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# Lab on a Chip



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# PAPER



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# An economical in-class sticker microfluidic activity develops student expertise in microscale physics and device manufacturing<sup>†</sup>

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Educating new students in miniaturization science remains challenging due to the non-intuitive behavior of microscale objects and specialized layer-by-layer assembly approaches. In our analysis of the existing literature, we noted that it remains difficult to have low cost activities that elicit deep learning. Furthermore, few activities have stated learning goals and measurements of effectiveness. To that end, we created a new educational activity that enables students to build and test microfluidic mixers, valves, and bubble generators in the classroom setting with inexpensive, widely-available materials. Although undergraduate and graduate engineering students are able to successfully construct the devices, our activity is unique in that the focus is not on successfully building and operating each device. Instead, it is to gain understanding about miniaturization science, device design, and construction so as to be able to do so independently. Our data show that the activity is appropriate for developing the conceptual understanding of graduate and advanced undergraduate students (n = 57), as well as makes a lasting impression on the students. We also report on observations related to student patterns of misunderstanding and how miniaturization science provides a unique opportunity for educational researchers to elicit and study misconceptions. More broadly, since this activity teaches participants a viable approach to creating microsystems and can be implemented in nearly any global setting, our work democratizes the education of miniaturization science. Noting the broad potential of point-of-care technologies in the global setting, such an activity could empower local experts to address their needs.

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# Introduction

Microfluidics embodies the convergence of knowledge from fields as broad as fluid dynamics, materials science, chemistry, and manufacturing. Those contributing fields are often conceptually challenging for students in their own right (*e.g.*, fluid dynamics<sup>1–5</sup> and manufacturing<sup>6,7</sup>). Perhaps unsurprisingly, their intersection in microfluidics amplifies

learning challenges as students must synthesize concepts from multiple fields along with new behaviours that emerge from miniaturization.<sup>1,4</sup> To aid learning, many creative approaches to microfluidic instruction and activities have emerged, especially those developed for true novices with limited technical background.<sup>8–10</sup> Frequently, however, the focus of the design and reporting of the resulting activities is on explaining the device technologies used in the activities.<sup>8</sup> While such details are important, creating credible learning activities and substantiating their broader adoption requires attention and documentation to a variety of aspects of the educational experience.<sup>11</sup> These include information on the environment, the learner, scheduling, educational strategies, and more that go beyond the devices themselves and the technology used to create them.

To that end, the guidelines for reporting evidence-based practice educational interventions and teaching  $(GREET)^{11}$  have been developed to help guide the analysis and communication of educational activities. GREET is content agnostic, focusing instead on information about a learning activity that is necessary for educators to evaluate and

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implement the activity in their own classroom.<sup>12</sup> As a parallel to experimental research, GREET gives structure to help evidence-focused educators to assess credibility, evidence, and reproducibility before relying on a publication. In the process of creating this paper, we intended to use GREET as the basis for reporting the learning activity that we created. In parallel, for ESI:† comparison table we also used GREET to analyse existing publications of microfluidic educational activities.<sup>13–26</sup> Doing so allowed us to identify three key gaps at the intersection of existing microfluidics education activities and the way those activities are reported.

First, in prior literature, we see strong parallels between the level of learning that an activity targets and the resource intensiveness of the activity. Among the published work in microfluidic learning, it remains difficult to balance device and required resources with advanced cost learning.<sup>14,15,17,19,22–24</sup> Less resource intensive activities tend to target less advanced learning. This is unfortunate for two reasons. First, it limits access to effective advanced miniaturization science education, especially in low resource settings where microsystems could be very impactful, for example in creating new point-of-care diagnostics.<sup>27</sup> Second, it tacitly links the knowledge and concepts of the field with resource intensive settings and technical complexity.

The second gap is that few existing publications link specific learning objectives and methods of assessment to provide evidence the learning outcomes have been achieved.<sup>13,14,16,17</sup> Such evidence is critical to supporting the credibility of new activities as achieving their intent, which is critical to others adopting them.<sup>28–30</sup> Many of the papers we reviewed do an excellent job in creating engaging activities that elicit a high level of learner satisfaction or enjoyment. Unfortunately, we observe that few papers in our review include evidence of learning, although most include evidence of student satisfaction. While satisfaction is important, explicit evidence of learning is critically important to improving the quality of microfluidics education by producing credible, generalizable, and clearly implemented educational activities.

The final gap we discovered was that few activities in our review identify a specific theory of learning, with no activities that we identified building on misconceptions as a specific theory of learning.<sup>31-33</sup> Misconceptions reflect instances where a student's mental schema for organizing knowledge is incorrect, which disrupts existing, and any future, understanding in non-obvious ways.<sup>33-35</sup> Chi & Roscoe<sup>32</sup> (pp. 3-4) provide a list of example misconceptions including "electrical current is stored inside a battery" and "coldness from ice flows into the water, making the water colder". In effect, they are a specific modality for how failures in conceptual understanding occur. Beyond being non-obvious to instructors, the central challenge with misconceptions is that they are often robust. That is, more or new information alone is likely to be assimilated into students' existing organizational schemas rather than inducing a change in organizational structure that properly accommodates it.<sup>33</sup> Therefore, attention must be paid to unlearning of existing schema prior to relearning a pre-requisite or new concept.

While misconceptions specific to miniaturization science are still being established,<sup>2-4</sup> many of the areas of knowledge upon which it builds have established research on students' misconceptions. These include transport processes,<sup>6,7</sup> fluid dynamics<sup>24</sup> (p. 172), and manufacturing<sup>25</sup> (p. 172), which suggests that misconceptions in miniaturization science are likely a challenge as students may retain existing misconceptions and rely on those misconceptions to organizing new, miniaturization-related, knowledge improperly. In addition, prior work has noted that many misconceptions share similar features, one of which is that the phenomena in question is typically invisible to the human eye<sup>33</sup> (p. 172). This feature of invisibility is nearly inherent in miniaturization science education.

These gaps in the existing microfluidic educational activities are not gaps in disciplinary knowledge, but instead represent gaps in reporting such activities, and a novel opportunity to improve the quality and impact of new microsystems learning publications. To that end, in this paper we report on a novel microfluidic learning activity in a way that shows how similar papers can address these three gaps. Specifically: (1) we use a low-resource, but technically advanced, method of device construction to support accessible deep learning about fundamental principles of device function. The layered thin film construction method we use is established in literature, does not require clean facilities, uses commercially available materials and tools, can be constructed by students, and can be used to demonstrate other types of devices in the future.<sup>14,36,37</sup> (2) We explicitly ground the activity in an appropriate theory of learning, explicitly identify learning outcomes, and apply instructional and assessment methods aligned with the outcomes and learning theory. (3) The instructional methods elicit and guide students in correcting misconceptions through hands-on activity and opportunities to correct errors. We assess student learning using a pre-post design based on known conceptual difficulties (e.g., diffusion and other emergent processes<sup>31,33</sup>) specific to the activity. Throughout this article we aim to provide a model for scholarly reporting of educational activities in microfluidics that can improve how others credibly evaluate and replicate microfluidics learning activities in their own classroom. We do so by using the GREET guidelines<sup>30</sup> to structure our reporting as well as including a ESI<sup>†</sup> (comparison table) summarizing each of the components of the activity using those guidelines.

