Teacher's Preparatory Guide

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Design and Production of a Microfluidic Chip[1]

Purpose This lab is designed to introduce students to a modern chemistry technique: microfluidics. Students will understand how microfluidics work and explore the possible uses for it.

Objectives Design and create microfluidic devices that can successfully mix two fluids and create concentration gradients to solve a problem.

Time required Teacher preparation is three hours. Students will need between three and five 45 minute class periods to complete the lab activity. Reserve 10 minutes of lab time each class period for clean-up.

Level High school introductory honors chemistry class.

National Science Education Standards
1. HS-ETS-ED-B. Analyze input and output data and functioning of a human-built system to define opportunities to improve the system's performance so it better meets the needs of end user while taking into account constraints.
2. HS-ETS-ED-D. Plan and carry out a quantitative investigation with physical models or prototypes to develop evidence on the effectiveness of design solutions, leading to at least two rounds of testing and improvement.

Florida State Standards
SC.912.P.8.11 Relate acidity and basicity to hydronium and hydroxyl ion concentration and pH

Teacher Background and Resources
Microfluidics is a modern analytical chemistry technique in which micro amounts of liquids are analyzed on a chip. This process allows for quick turn-around time and the ability to test multiple parameters simultaneously; which in turn reduces time spent preparing samples for testing, costs, and material waste.

The following internet resources are helpful for both teacher and student.

Microfluidics Videos:

http://www.youtube.com/watch?feature=endscreen&NR=1&v=5QVwljd04Kw

http://www.youtube.com/watch?v=BIXvgU1ud_c&feature=fvwrel

Lab-on-a-Chip TED talks:

Frederick Balagadde:
http://www.ted.com/talks/frederick_balagadde_bio_lab_on_a_microchip.html

George Whitesides:
http://www.ted.com/talks/george_whitesides_toward_a_science_of_simplicity.html

Supplemental reading:

http://www.popsci.com/scitech/article/2009-08/lab-chip-can-carry-out-1000-tests-once

http://www.sciencedaily.com/releases/2012/05/120510095616.htm

http://www.sciencedaily.com/releases/2012/10/121026100949.htm

http://www.sciencedaily.com/releases/2012/03/120327124858.htm

Microfluidic Lab video links:

Video lab guide http://mrsec.wisc.edu/Edetc/nanolab/index.html

Materials

- Computer and access to Microsoft Powerpoint or another graphic design program
- Clear Shrinky Dink film (Grafix, KSF50-C, amazon.com)
- Laser printer (we used a Hewlett Packard LaserJet 4100N)
- Scissors
- Colored pencils/markers
- Mineral or vegetable oil
- Crystallization dish (125 x 65 mm) or 500 mL glass beaker
- Hot plate
- Thermometer with capability to measure above 150 C
- Tweezers or forceps
- Glass plates
- Soap
- Glass microscope slides (75 x 50 mm and 25 x 75 mm)
- PDMS chemicals (Sylgard 184 Silicone Elastomer Kit, amazon.com)
- Plastic cup
- Wood stick/stir rod or plastic fork
- Vacuum desiccator or bell glass jar with pump
- Oven (60°C)
- Razor blade or scalpel
- Biopsy punch (2-mm diameter, sold for piercing, available on amazon.com for <$10)
- Scotch tape to clean the PDMS
- Double sided Scotch tape (wider tape is ideal)
- Plastic Petri dish (150-mm diameter, amazon.com)
- Transfer pipettes
- Food coloring or other colorimetric indicator (provide very concentrated solution)
- Acid and base solutions, weak (example: 100 mM NaH_2PO_4 and 100 mM Na_2HPO_4)
- Universal pH indicator solution, concentrated
- Small syringes (tapered tip or oral irrigator syringes, amazon.com)
- Tubing (Tygon PVC tubing OD = 3/32" and ID = 1/32" or similar, amazon.com

Advance Preparation Prepare the three microfluidic chip designs as given in the guided inquiry page. A template for printing is provided. Follow the student directions to make the chip. This should be done at least two days in advance to allow time for troubleshooting as necessary and to allow the molds to completely harden. A complete set of the three chips should be provided to each group of students. Alternatively, providing extra time, the students can prepare their own starter microfluidic chips. The will help them to produce their own designed chip more effectively.

