BIO 3320

ADVANCED HUMAN PHYSIOLOGY LABORATORY MANUAL

By Dr. C. Hays, Dr. A. Rao, Dr. P. Stranahan and Dr. J. Simpson
Illustrated by Dr. A. Rao

Department of Biology
Metropolitan State College of Denver

TABLE OF CONTENTS

*Note - the asterisk means that these labs require formal laboratory reports

PREFACE

The experiments and exercises in this laboratory manual will allow you to experience, first hand, the physiological principles that are presented in lectures. The experiments will involve the usage of some expensive equipment and computers. Care must be taken to be careful with the equipment and to clean up thoroughly after you complete your experiment.

In order to maximize the chance of success with the experiments, you **MUST** read the required reading material **BEFORE** coming to lab. This will provide you with the background knowledge necessary to perform the lab correctly and with precision. You must answer the background concept questions BEFORE lab. Have an idea what to expect before performing the experiment. Always plan ahead. Record all results immediately - do not rely on your memory to fill in your results later. Even with the utmost of care in performing the experiments, you won't always get classical textbook results. There are many sources of error, such as equipment error, set-up errors or variability of the subject. The results that you obtain should be interpreted and compared with the results that others obtain. Understand why your results may differ.

You should be able to answer all questions and objectives listed for each laboratory. Often, the answers are obtained only after your required reading and may require additional research beyond your textbook. Some questions will be answered as you perform the laboratory experiments. These questions are good examples of what to expect on laboratory examinations.

LABORATORY SAFETY

1. No food or drink or smoking is allowed in lab. Tie back long hair, restrain loose clothing and jewelry.
2. Locate exits, fire extinguisher, first aid kit, sharps container and eyewash station in your laboratory.
3. There are NO open lab hours. All labs can be done within the allotted amount of time, providing you prepare for the lab.
4. Some labs will use student subjects. NO ONE IS REQUIRED TO BE A SUBJECT. Do not volunteer if you have a heart or respiratory disorder. If the subject ever feels any discomfort as part of the procedure, stop the experiment immediately and inform the instructor. Inform your instructor if you have a special medical condition on special medication or are pregnant.
5. Do not use your mouth to ingest chemicals or pipette any substance.
6. Wash your hands thoroughly after each lab. Additionally, clean your work area after each lab.
7. Inform the instructor IMMEDIATELY if you spill any chemicals on yourself or in your eyes or in your mouth. Inform the instructor if any chemical is spilled anywhere in the room.
8. Safety glasses must be used for all experiments involving chemical solutions.
9. Disposable gloves must be worn when handling body fluids (e.g. blood lab, urinalysis).
10. Consult with the instructor for instructions when disposing of sharp, or contaminated supplies, or for the disposal of chemicals.

FORMAL LABORATORY REPORT FORMAT
Due exactly one week from laboratory!
Reports must be typed.

I. INTRODUCTION

Describe the purpose of the experiment - i.e. what you are trying to demonstrate. Include in this section any pertinent background information necessary to perform the experiment.

II. MATERIALS AND METHODS

Describe the procedure you used to set up the experiment, sketch the way the subject was connected to the equipment, and HOW you conducted the experiment.

III. RESULTS

This section should include all results obtained from the experiment. Use a graph as applicable. Include any source of problems you had in conducting the experiment.

IV. DISCUSSION

Analyze and evaluate your data. Compare your data to results found in reference books. Explain why your results may be different than "textbook" results. Did errors occur? If you could redo the experiment, would you do anything differently? If so, what?

V. REFERENCES

Include a bibliography of references you used. A minimum of two references are required.

FUNDAMENTALS OF CHEMISTRY

I. Required Reading: Review your Chemistry Notes, as needed.

II. Terminology:

Atmospheres
Ionization constant
Millimole
Molar
Mole
Osmolality
Osmolar concentration
Osmolarity
Osmole
Osmotic pressure
pH

III. Objectives:

1. Memorize metric units of length, volume and mass. Be able to perform conversions.
2. Understand solution concentrations when expressed as percent solutions.
3. Define and be able to incorporate the terms moles, millimoles, and molar concentration when working problems.
4. Define osmotic pressure. Understand all units used when discussing osmotic pressure.
5. Define osmolality and osmolarity.
6. Perform conversions between atmospheres and mm Hg.
7. Explain what pH measures. Be able to interpret pH values.

IV. Background Information and Concept Questions:

<u>Units of Measurement</u>

The following are conversions to the metric system and commonly used metric equivalents.

Length:
1 inch = 2.54 centimeters (cm)
1 centimeter = 10 millimeters (mm)
1 millimeter = 1000 micrometers (um)
1 micrometer = 1000 nanometers (nm)
1 micrometer = 10,000 Angstrom Units (A)
1 centimeter = 0.01 meters (m)
1 decimeter (dm) = 0.1 meters

Volume:
2.11 pints = 1 liter (L)
33.8 fluid ounces = 1 liter
1 liter = 1000 milliliters (ml)
1 milliliter = 1 cubic centimeter (cm^3 or cc)
1 microliter = 0.001 milliliters = 0.000001 liters
1 microliter (ul) = 1 cubic millimeter (mm^3)
1 deciliter (dl) = 0.1 liters

Mass:
0.0022 pounds (lb) = 1 gram (g)
1 kilogram (kg) = 1000 grams
1 milligram (mg) = 0.001 grams
1 microgram (ug) = 0.000001 grams
1 nanogram (ng) = 10^{-9} grams
1 picogram (pg) = 10^{-12} grams

Percent Solutions

Solution concentrations may be expressed in terms of **100 units** of total volume or weight. The volume equivalent of one gram is one milliliter.

3.5% NaCl = 3.5 grams NaCl/100 ml of solution
3.5 mg% NaCl = 3.5 milligrams NaCl/100 ml of solution

Moles and millimoles

A mole refers to Avogadro's number (6.0228×10^{23}) of molecules. The molecular weight of a substance is the weight, in grams, of one mole of that substance.

1 mole = grams solute/molecular weight of that solute
1 mole = 1000 millimoles (mmole)
1 micromole = 10^{-6} moles

1 mole of NaCl = 58.5 grams of NaCl
1 mole of atomic nitrogen = 14.0 grams nitrogen
1 mole of O_2 = 32.0 grams of O_2

Molar concentrations

Quantity of solute in 1 liter of solution can be expressed with the term molar.

1 Molar solution = 1 mole/liter of solution
Molar concentration = grams of solute/liter/molecular weight

Osmotic pressure

Osmotic pressure is the amount of pressure required to prevent osmosis. It is an indirect measurement of the water and solute concentrations of a solution. A solution with a high osmotic pressure would have a high solute concentration and low water concentration. Units used are osmoles, atmospheres and mm Hg.

1 osmole = 1 mole of solute particles.

If a molecule dissociates into two ions (2 particles), then 1 mole = 2 osmoles.

If a molecule dissociates into three ions (3 particles), then 1 mole = 3 osmoles.

Expressed in terms of concentration, 1 osmole dissolved in 1 liter of solution is a 1 osmolar solution.
Osmolar Concentration (osmoles/L) = iC

i = ionization constant
C = Molar concentration

Osmolality and Osmolarity

Osmolality is the osmolal concentration when the concentration is expressed as osmoles per kilogram of water.

Osmolarity is when the osmolal concentration is expressed as osmoles per liter of solution. This term is used more commonly in physiology.

Atmospheres and mm Hg

One mole of a non-electrolyte exerts an osmotic pressure of 22.4 atmospheres at O ^{o}C.
atm = iC (22.4); iC = osmoles/Liter, as above

mm Hg = atmospheres x 760 mm Hg/atm

1. How many centimeters are there in 3.5 inches?

2. How many mm are there in 9.4 cm?

3. How many cm are there in 63 mm?

4. How many micrometers are there in 0.40 cm?

5. How many cm are there in 800 Angstrom units?

6. The diameter of a gold atom is 3 Angstrom units? What is its diameter in inches?

7. Calculate the percent by weight of each element in magnesium chloride, $MgCl_2$.

Magnesium: _____ %

Chlorine: _____ %

8. What is the weight of a mole of Bromine atoms (Br)?
Of Bromine molecules (Br_2)?

9. What is meant by a solution that is 12% glucose?

10. What is meant by a solution that is 10 mg% $CaCl_2$?

11. The pituitary gland weighs 2.7 mg per 100 grams of body weight. Express this as a percent concentration.

12. How many milligrams of NaCl would you add to a beaker that is to contain a volume of 50 ml, if you wanted the NaCl to have a concentration of 10 mg%?

13. A beaker of water contains 14 mg of solute in 600 ml of solution. Express this as a percent concentration.

14. What is the molar concentration of a 0.9% NaCl solution? Answer in molar (M) AND in millimolar (mM).

15. What is the molar concentration of a 3.6% NaCl solution. Answer in molar (M).

16. What is the osmolarity (in osmoles/Liter) of a 1% NaCl solution? (MW = 58.5, i = 2) Also, try expressing the osmotic pressure in atmospheres. In mm Hg?

17. What is the osmolarity (osmoles/Liter) of a 5% glucose solution? (MW = 180, i = 1)

BUFFERS

I. Required Reading: Guyton and Hall, Textbook of Medical Physiology, 10th Edition.

II. Terminology:

Acetate buffer system
Acid
Base
Bicarbonate buffer system
Buffer
Carbonic anhydrase
Henderson-Hasselbalch equation
Metabolic acidosis
Metabolic alkalosis
pH
Phosphate buffer system
pK
Respiratory acidosis
Respiratory alkalosis

III. Objectives:

1. Describe how a buffer system works. Explain are these buffer systems so important in the body.
2. Explain, in detail, how the Bicarbonate Buffer System works. Know the equation. Explain how this buffer system works when strong acid or base is added.
3. Define metabolic acidosis and alkalosis.
4. Define respiratory acidosis and alkalosis.
5. Know the Henderson-Hasselbalch equation, and be able to use it in performing calculations.
6. Define acid and base.
7. Define pH. Explain how pH relates to the terms acid and base and how it relates to hydrogen ion concentrations. Be able to calculate a pH with given hydrogen ion concentrations.
8. Describe the role of the urinary and respiratory systems in maintaining acid-base balance.
9. Write the equations for the acetate and phosphate buffer systems.

IV. Background Information and Concept Questions:

To ensure body homeostasis, the pH of blood, intracellular and interstitial fluid must remain within a narrow range of normal limits. Regulation of hydrogen ion concentrations must be precisely regulated. When there is a change in hydrogen ion concentration, buffer systems react very quickly to minimize these changes. A buffer can reversibly bind hydrogen ions. Buffer

systems work efficiently so that either acids or bases may be added to the buffer system without markedly changing the pH.

1. Why must hydrogen ion concentrations be precisely regulated in the body?

2. Define acid and base.

3. Express hydrogen ion concentration in terms of the pH formula.

4. Calculate the pH of a solution of HCl, in which the hydrogen ion concentration is 0.0063 mole/liter.

5. Define acid and base in terms of pH.

6. What is the normal pH of arterial blood, venous blood, interstitial fluid and intracellular fluid based on values found in the textbook?

7. The most important buffer system is the bicarbonate buffer system. Write the entire equation of this system, assuming that the enzyme carbonic anhydrase is present. Explain how this buffer system might work if there was a strong acid, such as HCl was added. Explain how this buffer system might work if there was a strong base, such as NaOH added.

8. Define metabolic acidosis and metabolic alkalosis.

9. Define respiratory acidosis and respiratory alkalosis.

10. Which direction would the equilibrium shift for the bicarbonate buffer system, if you were to hold your breath (i.e. increase CO_2 levels) for as long as you could? What would happen to the pH during this condition?

11. Which direction would the equilibrium shift for the bicarbonate buffer system, if you were to hyperventilate (i.e. blow off carbon dioxide)? What would this do to the pH?

12. Which direction would the equilibrium shift for the bicarbonate buffer system, if you were to increase the bicarbonate ion concentration in the body? What would this do to the pH?

13. In the body, the kidneys regulate the bicarbonate ion concentrations in the blood, and the respiratory system regulates the carbon dioxide concentration of the blood. These two systems work together to regulate the pH of the blood. If excess acid was in the bloodstream, how might these two systems correct the condition? If excess base was in the bloodstream, how might these two systems correct the condition?

14. Explain why your rate of respiration affects the pH of your blood.

15. Increasing the carbon dioxide concentration in your body, does what to the hydrogen ion concentration? To the pH?

16. Increasing the bicarbonate ion concentration causes what change in the hydrogen ion concentration? To the pH?

17. The phosphate buffer system is composed of what two ions? What would happen if a strong acid, such as HCl, was added to the phosphate buffer system?

18. What would happen if a strong base, such as NaOH, was added to the phosphate buffer system?

19. The Henderson-Hasselbalch equation is used in physiology when studying acid-base balance of the body. It can be used to estimate the pH of buffered solutions, or to calculate the ratio needed of $[A^-]/[HA]$ in order to obtain a certain pH.

Henderson-Hasselbalch Equation:

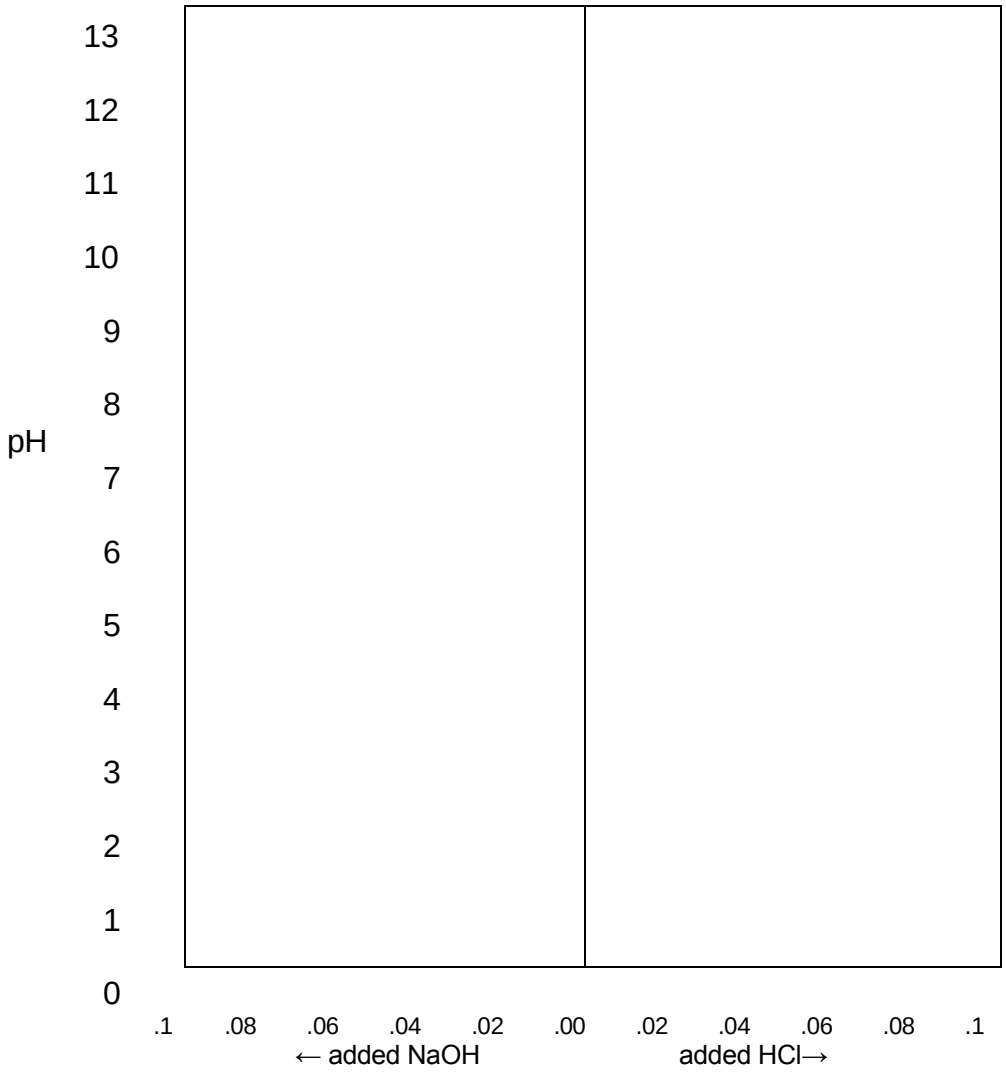
$pH = pK + \log [A^-]/[HA]$

pH = pH of buffer solution prepared from a weak acid (HA) being added to the salt, NaA.
pK = log (1/K)
K = dissociation constant of the acid
$[A^-]$ = concentration of conjugate base of the acid or salt of the acid $\{HA = H^+ + A^-\}$
[HA] = concentration of weak acid

Complete the following chart:

pH	**pK**	**[HA]**	**[A^-]**	**[Buffer]**
	7.0	20 mM	2 mM	
8.5		6 mM	100 mM	
10.1		25 mM	16 mM	
5.3	6.5	10 mM		
11.2	5.4		26 mM	26.00004 mM
7.4		30 mM	30 mM	

20. The buffering capacity of a system, based on the Henderson-Hasselbalch equation, is dependent upon the pK of the buffer system and the concentration of the weak acid and the salt of the mixture. The practical pH range of a buffer is approximately one pH unit on either side of the pK of the weak acid. Study the chart below illustration the effectiveness of an acetate buffer system, with a pK of 4.75, in maintaining pH when the pH is close to the pK of the buffer. This chart shows the effect of the acetate buffer when increasing amounts of HCl and NaOH (shown in moles per liter of mixture) are added. Compare the effectiveness of maintaining pH of the buffer, shown in solid line, with the lack of maintenance of pH without the buffer system, shown in dashed-line.



