlabs            | Dental SG Resin |
| Part #6    |                       | 1      | 486.00/L        | 0.34            | Formlabs            | Dental SG Resin |
| Part #7    |                       | 4      | 486.00/L        | 0.80            | Formlabs            | Dental SG Resin |
| Part #8    |                       | 2      | 486.00/L        | 1.45            | Formlabs            | Dental SG Resin |
| Part #9    |                       | 4      | 486.00/L        | 1.27            | Formlabs            | Dental SG Resin |
| Part #10   |                       | 1      | 486.00/L        | 0.97            | Formlabs            | Dental SG Resin |
| Part #11   |                       | 1      | 486.00/L        | 0.56            | Formlabs            | Dental SG Resin |
| Part #12   |                       | 1      | 486.00/L        | 0.30            | Formlabs            | Dental SG Resin |
| Part #13   |                       | 1      | 486.00/L        | 0.13            | Formlabs            | Dental SG Resin |
| Part #14   |                       | 1      | 486.00/L        | 0.08            | Formlabs            | Dental SG Resin |
| Part #15   |                       | 1      | 486.00/L        | 0.09            | Formlabs            | Dental SG Resin |
| Part #16   |                       | 1      | 486.00/L        | 0.09            | Formlabs            | Dental SG Resin |
| Part #17   | Tubing Tygon .02X.06  | 100    | 0.01 ft/roll    | 192.00/100 ft   | 1.92 https://www.coleparmer.com/i/masterflex-transfer-tubing-tygon-nd-100-80-microbore-0-020-id-x-0-060-od-100-ft/0641901 | Polymer |
| Part #18   | Micro Gear Motor, 2-Phase | 1 | 4-Wire – 5 rpm | 14.54 | 14.54 https://sea.banggood.com/Machifit-GM1024BY10-DC-5V-51530RPM-Micro-Gear-Motor-2-Phase-4-Wire-Stepping-Motor-All-Metal-Gearbox-p-1463680.html | Metal |
| Part #19   | PMMA breadboard       | 1      | 25.29/0.12 m²   | 0.12 m²         | 1.79 https://www.aliexpress.com/item/32968042304.html | PMMA  |
| Designator | Component | Number | Cost/unit - SGD | Total cost - SGD | Source of materials | Material type |
|------------|-----------|--------|-----------------|-----------------|---------------------|--------------|
| Part #20   | Solid 2 mm Diameter × 60 mm | 1 | 0.20 | 0.20 | https://www.sgbotic.com/index.php?dispatch=products.view&product_id=2718 | Metal |
| Part #21   | Solid 2 mm Diameter × 30 mm | 3 | 0.10 | 0.30 | https://www.sgbotic.com/index.php?dispatch=products.view&product_id=2832 | Metal |
| Part #22   | 1/2" Plastic Blunt Tip Dispensing Needle – 14G | 3 | 0.13 | 0.38 | https://shopee.sg/product/96555506/1881154404 | Polymer |
| Part #23   | 1/2" Blunt Tip Dispensing Needle – 23G | 6 | 0.12 | 0.69 | https://sea.banggood.com/50Pcs Set-12-Blunt-Tip-Dispensing-Needle-w-Clear-Protector-for-Syringe-Refilling-and-Measuring-Liquid-Industral-Glue-Applicator-Different-Gauge-p-1502090.html | Metal |
| Part #24   | 1/2" Blunt Tip Dispensing Needle – 18G | 3 | 0.12 | 0.35 | https://shopee.sg/product/85459119/7908644806 | Metal |
| Part #25   | 1.5" Blunt Tip Dispensing Needle – 18G | 1 | 0.24 | 0.24 | https://shopee.sg/product/85459119/7908644806 | Metal |
| Part #26   | Silicone tubing (ID0.5xOD1mm) – 1 m | 1 | 1.10 | 1.10 | https://www.aliexpress.com/item/33003631601.html | Polymer |
| Part #27   | 2 to 3 mm flexible shaft coupler | 1 | 1.74 | 1.74 | https://www.aliexpress.com/item/4000201861175.html | Metal |
| Part #28 – 20 mm | 1 | 486.00/L | 0.55 | | Formlabs Dental SG Resin | Other |
| Part #28 – 40 mm | 1 | 486.00/L | 0.74 | | Formlabs Dental SG Resin | Other |
| Part #29   | Photo Interrupter GP1A57HRJ00F | 2 | 2.95 | 5.90 | https://www.sgbotic.com/index.php?dispatch=products.view&product_id=451 | Other |
| OLED display | 0.96 Inch I2C Graphic OLED Display Module | 1 | 3.50 | 3.50 | https://shopee.sg/0.96-Inch-I2C-Graphic-OLED-Display-Module-128x64-White-Blue-Color-for-Arduino-i.189216777907938507 | Other |
| Arduino Micro | Arduino Micro/Arduino Nano | 2 | 4.01 | 8.02 | https://shopee.sg/Arduinleo-Pro-Micro-ArTmea32U4-5-V-f o r-Arduino-IDE-1.0.3-Bootloader-Re place-Pro-Mini-i.161535507.2423045509 | Other |
| Tactile Switch | Tactile Push Button switch | 3 | 0.04 | 0.11 | https://shopee.sg/200Pcs-6x6mm-Tactile-Push-Button-Straight-Switch-Micro-Momentary-Tact-Kits-i.1406258163715140290 | Other |
| Latching Push Button | Latching Type Push Button | 2 | 0.23 | 0.45 | https://shopee.sg/20-Pcs-1.2-in-Thr ead-Green-Red-Cap-SPST-Latching-Type-Push-Button-Switch-OFF-ON-i.130099651.2017623850 | Other |
| Power Adaptor | USB power Adaptor – 1A, 5 V | 1 | 4.20 | 4.20 | https://shopee.sg/USB-wall-charger-3PIN-UK-Plug-Single-Port-Power-5V-1A-Adaptor-for-all-phones-i.110927509.322481363 | Other |
| MicroUSB Cable | MicroUSB Cable | 1 | 2.35 | 2.35 | https://shopee.sg/Samsung-100-Origin al-Android-Data-Micro-USB-Cab le-1.2-m-Fast-Charging-S7-S6-i.82436100.1693846962 | Other |

