Displacement pipettes are valuable for manipulating tiny volumes of liquid. Their calibrated thumb-driven piston sucks up the desired volume of fluid into the pipet tip. The units of volume are in microliters (µL), also termed lambda (Λ). Since there are a million µLs in a liter, 100 µL is 0.1 mL. (Learn how set, read the pipet and interconvert these volumes!) The plastic tips are usually discarded after a single use. The pipettes (and sometimes their tips) are color coded according to their capacity: white for 10 µL, yellow for up to 200 µL, blue for up to 1000 µL.

The advantage of single-use disposable tips in genetic engineering is that there is no possibility of contamination with endonucleases which could destroy DNA samples. Also, the tips are much less expensive than single-use pipettes. However, the instruments are very expensive (about $200 each), and must be handled and cared for properly. NEVER allow fluid up into the body of the pipetter.

Illustrate a displacement pipette and include all of the following features:

body  lock ring  tip
thumb plunger  volume of aliquot (in window, practice reading)  tip ejector button
calibrated stop  thumb knob (to adjust volume)  ejection ring
blow out stop  shaft  capacity range indication

USE OF THE DISPLACEMENT PIPETTER

1. Select the pipetter whose range is appropriate for the volume you wish to measure out.
2. Set the volume as follows:
   a. Loosen the lock ring at the base of the thumb plunger so that it turns easily.
   b. Rotate the thumb knob until the desired number of µL appears in the window. [Do not go above the specified range of the pipetter or you may damage the instrument.]
   c. Snug the lock ring. (Do not over-tighten.)
3. Attach a fresh tip to the pipetter:
   a. Open the box of sterile tips.
   b. Press the pipetter firmly down onto one of the tips, tap once or twice to firm the fitting.
   c. Pull out the attached tip, immediately close the box to maintain sterility.
4. Draw up the desired volume of fluid:
   a. Before you insert the tip into the sample solution, gently depress the plunger to the calibrated (first) stop and hold in place. DO NOT GO PAST THE CALIBRATED STOP.
   b. Insert the pipetter into the sample solution so that the tip is just below the surface.
   c. Allow the plunger to slowly and smoothly return to its original position. Note that fluid is drawn up into the tip (no bubbles). NEVER LET FLUID ENTER PIPETTER BODY.
   d. Withdraw the pipetter from the sample vessel, flame and re-cap vessel to maintain sterility.
5. Deliver the fluid into the receiving vessel:
   a. Insert the pipet into the receiving vessel so that the tip a: is 3 mm below the surface of the receiving fluid, or b: touches the bottom of the empty vessel.
   b. Depress the plunger slowly and smoothly until the first stop is felt.
   c. Press more firmly on the thumb plunger to blow out the remainder of fluid. You should see a bubble expelled. KEEP THE PLUNGER HELD DOWN.
   d. Withdraw the pipetter from the vessel, dragging it along the side to leave any adhered fluid.
   e. Allow the plunger to return to its normal position.
6. Discard the used tip (unless you are to deliver additional aliquots of the same fluid):
   a. Hold the pipetter over a discard vessel.
   b. Depress the tip eject button with the thumb until the tip is ejected.

PRACTICE USING THE PIPETTER'S FEATURES

Per desk for practice by two students:

<table>
<thead>
<tr>
<th></th>
<th>20-200 µL pipetters</th>
<th>2+ non-sterile tips in a tray</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>2 x 13 x 100 mm test tubes, half full of distilled water</td>
<td></td>
</tr>
<tr>
<td></td>
<td>4 x 13 x 100 mm test tubes, empty</td>
<td></td>
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</tbody>
</table>

1) Loosen the lock ring, select the volume by rotating the thumb knob. **Do NOT exceed the range of the pipet as indicated on the handle top.** Relock (gently).
2) Gently press the plunger to the first calibrated stop, press more firmly to feel the blow out stop.
3) Pick up a tip from a non-sterile practice box by tapping the pipetter firmly once or twice into the tip, then discharge the tip to a different hole in the box by pressing the tip ejector button.
4) Set the pipetter to 100 µL, and draw up this volume of H₂O₂, deliver into a clean test tube. Change setting to 10 µL, deliver to a second clean test tube. Note how small a volume this is.