Safety Information This procedure utilizes hot oil, a biopsy punch, and a scalpel/razor. The risk of burns and cuts are inherent in this lab. Proper use of tools, safety equipment, vigilance of the teacher and common sense by the students is sufficient to prevent injury and accidents. Students must adhere to all safety rules while working in the laboratory. Failure to do so will result in immediate removal from the lab.

Teaching Strategies Collaboration of ideas between students is crucial for success in this lab. Student groups should be limited to four students. Be mindful of material supply when deciding how many students will be in each group. At times, students prefer to work alone in the lab, however, manipulation of the equipment and material in this lab are easier done with at least one lab partner.
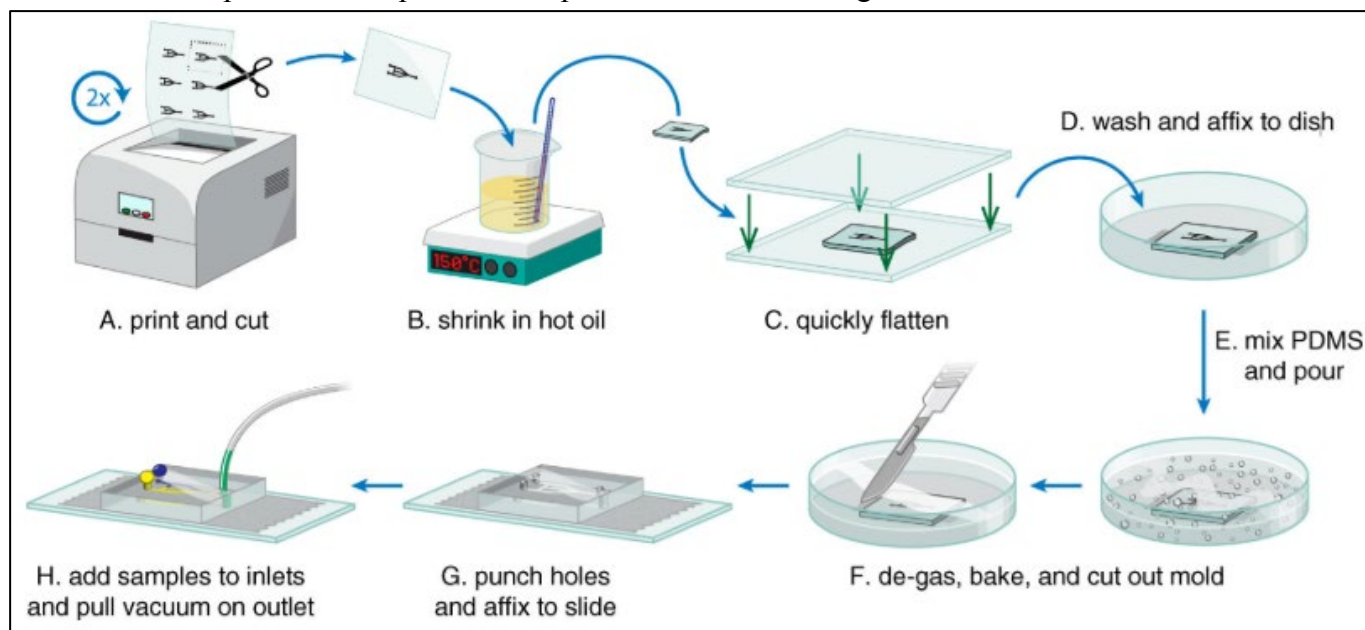
Directions for the activities

