# ADVANCED BIOTECHNOLOGY Pipetting skills

Focus question	How can we develop and practice micropipetting skills? Why is micropipetting important?
Learning target	Students practice micropipetting and check their accuracy.
Vocabulary	Micropipette

Adapted from "Pipetting by Design" cpet.ufl.edu

# LS3: Heredity: Inheritance and Variation of Traits

Performance expectation HS-LS1-1	<b>Classroom connection:</b> This activity helps build lab skills to be successful using other biotech techniques.		
<b>Performance expectation</b> HS-LS3-1	<b>Classroom connection:</b> Students practice micropipetting small amounts of liquids, a necessary skill in a biotechnology lab.		

# Science and engineering practices

Asking Questions and Defining Problems	<b>Classroom connection:</b> Students will micropipette colored water into cell well plates to make patterns. The patterns will encourage them to <i>ask questions and define problems</i> .		
Constructing Explanations and Designing Solutions	<b>Classroom connection:</b> Students will create the pattern assigned to them and <i>construct an explanation</i> to connect the pattern to biotechnology.		

# Disciplinary core ideas

LS3.A: Inheritance of Traits	<b>Classroom connection:</b> Students will practice moving small amounts of liquids to develop skills for performing other	
	biotech lab activities.	

# **NOURISH IN FUTURE**

### **Cross-cutting concepts**

Structure and Function	<b>Classroom connection:</b> Students will compare the accuracy of transferring small amounts of liquid using micropipettors and 1 mL disposable pipettes.	
Cause and Effect	<b>Classroom connection:</b> Using micropipettors instead of disposable pipettes results in greater accuracy for these activities.	
Science is a Human Endeavor	<b>Classroom connection:</b> Micropipettors are a technological advance that has influenced the progress of science.	

# **Prior knowledge**

Students may have used a disposable 1mL pipette before. It is difficult to control accuracy when using these pipettes. This activity shows students that micropipettors can transfer much smaller amounts of liquid and are much more accurate if the proper technique is used. It is possible to do the lab with the sterile pipettes that are included in the kit from BioRad. The ability to use micropipettes is a valuable skill for employment in any biotech lab.

# **Teacher preparation**

In order to develop a skill with accuracy, students need to practice. This activity allows students to practice with micropipettors and check their accuracy by measuring the mass of their results. This also gives students the opportunity to determine a percent error measurement.

- 1. Copy protocol instructions.
- 2. Organize students in groups of 3–4, with two pipettes each of different sizes, to work together to create the pattern. From the protocols (attached on additional pages) there are multiple amounts called for and each student will have ample opportunity to pipette. If a check for accuracy is desired after each student adds their assigned color/amounts, they can determine the mass at that point and measure to see where they are in terms of accuracy.

Final totals for each protocol (adjustable volume pipettes)	Final totals for each protocol (fixed volume pipettes)		
A = DNA = 5045µl = 5.045g	A = DNA = 2655µl = 2.655g		
B = corn = 5841µl = 5.841g	B = corn = 2760µl = 2.76g		
C = pGLO = 5217µl = 5.217g	C = soil = 2930µl = 2.93g		
D = a plasmid = 1580µl = 1.58g	D = a plasmid = 1910µl = 1.91g		
E = GMO = 4621µl = 4.621g	E = GMO = 2340µl = 2.34g		
F = soy = 5003µl = 5.003g	F = soy = 3560µl = 3.56g		

# Differentiation

Other ways to connect with students with various needs:

- Local community: Students may do a search to see what genetic modification resources are available in their community. Medical labs are using genetic modification techniques to target specific diseases, and agriculture companies (Corteva, Bayer, Syngenta, BASF) may have education and outreach departments that would send a speaker to your class. Lab techs in hospitals and veterinary clinics often use micropipetting skills and may be interested in sharing experiences with your class.
- Students with special needs (language/reading/auditory/visual): This lesson requires fine
  motor coordination and may result in frustration among students who are not detail-oriented
  or able to be precise. They may be employed as accuracy assessors. They could travel around
  the class to check progress and measure the mass of the plates to see how accurate each
  group is being.
- Extra support: Watch Basic Pipetting at: youtu.be/bex0itUMxmI
- Extensions: Students may want to do more than one protocol, or work on their own to check their accuracy.

# Assessments

Have students determine their percent error.