1. The Acetate Buffer System

The acetate buffer system is composed of the following weak acid, and its conjugate base:

$CH_3COOH \leftrightarrow CH_3COO^- + H^+$
(Acid) (Base)

This system reacts with a strong acid, as follows:

$HCl + CH_3COO^- \leftrightarrow CH_3COOH + Cl^-$

This system reacts with a strong base, as follows:

$NaOH + CH_3COOH \leftrightarrow CH_3COO^- + Na^+ + HOH$

Vernier Lab Pro pH Measurement Instructions

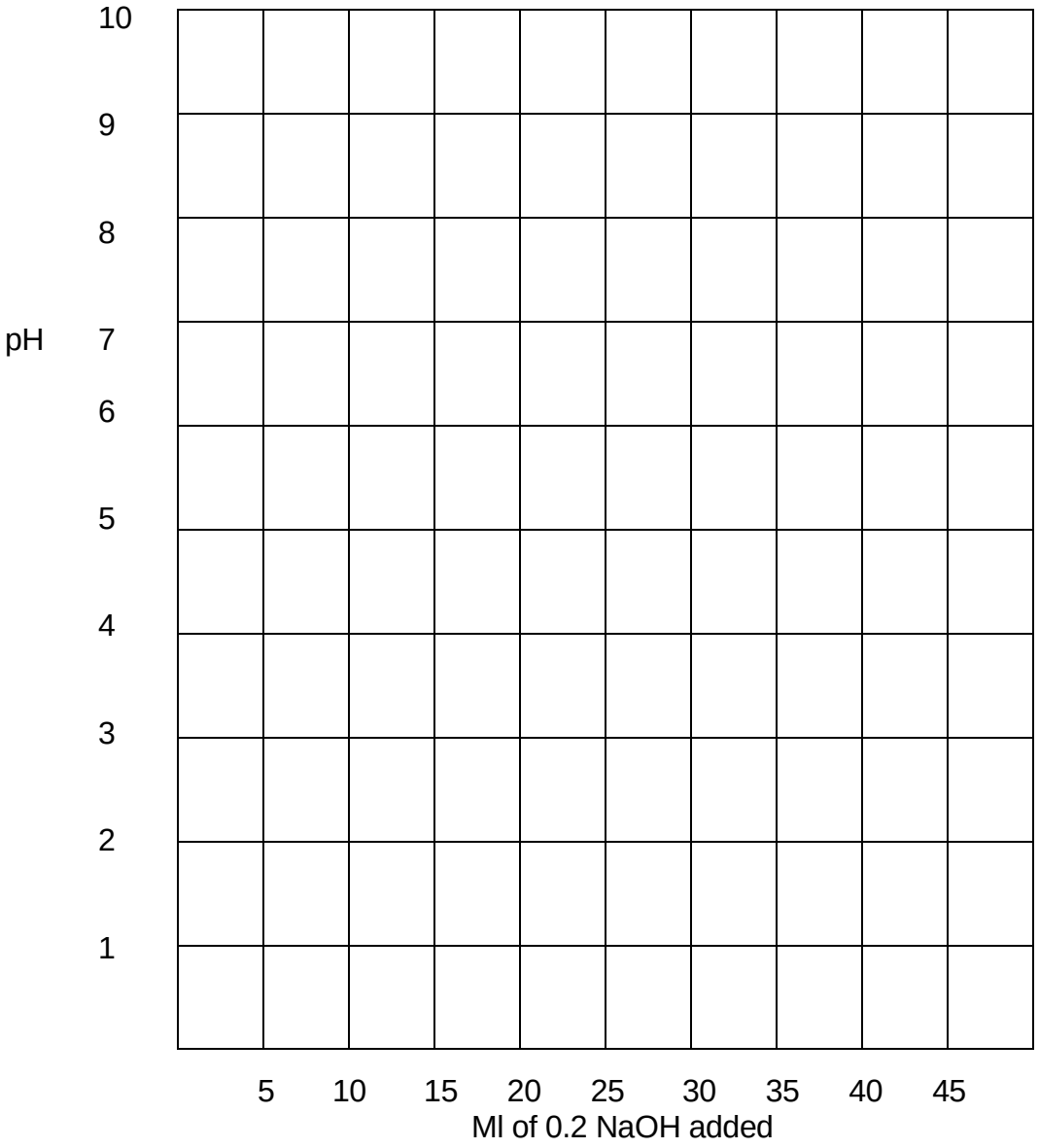
1. Click on the Logger Pro software after powering up the computer.
2. Go to Experiment on the toolbar.
3. Scroll down to Collection. Under the collection tab click on the Mode line and click on Selected Events. Click on the Use Time Column box and click Done.
4. On the toolbar in the upper right corner of the screen, click the Collect button to initiate data collection.
5. Dip the probe into the first solution. Let the pH stabilize and click the Keep button next to Stop in the upper right hand corner of the screen.
6. Remove the probe from the solution and rinse with deionized water.
7. *Do not click the stop button until all solutions are analyzed.*
8. Continue with this procedure, measuring the pH of each solution until you are finished.
9. Now click on the Stop button to finish data collection
10. Under the Analyze button scroll down and click on Zoom Graph Out to observe your data.

a. Pour 50 ml of 0.1 M acetic acid into a 250 ml beaker. Gently stir the solution, then use the pH meter to determine the pH of the solution. Carefully lower the specially prepared electrode into the solution. Do not let the electrode tip tough any solid surface. Then record the pH and turn the meter off or to "standby". Always rinse the electrode with distilled water after usage. Keep the electrode immersed in solution, except when it is being moved to another solution.

b. Then, add 4.0 ml aliquots of 0.2 M NaOH to the acetic acid solution, gently stir and record the pH after each 4.0 ml addition. Record your results below. Stop when pH reaches 10 or above.

Amount of 0.2 M NaOH	pH of Acetic Acid Solution
0	
4	
8	
12	
16	
20	
24	
28	
32	
36	
40	

c. Plot your results:



d. From your plotted results, determine the pH when the ratio of base/acid is 1.0. This is the inflection point of the graph. This pH is the pK of the acetate buffer system.

pK of acetate buffer system:

e. The actual pK of acetate buffer system is 4.75, how do your results compare to the actual value. Explain any discrepancies.

B. The Bicarbonate Buffer System

The bicarbonate buffer system involves the following weak acid and conjugate base:

$H_2CO_3 \leftrightarrow HCO_3^- + H^+$
(acid) (base)

This system reacts with a strong acid, as follows:

$HCl + CHO_3^- \leftrightarrow H_2CO_3 + Cl^-$

This system reacts with a strong base, as follows:

$NaOH + H_2CO_3 \leftrightarrow HCO_3^- + Na^+ + HOH$

CELL TRANSPORT MECHANISMS

I. Terminology:

Active transport
Brownian motion
Co-transport
Diffusion
Facilitated diffusion
Hyperosmotic
Hypertonic
Hypo-osmotic
Hypotonic
Kelvin
Ionization constant
Isosmotic
Isotonic
Osmolality
Osmolarity
Osmole
Osmosis
Osmotic pressure

II. Objectives:

1. Recall and define concepts of diffusion, facilitated diffusion, osmosis, osmotic pressure, active transport and co-transport.
2. Compare isotonic, hypotonic, and hypertonic solutions as to their effect on a red blood cell.
3. Account for the processes (what is the driving force) that allow movement of substances across the cell membrane by all of the mechanisms listed in number 1.
4. Given appropriate information on concentration gradients, determine which way substances will move through a selectively permeable membrane.
5. Perform calculations determining osmotic pressure and osmolarity.
6. Perform conversions between units of atmospheres and mm Hg.

III. Background Information and Concept Questions:

1. Diagram the structure of a cell membrane.

2. List four factors that affect the net rate of diffusion across a cell membrane.

3. Define osmosis. Give an example of osmosis occurring in the body.

4. Define osmotic pressure. If the concentration of solutes is greater on side A of a membrane, and lower on side B of a membrane, which side has higher osmotic pressure?

5. Define osmole, osmolality and osmolarity.

6. Osmotic pressure depends on the absolute temperature which influences the rate of molecular movement and upon the difference in concentration (osmoles/liter) between the two sides of the membrane. Expressed mathematically,

$\pi = \Delta CRT$

π = osmotic pressure in atmospheres
(atmospheres x 760 mm Hg/atm = mm Hg)
Δ C = concentration difference across the membrane expressed as osmoles/liter of solution
R = .082 liter atm/osmole (the ideal gas constant)
T = Kelvin = 273 + ^{o}C

7. Osmolarity = i x molar concentration

i = ionization constant = number of moles of particles resulting when one mole of the compound is dissolved in water.

Compound	i	Molecular Weight
NaCl	1.8	58.5
Glucose	1.0	180
Sucrose	1.0	342
Urea	1.0	60

If NaCl is mixed with water, complete dissociation of sodium and chloride ions occurs. If i = 1.8, this means that 1.8 moles of particles result from each mole of NaCl added to water. If glucose does not dissociate when mixed with water, 1.0 mole of glucose gives _____ mole(s) of particles.

8. What happens to a red blood cell if it is placed in an isotonic solution? A hypertonic solution? A hypotonic solution? How do these terms differ from the terms isosmotic, hyperosmotic, hypo-osmotic?

9. What is the approximate osmolarity of plasma? Interstitial fluid? Intracellular fluid?

10. Which ions comprise the majority of the total osmolarity of plasma? Interstitial fluid? Intracellular fluid?

C. Osmosis

Osmosis in animal cells

Osmosis can be demonstrated with red blood cells. The cell may hold its shape when placed in an isotonic solution, or swell - even to the point of bursting when placed in a hypotonic solution, or shrink (crenate) when placed in a hypertonic solution

1. Obtain 3 microscope slides and divide them in half with a grease pencil line. In the corner, label the six slide areas a-f. Obtain 6 cover slips and 12 toothpicks.

2. Place a SMALL drop (a toothpick will aid in obtaining a small drop) of each of the following solutions on the slide with the corresponding label.

a = 0.015 M NaCl (0.09%)
b = 0.15 M NaCl (0.9%)
c = 0.6 M NaCl (3.6%)
d = 0.15 M Sucrose (5%)
e = 0.30 M Sucrose (10%)
f = 0.30 M Urea (1.8%)

3. Dip a toothpick into the blood sample and stir blood into the solutions. Use a new toothpick each time. Cover with a cover slip.

4. Observe the red blood cells on the slide under 40 x objective. Record your results: [Hint - for the NORMAL size and shape of RBC, observe solution "b" first]

Solution	Size/Shape of RBC	Isotonic	Hypotonic	Hypertonic
a				
b				
c				
d				
e				
f				

5. Which solutions prove to be isotonic to red blood cells?

6. If you see NO cells in a solution, what does this tell you about the solution?

3. Calculate the osmolarity of each solution.

Osmolarity of 0.3 M Sucrose:

Osmolarity of 1.2 M Sucrose:

4. Calculate the osmotic pressure in mm Hg developed by each solution.

Osmotic pressure for 0.3 M Sucrose: made

Osmotic pressure for 1.2 M Sucrose:

5. Why was the rate of osmosis different between the 0.3 M sucrose and the 1.2 M Sucrose?

DNA FINGERPRINTING LAB

Student Manual Background Information

I. Terminolgy
   DNA Base Pairs
   Introns
   RFLPs
   Gel Electrophoresis
   Probe

We are in an exciting period in the history of man – the era of biotechnology. This term, translated literally as “life technology” applies our knowledge of living organism for the practical and beneficial use of mankind. While we tend to think of biotechnology as a modern buzzword, man has for thousand of years, utilized biotechnology in such activities as making bread, brewing, wine making, the production of soap, the treatment of waste and the enhancement of flavor in foods such as cheese. Today, the knowledge and techniques of molecular biology are being applied to almost every field of biology - from paleobiology to botany, immunology to neurophysiology. Today’s buzzwords include gene cloning, gene splicing, genetic engineering and recombinant DNA.
The ever-growing biotechnology industry is profoundly influencing our lives and culture. Microorganisms have been programmed as mini-factories to produce (more efficiently and less costly) drugs such as insulin (Humalin), growth hormone, hepatitis vaccine (Recombivax and Engerix-B), interferon, and interleukin-I. Genen transfers enable us to produce plants having resistance to frost, drought and many parasites and pathogens.

The worldwide effort to map the human genome has far-ranging implications for diagnosing and treating genetic disease. The techniques of genetic fingerprinting are being used to isolate and identify the disease genes of Huntington’s chorea, Duschenne’s Muscular Dystrophy, cystic fibrosis and the gene responsible for Neurofibromatosis (the famed Elephant’s Man disease). DNA fingerprinting techniques are also being used to test human cell lines for authenticity, determine animal pedigrees and to identify humans in cases involving disaster, paternity and crimes.

DNA FINGERPRINTING

Humans have a huge number of base pairs of DNA, approximately 3 billion. Only 2-3% of these base pairs actually code for genes. The function of the remaining DNA, called introns or intergenic sequences, is unknown. Scientists have discovered that most of the intergenic DNA consists of sequences that tend to repeat many times throughout the DNA. These repetitive sequences are called Restriction Fragment Length polymorphisms, or RFLP’s. Mutation within the RFLP sequences cause variation from person to person. The sequence, length

and number of these repeating fragments are as unique to each person as the fingerprints on their hands. When restriction enzymes are used to cut RFLP sequences, a collection of fragments is produced that is unique to each individual. Since only one base difference can alter the restriction site location, DNA identification can be quite accurate. The likelihood of two persons, other than identical twins, having the same RFLP patterns is 1 in 30 billion.
Analyasis of DNA band patterns in a criminal case is a comparative technique. The DNA band patterns generated from the specimens collected from the scene of the crime are compared with DNA taken from the suspects.
The DNA fingerprinting process begins by taking the typically small amount of DNA extracted from evidence (tissue, hair follicle, Blood), duplicating or amplifying it millions of times, cutting it into fragments using specific restriction enzymes. The DNA fragments, RFLP's , are then separated by size using gel electrophoresis which produces band patterns that are unique to each individual. These band patterns are like human "bar codes", resembling the bar codes seen on products in grocery store. After electrophoresis, the DNA in the gel is denatured with heat or high pH to separate the double stranded helix. This allows the DNA to be blotted onto a membrane that will serve as a replica of the gel pattern. The membrane is exposed to a probe, a specific single stranded sequence of DNA that is tagged, or labeled, with a radioactive isotope. The probe hybridizes, or binds only to fragments on the membrane that contain DNA sequences that are complimentary to the sequence of the probe. The radioactive label makes it possible to detect the size and position of the fragments by exposing the membrane to x-ray film. Only the fragments on the membrane that have hybridized to the radioactive probe are visualized as a line or band on the x-ray film. It is this probed, electrophoretic pattern that holds the power to convict or eliminate a subject.

It is important to understand that the procedures performed in this laboratory are only a portion of the complete DNA fingerprinting process.

Information taken from Fotodyne Safekit 105 & 106 used for this lab.

MEMBRANE POTENTIALS and ACTION POTENTIALS

I. Required Reading: Guyton and Hall, Textbook of Medical Physiology, 9th Edition, p. 43-66.

II. Terminology:

Action Potential
Conductance
Depolarization
Hyperpolarization
Membrane Potential
mhos
Millivolt
Nernst Potential
Nerve
Nerve impulse
Neuron
Permeability
Refractory Period (absolute and refractory)
Repolarization
Sodium-Potassium pump
Threshold
Voltage-regulated ion gate

III. Objectives:

1. Recall the concepts of diffusion, facilitated diffusion, osmosis, osmotic pressure, active transport and cotransport.
2. Understand the sodium/potassium pump and how it contributes to the resting membrane potential.
3. Know values for intracellular and extracellular fluid for K^+, Na^+, Cl^-, and HCO_3^-.
4. Explain the "all or none" phenomenon as it relates to an action potential.
5. Describe the stages of an action potential.
6. Understand what is meant by voltage-regulated ion gates.
7. Compare the length of time that the Na^+ channels are open to that of K^+ channels for an action potential.
8. Memorize the Nernst Equation and the equation used to calculate membrane potentials relative to membrane permeability. Understand the application of these two equations.
9. Explain the factors contributing to the maintenance of a resting membrane potential.
10. Explain the significance of refractory periods (absolute and relative).

IV. Background Information and Concept Questions:

1. Diagram the structure of a cell membrane.

2. What is meant by "gating" of protein channels in reference to the passage of sodium and potassium ions across the cell membrane?

3. Draw a diagram of the sodium-potassium pump in action.

4. List four factors that affect the net rate of diffusion across a cell membrane.

5. List the chemical composition of extracellular and intracellular fluids of the following ions. Use the Guyton & Hall text as your source.

Ion	Extracellular fluid	Intracellular Fluid
K^+		
Na^+		
Cl^-		
HCO_3^-		

6. The Nernst Equation is used to calculate the Nernst or equilibrium potential for any ion. The Nernst potential refers to the electrical potential required to prevent diffusion of an ion across the cell membrane. The Nernst potential of each ion on each side of the membrane contributes a portion to the total membrane potential. Nernst potentials must always be considered in combination with the permeability of the membrane to each ion. For example, if the cell membrane is not permeable to a particular ion, the concentration difference of that ion across the cell membrane does NOT contribute to the total membrane potential. What is the Nernst equation for a univalent ion (such as sodium or potassium) at normal body temperature? What does this equation demonstrate? [When using this equation, we assume that the Nernst potential is being calculated for the inside of the membrane. The sign of the potential is positive if the ion being considered is a negative ion; and negative, if the ion being considered is a positive ion. If dealing with an ion with a valency other than one, divide 61 by the valency of that ion. e.g. 61/2 for calcium^{2+}.]