# Learning activity

We use this section to describe the activity itself separately from how we studied its efficacy – including the intended audience, the underlying microfluidic device construction technology, the specific devices constructed in our activity, the theory of learning used in designing the activity, and the details of the activity itself. Additional information necessary

Lab on a Chip

to support implementation of the learning activity by others; including videos, instructions, materials lists, and handouts; can be found in the ESI.<sup>†</sup>

# Intended audience

The intended audience for this activity is an advanced undergraduate and/or graduate level engineering miniaturization course on microsystems theory, design, and manufacturing. The course is developed for early career Ph. D. students, but is cross listed for enrolment by master's and undergraduate students as well. We expect that students enrolled in the course will have completed, at minimum, typical required undergraduate engineering courses covering the concepts and mathematics of thermodynamics, fluid dynamics, and heat and mass transfer. We also expect most students to have some familiarity with the existence and uses of microsystems. We anticipate students will have a broad range of knowledge about the theory, function, and design of the types of microfluidics devices they interact with in the learning activity - from none to extensive. Student's goals and motivation for enrolling in the course vary. Some were enrolled in the course based on it aligning with their research area or program of study, others enrolled based on intrinsic personal interests or goals.

Because we are interested in an activity that is broadly useful, we considered the accessibility of resources as part of the criteria when designing the activity and establishing an appropriate student audience. That is, educational activities that necessitate extensive microfabrication resources limit who can implement them. While an increasing number of institutions have access to the tools, facilities, and staff needed to create microdevices, traditional photolithography manufacturing remains an expensive process.<sup>38</sup> World-wide, the majority of higher education institutions still do not have these types of facilities.<sup>39,40</sup> That lack of resources not only directly limits access to effective miniaturization science education,<sup>20</sup> it stands in direct opposition to needs in low resource settings that microsystems could address if there was greater awareness and functional knowledge (e.g., creating new, low-cost, shelf stable, point-of-care diagnostics).<sup>27</sup> Further, we perceive that increasing complexity of learning outcomes for a given activity often correlates with an increase in the sophistication of device technology used to teach it.8 Given that the available resources to implement such technologies are increasingly independent of the value of microdevice understanding, we considered it in establishing our intended audience. Specifically, we designed the activity and selected device technology so as to not require the intended audience to have significant existing resources for fabricating microdevices, but still create interaction between students and sophisticated microdevice technologies.

Our empirical studies are based on evaluating the learning activity's efficacy with both the intended audience and with an audience with lesser experience with pre-requisite materials. Doing so allows us to evaluate the activity's efficacy with its intended audience as well as the limits and effect of pre-requisite experience on learning through the activity.

# Theory of learning

As described in the introduction, we are particularly interested in *misconceptions*, a particular type of 'learning failure'.<sup>30–34</sup> Rather than a lack of knowledge, misconceptions are characterized by mis-categorization of knowledge.<sup>32</sup> By extension, a theory of learning appropriate to address misconceptions cannot begin from an assumption that a lack of knowledge is the problem and new information is the solution. The theory of learning established in research, and which we adopt in the learning activity, to address misconceptions is conceptual change.<sup>32,41,42</sup>

Conceptual change is a multi-stage process of learning that actively changes categorization of knowledge.<sup>1,32,41</sup> The process of conceptual change begins with an experience where students are likely to rely on an existing misconception and then struggle or make a mistake because of that reliance (*i.e.*, eliciting). Eliciting is followed by the student engaging in a reflective process of reflection, explanation of the error, and reformation of the conceptual understanding (i.e., a conceptual shift). If a specific misconception is known a priori, instructors can create activities that ask probing questions to elicit and correct them. Such activities often address both misconceptions that are known and misconceptions that are unknown to instructors. In any case, activities are most effective when they are designed to elicit misconceptions, make the resulting errors visible to the student, and guide students to independently construct a correct conceptual basis.

The nature of how student errors are treated in learning activities is specifically important to microfluidics education and a shift towards conceptual change. In our review of microfluidics learning activities (ESI:† comparison table), we note that the many existing publications explicitly or implicitly rely on an information transmission theory of learning. That theory of learning is seen in designing activities that centre on proper device function so as to demonstrate concepts through observation of certain fluid phenomena. For example, Gerber et al.,<sup>20</sup> uses the percentage of error free devices and the lack of need for correction as evidence for a construction method in an activity that sought to demonstrate the results of correct device function. Similarly, proposals for educationally appropriate methods focus on the fault tolerance and simplicity of constructing working devices.43 Additionally, Hemling et al.18 follow a trend in many such papers in providing premade devices and instructions for teachers meant to eliminate the potential for errors. From a technical standpoint, activities that are designed to ensure working devices are rational. From a learning perspective ensuring device function is only rational if the activity's theory of learning is information transmission-driven. In such a theory, correctness of

information is foundational, because the goal is to transmit correct information.<sup>42</sup> However, we know that students have misconceptions about these topics, and by extension such a learning theory will build on misconceptions rather than address them. Therefore, activities that ensure device function can reduce learning by reducing potential for the conceptual reformation necessary to achieve the learning needs of more advanced students.<sup>1,32</sup> Within our theory of learning, assembly errors, functional faults, error correction, and device construction are *beneficial*. That is because they represent components of the learning experience that can initiate cycles of reflective learning and conceptual change, especially when errors make misconceptions visible because they result from a students' activation of a misconception they rely on.

### Device technology

The device fabrication technology that we selected uses multiple laser cut dry film adhesive layers to construct microfluidic devices.<sup>36,44</sup> For further review of the technology, we suggest Delgado, et al.<sup>37</sup> as a fundamental resource for the capabilities and details of this technology. To assemble the devices, students simply peel off the backing layers from the double-sided tape layers and stack them together. While each layer must be carefully aligned to the previous layer to ensure proper device operation, the transparent nature of each layer greatly simplifies this process and subsequent troubleshooting. To aid in alignment, the tape layers and PDMS fluidic interface layer had the same outline, such that students could also align the outside edges of each layer if preferred. In more complex multilayer devices, alignment squares have been added as markers to further guide students for proper orientation of the tape layers.

One possible concern was the high adhesive strength of the tape. While it facilitates bonding and prevents leaking between device layers, we anticipated that each layer could only be applied once and could not be removed if misaligned. In anticipation of mis-assembled devices, extra kits for each device were available for each team. We also created fully assembled devices, which could be reviewed by each team at any time, and later used if assembly of their device was not successful.