### **Rubric for self-assessment**

Skill	Yes	No	Unsure
Our group pipetted the correct amounts into the cell well plate and we calculated our percent error.			
Our pattern helped me to ask questions and define problems related to biotechnology.			
We were able to connect our pattern to a biotechnology-related topic.			

# Protocols for adjustable volume pipettes

### **Protocol A**

Micropipette the indicated volumes into designated wells on the 96 well plate.

#### Using the red water

- 20 µl: B1, B2, B3, B11
- 26 µl: D1, D3, D10, D11, D12
- 27 µl: E1, E3, E10, E12
- 28 µl: F1, F2, F10, F12
- 29 µl: C1, C3, C10, C12

#### Using the blue water

- 38 µl: B5, B8
- 36 µl: D5, D7, D8
- 48 µl: E5, E7, E8
- 59 µl: F5, F8
- 67 µl: C5, C6, C8

#### Using the red water

- 90 µl: B1, B2, B3, B11
- 132 µl: D1, D3, D10, D11, D12
- 127 µl: E1, E3, E10, E12
- 103 µl: F1, F2, F10, F12
- 112 µl: C1, C3, C10, C12

#### Using the blue water

- 126 µl: B5, B8
- 102 μl: D5, D7, D8
- 96 µl: E5, E7, E8
- 138 µl: F5, F8
- 114 µl: C5, C6, C8

### **Protocol B**

Micropipette the indicated volumes into designated wells on the 96 well plate.

#### Using the yellow water

- 20µl: E5, E6, E9, E12
- 27µl: F4, F7, F9, F10, F12
- 45µl: G4, G7, G9, G11, G12
- 38µl: H5, H6, H9, H12

#### Using the green water

- 119µl: A2, A3, A7, A8
- 48µl: B1, B4, B7, B9
- 67µl: C1, C7, C8
- 76µl: D2, D3, D4, D7, D9
- 55µl: E2, E3, E4

#### Using the yellow water

- 70µl: E5, E6, E9, E12
- 110µl: F4, F7, F9, F10, F12
- 116µl: G4, G7, G9, G11, G12
- 93µl: H5, H6, H9, H12

#### Using the green water

- 129µl: A2, A3, A7, A8
- 118µl: B1, B4, B7, B9
- 107µl: C1, C7, C8
- 96µl: D2, D3, D4, D7, D9
- 88µl: E2, E3, E4

# **Protocol C**

Micropipette the indicated volumes into designated wells on the 96 well plate.

#### Using green water

- 20µl: B7, C7
- 47µl: D1, D2, D7
- 28µl: E1, E3, E7
- 59µl: F1, F2, F7, F8, F9
- 35µl: G1, H1

#### Using blue water

- 28µl: B5, B11
- 62µl: C4, C6, C10, C12
- 51µl: D4, D10, D12
- 90µl: E4, E5, E6, E10, E12
- 79µl: F5, F11

#### Using green water

- 70µl: B7, C7
- 116µl: D1, D2, D7
- 110µl: E1, E3, E7
- 85µl: F1, F2, F7, F8, F9
- 93µl: G1, H1

#### Using blue water

- 118µl: B5, B11
- 96µl: C4, C6, C10, C12
- 88µl: D4, D10, D12
- 199µl: E4, E5, E6, E10, E12
- 107µl: F5, F11

# **Protocol D**

Micropipette the indicated volumes into designated wells on the 96 well plate.

#### Using blue water

- 20µl: A6, A7
- 39µl: B5
- 28µl: C4, D4, E4
- 42µl: F5
- 51µl: G6, G7

#### Using red water

- 100µl: A8
- 111µl: B9
- 120µl: C10, D10

#### Using green water

- 140µl: E10
- 176µl: F9
- 115µl: G8

#### Using blue water

- 70µl: A6, A7
- 81µl: B5
- 76µl: C4, D4, E4
- 92µl: F5
- 101µl: G6, G7

### **Protocol E**

Micropipette the indicated volumes into designated wells on the 96 well plate.

#### Using red water

- 20µl: B2, B3, B4, B11
- 35µl: C1, C10, C12
- 47µl: D1, D3, D4, D10, D12
- 28µl: E1, E4, E10, E12
- 59µl: F2, F3, F11

#### Using yellow water

- 88µl: B6, B8
- 26µl: C6, C7, C8
- 45µl: D6, D8
- 79µl: E6, E8
- 27µl: F6, F8

#### Using red water

- 70µl: B2, B3, B4, B11
- 116µl: C1, C10, C12
- 110µl: D1, D3, D4, D10, D12
- 135µl: E1, E4, E10, E12
- 179µl: F2, F3, F11

#### Using yellow water

- 118µl: B6, B8
- 96µl: C6, C7, C8
- 89µl: D6, D8
- 129µl: E6, E8
- 107µl: F6, F8

### **Protocol F**

Micropipette the indicated volumes into designated wells on the 96 well plate.