7. The resting membrane potential of a cell depends on the concentration difference of important ions across the cell membrane as well as the permeability of the membrane to the passage of the ions. If a cell membrane were only permeable to potassium ions, use the Nernst equation to calculate the membrane potential caused by diffusion. (Use values from your chart on #5) Explain your answer. What does the negative refer to in your answer. If the cell membrane were only permeable to sodium ions, use the Nernst equation to calculate the membrane potential caused by diffusion? Explain your answer. What does the positive mean in your answer?

8. Again, many ions contribute to the total membrane potential. In calculating the membrane potential, one must consider the concentrations of the ions across the cell membrane, the permeability of the membrane to each ion, and the polarity of the electrical charges of each ion (positive ions moving towards negative areas and negative ions moving towards positive areas). Conductance (g, with units of mhos/cm^2 - mho is pronounced mo, and is "ohm" backwards; a unit of conductance) can be used to indicate the membrane permeability to each ion. The higher the conductance, the more permeable the membrane is to that ion. Thus, the membrane potential can be calculated by the following equation:

$$EMF\ (mV) = \frac{g_K EMF_K + g_{Na} EMF_{Na} + g_{Cl} EMF_{Cl} + g_{Ca} EMF_{Ca} \ldots}{g_K + g_{Na} + g_{Cl} + g_{Ca}}$$

9. How does the sodium/potassium pump contribute to the resting membrane potential?

10. If depolarization reaches a threshold level, the membrane will depolarize completely as an "all or none" state, termed an action potential. Diagram a typical action potential illustrating the relationship of millivolts and time. Include the threshold level in your diagram.

11. Within 1 to 5 milliseconds after depolarization, the resting membrane potential is restored. Explain the events responsible for this repolarization.

Membrane Potential Calculations

1. Assume a cell membrane contains 100 mM KCl inside of the cell and 10 mM KCl outside of the cell.
   a. Calculate the Nernst Potential for potassium ions at normal body temperature.

   b. Calculate the Nernst Potential for chloride ions at normal body temperature.

2. Assume that the above cell membrane is permeable ONLY to potassium ions. Explain how chloride ions contribute to the membrane potential.

3. What would the actual, measured membrane potential of the above cell be, due to diffusion? (The cell is only permeable to potassium ions.)

[Potassium ions will diffuse until the electrical potential build up and prevents continued diffusion. Additionally, as the potassium ions diffuse out of the cell, a small excess of positive ions collect near the membrane and repels further transfer of positively charged ions into the region.]

4. Complete the following table:

Concentration (mMKCl)		Calculated Nernst Potential	
Inside	**Outside**	**EMF_K**	**EMF_{Cl}**
1	100		
3.2	100		
10	100		
32	100		
100	100		
320	100		

5. What would the actual measured membrane potentials be in the above problem, if the membrane were a cation selective membrane (only cations can pass)? What would the actual measured membrane potentials be in the above problem, if the membrane were an anion selective membrane?

6. Fill in the Nernst Potentials on the following table:

Ions	**Inside**	**Outside**	**Nernst Potential**
Na^+	10 mM	120 mM	EMF_{Na}=
Cl^-	5 mM	100 mM	$EMF_{Cl=}$
K^+	150 mM	5 mM	EMF_K=
Ca^{2+}	.01 mM	5 mM	EMF_{Ca}=

7. Calculate the above (#6) cell's membrane potential with the following conductance information:
g_{Na} = 1 umho/cm^2
g_{Cl} = 40 umho/cm^2
g_K = 50 umho/cm^2
g_{Ca} = 0 umho/cm^2

8. Using the same conductance values in #7, calculate the membrane potential when the potassium concentration outside of the cell changes to 2 mM, with all other values unchanged.

9. Using the same conductance values in #7, calculate the membrane potential of the same cell in #6, when the sodium ion concentration outside of the cell changes to 110 mM, with all other values unchanged.

10. Using the same Nernst Potentials calculated in #6, and the same conductance values in #7, show how increasing sodium conductance can depolarize the membrane potential, by calculating the membrane potential for a cell with a conductance, g_{Na} of 6, 60, 600 umho/cm^2.

11. To illustrate the ionic distribution during an action potential, consider a resting neuron with an intracellular concentration of 140 mM Potassium ions and 14 mM Sodium ions. Extracellular concentrations are 5 mM Potassium ions and 140 mM Sodium ions. Calculate the Nernst Potentials for Potassium and Sodium ions.

12. Upon stimulation of the above neuron, the permeability changes for the ions. Calculate the membrane potential based on the changing conductivity of the ions for each interval after stimulation.

Time after Stimulation (msec)	g_K	g_{Na}	Membrane Potential (mV)
0	0.8	0.005	
0.1	0.8	0.008	
0.2	0.9	0.1	
0.3	2.0	40.0	
0.4	10.0	10.0	
0.5	10.0	4.0	
0.6	10.0	0.1	
0.7	9.0	0.005	
0.8	5.0	0.005	
0.9	3.0	0.005	
1.0	2.0	0.005	
1.2	0.9	0.005	
1.5	0.8	0.005	

CARDIOVASCULAR PHYSIOLOGY

I. Required Reading: Guyton and Hall, Textbook of Medical Physiology,

II. Terminology:
Apical pulse
Arteriole
Artery
Atrioventricular valves
Auscultation
Bicuspid valve
Capillary
Cardiac cycle
Cardiac output
Collateral blood flow
Diastole
End-diastolic volume
End-systolic volume
Frank-Starling mechanism
Heart rate
Ischemia (ischemic)
Murmur
Pulse
Pulse deficit
Resistance
Semilunar valves
Sphygmomanometer
Stroke volume output
Systole
Thrombus
Tricuspid valve
Valsalva Maneuver
Vasodilation
Vein
Venous return
Venule

III. Objectives:
1. Name the four heart chambers. Name the four associated valves.
2. Define cardiac cycle, systole and diastole.
3. Describe and illustrate the pressure changes in the left atrium, left ventricle, and aorta through one complete cardiac cycle.
4. Define cardiac output.
5. Describe the controls of heart rate and describe how heart rate relates to cardiac output.

6. Define end-diastolic volume and factors that influence it.
7. Define end-systolic volume and factors that influence it.
8. Describe the origin of the "lub-dup" sound as your heart beats.
9. Contrast arteries, veins, capillaries, arterioles, and venules.
10. Describe the differences in blood velocity in large vs small blood vessels.
11. Explain what causes a pulse.
12. Define blood flow, blood pressure, and resistance. Describe their relationships.
13. Describe the measurement of blood pressure. What does systolic pressure correspond to? Diastolic pressure?
14. Define venous return and describe influencing factors.

IV. Background Information and Concept Questions:

1. One cardiac cycle occurs during one complete heartbeat. What are the two periods of the cardiac cycle?

2. At rest, the normal cardiac cycle lasts how long?

3. Name the four heart chambers.

4. Name the four heart valves, and describe their locations.

5. Diagram the following events of the cardiac cycle.
    a. Left atrial pressure changes
    b. Left ventricular pressure changes
    c. Aortic pressure changes

6. Based on your diagram, explain WHY the A-V and semilunar valves open and close.

7. What is the reason for the "lubb-dup" heart sounds?

8. Define end-diastolic volume, end-systolic volume and stroke volume output.

9. Define venous return. What is meant by the Frank-Starling Mechanism?

10. Define cardiac output. What is a normal resting value for cardiac output?

11. What is a normal resting heart rate expressed in beats per minute?

12. What affect does sympathetic stimulation have upon heart rate and force of heart contraction?

13. What affect does parasympathetic stimulation have upon heart rate and force of heart contraction?

14. Describe an artery, arteriole, vein, venule and capillary.

15. The velocity would be slowest in which of the above vessels.

16. Compare the blood pressures in the vessels listed in number 14.

17. What is the relationship between blood flow, pressure and resistance?

18. What unit is usually used for measuring blood pressure?

19. Describe the auscultatory method of measuring arterial pressure. What are normal values?

20. Which vessel serves as a blood reservoir for the circulation.

21. In terms of valvular malfunction, explain what causes a heart murmur.

22. A pulse is due to the expansion and recoil of a vessel wall as the heart contracts and relaxes. Which type of vessel has a palpable pulse? What is the average resting pulse for an adult?

V. Procedure:

Observe the cardiovascular animations, heart sounds, anatomy, diastole and systole on Martini's LaserDisc entitled Fundamentals of Human Anatomy and Physiology. Additionally, view the A.D.A.M. Interactive Physiology Cardiovascular CD.

A. Auscultation of heart

1. Obtain a stethoscope and clean the earpieces with an alcohol swab. Place the diaphragm of the stethoscope on the subject's chest and listen for the characteristic "lubb-dup" as the heart beats.

The first heart sound, the "lubb" is due to

The second heart sound, the "dup" is due to

2. Place the stethoscope over areas of the different heart valves to hear the individual valve sounds.

FIGURE OF VALVE LOCATION

3. Count the number of beats ("lubb-dups") in one minute.

Heart rate:

4. Which is longer, the interval between the first heart sound and the second, or the pause between the second sound of one heartbeat and the first sound of the next heartbeat?

B. Pulse

1. Several superficial points may be easily palpated to obtain a pulse. Try the following pulse points:

-Temporal artery (in the temple region)
-Common carotid artery (side of neck)
-Brachial artery (anterior elbow)
-Radial artery (lateral wrist)
-Ulnar artery (medial wrist)
-Popliteal artery (back of knee)

2. Which pulse point was strongest? Explain.

3. Which pulse pint was weakest? Explain.

4. A pulse deficit is any large difference between the actual counting of heartbeats, termed the apical pulse, and the peripheral pulse, such as the radial pulse. A large pulse deficit may be indicative of cardiac abnormalities such as arrhythmias, or cardiac failure.

To determine whether a large pulse deficit is present, one person should count the radial pulse while another counts the apical pulse for one minute on the same subject.

Radial pulse _____ pulse/min

Apical pulse _____ beats/min

Pulse deficit _____ /min

C. Blood Pressure

1. Obtain a stethoscope and swab the earpieces with alcohol. Obtain a sphygmomanometer or blood pressure cuff.

2. Measure the arterial pressure in the brachial artery by placing the cuff around the arm, above the elbow. Secure the cuff with the Velcro. Never leave the cuff inflated for longer than one minute! Place the stethoscope where you palpated the brachial pulse on the anterior elbow.

3. Tighten the cuff to 160 mm Hg. SLOWLY release the pressure valve. Watch the pressure gauge as you listen for the soft sounds of blood spurting through the partially occluded brachial artery. The first soft sound you hear is the first point at which a small amount of blood spurts through the partially occluded brachial artery. You will also notice the first "bounce" of the pressure valve needle at this point. Record this pressure as the systolic pressure.

Systolic pressure_____

Continue to release the pressure. When the artery is no longer restricted and blood flows freely, the sounds can no longer be heard and the pressure valve needle stops "bouncing". This pressure is the diastolic pressure.

Diastolic pressure_____

4. How do these values for arterial systolic and diastolic pressure relate to the pressures of the left ventricle during systole and diastole? Explain why there might be differences.

5. Are the subject's arterial pressures within normal limits?

6. Calculate the subject's pulse pressure by subtracting the diastolic pressure from the systolic pressure.

Pulse pressure _____

7. Calculate the subject's meat arterial pressure with the following equation:

MAP = diastolic pressure + pulse pressure/3

MAP _____

8. Since a sphygmomanometer only measures arterial pressure, a crude estimation will be done to determine venous pressure. Do you predict that venous pressure will be greater or lesser than arterial pressures?

9. At the chalkboard, estimate the location of the subject's right atrium, as he/she stands with his/her right side against the board. Place an "X" on the board at the location of the right atrium.

10. Observe the bulging of the veins on the dorsum of the subject's right hand, as it hangs by his/her side. The subject should slowly raise the arm, while the partner closely observes the bulging veins. At the exact moment when the veins collapse and the bulging disappears, mark this height on the board.

11. Measure the vertical distance in mm between the right atrium and the point where the veins collapsed. Determine the venous pressure with the following equation:

$$VP \text{ (in mm Hg)} = \frac{1.056 \times \text{mm measured}}{13.6}$$

1.056 is the specific gravity of blood,
13.6 is the specific gravity of mercury

VP_____

12. Based on the values you looked up in your textbook for normal venous pressures, how do your estimated venous pressures compare?

13. Because the walls of veins are so thin, increasing intrathoracic pressure will push on the walls of thoracic veins. Intrathoracic pressure may be increased by performing the Valsalva maneuver. Perform the Valsalva maneuver by taking a deep breath, then try to exhale with the glottis closed - i.e. without actually exhaling; as done when straining. Repeat the measurement of venous pressure as done above WHILE performing the Valsalva maneuver.

VP with Valsalva_____

14. Explain why this value is different than the estimation of venous pressure on number 11.

D. Various factors influencing heart rate and blood pressure

1. To illustrate the effect of nicotine, posture, exercise, and painful stimuli on heart rate and blood pressure, complete the following charts:

NICOTINE

Baseline		After smoking for 2 min		After smoking for 4 min	
BP	P	BP	P	BP	P

POSTURE

POSITION	BP	PULSE
Sitting		
Reclining		
Immediately upon standing		
After standing 3 minutes		

EXERCISE

Baseline	Immediately After exercise		2 min		3 min		after 5 min exercise	
BP	P	BP	P	BP	P	BP	P	BP	P

PAINFUL STIMULUS (Place left arm in ice water, as a painful stimulus, with cuff on right arm. Leave arm in water as measurements are performed.)

Baseline	1 min after immersion in ice water	2 min after immersion in ice water
BP ___ P _____	BP _____ P _____	BP _____ P

2. What was the effect of nicotine on blood pressure? Knowing that nicotine is a vasoconstrictor, can you account for your results?

3. In which position (sitting, reclining, standing) was the blood pressure the lowest? The highest?

4. Immediately upon standing, gravity pulls the blood downwards. If your measurement was quick enough, upper body blood pressure decreases. After the subject stood for there minutes, what changes in blood pressure were observed? What accounts for these results? Why do some people feel dizzy when the stand up too fast?

5. What as the effect of exercise on the pulse and blood pressure? Did the subject's values return to baseline levels after 3 minutes? Relate this answer to the subject's conditioning level.

6. There is variability as to how a person responds to a painful stimulus. How did the blood pressure and pulse change when the subject was immersed in the icy water?

E. Local Circulation

Vasodilation

1. Observe vasodilation of cutaneous blood vessels and skin flushing due to a build up of local metabolites by performing the following on a subject.

2. The subject should roll up both sleeves and place both forearms (posterior side down) on the table. What is the general color of the skin of the forearms? Describe the size and contour of any visible blood vessels.

3. Place a sphygmomanometer cuff on one arm, and inflate to 250 mm Hg for one minute. Record changes in this forearm (color, sensations, blood vessels) especially as they compare to the forearm without the cuff.

4. Release the cuff and observe immediate changes in the forearm and changes 1 minute after deflation.

Ischemia

5. Repeat the above experiment, but this time partially empty the blood out of the forearm before blocking blood flow. The subject should raise the cuffed arm and clench a fist as tightly as possible, while the partner rapidly inflates the cuff to 240 mm Hg. Now, the subject may relax the fist and place the forearm next to the other uncuffed forearm on the laboratory table.

6. Leave the cuff inflated for one minute. Then rapidly release the pressure. How long does it take for the forearm to return to a normal color?

Describe how the subject feels (temperature, pain, tingling, weakness...) while the cuff has stopped blood flow to the forearm for one minute.

What does the forearm look like (color, blood vessels) while the forearm is deprived of blood?

Relate this experiment to the effects of an ischemic episode in the brachial artery due to a thrombus. (Define ischemic, define thrombus).

Venous Congestion

7. Use a new subject, and place the cuff on the brachium and inflate it to 40 mm Hg. Keep the cuff inflated for five minutes and then release.

Describe the sensations felt (tingling, heat, weakness, pain...) and the physical appearance (blood vessels, color of fingertips, color of hand) of the cuffed limb during the five minutes.

Describe the sensations and physical appearance of the limb after the cuff was released.

Collateral Circulation

8. Locate the subject's radial (lateral wrist) and ulnar (medial wrist) pulses. To simulate ischemia to the hand, compress both arteries manually for five minutes while the subject rests with the elbow on the table at about a 45 degree angle. Describe the appearance of the hand as ischemia progresses.

9. Fortunately, many parts of the body have collateral circulation, or alternate routes for blood circulation if one blood vessel becomes blocked. The radial and ulnar arteries illustrate collateral circulation to the hand. Repeat the above experiment, but this time, only compress the radial artery. Did the hand appear ischemic as it did when both radial and ulnar arteries were blocked?

10. Repeat the above experiment, but this time, only compress the ulnar artery. Did the hand appear ischemic as it did when both radial and ulnar arteries were blocked? Explain the advantage of having both the radial and ulnar artery supplying blood to the hand.

ELECTROCARDIOGRAM AND VECTORCARDIOGRAM

I. Required Reading: Guyton and Hall, Textbook of Medical Physiology

II. Terminology:
Atrioventricular bundle
Atrioventricular node
Augmented unipolar limb lead
Bipolar limb lead
Chest lead
Einthoven's Law
Einthoven's triangle
Electrocardiogram
Lead
Mean electrical axis
Pacemaker
Purkinje fibers
Sinoatrial (=sinus) node
Vector
Vectorcardiogram

III. Objectives:
1. Describe the basis for electrocardiography. What are leads?
2. Describe the waves of a normal ECG in lead II, and explain what each wave represents.
3. Describe the placement of electrodes in obtaining an ECG for Leads I, II, and III.
4. Construct the mean electrical axis and a vectorcardiogram with any two standard limb leads of the ECG given.
5. Describe the initiation and conduction of impulses through the heart.