Day 1: - Have students brainstorm a response to the question: "What does microfluidics mean?"
- Introduce the concept of microfluidics. (An assortment of internet resources are provided in the background section).
-Review basic concepts of acid/base chemistry, neutralization and pH. An in-depth discussion is not necessary for a successful design.
- Assign student groups
- Students complete Microfluidics 101 Guided Inquiry
- Class discussion of laminar flow and the features of each starter chip
-Discuss design challenge, each student will bring their own design to their group the next day (to be completed as homework)

Day 2-3: -Students choose one design to implement within their group and will draw it with a graphic design program such as Power Point or Publisher if not already done so.
-Create and test the group chosen microfluidic device.

Day 4-5: -Group discussion of results obtained from the first microfluidic device. Brainstorm changes to first design to optimize performance.
-Draw the optimized design on the computer.
-Create and test the optimized design.

Conclusion of Lab: -Class discussion of the scientific method followed through the design and test process.
- Show the Ted Talk Videos provided
- Brainstorm ways that microfluidic devices can help to solve real world problems
- Discuss current research utilizing microfluidics

Procedure A complete detailed procedure is provided in the student guide.

A. print and cut
B. shrink in hot oil
C. quickly flatten
D. wash and affix to dish
E. mix PDMS and pour
F. de-gas, bake, and cut out mold
G. punch holes and affix to slide
H. add samples to inlets and pull vacuum on outlet

Cleanup Excess PDMS may be disposed of in the trash, it is non-toxic. Weak acid and weak base solutions

may be poured down the sink with running water.

Microfluidic Design Templates
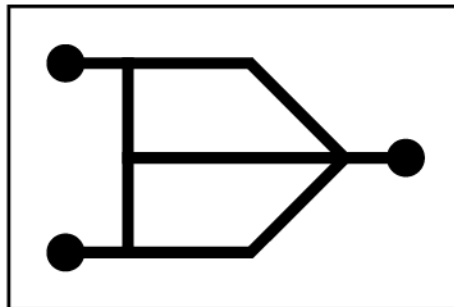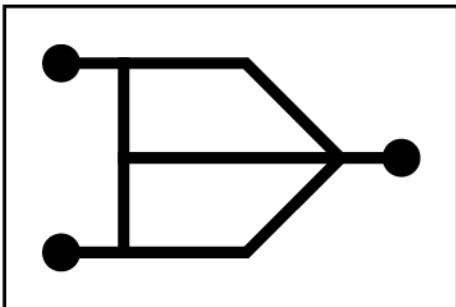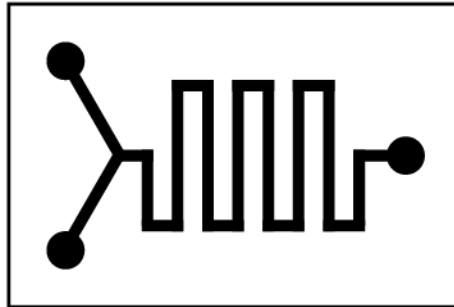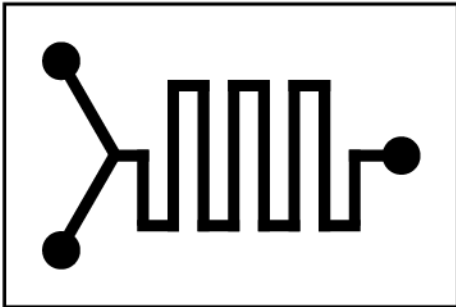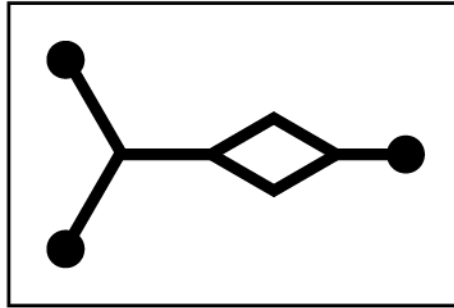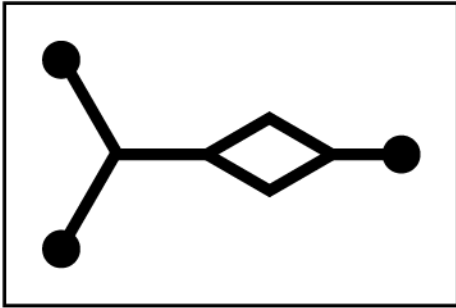
Microfluidics 101 Guided Inquiry
Design and Production of a Microfluidic Chip