(continued on next page)
Designator | Component | Number | Cost/unit - SGD | Total cost - SGD | Source of materials | Material type
--- | --- | --- | --- | --- | --- | ---
30AWG Wire | 8-Wire Colored Insulated P/N B-30–1000 30AWG Wire Wrapping Cable | 1 | 10.58 | 10.58 | https://www.banggood.com/DANIU-250-M--8-Wire--Colored--Insulated--PN-B--30--1000-30AWG--Wire--Wrapping--Cable--Wrap--Reel--p--1081298.html | Other
Heat Shrink Tube | Heat Shrink Tubing | 12 | 0.01 | 0.13 | https://shopee.sg/530pcs-Heat-Shrink-Tubing-Insulation-Shrinkable-Assortment-i.19051557.248935763 | Other
Electronic Breadboard | Fa 400 Points Solderless Breadboard | 1 | 1.79 | 1.79 | https://shopee.sg/Fa-400-Points-Solderless-Breadboard-Breadboard-PB-Test-Board-1.70684265.1633983594 | Other
Arduino Jumper Wire | Arduino Jumper Wire for Breadboard (M–M) | 4 | 0.04 | 0.17 | https://shopee.sg/40-Pcs-Dupont-Cables-M-F-M-F-Jumper-Breadboard-Wire-GPIO-Ribbon-Pl-Arduino-no-i.16732896.1164631824 | Other
Jumper Wire | Jumper Wire | 7 | 0.01 | 0.10 | https://shopee.sg/Autor-560Pcs-Jumper-Wire-Kit-14-Lengths-Assorted-Preformed-Breadboard-FTO-i.988018.2456910708 | Other
M2 Screws | M2x4mm Stainless Steel Hexagon Socket Head Cap Screw | 17 | 0.04 | 0.68 | https://www.aliexpress.com/item/40005722727992.html | Metal
Pin Header | Single Row Break Away Pin Header | 4 | 0.04 | 0.15 | https://www.sgbotic.com/index.php?dispatch = products.view&product_id = 86 | Other