#### Justification for this technology

When selecting a device technology, we considered three aspects to be critical to meeting the needs of our intended audience and achieving the learning goals we intended. Those three primary aspects were the technical sophistication experienced by students, the resource and financial accessibility, and the level of manual interaction and potential for troubleshooting by students.

First, the types of devices that can be constructed invoke more advanced concepts than most prior methods. Using this construction technology, our activity adds new concepts on layer-by-layer assembly of 3-dimensional devices, mechanical motion (valves), and splitting flows to enhance diffusive mixing. Specifically, prior research shows many functional devices that can be created using thin film layered plastics including devices for cell culture, integration with optical biochips, and electrochemical biosensors.<sup>45–47</sup> Given our intended audience, the ability to create behaviours based on advanced fluidic concepts is critical to building on pre-requisite knowledge. Further, the ability to have such phenomena occur at a human visible scale provides opportunities to address misconceptions that have been retained from introductory fluids courses or developed in initial learning about microdevices.

Second, the device construction technology is highly accessible. Here we define accessible for microfluidics learning activities as minimizing both incremental cost and necessary resources without reducing the sophistication of device construction and function. That compromise remains a challenge in accessing microchips and microdevices suitable for our audience.<sup>28,42</sup> This technology is part of a suite of advances in xerography,<sup>36,48</sup> laser-cutting adhesive tapes,<sup>46,49,50</sup> and 3D printing<sup>51-53</sup> that have the potential to enable microsystems to be created with accessible, inexpensive materials in nearly any environment. This technology uses off the shelf materials (Table S1<sup>†</sup>) and requires minimal advanced capacities. Laser cutting the individual layers, which can be outsourced to many commercial services, is the most resource intensive part of device construction and outsourcing does not diminish learning. No cleanroom facilities are required to use this technology for learning. Notably, however, the technology does retain the layer by layer construction techniques that are common in more advanced micro fabrication - enabling students to gain experience with core challenges in manufacturing sophisticated microscale devices such as layer alignment and interfacing.

Third was the ability for students to physically touch the devices and actively manipulate parts during the assembly process. Because the layers of the device are macro scale, even if the functional elements are micro scale, this technology allowed students to directly perform device assembly. Doing so removes several layers of abstraction as opposed to more automated approaches for creating micro devices. We see the difference as similar to learning subtractive manufacturing using a manual 3-axis knee mill as opposed to learning by programming a CNC machine in CAM software. Further, the physical interaction further benefited by being construction-fault tolerant. That is, errors can often be observed and addressed during device construction or testing - which we did not initially believe to be the case with this device technology. We see this as both a resource benefit and particularly beneficial given our theory of learning. This was an aspect we had not considered during our initial planning, but now see as critical. As will be discussed in the study results, both students and instructors noticed that this (1) ensured the ability of students to create functional devices and (2) actively created learning when

# Devices

Using the multi-layered dry film device technology, we designed four devices for students to build in a learning activity. We chose the devices for students to build specifically because they emphasize concepts where microfluidics phenomena fundamentally differ from macrofluidic behaviour. For example, diffusive mixing under laminar flow as opposed to turbulent mixing and flow. Changes in mixing mechanisms in particular represents a shift where we expect students to likely have existing macrofluidic understanding that is at risk of inducing misconceptions about microfluidics. We also selected for devices that necessitate engagement with both practical and advanced fluidic concepts including manufacturing tolerances, mechanical motion (valves), and techniques to amplify basic concepts - e.g., splitting flows to enhance diffusive mixing.

The four devices that we selected for students to build and test include a T-mixer (Fig. 1A and G), a droplet generator (Fig. 1B), a multi-layered F-mixer (Fig. 1D and H), and a microfluidic valve (Fig. 1C). These four devices were selected because they have varying complexity in terms of the number of layers, required precision, and principle of operation. Importantly, the functional principles of these devices are well established and similar devices have been previously reported using this and many other device construction technologies.54-57 The specific dimensions and details of each device were created by the course instructor in advance, however it is possible for instructors to modify the activity to have students create their own devices or design parameters. The devices were sized such that they all experienced key phenomena associated with microscale structures, but remained large enough for function and flow to be seen with the naked eye. While the devices were scaled up for the students to see the phenomena on their own, the materials and complete set up remained compact enough to be encapsulated within a portable box and perform the activity on the footprint of a standard classroom desk (Fig. 1E and F ).

#### **Classroom implementation**

The classroom activity we created involves both constructing and testing all four devices described above. We describe the activity here as it was implemented in a graduate student course on microsystems. The activity occurs over three (initially two on the first offering) 75 minute class sessions, described in detail below. Students completed a pre-



**Fig. 1** Devices, kit components, test equipment, and examples of testing for four layered microfluidic devices. A–D) Four different devices of increasing complexity using the same device technology (note: the white scale bar at the bottom of A–D is 1 cm) E) a flat lay of all of the components included within the kit. F) Test equipment highlighting use of multiple colours and varying fluid properties to induce observable device function. G and H) demonstration devices being tested by authors.

assessment during the first activity day prior to beginning construction and a post assessment on the third activity day after completing testing. Finally, one week after the activity, students completed a reflection about what they had learned and observed. While primarily used here to evaluate the learning efficacy of our activity, we strongly encourage other instructors to keep the assessments and reflection. To adjust for available time, schedule, and focus, interested instructors can easily combine the three parts of the activity, change between teams or individuals, or adjust the number of devices created to fit their particular instructional needs. The necessary materials to replicate this activity; including an instructor guide, student handouts, bill of materials, assessments, and tips and tricks for faculty; appear in the ESI.<sup>†</sup> The specific learning objectives (LOs) that we established for students participating in the activity were:

LO.1) Apply theoretical knowledge about device function to physically construct and test devices.

LO.2) Experience, reflect, and refine their individual conceptions of macro and microfluidic differences.

LO.3) Evaluate failure modes from device manufacturing discussed theoretically in class.

LO.4) Link fundamental concepts of microfluidics to each device's physical function.

In the activity, we situate the instructor as a support person that teams can use to assist in live problem solving of realistic problems they encounter in device construction and testing. Students receive materials and support that reinforced general device principles without proscriptive information on how to handle all eventualities - they are expected to lead the implementation rather than be instructed in its step by step application. The instructor primarily answers questions, often with questions, or asks questions based on observations of teams' work. Materials are designed to emphasize parallel skills such as troubleshooting, key physical concepts, and translating 2-dimensional drawings of device layers into 3-dimensional final shapes that are not directly addressed in didactic instruction. This role provides greater opportunities for students' misconceptions to become apparent to them, which is necessary to confront and manage conceptual change through experimentation and failures (e.g., misassembled devices, misconnected fluidic ports). In doing so we aimed to specifically engage the processes that support conceptual change described previously.32

The first day of the implementation was focused on device construction. Students self-organized into two- to four-person teams and were provided with kits that included all parts and materials needed to construct devices. Students were also provided with handouts that included basic instructions and pictures on how to assemble each device (Fig. 2). The handouts suggested constructing the devices in the order of increasing complexity and layer count (i.e., T-mixer [3 layers], droplet generator [4 layers], F-mixer (4 layers), and valve [5 layers]). As expected, students encountered known challenges with this device technology including alignment, orientation, and the one-time nature of applying layers. Prior literature often identifies challenges in device assembly as obstructive to carrying out such activities and to be avoided. As noted, we treat such challenges differently - as opportunities to experience and learn about how practical details affect device construction and function. In anticipation of misassembled devices, extra kits for each device were available



**Fig. 2** Example step-by-step visual instructions provided to teams. A) All step by step instructions begin by showing all the key layers of the device followed by B–H) visual instructions of general assembly steps and finishing with I) a final top down view of the device.

for each team. We also provided correctly assembled devices, that teams could observe at any time and use on day two or three if assembly of their device was not successful.