IV. Background Information and Concept Questions:

1. What and where is the sinoatrial (sinus) node?

2. Explain why the sinus node self-excites.

3. Trace the electrical pathway of conduction through the heart.

Indicate, where in the pathway, a delay occurs.

4. Why is the sinoatrial node considered to be the heart's pacemaker?

5. What are Purkinje fibers?

6. Explain the significance of one-way conduction through the A-V bundle.

Cardiac electrical impulses cause current to spread through surrounding tissues, all the way to the surface of the body. Although these electrical currents gradually diminish as they extend into surrounding fluids, they may be detected by placing electrodes on the skin. The recording of these currents is an electrocardiogram (ECG or EKG). Since the contractions of the heart, are preceded by the electrical events, the ECG may be very helpful in studying the electrical and mechanical events of the heart.

7. Sketch a normal electrocardiogram. Label the P wave, QRS complex, and T wave.

8. Explain what each of the above waves represent.

9. Fill in the normal voltages and durations for a Lead II ECG:

P wave	millivolt
QRS complex	millivolt
T wave	millivolt
P-Q or P-R interval	second
Q-T interval	second
Interval between 2 successive QRS complexes:	second
Normal heart rate (bpm) (60/above QRS complex interval = heart rate)	bpm

10. At least two electrode attachments are required for an ECG. A third position is used as a ground post. Three standard bipolar limb leads are used. Bipolar means that two electrodes are placed on the limbs. Describe the electrode placement for leads I, II, and III below:

Lead I: Negative terminal - _____
Positive terminal - _____

Lead II: Negative terminal - _____
Positive terminal - _____

Lead III:Negative terminal - _____
Positive terminal - _____

11. Sketch the triangle around the heart, called Einthoven's triangle.

12. Extending the apices of this triangle represents the vantage point that each electrode has for "seeing" electrical activity from the heart. The lower apex of the triangle extends to the left leg, the two upper apices point to the two arms. Einthoven's law states, that if any two of the three bipolar limb ECG leads are known at a given moment, the third can be determined mathematically. What is the mathematical formula for Einthoven's Law?

13. Chest leads, or precordial leads, require that the positive electrode be

placed on the anterior surface of the chest over the heart, in one of six positions, and the negative electrode must be connected to the right arm, left arm and left leg simultaneously. Using Figure 11-8, sketch the placement of the positive electrodes for leads V_1 - V_6.

14. With augmented unipolar limb leads, two limbs are connected to the ECG's negative terminal, and a third limb is connected to the positive terminal. What is the location of the positive terminal for the following leads?

aVR _____

aVL _____

aVF _____

15. Cardiac electrical potential gradient magnitude and direction can be described with vectors. According to your textbook, define vector. Include in your definition the meaning of the direction of the arrowhead and the length of the arrow. A positive vector in a lead will cause the reading of the ECG to be above the isoelectric line, and a negative vector will cause the reading of the ECG to be below the isoelectric line.

16. An electrical potential gradient has direction (angle) and magnitude.

Looking at an ECG, the information shows the gradient as projected on a fixed angle and distance view. In order to talk about direction, a reference frame must be used. With the horizontal axis as 0 and 180 degrees, draw a 360^{o} scale of vectors, indicating 0, 90, 180, 270, 360 degrees.

17. The mean electrical axis represents the preponderant direction of current flow (from negative to positive) during heart (ventricular) depolarization. Draw a diagram and indicate the mean electrical axis of the ventricles for bipolar limb leads I, II, and III.

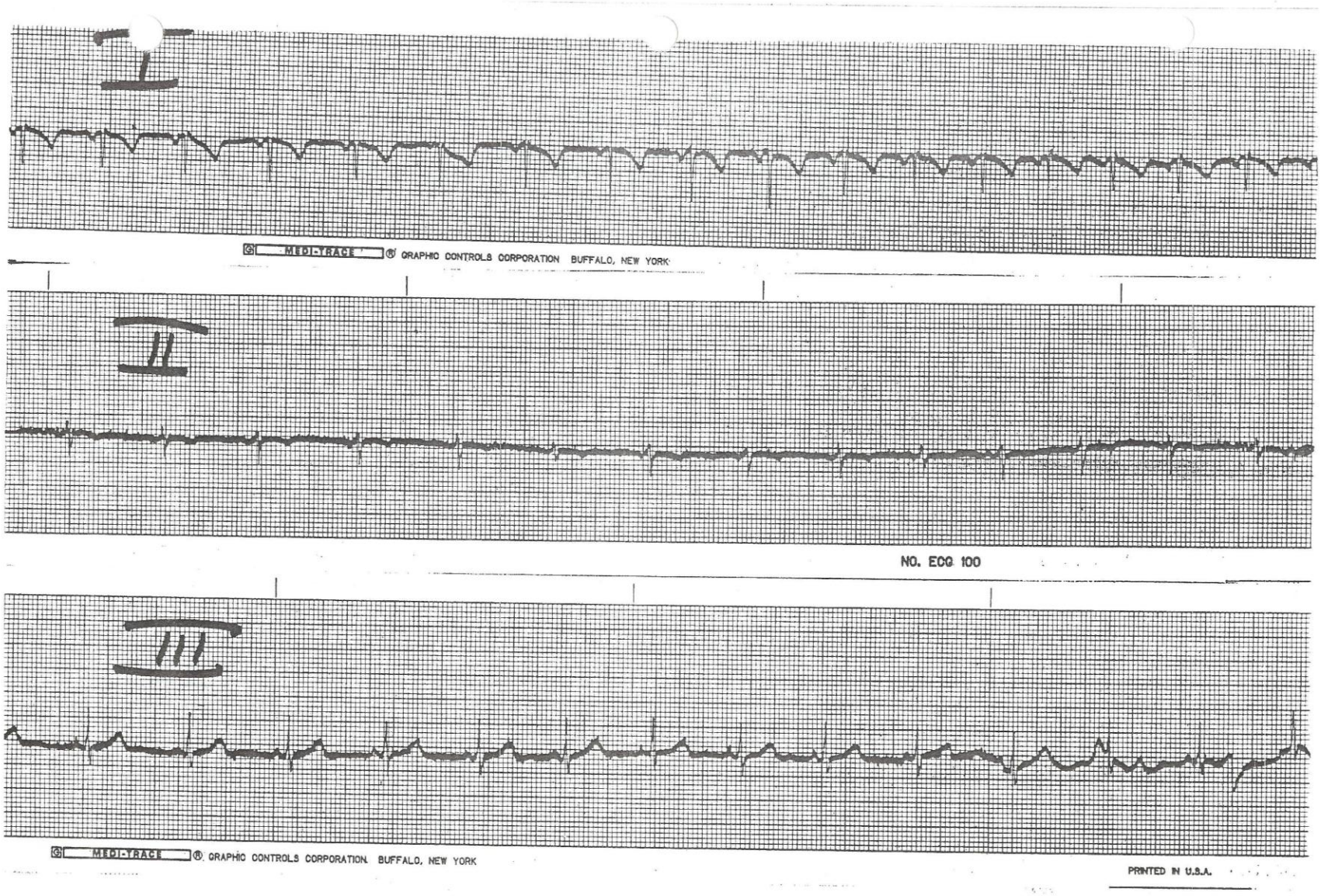
EKG Instructions
For the Vernier LabPro

1) Connect one end of the USB cable to the Vernier LabPro module and the other end to the USB port on the back of the computer.
2) Connect the EKG analog sensor cable to channel 1 on the LabPro module.
3) Plug the Vernier LabPro module into a power source.
4) Plug the computer into a power source and turn it on.

5) Place one electrode gel patch on the left ankle, one on the right ankle, one on the right wrist and one on the left wrist. (Remove your watch during this portion of the exercise).
6) Electrode placement for each lead:

Lead I:	green-	right wrist
	red-	left wrist
	black-	right ankle
Lead II:	green-	right wrist
	red-	left ankle
	black-	right ankle
Lead III:	green-	left wrist
	red-	left ankle
	black-	right ankle

7) Click on the Logger Pro Icon on the desktop.
8) Close the Tip of the Day and you are ready to start.
9) Under Experiment on the tool bar go to Extend Collection. Click on this two separate times to extend the collection of data set to 10.125 seconds.
10) Click on the Collect button in the upper right hand corner of the screen to start collection of data.
11) Between subjects or when changing leads go to experiment and click on Clear Latest Run to erase the previous set of data collection.


I
MEDI-TRACE ® GRAPHIC CONTROLS CORPORATION BUFFALO, NEW YORK
II
NO. ECG 100
III
MEDI-TRACE ® GRAPHIC CONTROLS CORPORATION BUFFALO, NEW YORK
PRINTED IN U.S.A.

10/28/1999 05:13:25
70 years Male

PSL MEDICAL CENTER
Dept: ICU
Room: 11
Oper: JLW

Rate	64
PR	220
QRSD	103
QT	419
QTc	432

--AXIS--

P	8
QRS	-5
T	116

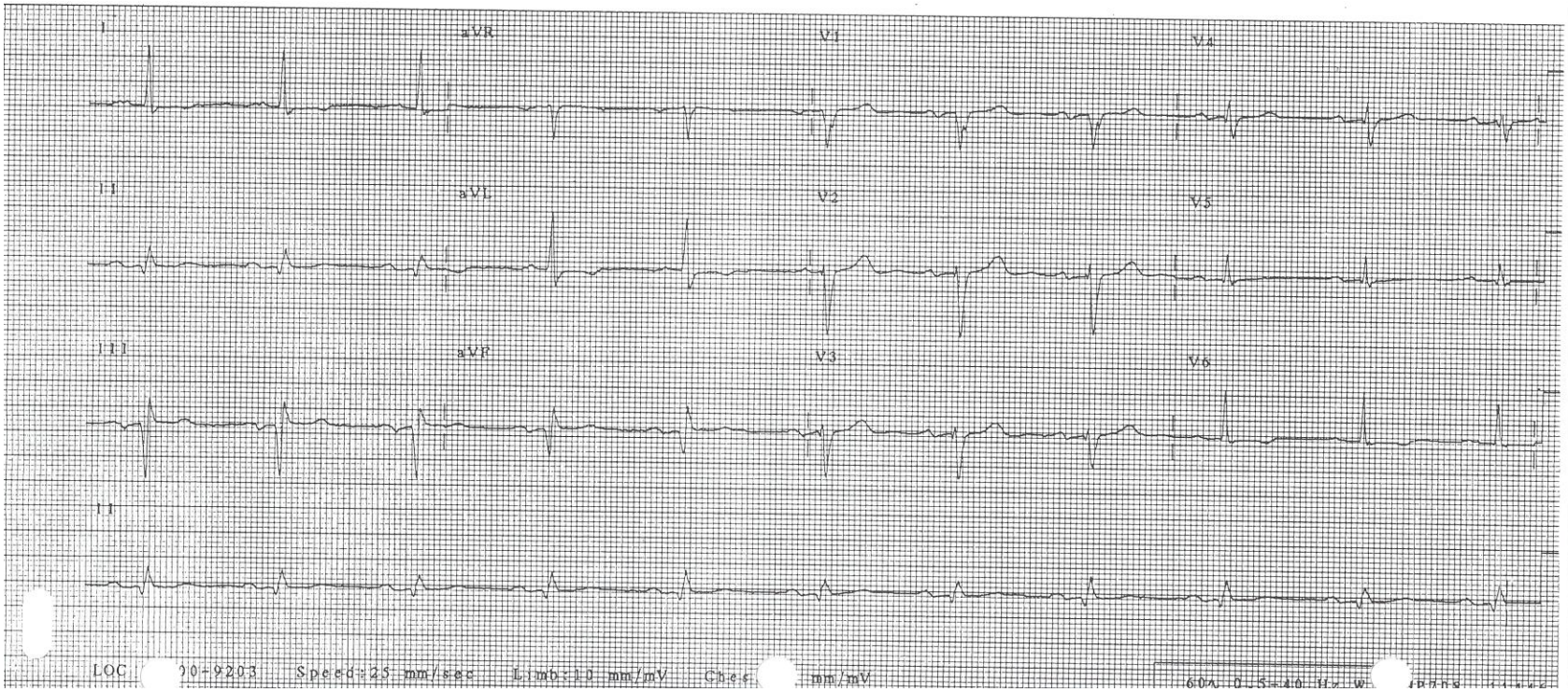
. Sinus rhythm, rate 64.....Normal P axis, rate
. First degree AV block.....PR > 220 mS age>60 V-rate 51- 90
. Left atrial enlargement.....P' -.10 mV and 40 mS in V1
. Old Inferior infarct.....Significant Q-waves in II,III,aVF
. Old Anterior infarct.....Q waves V2-V4
. Nonspecific Lateral T wave abnormalities.....T waves -.10 mV I,aVL,V5,V6

DIAG.
CP

Requested by:
ATCHLEY

- ABNORMAL ECG -

PRELIMINARY-MD MUST REVIEW



09/10/1999 23:15:57
73 years Female

H.

PSI. MEDICAL CENTER
Dept: ICU
Room: 10
Oper: JLW

Rate	64
PR	166
QRSD	119
QT	494
QTc	510

--AXIS--

P	56
QRS	-45
T	-77

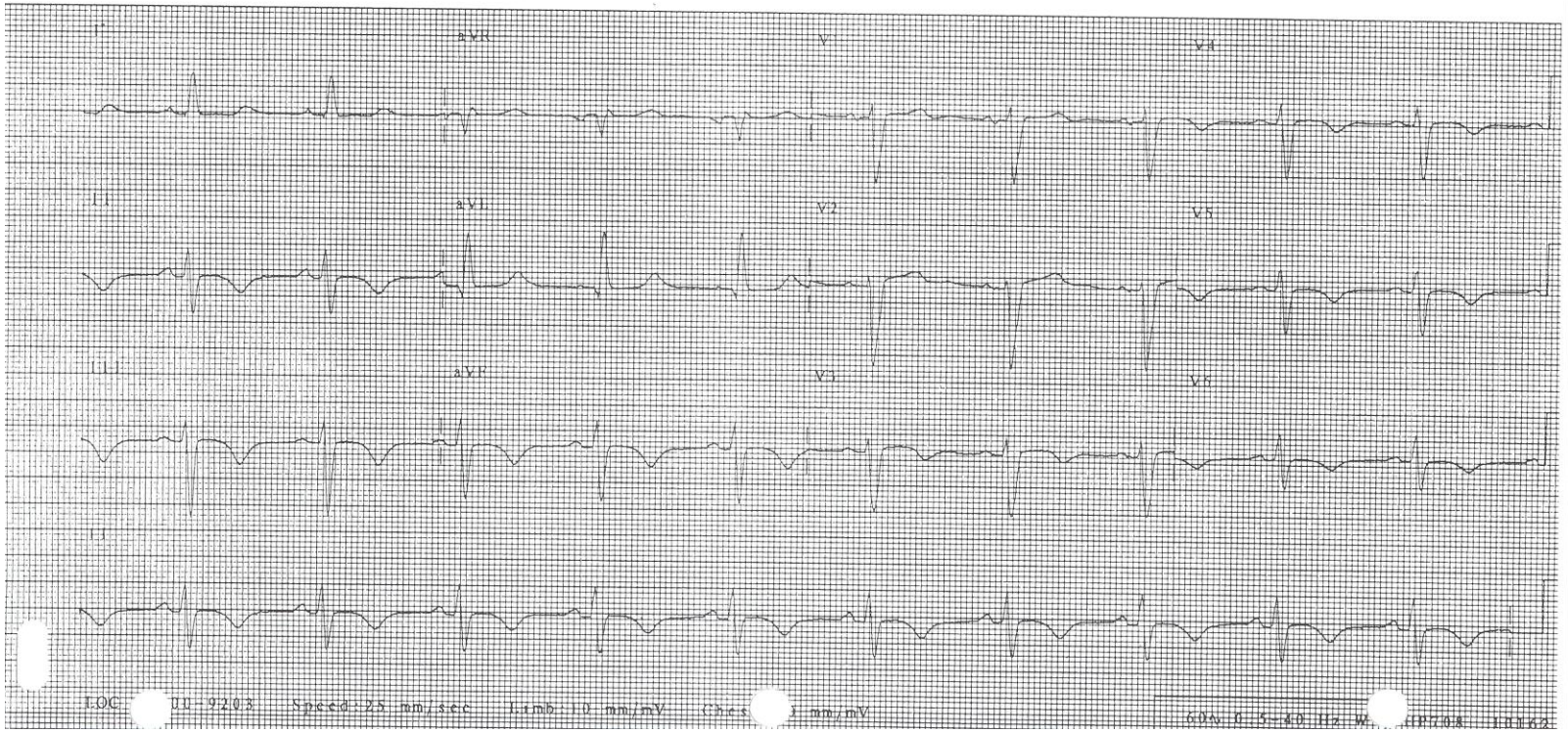
. Normal sinus rhythm, rate 64.....Normal P axis, PR, rate & rhythm
. Left axis deviation, consider LAFB.....Axis -40 deg, S>R & no Q 2,3,F
. Late transition.....QRS negative in V5 or V6
. QT interval long for rate.....QTc > 470 mS
. Diffuse T wave abnormalities,.....T waves -.30 mV ANT/LAT/INF
. . Possible ischemia.....T > -.30 mV

DIAG.
CP

Requested by:
JANTZ

- ABNORMAL ECG -

PRELIMINARY-MD MUST REVIEW



20 yrs Male [redacted] 13 JUL 1999 23:14:10

523291618

pg 1 of 2

PR
QRSD 245
QT 417
QTc 569

--AXES--
P
QRS 234
T 98

+ Atrial fibrillation with V. response of 112 [Now Present]
+ Ventricular premature complex [Now Present]
= Marked posterior QRS axis [Remains]
= Nonspecific intraventricular conduction delay [Remains]
+ Probable LVH with ST-T abnormalities [Now Present]
+ Inferior injury (ACUTE INFARCT) [Now Present]
+ Anterior injury (ACUTE INFARCT) [Now Present]