Costs in USD were converted from SGD (exchange rate: 1 SGD = 0.72 USD).
Total Cost: 70.88 SGD ≈ 51.03 USD

5. Build instructions

The main bulk of the pump was 3D printed using a digital light processing (DLP)/LCD printer. The rest of the components were purchased online (Bill of Materials). The additional tools required for fabrication and assembly included:

- M2 tap
- Metric allen key set
- Wire stripper
- Soldering iron and solder
- Sandpaper
- 1.5-mm and 2-mm drill bit
- Hand drill
- Vaseline or multipurpose grease

5.1. Hardware preparation

5.1.1. 3D Printing: Selection of photoresin and printer

For the 3D printing of the parts, we used two different types of photoresin. The choice of resin was vital to ensure sufficient strength and hardness of the cured resin. We first used the Hard-Tough resin by eSUN (Shenzhen, China, 62 USD/kg) with the shore hardness of 81D and the ultimate tensile strength of 55–60 MPa. We printed this resin using a low-cost (~ 300 USD), LCD-based printer (Phrozen Sonic Mini, Phrozen, Taiwan) (Fig. 6A–C). We also used an autoclavable resin (Dental SG, Formlabs, Massachusetts, United States, 340 USD/L). The parts consisting of this resin were used for applications involving cell culture. Dental SG was with the shore hardness of 67D and the ultimate tensile strength of 73 MPa. Dental SG was printed with Asiga Pico 2HD (Sydney, Australia) because it required higher light intensity than LCD-based printers. We empirically observed that DLP/LCD-based printers (e.g., Asiga Pico 2HD, Phrozen Sonic Mini) created the parts with higher print fidelity than the laser-scanning printers (e.g., Form 2 or Form 3) (Formlabs, Massachusetts, Massachusetts, United States).
Due to the availability and the cost of the DLP/LCP-based printers, the use of DLP/LCD-based printers was adequate for the demonstrated work.

5.1.2. 3D printing of modules

1. Orient all modules on the build plate as shown (Fig. 7). All modules were designed to be directly printed without support. **Note:** It is important to orient the parts as indicated to ensure the fidelity of printing. The features that required accuracies (e.g., through-holes) were oriented parallel to the Z-axis. Because of the printing mechanism of DLP printers, features oriented perpendicular to the Z-axis may result in low print fidelity due to inadvertent curing of the polymer [29]. 3D Printing of parts with support structures is also not recommended because support structures would affect the dimensional tolerance of the parts.

2. Set the XY-offset for the burn-in layer to 0.1 mm and Z layer height to 50 μm in the DLP printer.

3. Remove 3D-printed parts from the build plate using a scraper after printing.

4. Rinse the uncured resin in isopropyl alcohol (IPA). Use a syringe to flush uncured resin from hard-to-reach places (i.e., corners) if necessary.

5. Rinse it under running water to remove residual IPA.

6. Place 3D-printed parts in an oven at 60 °C for at least 30 min for post-curing.

7. Place 3D-printed parts under ultraviolet (UV) (405 nm) for 3 min for post-curing.

8. If necessary, use sandpaper (1000 grit) to sand down unwanted sharp edges.

9. If necessary, use a 1.5-mm or 2-mm drill to ream holes. M2 pilot holes were designed with a diameter of 1.5 mm.

10. Use an M2 tap to tap pilot holes. The M2 tap may be coupled to a hand drill to speed up the operation. Multi-purpose grease may be added to facilitate tapping. Parts #1, #6 and #7, #13, #14 required tapping. More details on holes to tap are illustrated (Fig. S1)

5.1.3. Preparation of PMMA breadboard

1. Laser-cut a 3-mm (thickness) clear PMMA sheet using the provided laser-cut file. The size of the PMMA breadboard may be customized by the user.

2. Tap the pilot holes on the PMMA sheet with a M2 tap. The M2 tap can be coupled to a hand drill to speed up the operation. A multi-purpose grease can be added to facilitate tapping.