On the second day of the activity, the groups start testing the devices they created using syringes, ancillary tubing, mineral oil, and water with food safe dyes added. Groups were encouraged, but not required, to test their devices in a different order than assembled. We specifically suggested testing the two mixers back to back to facilitate visual functional comparisons. Students were free to use either gravity fed fluid flow or hand actuated syringes for the mixing devices. To induce gravity fed flow, students could either hold an open syringe partially filled with water above the device, or place the device in a ring stand. The coloured dyes aided in visualizing the flow of fluids, especially as they mixed (see Videos S1 and S2<sup>†</sup>). Students found that hand actuation of syringes worked best for the droplet generator (Video S3<sup>†</sup>) and valve (Video S4<sup>†</sup>). One strength of the device technology in this use case is that fluid connections are interference based. A hole in the PDMS layer is slightly undersized relative to the tubing, and students push the tubing into the holes to create a sealed connection. This enables students to easily change fluidic connections, which proved to be useful when students needed multiple attempts to find the correct fluidic configuration for the more advanced devices, such as the droplet generator.

In the first offering of the course, we budgeted two days for the activity, but received feedback asking for more time as some students felt rushed. In the second offering of the activity, we added an extra day. We still recommended that students begin with construction, then begin testing, but the extra day allowed them to devote more time to each portion. On the third day, teams were also free to explore concepts and ideas of interest related to the microsystems.

# Study design

In developing and sharing a new intervention, we also designed studies to measure whether the activity meaningfully contributes to the learning goals we described above for our target population. Gathering such data is in keeping with increasing calls throughout STEM education for evidence-based interventions and researcher efforts to validate claims about learning.<sup>42</sup> Towards that goal, we undertook studies on varying participant cohorts to address the following research questions (RQ):

RQ.1) Can undergraduate students consistently and accurately construct the layered microfluidics devices used in this learning activity?

RQ.2) How much learning do students with minimal to no prior education in microfluidics experience when completing our microfluidics fabrication activity?

RQ.3) How much learning do students with ongoing microfluidics education experience when completing our microfluidics fabrication activity?

RQ.4) What insights about the process of learning about scaling phenomena can be observed in student self-reflections and instructor observations on our microfluidics fabrication activity?

The research questions align with our learning objectives, but not in a one-to-one way (i.e., RQ.1 is not specific to LO.1). The first research question is a precursor to all four LO's - specifically can participants generate working devices with minimal direction (i.e., through knowledge, inquiry, and troubleshooting) during the activity. This RQ is a foundation for the conceptual learning the activity targets and demonstrates the activity is feasible. RQs 2 and 3 are specifically aligned with LOs 1 and 2. They address whether participants with less than (RQ.2) or the expected (RQ.3) amount of pre-requisite education achieve the intended conceptual learning about microfluidics. This helps inform the application of the activity to students with different backgrounds. RQ.4 goes beyond our specified LOs and looks at what other insights about the learning process that we can use to improve the activity or drive future research. As good educational practice, we see leveraging student's self-reflections and the instructor's observations to better understand challenges and misconceptions associated with microscale phenomena as an organic opportunity to better understand how the activity works (or doesn't) and not just whether or not it does. We used two data collection efforts to answer these questions. The first effort was performed in a microsystems course while the second was carried out with a general undergraduate population. Both data collection efforts operated under protocols approved by Georgia Tech's Institutional Review Board (IRB) (Protocol H22089). Because the two efforts used similar methods and measures, we describe them concurrently in each section.

# Participants

Both studies occurred in the Wallace H. Coulter Department of Biomedical Engineering at Georgia Institute of Technology and Emory University. Although a single department, biomedical engineering is part of both universities. It is part of the College of Engineering at Georgia Institute of Technology and is part of the School of Medicine at Emory University. Both institutions are classified as very high activity, doctoral granting, selective research more undergraduate admission universities. Our data were collected in the spring of 2022, fall of 2022, and spring of 2023. As noted, we collected data from two participant groups, both of whom reflect the intended audience of the activity - albeit in slightly different ways.

The first group of participants were undergraduates that were not enrolled in the course containing the in-class activity. These students reflect the lower boundary of prerequisite knowledge we anticipated would be necessary to learn effectively from the activity. That is, some classroom training in the conceptual, practical, and mathematical concepts of macroscale thermal fluids phenomena with the possibility, but low likelihood, that they have other experience via research. The participants were recruited via emails sent to students in the pre-requisite class as well as a broader population of biomedical and other engineering students. In total, 17 individuals from this recruitment method responded and completed the study. Of those who participated, 8 (47%) had completed a course in scaling phenomena, 1 (6%) was currently enrolled in such a course, and 8 (47%) had no formal education in microfluidics or scaling phenomena. The participants who completed this study received a \$10 gift card for their participation. They participated in a version of the learning activity that reduced the breadth of devices from four to two. They received an 18 minute video introducing microfluidics concepts relevant to all four devices prior to the study session. In the study session, they completed a reduced scope version of the activity - constructing the two mixing devices, testing them, and then testing two additional demonstration devices over the course of a single, approximately one hour, session. Our goal with the first group was to understand device fabrication success and failure rates (RQ.1) as well as how prior fluids education impacted learning (RQ.2).

The second group of participants consisted of students that enrolled in a graduate level course on microsystems in the university's biomedical engineering department during the spring of 2022 and spring of 2023. The spring 2022 course enrolment was 11, and consisted of 9 graduate students and 2 undergraduate students. The spring 2023 course enrolment was 33, and consisted of 11 graduate students and 22 undergraduate students. Nearly all of the students consented (11 and 29) to their data being used in

the study using typical IRB consent procedures. Of these students, 34 had no prior experience with microdevices, 3 had 0-1 years of research experience with microdevices, and 7 had more than 1 year of research experience with microdevices. We assume based on course and degree prerequisites that these students have completed basic course work in fluid phenomena. During the first three weeks of class, the course covered the following via lectures with worksheets to support structured note taking: an introduction to biomedical microsystems; scaling laws; photolithography; soft lithography; surface micromachining; microcontact printing of proteins; as well as the construction and use of multilayer microfluidics. In the fourth and fifth week of the course, students participated in the full scope of the learning activity. The goal with the second group was to more fully characterize the impact of prior education and experience on microfluidics with the intended audience (RQ.3), as these students participated in coursework prior to the activity.