--CONTINUED ONTO NEXT SHEET--

- ABNORMAL ECG -

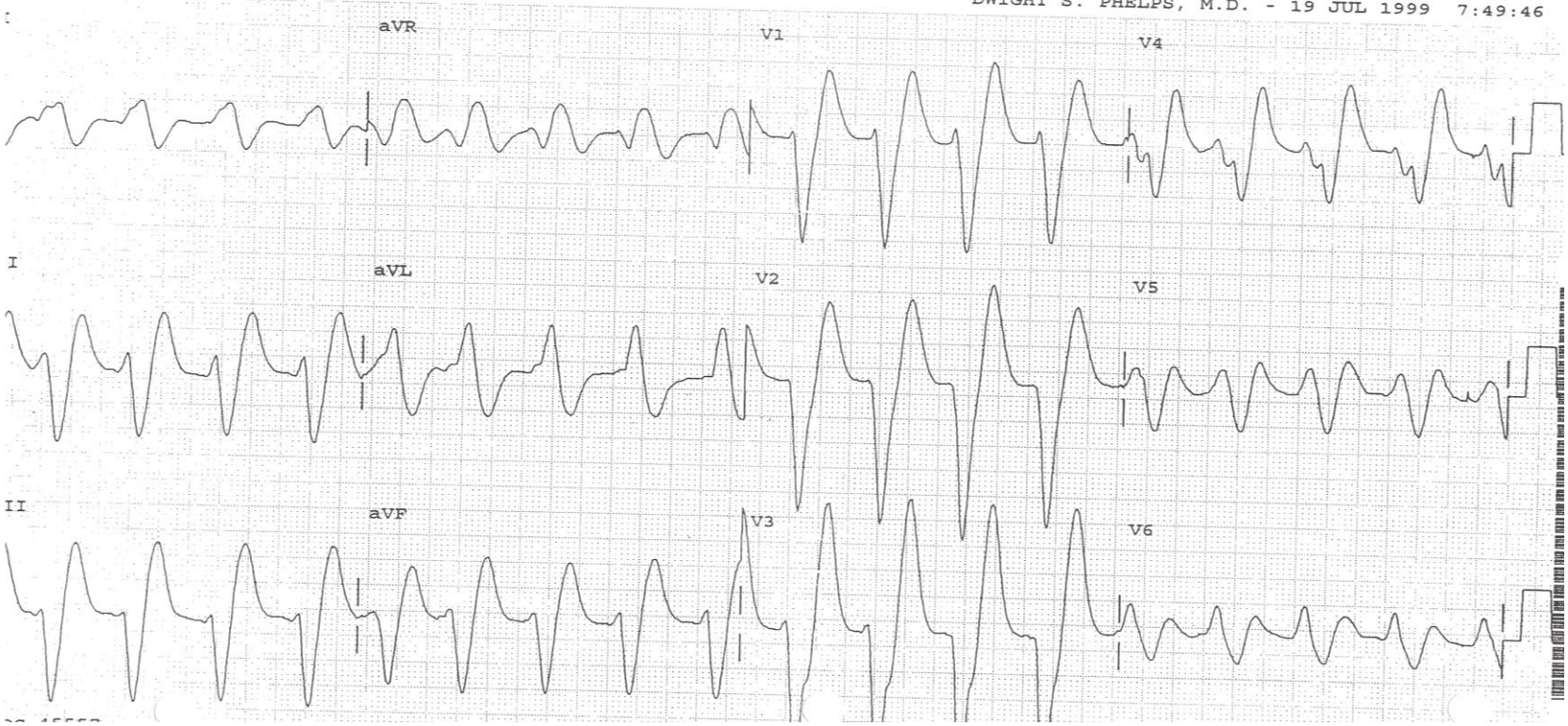
KPID
000620607

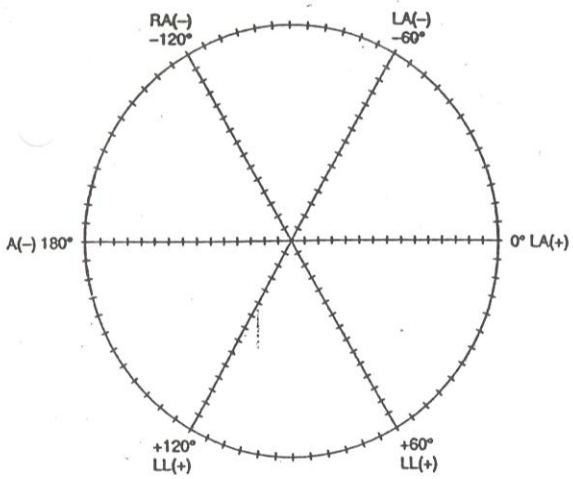
Requested by
PHELPS
Tech EK
Room 2

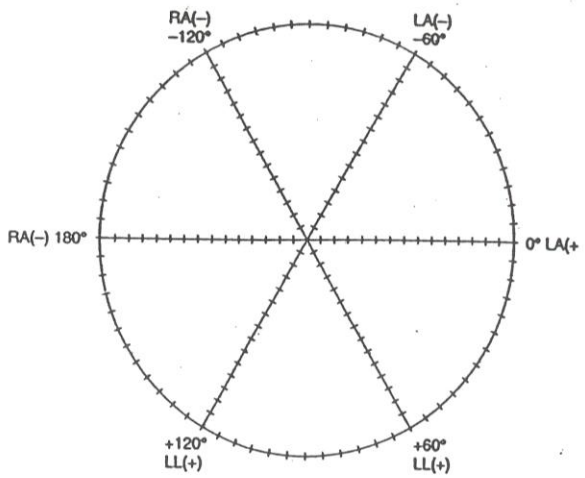
C-HP708

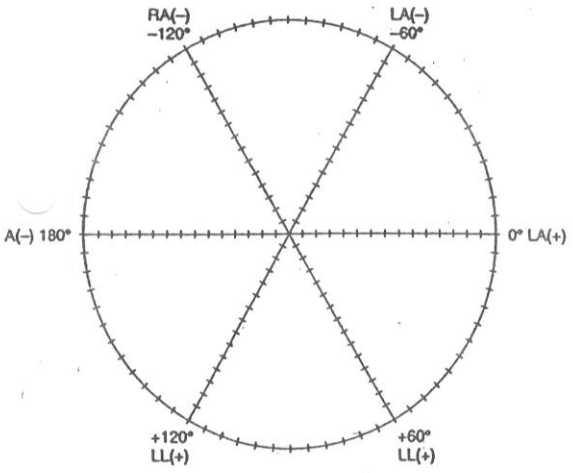
COMPARED TO: 13 JUL 1999 22:41:38, CONFIRMED BY DSP - AB
SAINT JOSEPH HOSPITAL - ED ZONE ONE

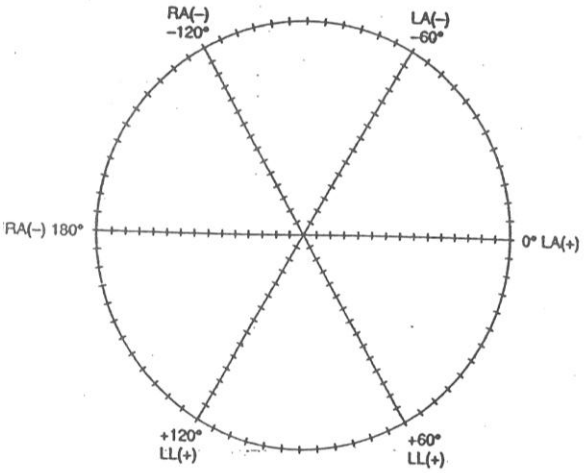
DWIGHT S. PHELPS, M.D. - 19 JUL 1999 7:49:46

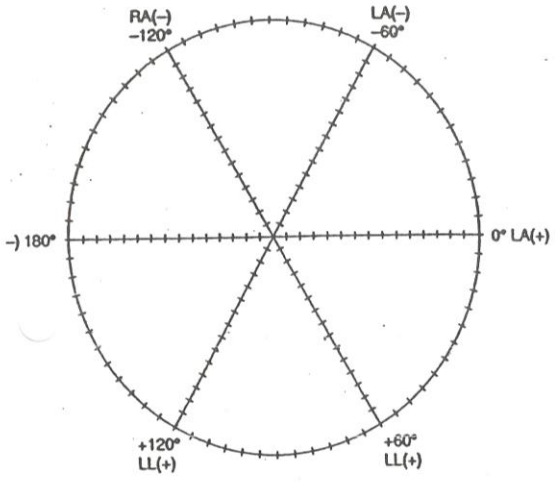


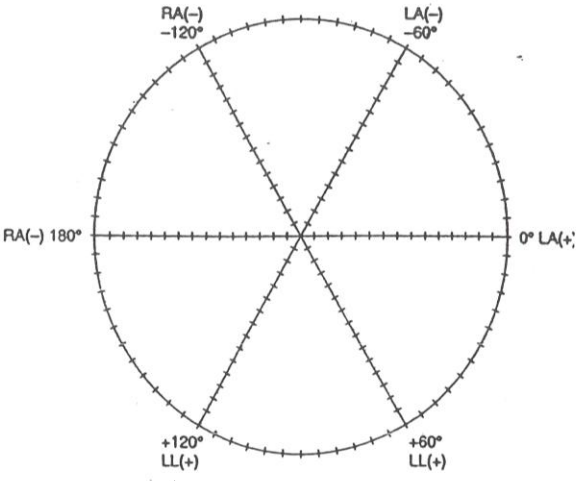

RA(–)
–120°
LA(–)
–60°
A(–) 180°
0° LA(+)
+120°
LL(+)
+60°
LL(+)

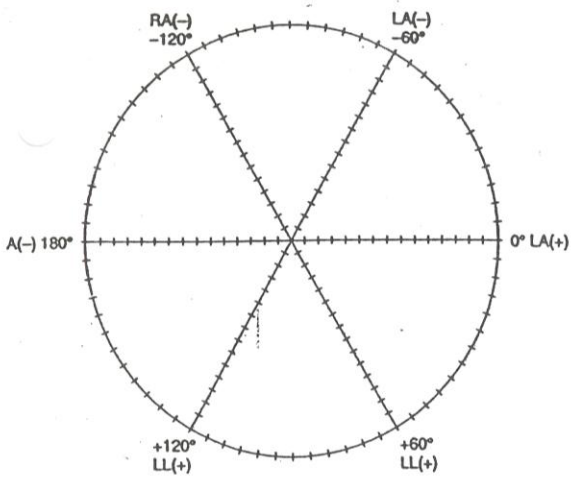

RA(–)
–120°
LA(–)
–60°
RA(–) 180°
0° LA(+
+120°
LL(+)
+60°
LL(+)

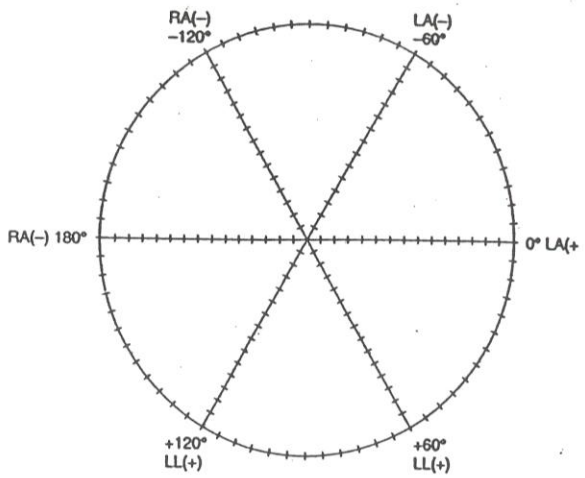

RA(–)
–120°
LA(–)
–60°
A(–) 180°
0° LA(+)
+120°
LL(+)
+60°
LL(+)

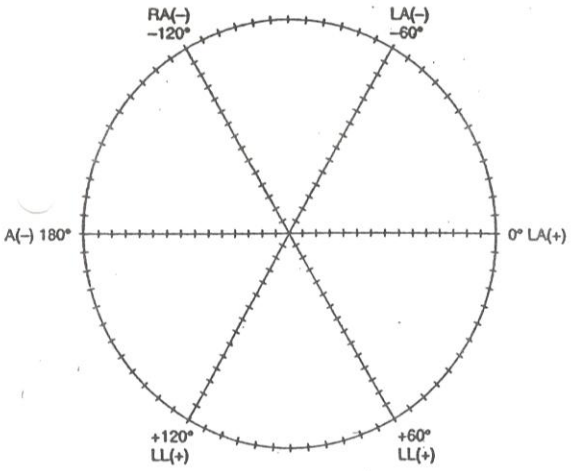

RA(–)
–120°
LA(–)
–60°
RA(–) 180°
0° LA(+)
+120°
LL(+)
+60°
LL(+)

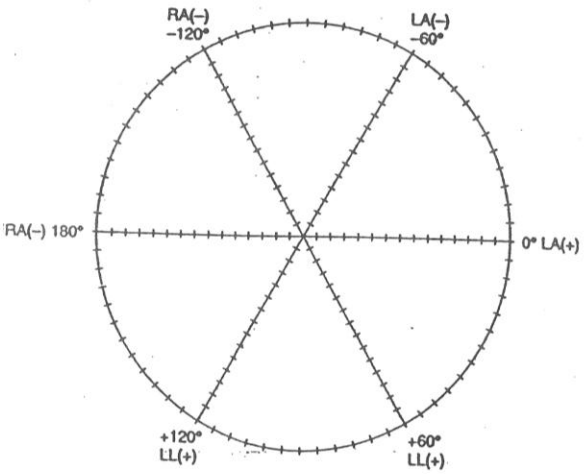

RA(–)
–120°
LA(–)
–60°
–) 180°
0° LA(+)
+120°
LL(+)
+60°
LL(+)

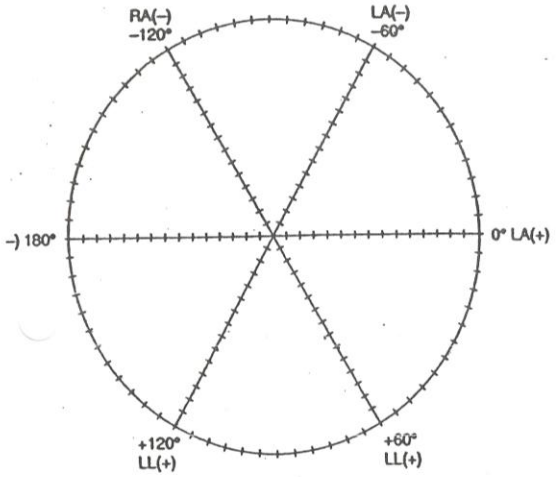

RA(–)
–120°
LA(–)
–60°
RA(–) 180°
0° LA(+
+120°
LL(+)
+60°
LL(+)

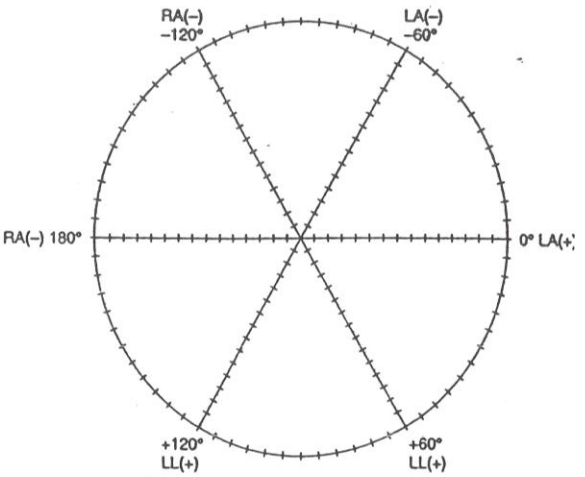

RA(–)
–120°
LA(–)
–60°
A(–) 180°
0° LA(+)
+120°
LL(+)
+60°
LL(+)


RA(–)
–120°
LA(–)
–60°
RA(–) 180°
0° LA(+
+120°
LL(+)
+60°
LL(+)


RA(–)
–120°
LA(–)
–60°
A(–) 180°
0° LA(+)
+120°
LL(+)
+60°
LL(+)


RA(–)
–120°
LA(–)
–60°
RA(–) 180°
0° LA(+)
+120°
LL(+)
+60°
LL(+)


RA(–)
–120°
LA(–)
–60°
–) 180°
0° LA(+)
+120°
LL(+)
+60°
LL(+)


RA(–)
–120°
LA(–)
–60°
RA(–) 180°
0° LA(+
+120°
LL(+)
+60°
LL(+)

PULMONARY FUNCTIONS

I. Required Reading: Guyton and Hall, Textbook of Medical Physiology, 9th Edition, p. 477-488 537-544.

II. Terminology:
Alveolar pressure
Asthma
Atelectasis
Atmospheric pressure
Bronchial sounds
Cyanosis
Dyspnea
Elastic recoil
Emphysema
Expiratory Reserve Volume (ERV)
Functional Residual Capacity (FRC)
Hypercapnia
Hypoxia
Inspiratory Capacity
Inspiratory Reserve Volume
Pleural pressure
Pneumonia
Residual Volume (RV)
Spirometry
Surface tension
Surfactant
Tidal Volume (TV)
Total Lung Capacity (TLC)
Tuberculosis
Vesicular sounds
Vital capacity (VC)

III. Objectives:
1. Discuss the mechanics of pulmonary ventilation.
2. Define surface tension, and explain the role of surfactant.
3. Diagram respiratory excursions during normal breathing, and during maximal inspiration/expiration. Accurately quantify all respiratory volumes and capacities.
4. Understand how spirometry is used for studying pulmonary functions.
5. Describe the sounds for bronchial and vesicular breathing.
6. Give examples of restrictive and obstructive lung diseases.