![Fig. 6. A) Photograph of Variant B printed using low-cost resin (eSUN hard and tough resin) and low-cost LCD-based printer (Phrozen Mini Sonic). B, C) Time-lapse photographs of Variant B in operation.](image)

![Fig. 7. Illustration of the suggested orientation of the 3D-printed parts on the build plate.](image)
5.2. Assembly of hardware

5.2.1. Assembly of variant A

Secure Part #1 onto Part #19 (PMMA breadboard) (Fig. 9A).
Secure 2 × Part #2 onto Part #1 using M2 screws (Fig. 9B).
Place the desired number of silicone tubing (maximum of four) between the anchor points (Fig. 9C).
Assemble the fixture separately (Fig. 9D). Lubricate moving parts with Vaseline or multipurpose grease. If necessary, M2 screws can be added to secure Part #6 to Part #20 (Fig. 9D, white arrows).
Place the fixtured assembled in Step 4 over Part #1. Secure the fixture with M2 screws onto PMMA breadboard (Fig. 9E).
Wind both ends of the silicone tubing around the anchors (Fig. 9F, white arrow). A setup with 4 channels is also illustrated (Fig. 9G).
Drill two 1.5-mm holes on the 2-mL Eppendorf tube cap to allow tubings to go through, one for the inlet, one for the outlet. Secure Part #9 on the PMMA sheet with M2 screws and mount the Eppendorf tube on Part #9.

5.2.1. Assembly of Variant B

(1) Extract the shafts from a blunt tip dispensing needle (Part #22–25). The details are described (Fig. 10A-D).
(2) Use a pen knife to remove the plastic Luer adapter carefully (Fig. 10A-B). (Use pliers to hold the blunt tip needle when cutting with the pen knife for safety).
(3) Use a pen knife to gently scrape the epoxy remnants found on the metal needle (Fig. 10C-D).
(4) Repeat Step 1 for Part #25.
(5) Measure out 10 mm and use a pen knife to cut the tip of the plastic needle (Part #22) (Fig. 10E-G).
(6) Slot the plastic tip needle over the metal needle (Fig. 10H). The 14G plastic needle fit over the 18G stainless steel needle with little or no tolerance. The outer diameter (OD) of the 18G stainless steel needle (Part #23) was 1.25 mm, and the inner diameter (ID) of the 14G plastic needle (Part #22) was ~1.30 mm. Vaseline or multi-purpose grease was applied to lubricate the parts.
(7) The remaining steps for the assembly of Variant B are described (Fig. 8B).

5.2.3. Assembly of flowrate calibration tool

Use a pen knife to remove part of the plastic frame in the commercially available photo interrupter, GP1A57HRJ00F (Fig. 11A-B).
Solder the 30AWG wires onto the sensor. Use a heat gun to securely wrap the heat shrink tubes over the exposed wires (Fig. 11C). Repeat this to fabricate two sets of emitter-detector pairs. A lighter can be used as an alternative to a heat gun.
Solder the jumper wire headers to the other end of the 30AWG wires (Fig. 11D). Cover the exposed wires using heat shrink tubes.
Insert the sensor pairs into Part #28 (Fig. 11E). Slot the Tygon tubing (Part #26) between the emitter and detector.
Use the 23G blunt dispensing needle to interface between the Tygon tubing and silicone tubing. Remove the Luer adaptor as shown (Fig. 10A-D).

Fig. 8. Schematic of the exploded part-view: A) Variant A and B) Variant B. The numbers in the schematics represent the respective part numbers.
Fig. 9. Photographs illustrating the steps of the assembly. A) Photograph of Part #1 secured to Part #19 using M2 screws. B) Photograph of Part #2 secured to Part #1 with M2 screws. C) Photograph showing the placement of Part #17 between the anchors. D) Photograph of the assembled fixture consisting of Parts #4–6, #18, #20–21, #27. The white arrows demarcated the location of the optional M2 screws. E) Photograph of the fixture secured over Part #1 and Part #2 with M2 screws. F) Photograph demonstrating the winding of silicone tube around the anchors. G) Photograph showing a four-channel pump under operation. H) Photograph of Part #9 mounted on Part #19, securing an Eppendorf tube.