#### Learning data collection

Data on the learning effectiveness of the activity was collected using the same short answer knowledge test for both groups. The microdevice knowledge test was composed of 9 open ended short answer questions with topics related to device physics and manufacturing (ESI<sup>+</sup>). The physics questions were conceptual and related to scaling, operation of microfluidic valves, fluid mixing at the microscale, droplet creation, and the effects of air bubbles in microfluidics. The manufacturing questions were designed to link device fabrication and design (e.g., draw cross section and top views of a mixer) and included asking students to describe the role of tolerances in microdevice manufacturing. Hence, the knowledge test included questions from all learning goals. In addition to providing answers, participants were asked to rate their confidence in their answers on a scale of 1 (least) to 5 (most confident). When the activity was performed in the graduate course, two of the authors (the course instructor and graduate research assistant) scored the assessments. For the data collected outside of the course, the assessments were scored by the graduate research assistant only. Each question was scored on a scale of 0 to 10, with a maximum score of 90. For both groups, the same assessment was administered immediately before and after the activity.

The participants in the graduate course also completed a guided reflection (RQ.3, RQ.4). Students were asked: 1) which devices did you understand better? 2) Which devices are you more confused about? 3) What are three things that you learned that you found most important? 4) How did your understanding of microfluidic mixing change? The reflection also included a prompt asking students to identify what they learned from the activity in their own words. The participants in the course completed it 1 week after the testing portion of the activity.

#### Data analysis

To ease the comparison of data, we reported confidence weighted scores. Specifically, the score for each question (0-10) was multiplied by the student's confidence in their answer (1-5). Confidence weighting helps to quickly identify student mastery of a concept, which can be defined as both a correct answer and the recognition that the answer is correct. Lower scores are associated with a confident incorrect answer (misconception) or a lack of knowledge (both confident and uncertain).<sup>58</sup> For completeness, the supplement contains figures showing individual student score and confidence changes and changes in test score questions and confidence (Fig. S2 and S3<sup>†</sup>). All data on student scores and confidence were analysed with OriginPro (OriginLab Corp). Changes in confidence weighted scores, test scores, and confidence were tested at the 0.001 significance level using the Wilcoxon signed-rank test.

# Results

# Participants consistently and accurately assembled multilayer devices (RQ.1)

As the activity was conceived of by microdevice experts, one concern was whether the techniques employed could be performed by untrained engineering students. To that end, we observed participants while they constructed the devices, and measured: 1) the success rates (only partial activity participants and not part of the course), 2) the time needed to construct the devices (only partial activity participants), and 3) the degree of misalignment (if any) in the final devices (partial activity participants and second course offering). The participants completing the partial activity had no prior training in microfluidics and therefore were naïve to the field of microfluidics in general. Notably, participants completing the partial activity were free to ask more questions and receive immediate feedback, enabling more frequent "checkins" on device assembly. The majority of participants enrolled in the microfluidics course also had very little microfluidics experience, although they had already received several weeks of instruction, and 16% had prior experience with microfluidics (see Participants section). The participants in the course completed the activity in a traditional classroom setting with instructors monitoring the class as a whole and performing periodic "check-ins" with the groups of students to address questions. For comparison, we also asked 5 individuals from our research laboratory to build three devices each, as they had prior experience constructing tapebased microfluidics, a sub-specialty of the field of microfluidics.

Our results show (Fig. 3) that both experts and novices had similar performance when constructing the devices. It was much easier to record the number of tries that it took for the students to create a functional device in the partial activity (n = 11) given the low student to instructor ratio. All five experts had working devices on the first try, while 83% of the devices built by partial activity participant novices



**Fig. 3** Observations of participants completing activity and quantitative measurements of student ability to construct devices. Different participants preferred to build the microfluidics A) with and B) without tweezers. C) All materials were included in a kit for students to complete D) some participants also elected to use gravity fed flow instead of syringes. E) All participants successfully constructed devise within 2 tries, 83% were successful on the first attempt (n = 11). F) Participants took longer to complete the devices working alone as compared to pairs (n = 11). G) The average *x*-translational and *y*-translational errors between partial and full study participants (novices, n = 15) and experts (5 experts, 3 devices each; n = 15) in microfluidic construction were similar. H) The rotational misalignment between experts (5 experts, 3 devices each; n = 15) in microfluidic construction and novice partial and full study participants (n = 15) were similar. Data for e and f are only from participants completing the partial activity.

resulted in a successful device on the first attempt and 17% of the devices worked on the second attempt. On average it took 8.5 minutes for novice partial activity participants to build both microfluidic mixers, and 6.9 minutes to build both in pairs. Translational errors were recorded for not only the partial activity participants, but also for second class cohort (total, n = 15) as it was simple to take a picture of the F-mixer created by each group. The x- and y- translational error for novices averaged 0.189 and 0.148 mm, respectively. The average x- and y- translational errors for the expert group was 0.097 and 0.187 mm, respectively. The average translational errors for both groups were much less than the maximum allowable misalignment of 1 mm. The average rotational misalignment for the experts and novices were 0.318° and 0.411°. Once again, for both groups the misalignment was less than the maximum allowable error of 1.6°. With this in mind, none of the devices that did not work on the first try were due to misalignment. They were instead due to placing the layers incorrectly (*i.e.* switching the order of the layers).

We would like to note one outlier condition in our partial activity participants that was difficult to quantify. One student in the partial activity group began the activity in a group of three, and decided to split off and build the microfluidic alone. This participant had difficulty manipulating and using the tape layers and spent approximately 22 minutes building a partial device. At this point, the individual elected to re-join the team, and was able to successfully complete the remainder of the activity, within the total 1 hour allotted time limit. In the classroom setting, some additional errors did occur, owing to the larger sample size and also less instructor attention due to the student-to-instructor ratio. The most common challenges associated with assembling the devices were correctly orienting and aligning the layers, and ensuring that the layers were completely flat and able to provide a water-tight seal. While we were not able to quantify the exact number of initial failures, all teams that had a non-working device on their first attempt were able to successfully construct a working device on the second attempt. Hence, taken in total, there was a 100% success rate among the 44 students who participated in the class activity.

