

3D-Printable Micropipette

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Abstract

Open source development of lab equipment is being facilitated, in part, by growing availability of 3D printing which allows the production of simple lab tools. We developed 3D-printable parts that, along with a disposable syringe, form a micropipette. Once assembled the pipette requires no calibration or validation with a scale. Our printed micropipette is assessed in comparison to a commercial pipette demonstrating comparable performance in accuracy and precision and approaches ISO standard.

Introduction

The open source development model, initially applied to software, is thriving in the development of open source scientific equipment due in part to increasing access of 3D printing [1,2]. Additive manufacturing methods have existed for decades although the recent availability of inexpensive desktop printers [?, 7] have made it feasible for consumers to design and print prototypes and even functional parts, and consumer goods [3]. Proliferation of free CAD software[OpenSCAD, Blender, SketchUp] and design sharing sites [?, 7–9] have also supported the growth and popularity of open designed parts and projects. Open design 3D-printable lab equipment is an attractive idea because, like open source software, it allows free access to technology that is otherwise inaccessible due to proprietary and/or financial barriers. Open design tools create opportunity for scientists and educational programs in remote or resource limited areas to participate with inexpensive and easy to make tools [1]. Open source development also enables the development of custom solutions to meet unique applications not met by commercial products that are shared freely and are user modifiable [3, 4]. Some advanced, noteworthy, open-source scientific equipment include a PCR device [5], and a two-photon microscope [6] although simple tools are often more impactful as they can serve a wider community. Some simple and clever printable parts that have emerged are ones that give a new function to a ubiquitous existing device, such as drill bit attachment designed to hold centrifuge tubes, allowing a drill to be used as a centrifuge [10]. Although this may make a rather crude centrifuge it may be an adequate method which only costs pennies compared to commercially available centrifuges. Other examples of open-design research tools that utilize 3D printed parts include optics equipment [11], microscopes [12, 13], syringe pumps [14], and reactionware [15] to name a few.

One example of a everyday scientific tool that provides opportunity for an open design solution is the micropipette. Micropipettes are an indispensable tool used routinely in lab tasks and can easily cost \$1000 USD for a set. Often a lab will require

several sets each for a dedicated task. Some pipettes are even re-calibrated for use with liquids of different properties. A open design pipette that can be made for cheap can cut costs for labs as well as allow a option for educational settings.

Air displacement pipettes use a piston operating principal to draw liquid into the pipette [?]. In a typical commercial pipette the piston is made to be gas-tight with a gasketed plunger inside a smooth barrel. Consumer grade fused deposition modeling (FDM) printers are unable to build a smooth surface do to the formation of ridges that occur as each layer is deposited. The ridges formed by FDM make it impractical to form a gas-tight seal between moving parts, even with a gasket. Existing printable open-design micropipettes get around this limitation by stretching a membrane over one end of a printed tube, which when pressed causes the displacement. The displacement membrane can be made from any elastic material such as a latex glove. There are a few open design printable micropipettes including a popular one which, in addition to the printed parts, uses parts scavenged from a retractable pen [16]. Because there is no built-in feature such as a readout for the user to set the displacement to a desired volume, this design requires the user to validate the volumes dispensed with a high precision scale. Without verification with a scale the volumes dispensed can only be estimated based on calculations of the deflection of the membrane.

We submit a new design whose major strength is the ability to adjust to any volume according to built in scale. Our open design 3D-printable micropipette works by actuating a disposable syringe to a user adjustable set-point. This allows the user to set the pipette to a volume by reading the graduations on the syringe barrel. The scale marks on the syringe can be used to adjust the dispensed volume accurately without calibrating with a scale. Our pipette also offers a simplified assembly requiring no glue, tape or permanent connections.

Design

Our printed pipette is designed to actuate a 1 mL or 3 mL syringe to a user set displacement. A syringe is used because the current state of FDM leaves ridges on the surface of printed parts making a airtight connection impossible. Micropipette designs use a piston operating principle to draw fluid into the device. Previous 3D printed designs have gotten around this limitation by using a flexible membrane to make an airtight tube. Then when the membrane is depressed the air is displaced allowing liquid to be drawn in. This allows a functional pipette to be created with very few easily sourced parts.