Name: _____   Class Per: _____

Microfluidics deals with the precise control of fluids on the microscale. Often at the microscale, smaller amounts of reagents are used so experiments are faster and cheaper. Today you will be using three simple microfluidic devices to discover what makes microfluidics so cool!

## Microfluidics Device Exploration

Prediction:
In the chart below predict what you think will happen if a drop of blue food coloring and a drop of yellow food coloring are placed in the two inlet holes of the device. Use colored pencils to draw your prediction and explain your reasoning. Repeat for all three devices.

Lab Observation:
1. Using pipettes, place a drop of blue food coloring and a drop of yellow food coloring in the inlet holes of the microfluidics device.
2. Insert the tip of the syringe (or tubing connected to the syringe) into the single outlet hole in the device.
3. Pull back very slowly on the syringe, until the food coloring is pulled through the device. Pulling too fast or too much will affect your observations.
4. Draw your observation in the chart below using colored pencils. Explain why you think this happened and answer the questions. Repeat for all three devices.
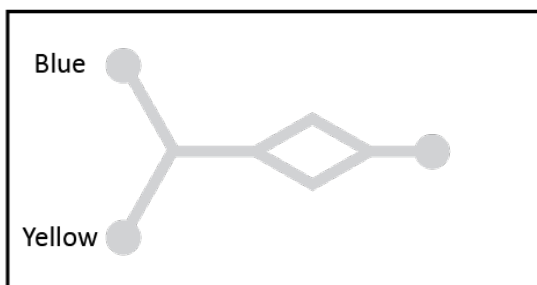
Device 1
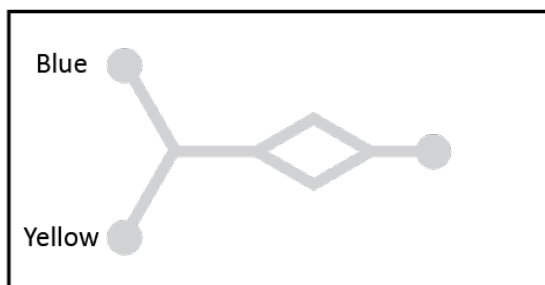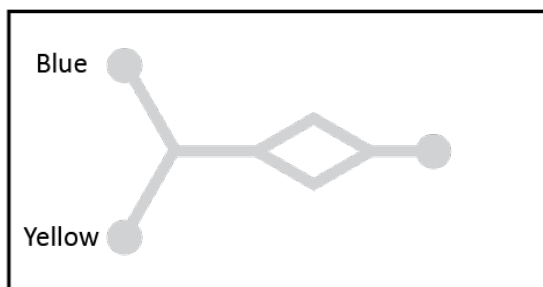
Prediction

Why do you think this?

Lab observation

Why do you think this happened?

Does this device show any mixing of fluids?

Device 2

Prediction

Blue

Yellow

Why do you think this?

Lab observation
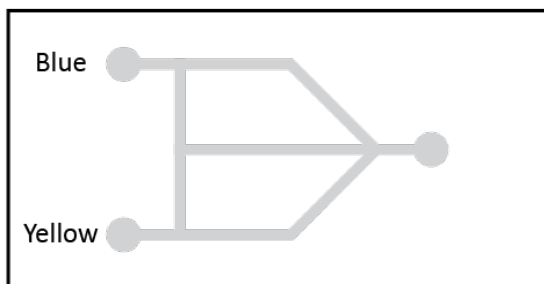

Blue

Yellow

Why do you think this happened?