# Confidence weighted knowledge test scores were significantly higher after the activity (RQ 2 & 3)

After completing the full activity, participants displayed significantly increased, high, confidence-weighted scores related to microdevice physics (Fig. 4A and B) and microdevice manufacturing (Fig. 5A and B). This data shows that participants experience measurable increases in their understanding of microsystems physics and manufacturing by completing the activity. That is despite a meaningful number of students achieving perfect scores (50 out of 50) on the post test, creating a measurement ceiling, as well as several students scoring zeros on the pre-test, creating a measurement floor. Students completing the partial activity also displayed measurable, statistically significant increases in their learning, albeit with a statistically significant lower



**Fig. 4** Participants experienced significant increases in confidence weighted physics scores after completing the full activity, while conceptual understanding remained limited for participants in the partial activity. Questions for the assessment included both concepts from lecture as well as those outside of lecture learned from completing the activity. For participants in the partial activity, an 18 minute video prior to the activity was substituted for the lectures. A) The graduate participants in the full activity showed significant changes in the scores among all topics. B) The same can be said for the undergraduate participants only exhibited statistically significant learning in valve and mixing concepts. In each box and whisker plot, the boxes represent 25–75% of the scores with a median line, and the error bars indicate the minimum and maximum scores.

magnitude than the full-activity participants. A one-way analysis of variance (ANOVA) revealed that there was no statistical difference between post-assessment expertise between physics and manufacturing related questions,



**Fig. 5** Participants experienced significant increases in confidence weighted manufacturing scores after completing the full activity, while limited significant learning was shown for those in the partial activity. A) Graduate participants had significant learning from pre- to post-assessment scores when completing the full activity. B) Undergraduate participants in the full activity had a similar cross category significant increase in learning, but higher manufacturing pre-assessment scores in comparison to the physics pre-assessment scores. C) Participants in the shortened activity had manufacturing pre-assessment scores that were lower on average as compared to physics-based questions. After completing the activity, post-assessment scores indicated mastery of these concepts. In each box and whisker plot, the boxes represent 25–75% of the scores with a median line, and the error bars indicate the minimum and maximum scores.

suggesting that the activity promoted similarly high levels of knowledge and confidence across categories. In comparison, the participants who completed the partial activity showed a significant increase in device physics scores related to valves

and mixing, with no significant changes in the categories relating to scaling, droplets, and air bubbles (Fig. 4C). With regards to manufacturing, participants completing the partial activity showed significant increases in the mixer and valve manufacturing, with no significant change in tolerances or droplet generation (Fig. 5C).

The pre-assessment knowledge test helped establish both the knowledge level and confidence of the participants from concepts covered in the lecture as well as concepts that they may have encountered in their own research. There was a significant spread in the participant's confidence weighted pre-assessment scores that appeared for both new concepts and for concepts covered in the course for the participants in the class (Fig. 4A and B; Fig. 5A and B). A two-way ANOVA showed that participants enrolled in a graduate degree taking the course had significantly higher scores in physics when compared to their undergraduate counterparts in the class. This can likely be attributed to the graduate participant's prior exposure to physics related concepts in their past education and research training. The two-way ANOVA showed no statistical difference between the manufacturing and physics pre-assessment scores between the undergraduate populations who were enrolled in the course and those who had received only a brief 18-minute long video prior to the activity. Hence, despite not having had the full classroom experience, the undergraduates with the partial activity had a similar performance. This somewhat surprising result supports the idea that lecture-only based education, even when presented in an active format with levity, frequent questions from the instructor, note-taking, and interesting video visuals still has limitations in building expertise. It is interesting to note that learning in the partial activity did not always align to the introductory video. For example, scaling was discussed in the partial-activity introductory video, yet there was not a significant change in learning (Fig. 4C). Overall, the pre-assessment data also showcased the variability in the student's knowledge, with a moderate degree of understanding in some areas, such as air bubbles and scaling, but lower expertise in mixing and droplets Fig. 4.

We also examined individual performance of the individuals in the activity as stratified by the participant group (Fig. S1†). These results largely matched the results that would be expected as presented above. Every individual saw an increase in their score after the activity, and statistically significant, larger increases were experienced by participants in the full activity. When considering the score of the entire assessment, there was not a statistically significant difference between pre-assessment scores among the groups.

#### Participant reflections indicate activity supports individualized development of mastery (RQ 2 & 3)

The guided open-ended short answer reflection enables participants to self-identify what aspects of the activity were



**Fig. 6** Responses (n = 40) to open-ended short answer questions asked 1 week after the activity suggest that the activity was memorable and helps develop mastery. A) Most participants reported a deeper understanding of all the devices built. B) Most participants were not more confused about devices, however 7.5% reported more confusion on the droplet generator, 5% reported more confusion on the valve, and 2.5% reported more confusion on the F-mixer. C) The diversity of comments shows that each participant valued different aspects of the activity. This suggests the activity supports individualized development of mastery that helps fill key knowledge gap.

the most memorable for them. Over 60% of participants commented on understanding all four devices better after the activity (Fig. 6A), whereas 80% of participants commented on an improved understanding of the F-mixer. This may be related to the F-mixer's high geometric complexity and nonintuitive physical behaviour due to the split and recombined flows for mixing.

In effort to gather feedback on opportunities for improvement, especially related to participant learning and clarity of concepts, our guided reflection included a prompt in which participants could comment on whether the activity increased their confusion of specific devices. 7.5% of participants reported additional confusion on the droplet generator, 5% on the valve, and 2.5% on the F-mixer (Fig. 6B). One learner commented that they were more confused about traditional fabrication approaches. We interpreted this comment to relate to prior class lectures that preceded the activity related to the construction of microsystems using lithography, thin-film deposition techniques, and etching. Although the exact reason for this confusion is unknown, our theory of learning attempts to make misconceptions visible to students. Therefore, we see it as unsurprising that the activity may have elicited misunderstandings of traditional fabrication methods – even if that knowledge was not directly relevant to the activity.

While our primary evidence of learning comes from the test scores, participants' comments explicitly link the activity to our intended learning. This is useful in part because the size of course made it unfeasible to use a control group who did not complete the activity. Comments from participants identify that key aspects of the activity support improved understanding of microsystem function, device construction, and increased conceptual mastery. The participant responses directly after the activity and 1 week later are available in Tables S2 and S3,† respectively. The lead author also grouped the comments by category (Fig. 6C). The categories were evaluated by a second author, and both authors met to finalize each grouping. The most frequently occurring categories included concepts of device operation, concepts of device physics, device design, and troubleshooting. We see one participant's stated comment as both typical and relevant - "[I] did not realize microfluids are always more laminar", showcasing a correction of a prior misconception (Table S2<sup>†</sup>). In addition to conceptual learning, comments (Table S3<sup>†</sup>) also suggested an increased understanding of the societal need for microdevices. Some students simply noted a more positive experience with microdevices, as one student stated "I like microfluidics". We note that responses were similar in both tables, yet comments further removed from the activity (Table S3<sup>†</sup>) tended to be more sophisticated. However, this may be due to the difference in the prompt ("what did you learn?" vs. "what three things did you find most important?"). The similar responses suggest that the activity was impactful as students were still able to identify key lessons learned 1 week after the activity.