The core of the design is two printed parts, the body and the plunger which are able to be printed on a consumer grade FDM printer. A 1 mL BD syringe twists to lock in the body part and is held into place by the syringe flanges. The plunger part slides freely in the body part and actuates the syringe by pushing the thumb press. The pipette is spring loaded towards a set point which is adjustable by a set-screw. When the plunger is depressed the system locks when it reaches the latched position, where it is ready to draw in fluid. The plunger is held in place with a latching button design which is released with a lever drawing in fluid. The displacement is equal to the distance between the set position and the latched position. The pipette can also be pressed past the latched position to 'blow-out' the transfered fluid completely from the pipette tip. Additional materials required for assembly include a spring, a nut and bolt and washers. Attempts to make a printable luer lock adapter for tips was abandoned as the surface of printed parts is too rough to make an air tight seal with the luer or pipette tip. Instead a combination of a barbed luer adapter and elastic tubing was used to attach the pipette tips.

Our printed pipette mimics commercial pipettes's design, function, and user

Table 1. Parts list and cost to build the pipette

	Unit price	Source	Part number
Filiment	\$1.63	Makerbot	NA
1 mL Syringe	\$0.15	BD Biosciences	309628
3 mL Syringe	\$0.73	BD Biosciences	309657
Bolt	\$0.12	McMaster-Carr	91287A026
Nut	\$0.01	McMaster-Carr	90591A121
Spring	\$4.14	McMaster-Carr	94125K542
Washers	??	McMaster-Carr	??
Total	\$6.78		

Figure 1. CAD renderings of printable parts. Two parts are printed: The plunger part (A) slides inside the body part (B).

operation, making it intuitive to use. The button is pressed with the thumb until the latched position is reached. Then the tip immersed in fluid and drawn in by actuating the lever with the palm or fingers. Conveniently this design can also reach to the bottom of a 15 mL conical tube allowing tasks such as aspirating supernatant fluid from a cell pellet.

Adjusted Syringe Graduations

Our pipette uses the air-displacement design, where a pocket of air is used to draw liquid into the pipette. As air is an expansible fluid, this pocket of air expands due to the weight of the liquid pulling on it. Due to this effect the graduations on the syringe are not accurate, as they are designed for measuring liquid within the syringe. At larger volumes this effect is more pronounced resulting in the volume measured being greater than the amount of liquid pulled into the syringe. We remedied this by creating a new scale to account for the expansion. The scale is printed on a transparency sheet and is pasted on the syringe for accurate measurements.

Materials and Assembly

Validation

The printed pipette was validated and compared ISO 8655 and to a commercial pipette. The pipette was tested to see if the set point would drift between during use for example small, incremental movements of the screw or other parts causing the pipetted volume to drift. To test the pipette the same volume was pipetted 50 times without moving or re-adjusting the set point. A small amount of paraffin wax was applied to the screw to prevent it from slipping during each actuation. The printed pipette's accuracy and precision was characterized and compared to a commercial pipette. The printed

Figure 2. CAD renderings of assembled pipette and function. The pipetted actuates the syringe to three positions. (i) **The set position.** The position of the screw determines the total displacement. The pipette is spring loaded to return to this position (ii) **The latched position.** When the plunger is pressed the pipette locks at this position. The tip is then placed in a liquid and the button pressed to release the pipette back to the set position, drawing in liquid. (iii) **The blow-out position.** The fluid is transferred by pressing the plunger past the latched position to blow-out all the liquid.

Figure 3. Photos of pipette. The pipette is composed of two printed parts, a 1 mL syringe and some additional hardware.

pipette and a series of commercial pipettes were tested at volumes of 10 μ L, 20 μ L, 30 μ L, 50 μ L, 200 μ L, 300 μ L, 500 μ L and 1000 μ L. The printed pipette was adjusted to the the target volume by eye from the syringe graduations, and the resulting volume was measured and repeated 5 times to account for variability. Measurements were taken from the commercial pipette in a similar way.