Does this device show any mixing of fluids?
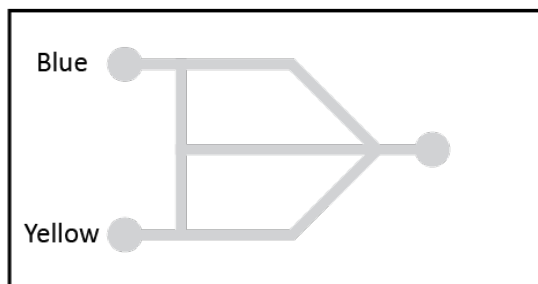
Device 3

Prediction

Blue

Yellow

Why do you think this?

Lab observation

Blue

Yellow

Why do you think this happened?

Does this device show any mixing of fluids?

Laminar Flow

Laminar Flow Definition: *Non-turbulent fluids flow in parallel layers.*
Very little lateral mixing occurs between adjacent layers.
Fluids flow "smoothly."

Reynolds number (Re) = (density)(velocity)(channel diameter)
                                                    viscosity

Re < 2000 → laminar flow
Re > 4000 → turbulent flow

Apply the Concept:

1. ***Circle*** all the areas in the three microfluidic designs where different fluids meet but do not mix.

2. What features of a microfluidic device promote laminar flow? How do you know?

3. If a scientist would like to have two solutions completely mix in a microfluidic device, what would you tell them to include in their microfluidic design? Why?

4a. Calculate your Reynolds number in water versus molasses (use the values provided below).

	Density (g/m^3)	Viscosity (g/(m•s))
Water	1×10^6	1
Molasses	1.4×10^6	1×10^4

Assume your velocity = 0.2 m/s, and your 'diameter' is roughly = 0.25 m.

4b. For comparison, the Re of a blue whale in water is roughly 1×10^8. And the Re of a bacterium in water is about 1×10^{-5}. How would your motion in water versus molasses compare to the motion of these organisms in nature?

Microfluidic Design Challenge

Challenge: Your task is to create a microfluidic device that will take a weak acid (100 mM NaH_2PO_4) and a weak base solution (100 mM Na_2HPO_4), and create five different pH values (1-2, 3-5, 6-8, 9-11, 12-14) in a single device. A universal pH indicator has been mixed into the acid and base solutions and will report changes in pH by changing the color of the solution at different pH values. Design a microfluidic device that is no larger than 6 by 8 cm. Make sure your design is in black and white and has ample space around it (about 5 cm) to easily cut out.

Use this blank space to brainstorm a design with your group

Microfluidics 101 Guided Inquiry
Design and Production of a Microfluidic Chip

Teacher's Guide

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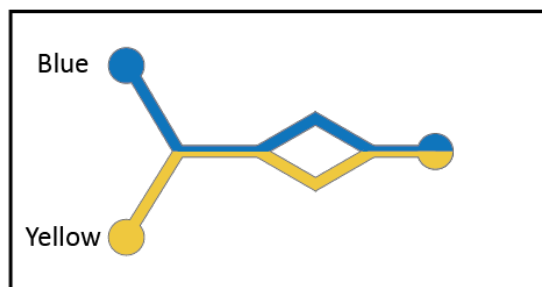
Device 1
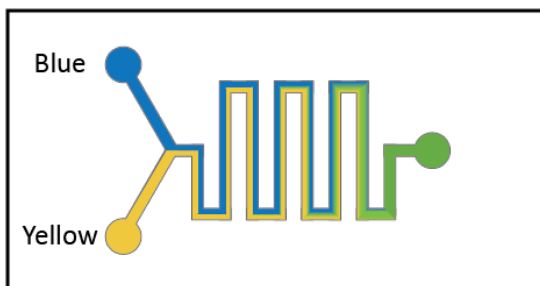

Why do you think this?