We also examined the course instructor opinion surveys (CIOS), which are conducted near the conclusion of every course. Responses are anonymous, and include 16 multiple choice questions about the student effort, quality of the course, and quality of the teaching. There are also 5 optional short answer questions for students to provide written effort about effort, best course features, suggestions for and improvements, instructor strengths, instructor improvements. For both offerings of the course, 40 students collectively elected to complete the survey, and the in-class activity was mentioned in 22 responses to the feedback questions (details of survey and comments provided in ESI<sup>+</sup>). Responses were very positive or requested more in-class activities. The most enthusiastic comment noted that this was the "...Highlight of my academic career...", and no negative feedback was provided related to the activity. Overall, this suggests that the activity was impactful to the participants even at 10-14 weeks after its conclusion.

#### Instructor observations on student learning (RQ.4)

The final section of our results relies on the observations the two instructors made while implementing the learning activity. Rather than direct evidence of learning, these observations provide three areas of relevant information to those wishing to implement or extend this activity: (1) our practical observations about students' learning and struggles during the activity, (2) observations and suggestions for effective instructor facilitation, and (3) observations during the activity that suggest areas where deeper research on microfluidics learning may be useful.

In the graduate class, we perceived the participants as actively engaged with the activity and motivated by the goal creating working devices. Participants of were, predominantly, able to complete the activity independently with the materials provided and their knowledge. We base this assertion on the limited clarifying questions that participants asked and observations of participant progress. The questions that were asked were primarily questions related to constructing the devices. Some participants had particular difficulty with abstracting how a three-dimensional shape can be constructed from a stack of two-dimensional layers. This would manifest as a general confusion in how to assemble layers together to create a working microdevice. Similarly, some participants inadvertently removed the entire tape layer instead of the backing film when assembling a device, and assumed that the residual adhesive left on the coverslip was the microfluidic layer. Other participants believed that device assembly was always error free, and were surprised when assembled devices with errors such as layer misalignments were inoperable. We also see the particular challenges and errors as notable as to where practical learning about microfluidic devices was occurring.

After completing the devices, some participants expressed surprise that syringes and ancillary components were needed for microfluidics. Our perception from addressing those questions was that prior, hands-off, interactions with similar resulted in isolated systems had and idealized conceptualization of fluidic phenomena and devices that invoked them in the real world. That is, their prior contexts for learning (in our course and others) had prioritized explanations of the theories of function, at the expense of the reality of operations. The result was a lack of consideration for the structured operation of microfluidic devices and their potential to affect function - e.g., ancillary components and the potential for ancillary components to introduce problems. As an example, several participants became confused as to which ports to use for inlets and outlets. While highlighting an incomplete understanding of the device operation, we found that it could be remedied by suggesting that participants simply experiment with different ports, because using different ports would not damage the devices. This allowed participants to learn in an inquirybased way, which is better aligned with our intended theory of learning for the activity than a correction of prior

instruction. Finally, many participants were surprised that phenomenon that occur at the microscale could be visible with the naked eye and had assumed that microscopes would be necessary, which we interpret as evidence of a muddy understanding of scaling phenomena vs. scaling size.

We also saw other evidence of inquiry-based learning that we sought to encourage in the moment and plan to further encourage through activity revisions. Specifically, participants continued to perform informal experimentation after completing the testing in the end of the activity. For example, one team experimented with different pressures in the droplet generator and observed that certain flow rates were needed to create droplets instead of two streams of oil bisected by coloured water. Such experience is invaluable to developing students understanding of how small changes in fluid flows can dramatically change device operation, and helps highlight why some devices rely on microfluidic pumps to operate. Another team created a mini-competition for team members to apply greater pressure (and fluid) to one side of their T-mixer, resulting in a channel dominated by a single fluid rather than a split stream. Overall, student efforts to create aberrant or non-idealized function seem an opportunity for further exploration to support learning.

Lastly, through participants' reflections and our observations, we identified a number of learning challenges that likely deserve further exploration using more formal educational research methods. Specifically, we noted several patterns of conceptual misunderstanding about microsystems generally, and microfluidics specifically, which span from theory of device function to manufacturing. We use patterns of misunderstanding rather than misconception because we see our data as suggestive but insufficient to formally label these patterns of errors in student thinking. A common form was assumptions that the physical phenomena of macroscale function were similar, just at a smaller scale, in micro-scale devices. For example, some presumed that mixing was still turbulent, rather than diffusive, at microscale - with many not separating mixing into multiple phenomena without prodding. Others presumed that diffusive mixing was instantaneous or only occurred in static systems, an assumption which manifested as surprise that they could observe distinct progressively mixing fluid streams in a T-mixer where two fluid streams move as diffusion occurs across their interface. We observed some students have realizations about these phenomena when in certain configurations (the T- vs. F-mixer), the fluids would not mix at higher flow rates. At such flow rates, the lack of diffusive mixing resulted from decreased time for diffusion to occur. Observing the change in mixing as flow rate varied, but flow patterns did not, seemed to help students internalize key conceptual aspects of diffusion. Overall, we perceived participants' conceptual understanding of diffusion as a particular weakness. Others expressed a mental conception of scale-agnostic fluid mixing occurring via 'collisions' that is suggestive of a mental schema aligned more with atomic motion than expert conceptions of fluid behaviour. Still others, when confronted with device assembly errors, expressed surprise that such assembly was not inherently error free or self-correcting. While we cannot formally test this, our sense from teaching the broader class is that these conceptual misunderstandings were independent of students' ability to perform mathematical analysis of such systems.

In addition to mixing, we noted other patterns of conceptual misunderstanding related to microscale deformations and bubbles. Several participants thought that deformation of microfluidic device layers did not occur or was negligible - i.e., they assumed fluids interact with perfectly rigid devices. Participants had some difficulty understanding that valve layers of a microfluidic must deform in and out of plane for the device to operate. We observed several groups that were surprised to see devices deform to function, or groups that had trouble operating their valve because they inadvertently constructed it with the stiff carrier material rather than the flexible PDMS valving layer. Similarly, when working with the droplet generator, some participants miscategorized spherical objects in microfluidics and had difficulty recognizing the difference between droplets (made from immiscible fluids) and bubbles (made from a gas in a liquid). This was evidenced by answers participants gave to the question "how do air bubbles affect microfluidic operation?" One surprising observation was related to the droplet generator where several groups did not understand that droplets should be forming in the microfluidic device itself, and instead expected a single, centimetre-sized, oil in water droplet at the outlet of the device.

While this list is not comprehensive or rigorously evidenced, we hope it can be helpful to instructors to understand the conceptual challenges faced by students. In the discussion we address how these observations can seed future research on pre-requisite and novel conceptual difficulties that affect learning about miniaturization science. Based on our observations, we see participants encountering novel conceptual challenges related to scaling phenomenon as well as conceptual challenges with microfluidics driven by known conceptual difficulties in pre-requisite knowledge. We also see the potential for there to be much wider ranging difficulties that relate to common educational practices and classroom or lab simplifications – *e.g.*, surprise at the scale of ancillary equipment involved in testing a microfluidics device.