Fig. 10. A-D) Photographs illustrating the preparation of Parts #24 and 25. E-G) Photographs illustrating the preparation of Parts #22. H) Photograph showing the assembly of Parts #22 and #24.
5.3. Preparation of electronic components

1. Solder the jumper wires of Arduino to the latching push button using a soldering iron. Use heat shrink tubes to cover exposed wires (Fig. 12A).
2. Extend the wire connecting the motor to the Arduino board to ~1 m. Extend the wire by soldering additional 30AWG wires and subsequently use heat shrink tubes to cover the exposed wire (Fig. 12B). To connect to Arduino, solder the 30AWG wires onto pin headers. Use heat shrink tubes to cover exposed wires (Fig. 12C).
3. Connect the components as described (Fig. 13).

5.4. Programming of electronic platform

The code logic is illustrated (Fig. 14). The code runs on a state machine. It first prompts the user to input the desired rpm. To run the motor clockwise, push the red (forward) button. To run the motor counter-clockwise, push the reverse. To stop the motor from rotating, unlatch the forward or reverse buttons.

5.4.1. Programming the motor controller system

1. Extract “Peristaltic_Pump_Arduino_code.zip”.
2. Download and install Arduino IDE.
3. In Arduino IDE, navigate to Sketch > Include Library > Manage Libraries.
4. Install the libraries: “Adafruit_GFX.h” and “Adafruit_SSD1306.h”
5. Open Peristaltic_pump_Arduino_code.ino in Arduino IDE.
6. Connect Arduino Micro to the computer using a microUSB cable.
7. In Arduino IDE, navigate to Tools > Board. Select “Arduino Micro” (or respective Arduino platform)
8. Click “upload” and wait for the code to upload and wait for the “upload completed” message.
Fig. 13. A) Wiring schematic of the motor controller system. B) Photograph of wired motor controller system. C) Wiring schematic of the flowrate calibration tool. D) Photograph of the wired flowrate calibration tool.

Fig. 14. Diagram showing the program flow chart of the Arduino code.
5.4.2. Programming the flowrate calibration tool

1. Extract “Flowrate_calibration_tool_Arduino_code.zip”.
2. Open Arduino IDE.
3. Open “Flowrate_calibration_tool_Arduino_code.ino” in Arduino IDE.
4. Connect Arduino Micro to the computer using a microUSB cable.
5. In Arduino IDE, navigate to Tools > Board: Select “Arduino Micro” (or respective Arduino platform).
6. Click “upload” and wait for the code to upload and wait for the “upload completed” message.

6. Operation instructions

6.1. Operation of the pump

1. Input the desired speed by using the three pushbuttons (Fig. 15A). Each pushbutton toggles the digit as indicated by its respective red arrow. Pushing the left-most button once changes the number ‘0’ to ‘1′ (Fig. 15B). Pushing the same button again changes the number ‘1’ to ‘2’. The number returns to ‘0’ after reaching ‘9’.
2. Push either the red button or the green button after setting the desired speed. Push the red button if the desired direction of rotation is clockwise (Fig. 15C). Push the green button if the desired direction of rotation is counter-clockwise (Fig. 15D).
3. To stop the motor rotation, unlatch the red or green button.

6.2. Operation of flowrate calibration tool

It is advisable to calibrate the flowrate after each assembly as slight realignment between parts may alter the flowrate.

1. Connect one fluidic line from the pump to the flowrate calibration tool. Use clear water or water with blue dye for calibration.
2. Open Arduino IDE.
3. Connect the flowrate calibration tool to the computer using a USB cable.
4. In Arduino IDE, navigate to Tools > Serial Monitor
5. Ensure that the baud rate in the serial monitor is 9600.
6. Start the pump at the desired rpm.
7. The flowrate is displayed (in \( \text{L/s} \)) in the serial monitor when the water passes through the second sensor.

6.3. Operating instructions for Use inside incubator

The pump and the microfluidic device can be placed inside the incubator, while the electronic control board should be placed outside the incubator. Refer to the illustration for the operation of the pump with microfluidic devices that requires incubation (i.e., cell culture) (Fig. 16).