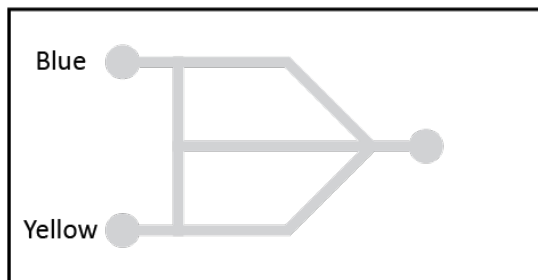Why do you think this happened?

Does this device show any mixing of fluids?

Device 2

Prediction	Lab observation

Why do you think this?

Why do you think this happened?

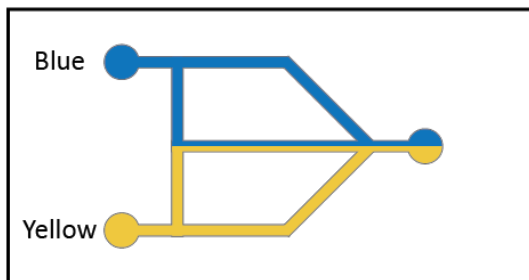Does this device show any mixing of fluids?

Device 3

Prediction	Lab observation

Why do you think this?

Why do you think this happened?

Does this device show any mixing of fluids?

Laminar Flow

Laminar Flow Definition: *Non-turbulent fluids flow in parallel layers.*
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Fluids flow "smoothly."

Reynolds number (Re) = (density)(velocity)(channel diameter)
                                    viscosity

Re < 2000 → laminar flow
Re > 4000 → turbulent flow

Apply the Concept:

1. ***Circle*** all the areas in the three microfluidic designs where different fluids meet but do not mix.   The correct fluid behavior is shown in the diagrams above.

2. What features of a microfluidic device promote laminar flow? How do you know?

   The primary feature is the small dimensions of the channels. If we consider the equation for Reynolds number above, every variable except channel diameter is relatively constant between the types of fluid motion we experience at the macroscale (e.g., a kitchen faucet or a garden hose) versus the conditions in our microfluidic device – at the microscale.

3. If a scientist would like to have two solutions completely mix in a microfluidic device, what would you tell them to include in their microfluidic design? Why?

   There are several ways to answer this question. A correct answer will be any design feature that exploits diffusion. For example, if we consider design #2 in the "Microfluidics Design Exploration" section, this serpentine design is essentially one long channel that maximizes the amount of time that the two solutions contact one another, therefore allowing diffusion enough time to completely mix the solutions.

4a. Calculate your Reynolds number in water versus molasses (use the values provided below).

	Density (g/m³)	Viscosity (g/(m•s))
Water	1×10^6	1
Molasses	1.4×10^6	1×10^4

Assume your velocity = 0.2 m/s, and your 'diameter' is roughly = 0.25 m.

   $Re_{water} = 5 \times 10^4$

(Note that these values are simple approximations)

4b. For comparison, the Re of a blue whale in water is roughly 1×10^8. And the Re of a bacterium in water is about 1×10^{-5}. How would your motion in water versus molasses compare to the motion of these organisms in nature?

There are many acceptable answers to this question. However, students should recognize that size greatly effects how objects behave in fluids. Resistance due to viscosity is the dominant force experienced by motile bacteria and when bacteria stop swimming, they immediately stop moving because they lose their momentum due to the viscosity of the medium. In contrast, when a whale stops swimming, its body glides slowly to a stop because the solution's viscosity has a smaller influence on its motion. Humans swimming in water feel very little influence from the liquid viscosity; to feel the influence of viscosity, we would need to swim in a very viscous liquid, such as molasses.