IV. Breon Spirometer

Introduction

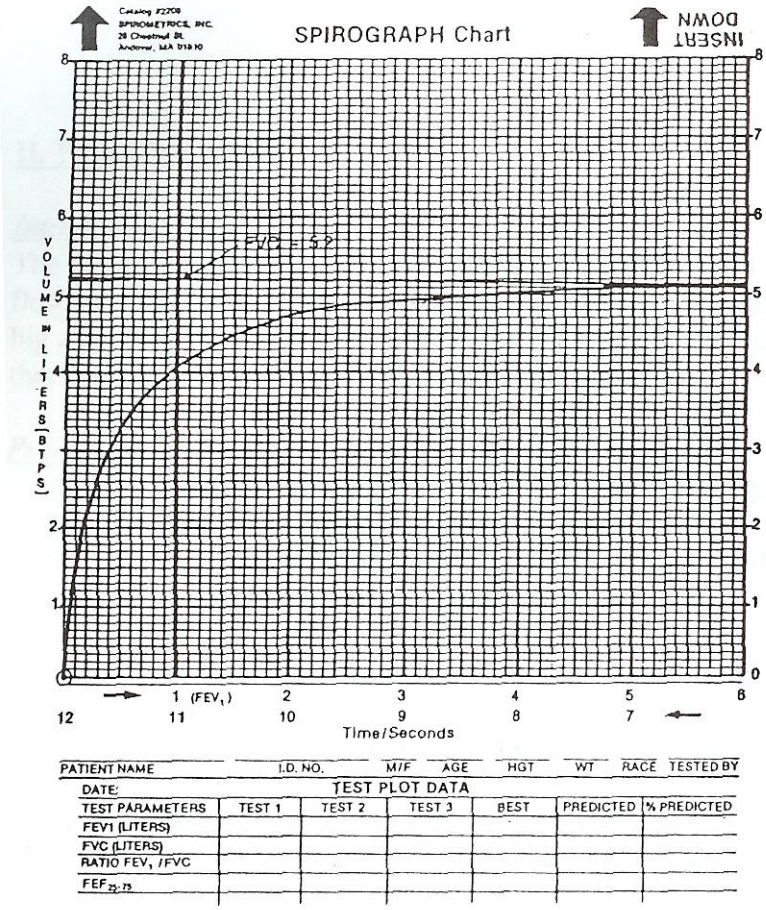
The spirogram does not diagnose any particular disease or condition. It indicates the severity of any ventilation impairment. It also provides a means of determining changes in pulmonary function from year to year once a baseline is established. It is a valuable monitor when a condition or disease pre-exists and it supplements other examinations such a hemoglobin concentrations, and urine analysis. The spirometer is evaluated against the norm for the particular subject's sex, height and age to determine their ventilation level. It can record measurements of the subjects Vital Capacity, Forced Vital Capacity, Timed Forced Expiratory Volume, and Forced Expiratory Flow.

Procedures

1. Place the noseclip over your nose, or plug your nose with your index finger and thumb.
2. Inhale as much air as possible (not from the breathing tube), and insert the mouthpiece into your mouth so that your lips form a tight seal around the mouthpiece. Press the manual start button, just prior to exhalation. This will activate the chart immediately. Exhale into the breathing tube as quickly and as completely as possible.
3. Try to exhale completely, then quickly remove the mouthpiece from your mouth and hold the breathing tube away from your face. As air leaves the spirometer, the chart returns to its original position, thus completing the test.
4. Three successive tests should be preformed and recorded. The best effort should then be used.

FORCED VITAL CAPACITY (FVC)

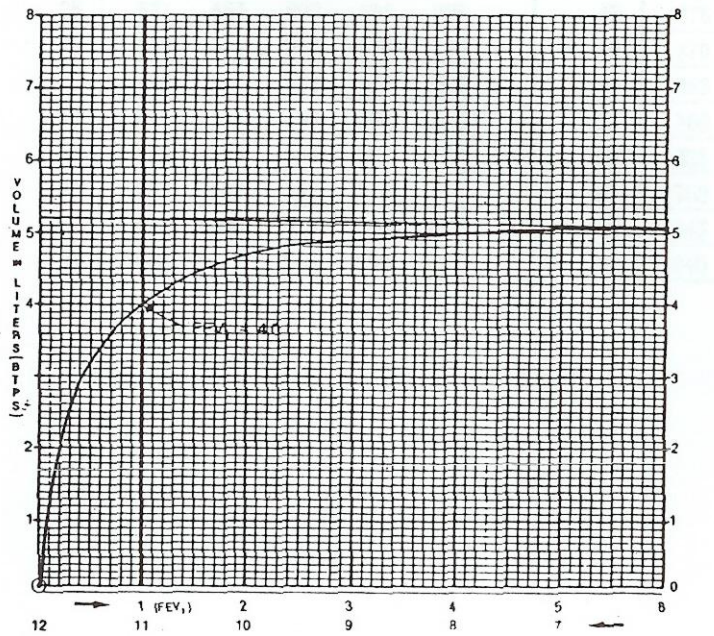
Vital Capacity (VC) and Forced Vital Capacity(FVC) are read directly from the spirograph chart. For Forced Vital Capacity this will be the point marking the highest excursion of the pen, whether or not time has run out. VC can be the same as FVC, but it also may not be the same. To determine if there is a difference between VC and FVC, run a test with the timer OFF (do not press the TEST switch) and let the pen scribe a vertical line while the patient is exhaling at his own rate, and then run another test with the timer ON while the subject is exhaling as rapidly as possible. What are your subject results? Compare your subject's results with the norms for their sex, height and weight.



FORCED EXPIRATORY VOLUME (TIMED-FEV_t)

A forced expiratory volume for a specified time (FEV_t) can be determined during the performance of FVC. Use a pencil to mark where the subject's spirogram crosses the one second line. This is the FEV_1 for the test subject. What were your subject's results? How did the results compare with the norms for a subject of the same sex, age and weight?

Calculate the the FEV1/ FVC from the data obtained on the spirogragh. How do your results compare with the results on the nomogram?



II. Peak Flow Meter

Introduction

The Peak Flow Meter is a portable device for the measurement of peak expiratory air flow rate. This is the fastest speed a person can blow air out of their lungs after taking as big a breath as possible. Peak expiratory flow rate is a simple measure of the flow of air that can tell you how well you are breathing.

Procedure

1. Attach the mouthpiece to the input side of the peak flow meter.
2. Make sure the sliding indicator is at the bottom side of the scale.
3. Grasp the peak flow meter at the bottom, and while standing, inhale as deeply as possible and place your mouth tightly around the mouthpiece. Make sure your lips form a tight seal.
4. Blow as hard and as fast as possible- a short sharp blast, causing the indicator to move up the scale.
5. The final position of the indicator is your peak expiratory flow rate.

Compare your subject's values with the norms for someone of your age, height, and sex.. How did the values compare with the norms? If your values were outside of the normal range speculate as to why that might be.

PREDICTED AVERAGE PEAK EXPIRATORY FLOW

NORMAL MALES*

AGE (YEARS)	HEIGHT 60"	65"	70"	75"	80"
20	554	602	649	693	740
25	543	590	636	679	725
30	532	577	622	664	710
35	521	565	609	651	695
40	509	552	596	636	680
45	498	540	583	622	665
50	486	527	569	607	649
55	475	515	556	593	634
60	463	502	542	578	618
65	452	490	529	564	603
70	440	477	515	550	587

NORMAL FEMALES*

AGE (YEARS)	HEIGHT 60"	65"	70"	75"	80"
20	390	423	460	496	529
25	385	418	454	490	523
30	380	413	448	483	516
35	375	408	442	476	509
40	370	402	436	470	502
45	365	397	430	464	495
50	360	391	424	457	488
55	355	386	418	451	482
60	350	380	412	445	475
65	345	375	406	439	468
70	340	369	400	432	461

PREDICTED VALUES
FEV_1/FVC FEMALE (Knudson – 1983)

AGE Yrs	HT ins	56"	58"	60"	62"	64"	66"	68"	70"
25	FEV.	2.4	2.6	2.8	2.9	3.1	3.3	3.4	3.6
	FVC	2.7	2.9	3.1	3.4	3.6	3.8	4.1	4.3
30	FEV.	2.3	2.5	2.7	2.8	3.0	3.2	3.3	3.5
	FVC	2.6	2.8	3.1	3.3	3.5	3.7	4.0	4.2
35	FEV.	2.2	2.4	2.6	2.7	2.9	3.1	3.2	3.4
	FVC	2.5	2.8	3.0	3.2	3.4	3.7	3.9	4.1
40	FEV.	2.1	2.3	2.5	2.7	2.8	3.0	3.1	3.3
	FVC	2.5	2.7	2.9	3.1	3.3	3.6	3.8	4.0
45	FEV.	2.0	2.2	2.4	2.6	2.7	2.9	3.1	3.2
	FVC	2.4	2.6	2.8	3.0	3.3	3.5	3.7	3.9
50	FEV.	1.9	2.1	2.3	2.5	2.6	2.8	3.0	3.1
	FVC	2.3	2.5	2.7	3.0	3.2	3.4	3.6	3.9
55	FEV.	1.9	2.0	2.2	2.4	2.5	2.7	2.9	3.0
	FVC	2.2	2.4	2.6	2.9	3.1	3.3	3.6	3.8
60	FEV.	1.8	1.9	2.1	2.3	2.4	2.6	2.8	2.9
	FVC	2.1	2.3	2.6	2.8	3.0	3.2	3.5	3.7
65	FEV.	1.7	1.8	2.0	2.2	2.3	2.5	2.7	2.8
	FVC	2.0	2.3	2.5	2.7	2.9	3.2	3.4	3.6
70	FEV.	1.6	1.7	1.9	2.1	2.2	2.4	2.6	2.8
	FVC	1.9	2.2	2.4	2.6	2.8	3.1	3.3	3.5

Based on "Knudson – 1983"

Micro Medical
P.O. Box 6, Rochester,
Kent ME1 2AZ ENGLAND
Telephone: Medway (0634) 43383

PREDICTED VALUES
FEV_1/FVC MALE (Knudson – 1983)

AGE Yrs	HT ins	60"	62"	64"	66"	68"	70"	72"	74"
25	FEV.	2.9	3.2	3.6	3.9	4.2	4.6	4.9	5.3
	FVC	3.3	3.8	4.2	4.6	5.0	5.5	5.9	6.3
30	FEV.	2.7	3.1	3.4	3.7	4.1	4.4	4.8	5.1
	FVC	3.2	3.6	4.0	4.5	4.9	5.3	5.8	6.2
35	FEV.	2.6	2.9	3.3	3.6	3.9	4.3	4.6	5.0
	FVC	3.0	3.5	3.9	4.3	4.8	5.2	5.6	6.0
40	FEV.	2.5	2.8	3.1	3.5	3.8	4.1	4.5	4.8
	FVC	2.9	3.3	3.7	4.2	4.6	5.0	5.5	5.9
45	FEV.	2.3	2.6	3.0	3.3	3.7	4.0	4.3	4.7
	FVC	2.7	3.2	3.6	4.0	4.5	4.9	5.3	5.7
50	FEV.	2.1	2.5	2.8	3.2	3.5	3.8	4.2	4.5
	FVC	2.5	3.0	3.4	3.8	4.3	4.7	5.2	5.6
55	FEV.	2.0	2.4	2.7	3.1	3.4	3.7	4.1	4.4
	FVC	2.4	2.9	3.3	3.7	4.2	4.6	5.0	5.4
60	FEV.	1.9	2.2	2.6	2.9	3.2	3.6	3.9	4.2
	FVC	2.3	2.7	3.1	3.6	4.0	4.4	4.9	5.3
65	FEV.	1.7	2.0	2.4	2.7	3.1	3.4	3.7	4.0
	FVC	2.1	2.6	3.0	3.4	3.9	4.3	4.7	5.1
70	FEV.	1.6	1.9	2.2	2.6	2.9	3.3	3.6	3.9
	FVC	2.0	2.4	2.9	3.3	3.7	4.1	4.6	5.0

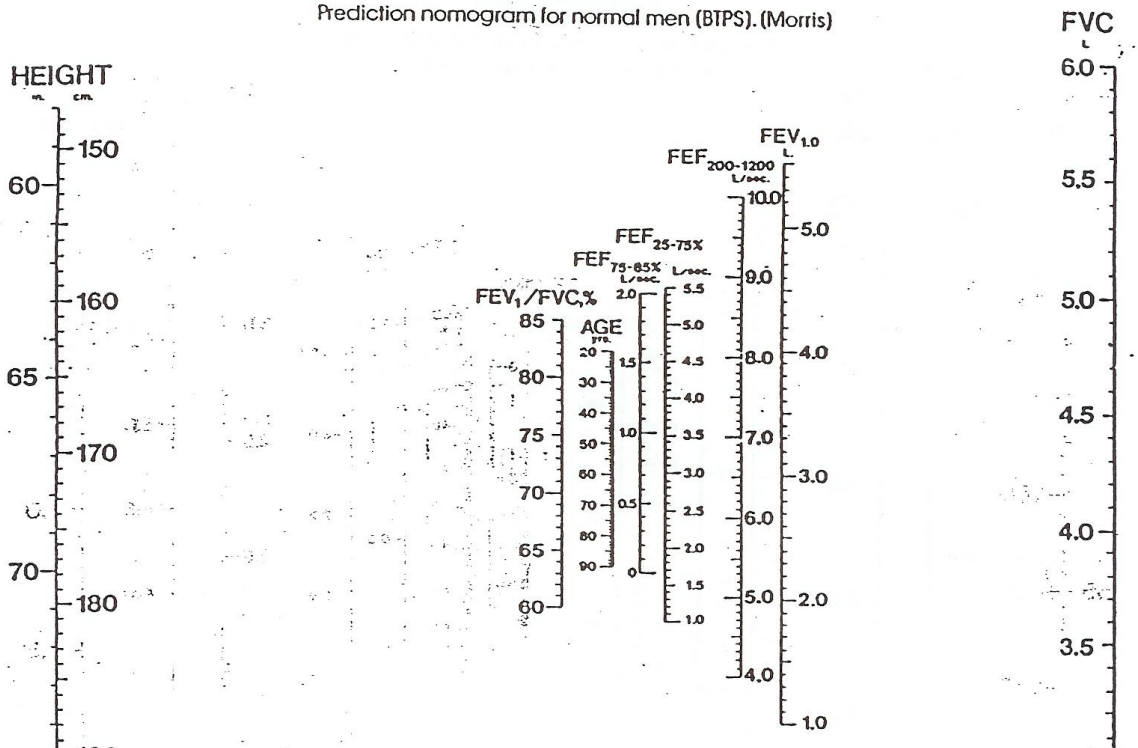
Based on "Knudson – 1983"

Micro Medical
P.O. Box 6, Rochester,
Kent ME1 2AZ ENGLAND
Telephone: Medway (0634) 43383

BREON
SPIROMETER
model 2400

Males

Prediction nomogram for normal men (BTPS). (Morris)



	R	SEE
$FEF_{200-1200}=0.109H_{in.}-0.047A+2.010$	0.44	1.66
$FEF_{25-75\%}=0.047H_{in.}-0.045A+2.513$	0.53	1.12
$FEF_{75-85\%}=0.013H_{in.}-0.023A+1.21$	0.57	0.48
$FEV_{1.0}=0.092H_{in.}-0.032A-1.260$	0.73	0.55
$FVC=0.148H_{in.}-0.025A-4.241$	0.65	0.74
$FEV_{1.0}/FVC,\%=107.12-0.3118H_{in.}-0.2422A$	0.40	7.79

BTPS=body temperature, ambient pressure, saturated with water
FEF200-1200=ratio of one-second forced expiratory flow
FEF25-75%=forced midexpiratory flow
FEF75-85%=forced end-expiratory flow
FEV=one-second forced expiratory volume
FVC=forced vital capacity

Reorder
Catalog #2016

BREON LABORATORIES INC.
90 Park Avenue, New York, N.Y. 10016

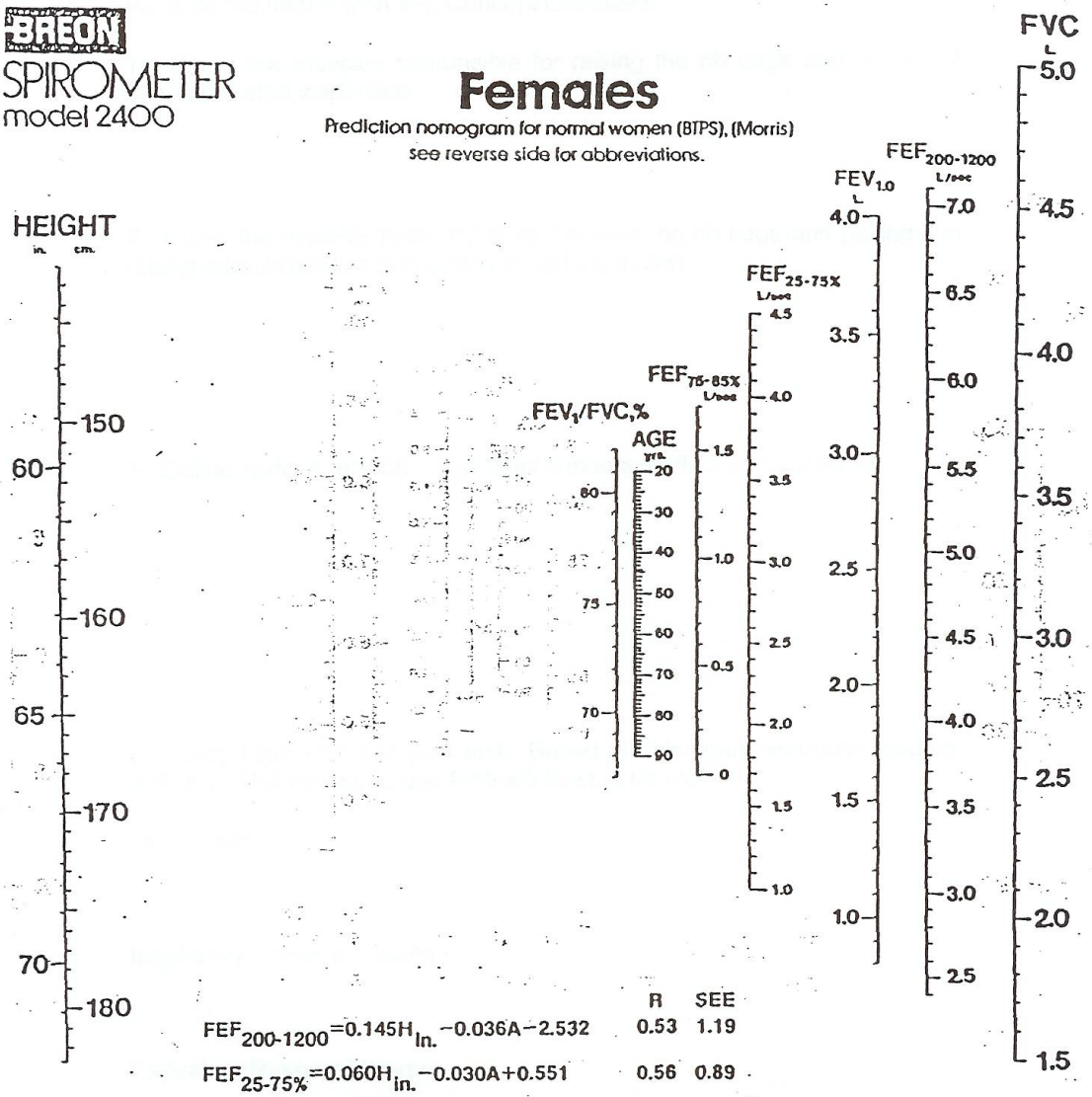
Spirometry in the Evaluation of Pulmonary Function—Morris et al. Western Journal of Medicine 125:110-118, Aug. 1976

BREON

SPIROMETER
model 2400

Females

Prediction nomogram for normal women (BTPS), (Morris)
see reverse side for abbreviations.