#### Using blue water

- 119µl: B1, B2, B10, B11, B12
- 102µl: C1, C3, C9
- 127µl: D1, D3, D9, D11, D12
- 115µl: E1, E3, E9, E12
- 132µl: F1, F2, F10, F11

#### Using red water

- 20µl: B5, B6
- 34µl: C5, C7
- 89µl: D5, D7
- 44µl: E5, E7
- 100µl: F5, F6

#### Using blue water

- 22µl: B1, B2, B10, B11, B12
- 31µl: C1, C3, C9
- 29µl: D1, D3, D9, D11, D12
- 22µl: E1, E3, E9, E12
- 40µl: F1, F2, F10, F11

#### Using red water

- 74µl: B5, B6
- 158µl: C5, C7
- 37µl: D5, D7
- 196µl: E5, E7
- 88µl: F5, F6

# Protocols for fixed volume pipettes

# **Protocol A**

Micropipette the indicated volumes into designated wells on the 96 well plate.

#### Using the red water

- 20 µl: B1, B2, B3, B11
- 205µl: C1, C3, C10, C12
- 30µl; D1, D3, D10, D11, D12
- 50µl: E1, E3, E10, E12
- 100µl: F1, F2, F10, F12

#### Using the blue water

- 40 µl: B5, B8
- 55µl: C5, C6, C8
- 20µl: D5, D7, D8
- 100µl: E5, E7, E8
- 200µl: F5, F8

### **Protocol B**

Micropipette the indicated volumes into designated wells on the 96 well plate.

#### Using the yellow water

- 20µl: E5, E6, E9, E12
- 30µl: F4, F7, F9, F10, F12
- 50µl: G4, G7, G9, G11, G12
- 100µl: H5, H6, H9, H12

#### Using the green water

- 200µl: A2, A3, A7, A8
- 55µl: B1, B4, B7, B9
- 100µl: C1, C7, C8
- 40µl: D2, D3, D4, D7, D9
- 120µl: E2, E3, E4

# **Protocol C**

Micropipette the indicated volumes into designated wells on the 96 well plate.

#### Using green water

- 50µl: B1, B2, B3, B4, B10
- 100µl: C1
- 70µl: D1, D2, D3, D4, D10
- 200µl: E4, E10
- 60µl: F1, F2, F3, F4, F10

#### Using blue water

- 70µl: B12, C12
- 100µl: D6, D7, D8, D12
- 210µl: E6, E8, E12
- 90µl: F6, F7, F8, F12

# **Protocol D**

Micropipette the indicated volumes into designated wells on the 96 well plate.

#### Using blue water

- 110µl: A6, A7
- 50µl: B5
- 90µl: C4, D4, E4
- 100µl: F5
- 200µl: G6, G7

#### Using red water

- 100µl: A8
- 200µl: B9
- 120µl: C10, D10

#### Using green water

- 120µl: E10
- 100µl: F9
- 110µl: G8

# **Protocol E**

Micropipette the indicated volumes into designated wells on the 96 well plate.

#### Using red water

- 20µl: B2, B3, B4, B11
- 50µl: C1, C10, C12
- 100µl: D1, D3, D4, D10, D12
- 30µl: E1, E4, E10, E12
- 60µl: F2, F3, F11

#### Using yellow water

- 100µl: B6, B8
- 150µl: C6, C7, C8
- 70µl: D6, D8
- 200µl: E6, E8
- 60µl: F6, F8

# **Protocol F**

Micropipette the indicated volumes into designated wells on the 96 well plate.

#### Using blue water

- 100µl: A1, A2, A3, A4, A8, A12
- 110µl: B1, B9, B11
- 50µl: C1, C10
- 30µl: D1, D2, D3, D4, D10
- 200µl: E4, E10, F4, F10
- 70µl: G1, G2, G3, G4, G10

#### Using red water

- 20µl: B5, B6, B7, B8
- 30µl: C5, C8, D5, D8
- 60µl: E5, E8, F 5, F8
- 200µl: G5, G6, G7, G8