Microfluidic Design Challenge

Challenge: Your task is to create a microfluidic device that will take a weak acid (100 mM NaH_2PO_4) and a weak base solution (100 mM Na_2HPO_4), and create five different pH values (1-2, 3-5, 6-8, 9-11, 12-14) in a single device. A universal pH indicator has been mixed into the acid and base solutions and will report changes in pH by changing the color of the solution at different pH values. Design a microfluidic device that is no larger than 6 by 8 cm. Make sure your design is in black and white and has ample space around it (about 5 cm) to easily cut out. Follow the lab instructions to make a device using Shrinky Dinks.
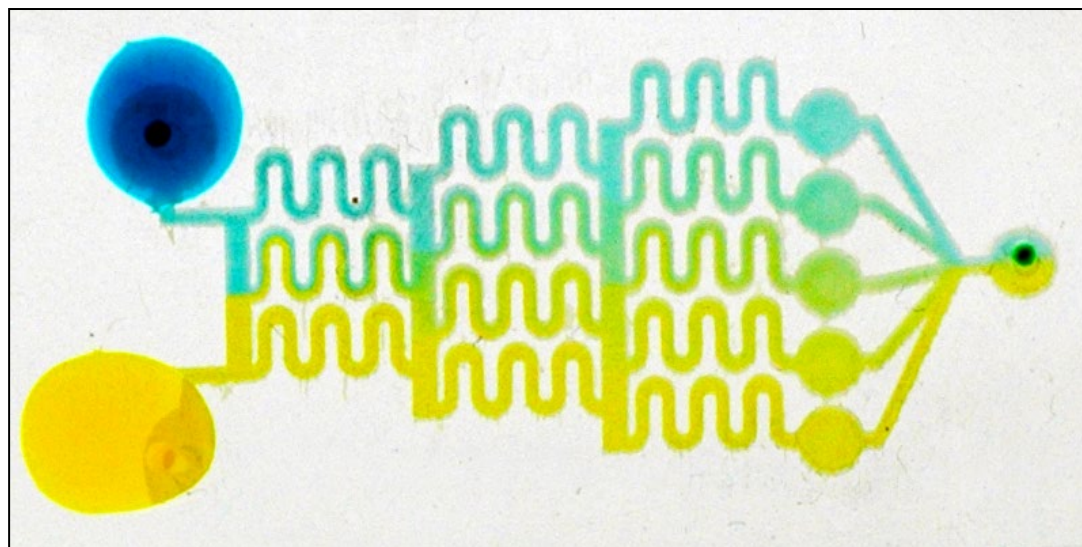
There are many designs that can correctly answer this design challenge. One example of a correct device is pictured above (Top-down view). Two inlets are shown on the left

where the weak acid and weak base solutions are added to the device (for clarity, the solutions covering the inlets here are drops of blue and yellow food dye). There is only one outlet on the far right, and the solutions flow through the device from left to right. The principle of this device's operation combines two key design concepts that are introduced in the 'Microfluidics Device Exploration' section. There are three thick vertical bars in this device whose behaviors match that seen in device #3 in the exploration activity. If we focus on the left-most bar, the blue and yellow solutions meet exactly in the middle of the vertical channel and do not mix due to laminar separation. The fluid exits the first vertical channel through three serpentine channels (similar to device #2 in the exploration activity). In the top and bottom serpentine channels, pure blue and yellow solutions are maintained. The middle channel, however, receives a 50:50 combination of blue and yellow solutions. These blue and yellow solutions mix through diffusion as they flow through the serpentine channel, emerging as a green solution at the end of the channel. At this point, three solutions with different pH values have been created (represented here as blue, green, and yellow solutions).

The same logic is repeated two more times for the rest of the device, resulting in five solutions with different pH values (shown here as five differently colored solutions in the five circular wells near the outlet).

References

1. Microfluidics for High School Chemistry Students
Melissa Hemling, John A. Crooks, Piercen M. Oliver, Katie Brenner, Jennifer Gilbertson, George C. Lisensky, and Douglas B. Weibel
Journal of Chemical Education 2014 *91* (1), 112-115
  DOI: 10.1021/ed4003018