7. Validation and characterization

7.1. Validation of pump performance

To characterize the flowrate of the pump, we connected Variants A and B to the flowrate calibration tool and operated the pumps at different rotational speeds (Fig. 17A-B). We characterized each Variant fitted with different tubings (e.g., ID = 0.5 mm, 1 mm, and 2 mm). Before measuring the flowrate, we pipetted 10 \( \mu \text{L} \) of water in the Tygon tubing and measured the distance that 10 \( \mu \text{L} \) of water occupied within the tube. We found that 10 \( \mu \text{L} \) represented 43 mm inside the tube. We used

Fig. 15. A-B) Illustration showing the pushbuttons and the corresponding digits that they control. The pushbuttons were used to set the desired rpm. C) Illustrations showing the latching of the red button that commands the motor to move “forward” (clockwise). D) Illustrations showing the latching of the green button that commands the motor to move “reverse” (counter-clockwise). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)
this relationship to convert the measured distance over time (mm/s) to the volumetric flowrate (µL/min) in the Arduino code. We conducted the test three times to check for repeatability in the flowrate. We observed that the pumps showed good repeatability in all three runs (Fig. 17A). For Variant A, the lowest demonstrated flowrate was 0.05 µL/min when the pump was fitted with the silicone tube (ID = 0.5 mm) operated at 0.01 rpm of the motor speed (Fig. 17B). The highest flowrate was 727.3 µL/min when fitted with the silicone tube (ID = 2 mm) operated at 9 rpm of the motor speed. The trendlines of each combination of Variant and tube were summarized (Fig. 17A). Each trendline has the equation in the form of:

\[ y = mx \]

where \( x \) was the angular velocity of the motor (rpm), \( y \) was the volumetric flowrate (µL/min) and \( m \) is the constant. This equation allowed the user to correlate flowrate to the rpm (unit to input to the pump). For Variant B, the lowest demonstrated flowrate was 0.022 µL/min when fitted with the silicone tube (ID = 0.5 mm) operated at 0.01 rpm of the motor speed (Fig. 17B). The highest flowrate was 143.0 µL/min when fitted with the silicone tube (ID = 2 mm) operated at 9 rpm of the motor speed. To observe the flow profile of the pumps, we took timelapse images (at the interval of 0.1 s) and plotted a displacement volume over time graph of both variants when fitted with the silicone tube (with ID = 0.5 mm) operated at 8 rpm.

![Fig. 16. A, B) Illustrations showing the suggested placement of the pump and the electronic control board.](image)

![Fig. 17. A) Graph showing the relationship between the rotational speed and flowrate Variant A and Variant B incorporated with different tubing sizes (IDs = 0.5 mm, 1 mm, and 2 mm). B) Graph showing the relationship between the rotational speed and the flowrate for Variant A and Variant B operating at 0.01 rpm, 0.1 rpm and 1 rpm respectively. X and Y axes of the graph are in the log scale. C) Graph showing the relationship between the displacement volume against the time for Variant A operating at 8 rpm. D) Graph showing the relationship between the displacement volume against time for Variant B operating at 8 rpm. The red columns indicated when the fluids did not move.](image)
(Fig. 17B–C). At 8 rpm, the time required for each cycle (i.e., revolution) was 7.5 s. For Variant A, each cycle displaced ~6.3 μL (Fig. 17B, dashed line). For Variant B, each cycle displaced ~2.25 μL. We observed three periods (highlighted in red) in each cycle without fluid displacement. This intermittent plumbing was characteristic of the peristaltic pumps. The pause in Variant A accounted for 1.2 s per cycle (17% per cycle), and the pause in Variant B accounted for 2.1 s per cycle (28% per cycle). This intermittent plumbing should be taken into account when operating the pump at low flowrates.