	R	SEE
$FEF_{200-1200}=0.145H_{in.}-0.036A-2.532$	0.53	1.19
$FEF_{25-75\%}=0.060H_{in.}-0.030A+0.551$	0.56	0.89
$FEF_{75-85\%}=0.025H_{in.}-0.021A+0.321$	0.63	0.45
$FEV_{1.0}=0.089H_{in.}-0.025A-1.932$	0.73	0.47
$FVC=0.115H_{in.}-0.024A-2.852$	0.71	0.52
$FEV_{1.0}/FVC,\%=88.70-0.0679H_{in.}-0.1815A$	0.39	6.84

Spirometry in the Evaluation of Pulmonary Function—Morris et al. Western Journal of Medicine 125:110-118, Aug. 1976

Reorder
Catalog #2016

BREON LABORATORIES INC.
90 Park Avenue, New York, N.Y. 10016

IV. Background Information and Concept Questions:

1. Name the muscles responsible for raising the rib cage and moving it outward during inspiration.

2. Name the muscles responsible for lowering the rib cage and pulling it in during quiet expiration and during forced expiration.

3. Define surface tension. How does surfactant affect surface tension?

4. Based on the figures in the text, define and list normal values for the following terms:

Tidal Volume -

Inspiratory Reserve Volume -

Expiratory Reserve Volume -

Residual Volume -

Inspiratory Capacity -

Functional Residual Capacity -

Vital Capacity -

Total Lung Capacity -

5. How do pulmonary volumes and capacities compare between adult males and females?

V. Procedure:

A. Pulmonary Volumes and Capacities

1. Record the subject's respiratory rate, at rest. Normal values are 12-18 breaths per minute.

Respiratory Rate at Rest

2. Using the wet spirometer (without the Spirocomp connection described below), determine the subject's Tidal Volume. The subject should inhale a quiet, normal breath, then exhale a normal breath into the spirometer with the cardboard mouthpiece. DO NOT FORCE YOUR EXHALATION. Normal TV is 500 ml. Repeat 2 additional times and average your results.

TV _____ TV _____

TV_____ Avg. TV_____

3. Measure the subject's Expiratory Reserve Volume by taking 3 quiet, resting breaths, then inhale quietly followed by an exhalation as forcible as you can. Normal ERV is 1000-1200 ml. Repeat 2 additional times and average your results.

ERV _____ ERV _____

ERV _____ Avg. ERV_____

4. Measure the subject's Vital Capacity. The subject should exhale all the air possible, by bending over while exhaling. Then, the subject should raise to an upright position while inhaling maximally. Insert the mouthpiece and exhale as forcibly as possible. Normal VC is 4500-4800 ml. Repeat 2 additional times and average your results.

VC_____ VC _____

VC _____ Avg. VC_____

5. The Inspiratory Reserve Volume can be computed by the following equation:

IRV = VC - (TV + ERV)

What is the subject's average IRV?

Normal IRV is about 2100-3100 ml.

URINALYSIS AND HORMONES

I. Required Reading: Guyton and Hall, Textbook of Medical Physiology, 9th Edition, p.315-322, 331-341, 980-982, 1017-1029,1037-1039.

II. Terminology:
Agglutination
Corpus luteum
Diabetes Mellitus
Estrogen
Filtration
Follicle
Glomerulonephritis
Glomerulus
Glycosuria
Hemoglobinuria
Hematuria
Human Chorionic Gonadotropin
Ketonuria
Insulin
Nephron
Progesterone
Proteinuria
Pyuria
Reabsorption
Renal calculi
Renal cast
Secretion
Urochrome

III. Objectives:
1. List the components of the nephron.
2. Describe the formation of urine and all of the changes that occur as the filtrate moves through the nephron.
3. Characterize the role of aldosterone and ADH in urine formation.
4. Explain the countercurrent multiplier system of the nephron.
5. Discuss the factors that regulate the composition of urine.
6. Discuss glomerular filtration and factors that favor and oppose filtration.
7. List the normal characteristics and constituents of urine.
8. Describe the ovarial changes and hormonal changes of the woman's monthly ovarian cycle.
9. Describe the function and gland (cells) that make the following female hormones: Estrogen, Progesterone, and Human Chorionic Gonadotropin.

10. Discuss the accuracy and immunologic basis of an "over-the-counter" pregnancy test.

IV. Background Information and Concept Questions:

1. Each kidney is composed of millions of microscopic functional units called what?

2. List three ways that the kidneys contribute to whole body homeostasis?

3. In reference to #1, the microscopic functional units are divide into what two major components?

4. The capillary network within the renal corpuscle is the _____, and the cup-like structure catching the filtrate is the _____.

5. Draw the functional unit of the kidney, and label all parts. Be specific on all of the names of the different tubules.

6. Urine formation results from what three steps?

7. Blood enters a glomerulus through which blood vessel, and exits through which blood vessel?

8. What is filtration? What determines whether substances are filtered?

9. The glomerular filtrate is similar in composition to blood, but lacks what two components?

10. The majority of the filtered organic nutrients and important ions are reabsorbed where? What is meant by reabsorption (from where, to where?)? Give an example of an organic nutrient absorbed here, and four different ions.

11. What are the roles of Antidiuretic Hormone (ADH) and Aldosterone, as they pertain to urine formation?

12. Give the location of tubular secretion and some examples of substances that are removed by this process.

13. Explain the function of the countercurrent multiplier system in the loop of Henle. In your answer, use the following terms: medulla, loop of Henle, concentration gradient, countercurrent, osmotic gradient, descending limp, ascending limp, permeable, peritubular fluid, and parallel.

14. Give an example of a substance that is reabsorbed passively, and one reabsorbed by active transport.

15. Although about 1/5 of your cardiac output is filtered through the kidneys, about how much urine does the typical person produce in 24 hours?

16. Diabetes Mellitus Type I, is typically due to hyposecretion of what hormone? What does this do to blood levels of glucose? What would this do to glucose levels in the urine?

CHARACTERISTICS OF URINE

Color - Freshly voided urine is pale yellow and clear. A pigment from bilirubin, called urochrome, is responsible for the yellow color. The color may be altered by vitamins, certain foods, blood and by specific gravity.

Odor - Freshly voided urine has a slightly aromatic odor, and will have an ammonia odor if left standing for awhile. Certain diseases, such as Diabetes Mellitus, can alter the odor, causing a sweet odor.

pH - The average pH of freshly voided urine is 6.0, but ranges from 4.5 - 8.0. Diet may affect the pH, with high protein foods and whole wheat products increasing the acidity, and vegetarian diet increasing the alkalinity. Infection may also alter the pH, typically by increasing it.

Specific Gravity - This density value compares the weight of the urine with distilled water, which has a specific gravity of 1.000 (1 ml weighs 1 g). Urine specific gravity normally ranges from 1.001 to 1.035. Dilute urine has a low specific gravity, concentrated urine has a high specific gravity.

Leukocytes - The presence of many leukocytes in the urine, pyuria, represents inflammation in the urinary tract.

Protein - Under most conditions, the presence of protein in the urine, proteinuria, is abnormal. If you understand the process of glomerular filtration, you should be able to explain why it is abnormal. Causes of proteinuria include kidney trauma, poisoning, increased blood pressure, bacterial toxins, and inflammation of the glomerulus (glomerulonephritis). Other, non-pathologic conditions may also temporarily cause proteinuria. Examples of non-pathologic conditions include pregnancy, overexertion, and high protein diet.

Glucose - Presence of glucose in the urine, glycosuria, is usually associated with pathology. Exceedingly high intake of glucose may temporarily overwhelm active transport reabsorption, but uncontrolled Diabetes Mellitus is one of the most common reasons for pathologic glycosuria. Excess and unusable glucose cannot be reabsorbed and spills into the urine.

Ketones - Excessive amounts of ketone bodies, ketonuria, results from abnormal metabolic processes producing excessive intermediates of fat metabolism. The result may be ketoacidosis, which results in characteristic "acetone" breath of the person. Ketonuria coupled with glycosuria is usually diagnostic for Diabetes Mellitus. Ketonuria can also be seen during starvation.

Erythrocytes and Hemoglobin - Hematuria, blood in the urine, and hemoglobinuria, the presence of hemoglobin in the urine from RBC hemolysis, are abnormal findings in voided urine. Causes included inflammation in urinary structures, glomerulonephritis, renal calculi (kidney stones), and non-pathologic contamination from a healthy menstruating female. Hemoglobinuria may be seen as a result of incompatible blood transfusions (Why?).

Cells - Erythrocytes and Leukocytes may be seen upon microscopic analysis of urine sediment if hematuria or pyuria is observed. Sloughed epithelial cells lining the urinary tract (especially those flushed from the vagina during urination in females) are a normal and common finding.

Unorganized sediment - Crystals are seldom of clinical significance when seen in small amounts in the urine. The type of crystal relates to the concentration of the crystalloid, its solubility and the pH of the urine. Acid urine tends to contain uric acid, and calcium oxalate crystals. Alkaline urine may contain calcium carbonate, ammonium ureate, and phosphate crystals. Large amounts of crystals may indicate infection along the urinary tract and may lead to renal calculi.

Casts - Casts are cylindrical "molds" made from tubular filtrate "sludging" in the renal tubules. They are pathologic, and may be seen with kidney disease such as glomerulonephritis.

17. Diagram the ovarian and hormonal changes involved in the woman's normal monthly ovarian cycle?

18. What is a follicle?

19. What is meant by the follicular phase of the ovarian cycle? The luteal phase?

20. Name the cells that make Human Chorionic Gonadotropin (HCG). What is the function of this hormone? During which part of pregnancy is HCG released?

21. Why is it important to prevent menstruation during pregnancy? Are there risks to women who have "spotting" (bleeding) during pregnancy? Explain your answer.

22. How accurate would an HCG pregnancy test be?

23. Describe the immunologic bases of agglutination.

IV. Procedure:

A. Urinalysis

1. View the video titled The Mammalian Kidney before proceeding with the urinalysis. View Fundamentals of Anatomy and Physiology LaseDisc, by Martini, for a better understanding of nephron physiology.

2. Wear disposable gloves throughout this laboratory exercise, including the pregnancy test described below. Obtain your own urine sample, and a sample of the unknown provided in laboratory. Dispose of all waste as instructed by your teacher.

3. Use visual observation, Chemstrips, and a refractometer to complete the following chart.

TEST	**NORMAL VALUES**	**SUBJECT URINE**	**UNKNOWN**
Color	Pale Yellow		
Transparency	Clear		
Odor	Sl. Aromatic		
Specific Gravity	1.001-1.030		
Leukocytes	Negative		
pH	4.5-8.0		
Protein	Negative		
Glucose	Negative		
Ketone bodies	Negative		
Hemoglobin	Negative		
RBCs	Negative		

4. Interpret your results as normal or abnormal. Interpret any abnormal results.

5. Fill a disposable test tube about halfway with the your own urine. (Do not do this test on the unknown.) Spin the urine in the centrifuge for 5 minutes. Remove the test tube, and take it to the sink. Over the sink, **rapidly** invert the test tube to pour out the supernate. Then, take one drop of the sediment (you may not see any sediment with the naked eye, but it's there) and place it on a clean microscope slide. Add a drop of Sedi-stain to make the components visible, and carefully place a coverslip over the drop. Microscopically observe the slide for cells and unorganized sediments (crystals). It is easier to view the crystals with a low illumination on the microscope, and they tend to congregate towards the edges of the coverslip. Record and sketch your findings.

B. Immunologic Pregnancy Testing

The simple over-the-counter pregnancy test kit tests for the embryo-produced hormone called HCG. Immunologic tests involve testing the urine with an antiserum containing antibodies to HCG. (These antibodies are obtained by injecting a laboratory animal with HCG, which will act as a foreign antigen, and elicit antibody production specific against HCG.) The antibodies are coupled to a color reagent. If HCG is present in the urine, a color will appear as agglutination of HCG with the specific color-labeled antibodies occurs. If HCG is absent, no reaction is seen.

1. Obtain unknown urine samples A and B. Follow the directions on the pregnancy kit and record your results.

UNKNOWN	PRESENCE OF HCG	ABSENCE OF HCG	PREGNANCY STATUS
A			
B			

2. How accurate is this test? Explain your answer.

3. During which part of pregnancy, can this test be used to determine pregnancy status?

4. Theorize reasons for false-negative results? Can there be false-positive results?

5. What immunological principle is demonstrated in this exercise?

REFLEX PHYSIOLOGY

I. Required Reading: Guyton and Hall, Textbook of Medical Physiology, 9th Edition, p. 565-581; 685-696.

II. Terminology:
Afferent neuron
Autonomic reflex
Axon
Babinski's sign
Consensual (indirect) pupillary light reflex
Dendrite
Depolarization
Direct pupillary light reflex
Efferent neuron
EPSP
Flexor reflex
Hyperpolarization
IPSP
Monosynaptic reflex arc
Muscle spindle
Neurotransmitter
Patellar reflex
Polysynaptic reflex arc
Postsynaptic neuron
Presynaptic neuron
Receptor
Reflex
Reflex arc
Soma
Somatic reflex
Spatial summation
Spinal reflex
Stretch reflex
Synapse
Synaptic cleft
Temporal summation
Threshold
Unlearned response
Withdrawal reflex

III. Objectives:
1. Define reflex. Trace the pathway of a typical reflex arc.
2. Theorize why reflexes are routinely checked during a physical examination.

3. Compare and contrast somatic and autonomic reflexes. Give examples of each.
4. Explain the difference in response time between a reflexes and unlearned responses.
5. Explain EPSP and IPSP.
6. Understand the reflex pathways of all reflexes performed in this exercise.
7. Describe spatial and temporal summation as they relate to EPSP and IPSP.
8. Detail neuronal transmission at the synapse.

IV. Background Information and Concept Questions:

1. Draw a typical motor neuron with dendrites, axons, and soma labeled.

2. Draw a diagram of transmission of the nerve impulse at a synapse. Include the terms synapse, synaptic cleft, transmitter vesicles, presynaptic neuron and postsynaptic neuron in your illustration.

3. Explain the role of calcium ions at the synapse.

4. Explain the role of neurotransmitters or synaptic transmitters.

5. If the postsynaptic receptor is excitatory, would the change as transmitter is released from the presynaptic terminal cause depolarization or hyperpolarization? The change in the resting membrane potential of this type is called an **excitatory postsynaptic potential**, or EPSP.

If the activated postsynaptic receptor causes inhibition, would the change caused by the release of neurotransmitter from the presynaptic terminal cause the resting membrane potential to depolarize or hyperpolarize? The change in the resting membrane potential of this type is called an **inhibitory postsynaptic potential**, or IPSP.

6. Postsynaptic potentials are graded - i.e. they are NOT all or none. The nerve impulse will continue, if the postsynaptic potential reaches a threshold level. Therefore, summation of postsynaptic potentials occurs. In fact, summation of EPSP and IPSP is necessary to produce excitation/inhibition. What is spatial summation in terms of its role in postsynaptic potentials?

7. What is temporal summation as it relates to postsynaptic potentials?

8. Summation of EPSP results in an action potential in the postsynaptic potential if what occurs?

9. Diagram and label the five essential components of a reflex arc.

10. The patellar or knee-jerk reflex is a monosynaptic stretch reflex. Diagram the components of this reflex arc.
Include in your illustration the terms muscle spindle (stretch receptor), motor neuron, sensory neuron (proprioceptor neuron), synapse, spinal cord, and effector.