Results and Discussion

Initially validation was preformed with the original graduations printed on the syringes. Due to the expansion of air the built in graduations are were not accurate, especially at larger volumes. Using our redesigned scale volumes were accurate. Our printed pipette meets ISO standards for accuracy and precision. (Table ??).

This pipette improves on existing open design pipettes in several ways. It requires only two printed parts, a syringe, and some hardware. It is able to reach into a 15 mL conical tube which is a routine requirement for cell culture. No permanent connections using tape or glue are required for assembly allowing worn out or broken part to be replaced easily. Assembly does not require a membrane that may wear or stretch and require tedious replacement. No validation or calibration is required. The pipette can be assembled and used to accurately pipette without first validating with a scale.

The only major limitation of this design compared to the briopipette is the option for a pipette tip ejector, although incorporation of a similar design in our pipette would not leave room for it to reach into a 15 mL conical tube. Our pipette requires a syringe and some additional hardware where briopipette's additional parts can be obtained virtually anywhere. The briopipette also allows step-wise shifts in displacement allowing the user to quickly change to a relative volume

Table 2. ISO 8655 for 100-1000 uL comparing a commercial pipette with our printed pipette used with existing 3 mL syringe scale and an adjusted scale

		Mean	Systematic Error	% Sys. err.	Random Error	% Rand. err.
1000 uL	ISO 8655, 100-1000 uL	1000	8.00	0.80	3.00	0.30
	Commercial Pipette	1002.98	2.98	0.30	1.72	0.17
	Printed Pipette	949.29	-50.71	-5.07	0.60	0.06
	Printed Pipette Scale	1003.57	3.57	0.36	0.89	0.09
500 uL	ISO 8655, 100-1000 uL	500	8.00	1.60	3.00	0.60
	Commercial Pipette	503.67	3.67	0.73	0.49	0.10
	Printed Pipette	475.99	-24.01	-4.80	4.75	1.00
	Printed Pipette Scale	503.62	3.62	0.72	1.64	0.33
200 uL	ISO 8655, 100-1000 uL	200	8.00	4.00	3.00	1.50
	Commercial Pipette	204.61	4.61	2.30	0.15	0.07
	Printed Pipette	186.55	-13.45	-6.72	1.31	0.70
	Printed Pipette Scale	201.87	1.87	0.94	1.47	0.73
100 uL	ISO 8655, 100-1000 uL	100	8.00	8.00	3.00	3.00
	Commercial Pipette	104.29	4.29	4.29	1.65	1.58
	Printed Pipette	94.02	-5.98	-5.98	4.81	5.12
	Printed Pipette Scale	101.00	1.00	1.00	1.05	1.04

Table 3. ISO 8655 for 30-300 uL comparing a commercial pipette with our printed pipette used with existing 3 mL syringe scale and an adjusted scale

		Mean	Systematic Error	% Sys. err.	Random Error	% Rand. err.
300 uL	ISO 8655, 30-300 uL	300	4.00	1.33	1.50	0.50
	Commercial Pipette	301.19	1.19	0.40	0.53	0.18
	Printed Pipette	286.91	-13.09	-4.36	0.42	0.15
	Printed Pipette Scale	299.11	-0.89	-0.30	0.48	0.16
200 uL	ISO 8655, 30-300 uL	200	4	2	1.5	0.75
	Commercial Pipette	200.06	0.06	0.03	0.46	0.23
	Printed Pipette	193.40	-6.60	-3.30	2.86	1.48
	Printed Pipette Scale	200.57	0.57	0.28	0.86	0.43
50 uL	ISO 8655, 30-300 uL	50	4	8	1.5	3
	Commercial Pipette	49.02	-0.98	-1.96	0.10	0.20
	Printed Pipette	49.62	-0.38	-0.76	1.26	2.53
	Printed Pipette Scale	48.73	-1.27	-2.54	1.11	2.27
30 uL	ISO 8655, 30-300 uL	30	4	13.3	1.5	5
	Commercial Pipette	29.08	-0.92	-3.06	0.09	0.31
	Printed Pipette	29.22	-0.78	-2.59	0.31	1.07
	Printed Pipette Scale	27.78	-2.22	-7.41	1.37	4.93
20 uL*	ISO 8655, 30-300 uL	20	4	20	1.5	7.5
	Commercial Pipette	NA	NA	NA	NA	NA
	Printed Pipette	18.70	-1.30	-6.48	0.38	2.01
	Printed Pipette Scale	17.94	-2.06	-10.29	1.87	10.42
10 uL*	ISO 8655, 30-300 uL	10	4	40	1.5	15
	Commercial Pipette	NA	NA	NA	NA	NA
	Printed Pipette	11.95	1.95	19.52	0.73	6.08
	Printed Pipette Scale	7.64	-2.36	-23.64	0.38	4.92