One of the major technical challenges of peristaltic pumps is the variation in flowrate after each assembly. As shown by the error bars (Fig. 17A), when the tubes were not disassembled and refitted between runs, there was little variation in the flowrates between the operations. However, the variation in the flowrates was prominent after disassembling and refitting the tubing in place (i.e., removing Part #12 to replace the tubing). We found that the tension applied to the tubing during assembly can vary the volume of fluid displaced per cycle, influencing the flowrates (Fig. 18A). In extreme cases, when excessive tension was applied to the tubing during assembly, flowrate could reduce as much as 32%. The reduction in the flowrates can be attributed to the constriction of the tube diameter under tension. With the reduced tube diameter, the volume of the fluids displaced per cycle also decreased. While we do not advise the users to apply excessive tension to the tubing during assembly, some residual tension on the tubing was inevitable during assembly, which resulted in the variation of the flowrates. We measured the flowrate after disassembling and refitting the tubing for eight consecutive turns to characterize the magnitude of variation (Fig. 18B). The standard deviation of the eight runs was found to be 5%. This experiment suggested that it is advisable to calibrate the flowrate using the flowrate calibration tool after each assembly, especially when the application requires identifying the flowrates.

7.2. Validation of pump operating inside the incubator

To validate that the pump can operate for an extended period inside an incubator, we connected the pump to a microfluidic device and cultured mouse fibroblasts (NIH/3T3) cells for seven days (Fig. 19A). The pump was able to operate for seven
days continuously with no noticeable issues in performance. The 3T3 cells on Day 7 are shown (Fig. 19B). Additionally, we performed a live/dead assay of 3T3 cells within a microfluidic device (Fig. 20A). The microfluidic device was fabricated as previously described [30]. A fluorescent image and the quantitative data of the live/dead assay of 3T3 on Day 3 was summarized (Fig. 20B-C). These observations supported the viability of the cells by the perfusion culture using the developed pump. Overall, the continuous operation of the pump inside the incubator for up to seven days showed that our pumps were amenable for cell culture applications and suitable for use in LoC and OoC applications.

7.3. Wear and tear

In the developed pumps, no ball bearing was incorporated. Incorporating ball bearings would inadvertently increase the overall footprint of the pump. As such, we did not include any bearing in the developed pump. Wear and tear are susceptible in parts such as Parts #3 and #4 for Variant A and Parts #16 and #22 for Variant B. To observe the occurrence of the wear and tear, we compared Part #16 that was in constant operation for 60 days to the newly printed counterpart (Fig. 21). Fig. 21C suggested that dark spots appeared in Part #16 under constant operation (as highlighted by the red arrow), presumably due to wear and tear.

![Fig. 20. A) Photograph showing the setup of the microfluidic device connected with the pump inside of the incubator. B) Box plot summarizing the results of the quantitative live/dead assay of 3T3 inside a microfluidic device on Day 3. C) Micrograph image 3T3 cells inside the microfluidic device colored with live (green)/dead (red) stains on Day 3. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)](image)

![Fig. 21. A) Photograph of a newly printed Part #16. B) Micrograph showing the side-view of the through-hole in the newly printed Part #16. C) Photograph of a used Part #16 that was in operation for 60 days. D) Micrograph showing the side-view of the through-hole in Part #16 used for 60 days. Scale bar: 500 μm.](image)
to the degradation of the grease after long periods of operation. However, the optical micrographs showed little variation in the circumference of the hole. While we observed little evidence of substantial wear and tear that warranted replacing the parts after 60 days, we note that the highlighted observation is specific to the employed resin (Formlabs Dental SG). Nevertheless, we foresee that the modularity and 3D printability of the parts permit easy replacement of the parts when needed.

**CRediT authorship contribution statement**

**Terry Ching:** Conceptualization, Methodology, Investigation, Validation, Visualization, Writing – original draft, Writing – review & editing. **Jyothsna Vasudevan:** Investigation, Visualization. **Hshin Yin Tan:** Investigation, Validation, Writing – review & editing. **Chwee Teck Lim:** Supervision. **Javier Fernandez:** Conceptualization, Methodology, Funding acquisition, Supervision, Resources, Project administration, Writing – review & editing.

**Declaration of Competing Interest**

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Javier regularly collaborates with some of the most important international media outlets. His opinions and views on scientific issues and environmental policies have been, for example, covered in personal interviews, documentaries, and articles of National Geographic, BBC, PBS, FOX, Euronews, Discovery Channel, The Guardian, Huffington Post, Scientific American...

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