1. Metric conversions: complete the following

a. 1 ml = <u>1000</u> microliters	$d. 1 \mu l = \underline{O}, \underline{O} \circ \underline{I} m l$
b. 10 microliters = <u>O⋅O∫</u> ml	e. 20 μl = <u>(). () 2</u> ml
c. 100 microliters = 0.700 ml	$f. 2 ml = 2000 \mu l$

- 2. Put the following volumes in order from largest to smallest.
 - 2.5 ml, 250 µl, 0.025 ml, 2.5 µl 2.5 nl 250 µl, 0.025 ml, 2.5 µl
 - b. 100 µl, 0.01 ml, 250 µl, 0.015 ml 250 µl, 100 µl, 0.015 ml, 0.01 ml
- 3. Explain the reason for each of the following rules:
 - a. Always use the micropipet within its designated range:

Using it out of range can damage + break the pipet.

b. Always use a disposable tip on a micropipet:

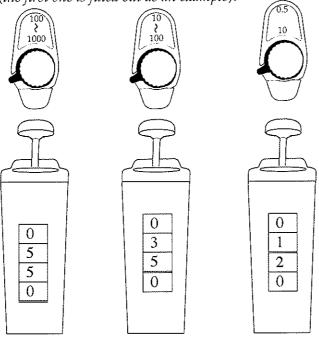
to be able to change tips - this prevents cross contamination.
c. Always hold a loaded micropipet in a vertical position:

So fluid down't leak into the piston.

d. Always release the micropipet plunger slowly:

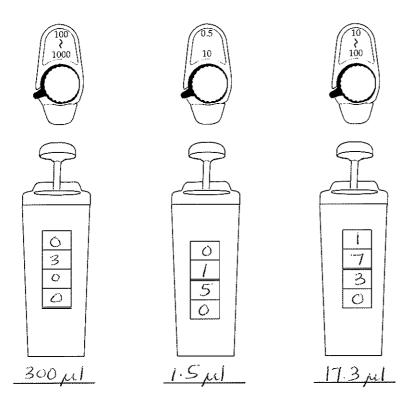
To avoid sucking liquid into the pipet shoft.

4. Under each micropipet, indicate the size of the micropipet, the set volume and the range for that micropipet (the first one is filled out as an example).



Size (P-?)	P-1000	P-100	P-10
Volume:	550 μl	35 ml	1.2 pl
Range:	$100 - 1000 \mu$ l	10-100 pd	0.5-10 pd

5. Select the appropriate micropipet and show what the dial should read to measure each of the following volumes: 1.5 μ l, 300 μ l, and 17.3 μ l. Also write the amount on the line beneath each drawing.



6. Volume comparisons

Practice measuring 1, 5, 10, 20, 100, and 500 μ l. On a piece of wax paper or Parafilm release the drops and visually compare the sizes. Draw the actual size of each droplet on the chart below. (If you wet the lab table before setting the wax paper down, the paper will not curl up as much.)

Work on your technique. Be smooth. Try to pick up and dispense the same drop several times

1.0 μl	5.0 μl	10.0 μl
ć	O	O
20.0 μ1	100 μl	500 μl
0		

7. Put one drop from an eyedropper or plastic transfer pipet on the piece of wax paper away from your other drops. **Estimate** its size in μ l. _____

8. Microcentrifuge instructions

- Close the caps on the tubes.
- •The rotor must always be balanced—you cannot, for example, insert only one tube into a microcentrifuge (microfuge). Spinning in an unbalanced arrangement like this would damage the motor of the instrument. Use an extra balance tube if necessary.
- After you have closed the lid of the centrifuge, give the tubes a 1-3 second spin. This will mix and pool all the reagents into a droplet in the bottom of each tube.
- Wait for the rotor to stop before opening the centrifuge lid.
- 9. Why is it important to balance a centrifuge before turning it on?

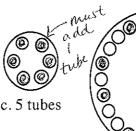
To avoid damaging the motor of the instrument

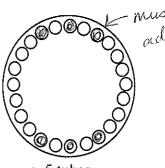
10. Show how you would arrange the given number of tubes in each centrifuge to balance the load. If you decide that you must add or remove tubes, explain.





b. 4 tubes





add I tube e. 5 tubes

11. Measurement Matrix

Label three empty microtubes A, B, and C, with a permanent marker pen.

Add solutions I, II, III, and IV to tubes A-C as shown in the matrix. To help you stay organized and prevent cross contamination, after pipetting each liquid place a check mark next to that space on your

Always use a new tip for each new liquid being added to the tubes or when adding a new liquid to a tube already containing some.

Micropipet Matrix

d. 10 tubes

Tube	Micropipet	Solution I	Solution II	Solution III	Solution IV	Total
A	P-10	$1.8~\mu l$	$4.1~\mu l$	3.7 μl		
В	P-100	34 μl	-	26 μl	12 μl	
С	P-1000	125 μl	142 μl		150 μl	

Spin the tubes in the microcentrifuge (microfuge) for a few seconds to pool the solutions. See centrifuge instructions in 8 above.

As a check of your pipetting accuracy, do the following exercise. Set the P-10 to 9.6 µl. Slowly attempt to suck in all of the fluid in tube A. The contents should just fill the tip--no air space at the bottom of the tip, no leftover fluid in the tube. Repeat with tube B and the P-100 (total 72 μ l) and with tube C and the \tilde{P} -1000 (total 417 μ l).

OBSERVATION

12. What is the approximate volume of a microcentrifuge tube? $1500 \mu l$

13. If practice agarose gels are available:

Load 15µl from tubes B and C and all of tube A into separate wells. How much fluid do the gel wells hold? 15 20 LU Use an empty well to find out. You may flush out the wells and load them again for practice.