# Discussion

These results are important and have interesting implications for two different audiences, educators and educational researchers, discussed below:

# For miniaturization science educators

One key aspect of this activity is the learning goal to have students experience, reflect, and refine their conceptions.

The goal of the activity is not to successfully build and operate each device - it is to gain understanding about miniaturization science, device design, and construction so as to be able to do so independently. As such, assembly errors and device operation errors themselves lead to meaningful learning experiences. We recommend that instructors focus on helping with troubleshooting, especially by asking students to explain their reasoning and observations and providing timely questions and advice. We repeatedly saw such guidance and asking questions was more productive at facilitating both learning and successful activity completion by the students than simply giving the "correct answer". In this manner, conceptual difficulties can be identified and corrected by students, rather than remaining hidden. This is why we recommend backup kits to assembled devices as well as backup working devices if a team is having a particularly difficult time. As engineers and scientists, we have a natural bias to prevent errors and fix problems, and therefore, may be tempted to make modifications to make the kits "fool proof" or easier to assemble. However, this again would be counterproductive to our learning goals, which is to elicit and correct conceptual difficulties.

This work highlights that miniaturization science theoretical learning does not require high resources, or that accessible technology necessitates simpler learning objectives. Instead, theoretical learning requires activities tuned to achieve learning goals that are appropriate for that audience. Our activity leverages a low-cost and established microsystems technique and needs little ancillary equipment. As such, it can be packaged into a small kit to be employed in nearly any classroom, and even mailed to provide an activity that complements virtual learning.

This technology could especially impact the education and implementation of miniaturization science worldwide. Point of care technologies designed using microdevice principles have the potential to significantly impact healthcare. Since these technologies can be designed to automatically perform complex laboratory steps with a minimal amount of equipment and power, they are especially suited to lowresource settings and more rural areas with less access to healthcare. Unfortunately, the scientists in these settings who have first-hand knowledge of both the needs and challenges related to working in their environment do not have access to the same resources and educational opportunities related to miniaturization science. Our activity could help address this issue by creating a significant learning experience on microdevices and teaching students a technique that could be broadly applied to local pressing needs.

#### For education researchers

The area of microfluidics and microsystems is a rich area for inquiry to better understanding learning processes as it operates at the intersection of multiple fields including fluid mechanics, mass transport, manufacturing, mechanics of materials, and more. We observed many student conceptual weaknesses, ranging from device function to manufacturing, as well as some specific to microfluidics. More work is needed to formally identify whether these conceptual challenges were misconceptions, that is, the ontological miscategorization of a concept.

Our work also highlights challenges that students can have when multiple physical processes occur simultaneously. Students are conditioned to look for single physical processes to occur, for example, that either static diffusion or fluid movements is occurring. We noted that some students had trouble combining the two concepts together, namely that fluid could both move and still experience diffusive mixing across a boundary. It is interesting to note that this confusion also mirrors some common simplifications used to teach difficult concepts in the classroom, such as teaching static diffusion or fluid movement and not in combination. Hence, these simplifications may be contributing to some later confusion.

# Conclusions

In this paper we describe the implementation of a microfluidics learning activity and provide evidence that the it achieves our intended learning outcomes. We do not have comparative evidence from a control group, a limitation we hope to address in the future. However, attributing the large gains in conceptual learning from test results to the activity as opposed to other sources is supported in two other ways. First, the shift in scores from the pre to post-test occurred after the topics had been covered in the lecture portion of the class. While the activity may be functioning symbiotically with the lecture, the lecture alone (as shown by the pre-test) created less Second, participants' reflection comments learning. described in the previous section link discoveries or realizations that occurred while completing the activity to known areas of conceptual difficulty and our specific learning activities. We also showed differences in learning gain between undergraduates and graduate students who otherwise took the same course. Therefore, we see the activity as appropriate for developing the conceptual understanding of graduate and advanced undergraduate students - with the caveat that further testing will always build a case for this claim. The activity is based on a technology for microfluidic device construction that is less resource intensive than typically used at this level of education to demonstrate micro scale fluidic phenomena that are of commercial and research interest. The technology also allows for greater hands on interaction with devices and their components, including hands on assembly.

Students with less prerequisite knowledge about fluid phenomenon can complete the abridged activity, but data shows they learn less than those who complete the full activity. When studied in a graduate level course on microfluidics, evidence shows that completing the activity

predicts large, significant, gains in conceptual understanding. Those gains were greater than those who completed the partial activity for both graduate and undergraduate students in the course, despite meaningful evidence of a measurement ceiling. Both studies (i.e., inside the course and outside of it) show significant improvements in confidence and confidence weighted scoring. Student reflections and instructor observations specifically provide evidence of increased self-awareness of conceptual mastery as well as changes in understanding about key concepts. Finally, instructor observations and student comments also show evidence of potential misconceptions, especially about dominant phenomena, when learning about the transition from macro to micro scale that deserve further exploration. For example, most students did not identify microscale mixing as predominantly diffusion based, and when queried struggled to differentiate different phenomena that created fluid mixing - i.e., mixing was a phenomenon rather than a type of phenomena. One limitation of the present study is that we were not specifically able to test for misconceptions, but instead identify patterns of misunderstanding that are possibly misconceptions. More specific studies are needed to determine microsystem specific misconceptions experienced by students.

The differences in learning between undergraduate and graduate students does show that the prior experience and knowledge may affect the amount of learning from our activity. However, given that our intended population was graduate students, we see use with the target population as a reasonable limitation. As a result, we conclude that it is reasonable for others to adopt and use this activity with similar populations of students, but should use caution when adapting it for students with much less prior knowledge or experience in thermal fluid phenomena. We note that suggestions for instructors contained in the discussion and ESI† have a pragmatic vs. empirical basis. However, those suggestions are also well aligned with established theories of learning, such as conceptual change and inquiry-based learning, that are the foundation of the activity.32 More broadly, our activity demonstrates that efforts to democratize the education in advanced miniaturization science can maintain rigorous conceptual learning even when focusing on a reduction in required requirements resources. Reducing resource while maintaining learning is important to the value of such learning activities, especially to scientists and engineers in low-resource settings where microfluidics and similar pointof-care technologies have enormous potential to address unmet needs.27 Further studies are necessary to confirm the portability of this activity to other learning environments. Because we see the accessibility and portability of the activity resources as a key benefit, testing in virtual settings and in low resource settings would be important to address any unforeseen challenges due to shipping, availability of materials, and less controlled environmental conditions.

# Author contributions

PD, SS, TF, & DRM planned and conceptualized the work. PD, CAL, AD, & DRM performed the experiments on all cohorts and analyzed data. PD, JJW, TF, & DRM interpreted the data and wrote the manuscript.

# Conflicts of interest

There are no conflicts to declare. GT IRB reviewed and determined this study to be exempt. All participants consented to participate. Data available upon request.

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