* The 20 uL and 10 uL volumes are out of the range in this case but we wanted to demonstrate that the pipette is capable of even smaller volumes.

For more information, see ??.

125

Supporting Information

126

References

1. Baden T, Chagas AM, Gage G, Marzullo T, Prieto-Godino LL, Euler T. Open Labware: 3-D Printing Your Own Lab Equipment. PLOS Biology. 2015;13(3):e1002086. Available from: <http://dx.plos.org/10.1371/journal.pbio.1002086>.
2. Pearce JM. Open-source Lab; 2014.
3. Fullerton JN, Frodsham GCM, Day RM. 3D printing for the many, not the few. Nature Biotechnology. 2014;32(11):1086–1087. Available from: <http://www.nature.com/doi/10.1038/nbt.3056>.

4. Pearce JM. Building Research Equipment with Free, Open-Source Hardware. *Science*. 2012 sep;337(6100):1303–1304. Available from: [citeulike-article-id:11241059\\$\\delimiter"026E30F\\$nhhttp://dx.doi.org/10.1126/science.1226328http://www.ncbi.nlm.nih.gov/pubmed/22984060http://www.sciencemag.org/cgi/doi/10.1126/science.1228183](http://dx.doi.org/10.1126/science.1226328).
5. Chai Biotechnologies Inc. Open PCR; 2016. Available from: <http://openpcr.org/>.
6. Rosenegger DG, Tran CHT, LeDue J, Zhou N, Gordon GR. A High Performance, Cost-Effective, Open-Source Microscope for Scanning Two-Photon Microscopy that Is Modular and Readily Adaptable. *PLoS ONE*. 2014;9(10):e110475. Available from: <http://dx.plos.org/10.1371/journal.pone.0110475>.
7. Makerbot Industries. Thingiverse; 2016. Available from: <https://www.thingiverse.com/>.
8. National Institutes of Health. NIH 3D Print Exchange. 2016;Available from: <http://3dprint.nih.gov/>.
9. GitHub Inc . GitHub. 2016;Available from: <https://github.com/>.
10. Garvey C. DremelFuge; 2009. Available from: <https://www.thingiverse.com/thing:1483>.
11. Zhang C, Anzalone NC, Faria RP, Pearce JM. Open-Source 3D-Printable Optics Equipment. *PLoS ONE*. 2013;8(3).
12. Baden T. Raspberry Pi Scope. 2014;Available from: <http://3dprint.nih.gov/discover/3dpx-000609>.
13. Walus K. A Fully Printable Microscope; 2014. Available from: <http://3dprint.nih.gov/discover/3dpx-000304>.
14. Wijnen B, Hunt EJ, Anzalone GC, Pearce JM. Open-Source Syringe Pump Library. *PLoS ONE*. 2014;9(9):e107216. Available from: <http://dx.plos.org/10.1371/journal.pone.0107216>.
15. Symes MD, Kitson PJ, Yan J, Richmond C, Cooper GJT, Bowman RW, et al. Integrated 3D-printed reactionware for chemical synthesis and analysis. *Nature Chemistry*. 2012;4(5):349–354. Available from: <http://eprints.gla.ac.uk/68744/>.
16. Baden T. Biopette: customisable, high precision pipette.; 2014. Available from: <http://www.thingiverse.com/thing:255519>.