11. Explain why a stretch reflex helps in maintenance of posture and balance.

12. The flexor or withdrawal reflex is a polysynaptic reflex. Diagram the components of this reflex arc. Label the terms receptor, sensory neuron, spinal cord, interneuron, motor neuron and effector.

13. A spinal reflex does not involve the higher brain centers. Give an example of a spinal reflex. Would this type of reflex be faster or slower than a reflex involving input from higher brain centers?

14. What is a somatic reflex? Give an example.

15. What is an autonomic reflex? Give an example.

16. Draw a diagram of the crossed extensor reflex. What parts are different from your diagram of the flexor reflex?

IV. Procedure:

A. Somatic Reflexes - **Spinal reflexes**

STRETCH REFLEXES

1. Patellar Reflex

The subject should sit on the laboratory table with the legs hanging and relaxed. Tap the patellar tendon with the reflex hammer. Repeat this several times, until a visual sense of the magnitude of this reflex is assessed.

a. This reflex evaluates which nerve?

b. Which region of the spinal cord?

c. Which skeletal muscle?

d. Repeat the patellar reflex while the subject concentrates on preventing the reflex. Is the response greater than or less than the first response? Explain your answer.

e. Repeat the patellar reflex while the subject clasps the edge of the laboratory table tightly and tries rigorously to pull it upward with both hands. Is the response greater than or less than the first response?

f. Muscular activity occurring simultaneously in other parts of the body results in responses greater than or less than the original response?

g. Repeat the patellar reflex after the subject is truly fatigued from running up and down the stairs. How did the reflex compare to that of the first patellar reflex that you performed?

h. If the subject was truly fatigued, was the reflex change due to the nervous system activity or muscle function?

2. Achilles Reflex

This reflex assesses the superior sacral region of the spinal cord and the gastrocnemius muscle, as the effector.

a. The subject should remove the shoe, and place their flexed knee on the laboratory stool with their foot hanging freely without support of the stool. Tap the Achilles tendon with the reflex hammer. What is the result?

b. What is the normal action of the gastrocnemius muscle?

3. Flexor or Withdrawal Reflex

a. If you touch a hot stove with your hand, the immediate response is the flexor or withdrawal reflex. Without actually performing this experiment, describe your response to this painful example?

b. Compare the number of neurons involved in the flexor reflex to that of the patellar reflex.

c. Describe the action of the effectors during this reflex.

d. Is your awareness of the heat/pain part of the flexor reflex? Explain your answer.

4. Crossed Extensor Reflex

This reflex consists of an ipsilateral flexor reflex followed by a contralateral reflex resulting in extension of the opposite limb. If you step on a nail with your right foot, you reflexively flex your right leg as you extend your left leg to support your weight.

a. The subject should sit with their arm resting on the laboratory table {palmer side up}. The subject should close his/her eyes. Suddenly prick the subject's index finger with a sharp pencil. What happens to the arm that was pricked?

b. What happened to the other arm?

c. Did the extensor portion of this reflex seem to be slow? Explain.

d. If the extensor portion of this experiment did not work, explain why the subject's anticipation or anxiety could have influenced the results?

B. Somatic Reflexes - Superficial Cord Reflexes

1. Plantar Reflex

While the above reflexes involved the spinal cord only, these reflexes depend on the spinal cord reflex AND upper motor pathways. As cutaneous receptors are stimulated in the sole of the foot, the normal reflex results in toe flexion. Damage to the corticospinal tract (upper motor neuron from cerebral cortex, through the medullary pyramids, and on to the spinal cord) causes and abnormal response, termed the "Babinski Sign" in which the toes flare and the big toe moves upward.

a. The subject should remove a shoe and rest the lateral side of the foot on a laboratory stool. Using the handle from the reflex hammer, firmly stroke the lateral border of the subject's sole from the heel to the base of the big toe.

DIAGRAM HERE

b. What is the response?

c. Is in normal or a Babinski sign?

C. Autonomic Reflexes

1. Pupillary Reflex

This reflex involves receptors for light in the retina of the eye, the Optic Nerve as the sensory neuron, the Oculomotor Nerve containing motor neurons to the smooth muscle of the iris - the effector. Since many CNS centers are involved in these responses, absence of normal pupillary reflexes often indicates severe damage to the brain stem.

a. In a dim area, measure the diameter of the subject's pupils. This value will be an approximation.

Right pupil - _____ mm Left pupil - _____ mm

b. The subject should place their hand vertically between their eyes to shield the contralateral eye from the light. Shine a penlight into the subject's right eye. What is the response?

Right pupil - _____ mm

This response is a direct pupillary light reflex.

c. Repeat step b., but this time observe the pupil of the left eye. What is the response?

Left pupil - _____ mm

This is the consensual reflex.

d. Is the consensual reflex ipsilateral or contralateral?

e. Explain the function of the pupillary light reflex.

f. Does this reflex activate the sympathetic or parasympathetic division of the autonomic nervous system?

2. Ciliospinal Reflex

This reflex is an ipsilateral reflex involving the sympathetic division of the autonomic nervous system.

a. While noticing the subject's eyes, gently stroke the subject's skin on the left side of the back of the neck.

b. What is the immediate reaction of the left pupil?

c. What is the immediate reaction of the right pupil?

d. What does this reflex demonstrate in terms of the integration of the sympathetic innervation of the left and right iris?

D. Unlearned Responses

Unlearned response times differ greatly from reflex response times. Far more neural pathways and many higher brain centers are involved in an unlearned response.

1. The subject should hold his/her hand out ready to catch a 12-inch ruler that will be dropped in a vertical position from 1 inch above the subject's hand. The response time can be represented by reading the number of inches that pass through the subject's fingertips when the ruler is caught.

Trial 1 _____ in

Trial 2 _____ in

Trial 3 - ____ in

Trial 4 - ____ in

Compare your response time with that of the simple patellar reflex.

2. Repeat this experiment, but this time decide on a simple signal word to alert the subject that the ruler is about to be dropped. If any other word is said, the subject should not catch the ruler. (If he/she does, disregard this result.) Record your results for four successful trials:

Trial 1 - ____ in

Trial 2 - ____ in

Trial 3 - ____ in

Trial 4 - ____ in

How did your response time compare to that in the trials in # 1?

3. Repeat the experiment, but add word association. As the ruler is dropped, a word such as "hot" should be said. The subject should respond with the word "cold" as he/she catches the ruler. The subject should not catch the ruler, if he/she cannot make a word association. Record your result for four successful trials of catching the ruler.

Trial 1 - ____ in

Trial 2 - ____ in

Trial 3 - ____ in

Trial 4 - ____ in

Number of times that the subject did not catch the ruler:

How did the response time compare to that of the trials in Number 1 and Number 2? Explain the large variation in reaction times seen in this last set of trials?

E. Flexicomp - Human Reflex Physiology

The involuntary knee-jerk (patellar) reflex is a monosynaptic arc that involves a receptor, sensory neuron, the spinal cord, a motor neuron and the effector (quadriceps femoris muscles). This reflex is a common component of a physical exam. An abnormal reflex arc could indicate injury or disease to the spinal cord, sensory or motor neurons.

The latent period of the reflex is the time between the stimulus and the onset of the effector muscle contraction. The maximum excursion (rotation) is the time from the stimulus to the maximum contraction. With the joint displacement transducer strapped to the knee joint, the maximum degree of rotation can be recorded.

GETTING STARTED

1. Flexicomp is a computer system designed to demonstrate some of the characteristics of the reflex arc. You will need a computer-interfaced goniometer ("gonio-" denotes relationship to an angle) and a percussion mallet (hammer).

2. Power up the Macintosh computer. Turn on McADDAM. Launch the Flexicomp program. Click the Flexicomp display.

3. Calibrate the Goniometer. Select **New** from the **File** menu. Select "Calibrate" and follow the instructions on your monitor.

PREPARING THE SUBJECT AND DATA COLLECTION

1. Have the subject sit on the edge of a table with their legs hanging freely. Strap the transducer to the right thigh and leg, over the clothes, so that the transducer box is facing outward and the hinge of the transducer is aligned with the knee joint.

DIAGRAM HERE

2. Data acquisition starts as the mallet hits the target (knee). The x-axis on the computer screen is time; the y-axis is the angle of the goniometer. You can collect up to 10 events. The statistics about each event will be displayed at the bottom, left side of the screen. When you are ready to begin data collection, press "Start" button.

3. Strike the subject's patellar ligament (tendon) hard enough to elicit a good response. Repeat the application of the mallet several times. If your technique is good, you should get similar results each time.

ANALYZE THE DATA

1. Select **Rotation** from the **Analyze** menu. The maximum rotation and latent period are shown in the control bar.
   To select a specific event, click the event number at the top of the window, or use the arrow buttons on either side of the strip of event numbers. Note that the left edge of the screen represents the instant the stimulus was applied.

2. Now select **Velocity** from the **Analyze** menu. The data curve represents the rate of change of the sample data.

3. Print Screen.

EVALUATE A VOLUNTARY REFLEX

Repeat this experiment, but this time have the subject face away from you as you randomly tap the hammer gently on the table, instead of their knee. The subject should respond as soon as they hear the tap on the table by gently kicking. Compare the latent periods for this set of data with that of the involuntary (knee-jerk) responses.

RECORD YOUR DATA

1 - INVOLUNTARY KNEE-JERK REFLEX

a. Latent period:_____sec

b. Time to maximum rotation:_____sec

c. Maximum rotation:_____degrees

2 - VOLUNTARY RESPONSE FROM HAMMER TAP ON THE TABLE

a. Latent period:_____sec

b. Time to maximum rotation:_____sec

c. Maximum rotation:_____degrees

3 - Propose possible reasons for any differences between latent periods of involuntary and voluntary reflexes. Compare several frames of data for both involuntary and voluntary reflexes. Which exhibits greater consistency? Why?

4 - Note that if a subject is tense, the secondary swings of the subject's leg will be diminished and the leg will stop swinging quickly. If the subject is coaxed into relaxing the secondary swings are not as diminished and the swinging continues for a longer period of time. Explain these differences.

SENSORY PHYSIOLOGY

I. Required Reading: Guyton and Hall, Textbook of Medical Physiology, 9th Edition, p. 583-587, 595-597, 623-632, 637-645, 663-669, 675-681.

II. Terminology:

Accommodation
Action Potential
Acuity
Adaptation
Astigmatism
Blind spot
Central fovea
Conduction of sound
Cone
Decibel
Emmetropia
Hyperopia
Modality
Myopia
Olfactory
Organ of Corti
Proprioception
Receptor potential
Refraction
Rhodopsin
Rod
Stereopsis
Taste bud
Taste pore
Threshold

III. Objectives:

1. Distinguish between general senses and special senses.
2. Recognize different general sensory receptors, and explain how they work.
3. Define adaptation. Why might it be desirable?
4. Determine the differences in density and distribution of tactile receptors as it relates to tactile localization, two-point discrimination, and overall sensitivity.
5. Name the parts of the eye and describe how they contribute to vision.
6. Explain how we can see objects and distinguish their color and depth.
7. Compare the relative location of the photoreceptors on the retina.
8. Define accommodation. Explain how and why near-point

accommodation changes with age.

9. Explain the presence of the blind spot and why it is not normally perceptible.
10. Explain the physiology behind the concept of afterimages.
11. Explain how the Snellen eye chart is used to test for emmetropia.
12. Describe the anatomy of the cochlea. Identify the receptors of sound.
13. Explain conduction deafness.
14. Explain how sound localization occurs.
15. Describe the shape of a taste bud. Name and locate the four taste zones.
16. Explain the contribution of olfaction to taste.

IV. Background Information and Concept Questions:

1. List five different types of sensory receptors. Include a brief definition of each.

2. List four ways of exciting a sensory receptor so that there is a change in the transmembrane potential.

3. Are receptor potentials graded or "all-or-none" potentials?

4. If a receptor potential reaches threshold, what will happen to the attached nerve fiber?

5. Explain how stimulus intensity is determined.

6. Describe adaptation of receptors.

7. Relate the term "dendrite", to the term "receptor".

8. List 6 different tactile receptors. Compare their locations.

9. Explain light refraction, as it pertains to the eye.

10. What does the lens do?

11. What is accommodation? How does it change with age?

12. What is emmetropia, hyperopia, myopia, and astigmatism?

13. What is visual acuity? Where on the retina is it the greatest?

14. List three ways of determining the distance of an object from the eye, i.e. depth perception.

15. Compare and contrast rods and cones. Include in your answer - amount of illumination, location on the retina, rhodopsin, color pigment, central fovea, convergence, vitamin A, black & grey, red, green and blue.

16. If you only have three types of color cones, how can the entire spectrum of colors be observed?

17. Based on relative receptor densities, explain why visual acuity is the sharpest when light rays focus upon the central fovea.

18. What and where is the blind spot? Why don't we notice it in our everyday activities?

19. What is the function of the Organ of Corti?

20. Describe 3 ways that the loudness of a sound can be determined.

21. What two factors determine one's ability to localize sound?

22. Name the receptors for taste. Name the four taste zones. Approximate their locations in a drawing.

23. Locate the receptors for olfaction.

24. General senses are those with receptors for touch, pressure, heat, cold, pain, and proprioception. Special senses include sight, hearing, equilibrium, smell and taste. Explain the differences in the terms "general" and "special".

V. Procedure:

View the LaserDisc by Martini, Fundamentals of Anatomy and Physiology, chapter 17 on sensory physiology.

A. General Sensory Physiology

Touch Receptors

1. Tactile Localization

This experiment will determine the ability of the subject to localize a tactile stimulus. Areas of the body with numerous touch receptors and corresponding small receptor fields will have accurate ability of localization.

Areas with few touch receptors, each having a large receptor field, will have poor localization of tactile stimuli.

a. The subject should have his/her eyes closed. As the laboratory partner touches the subject with a color marker, the subject should try to touch the exact point that has been touched with his/her own different-colored marker. The difference between the two dots, the error distance, should be measured in millimeters and recorded in the chart below. Repeat the experiment in each location three consecutive times.

BODY AREA	ERROR DISTANCE First trial	ERROR DISTANCE Second trial	ERROR DISTANCE Third trial
Palm			
Fingertip			
Anterior forearm			
Upper arm			
Upper back			

b. Did the ability to localize the touch by the marker improve with subsequent trials or worsen? Explain why this would happen.

2. Two-Point Discrimination

This test assesses the density of touch receptors. Areas with great densities of touch receptors have the best ability to discriminate two closely placed points as being separate - termed two-point discrimination. Areas with a low density of touch receptors have a poor ability to discriminate two closely placed points as being separate, and generally one cannot determine separation until the two points are widely spaced.

a. Obtain a caliper and wipe the tips with alcohol and cotton. The subject must close their eyes. On the skin areas that will be tested, start with the caliper arms completely together. The subject must determine whether he/she feels one point or two. Gradually widen the calipers, until the subject can no longer feel one point, and now can feel two. Measure the distance between the two points in millimeters, by measuring the caliper distance. Record your data.

BODY AREA	TWO POINT DISCRIMINATION (mm)
Face	
Palm	
Dorsum of hand	
Fingertips	
Lips	
Back of neck	
Back of leg	

b. Based on your results, rank order the body areas tested from those with the greatest density of receptors to areas with the smallest density of receptors. How do your results correlate with sensitivity of those body areas?

3. Adaptation of Touch Receptors

a. The subject should close the eyes throughout this experiment. A coin should be placed on the anterior surface of the subject's forearm. Glance at the time, when the coin is placed on the forearm. The subject should report when he/she can no longer feel the coin - i.e. when adaptation has occurred.

Duration of the sensation:_____

b. Repeat the experiment at a different site on the forearm. But do NOT remove the coin.

Duration of the sensation:_____

c. After adaptation has occurred at this site, add three more coins on top of the coin already present on the forearm. Glance at the time.

Does the awareness of the coins return?

d. How long until adaptation occurs for the four coins?

Duration of the sensation:_____

e. Explain why the sensation of the coins returns when three more are added? Do the four coins stimulate the same touch receptors as the single coin?

B. Special Senses

Vision

1. Blind Spot

a. Observe the figure below with the paper about 18 inches away from the eyes. Close the left eye, look at the "X" with the right eye. The right eye should be aligned with the "X". Slowly move the figure towards the face until the dot disappears. This occurs when the light rays from the dot focus on the blind spot. Measure the distance between the dot and the eye, when the dot disappears.

Right eye distance when dot disappears:_____cm

b. Now move the figure even closer to the eye. Does the dot reappear?

c. When the dot focuses on the blind spot, why does the image disappear? In other words, why is there a blind spot?

d. Repeat the experiment, with the figure turned upside down and the right eye closed. Measure the distance between the dot and the left eye, when the dot disappears.

Left eye distance when dot disappears: _____ cm

2. Afterimage

As light rays strike the rods of the retina, the rhodopsin splits into Vitamin A and opsin which leads to a nerve impulse. Before the rod can be restimulated, the rhodopsin must reform. This phenomenon is illustrated as positive and negative afterimages. As the rods continue to fire, a positive afterimage is formed. As the rhodopsin splits and then resynthesizes, a negative afterimage occurs.

a. Stare at a bright light for a few second, and then close your eyes for one minute.

b. The first image that you "saw" is a positive afterimage. Describe what it looked like.

c. The subsequent image that you "saw" is a negative afterimage. Describe what it looked like.

3. Near point accommodation

This test tests for the ability of the elastic lens to focus for close vision. This distance is very close to the eye in children and farther away in old age.

a. Hole a straight pin vertically in front of one eye with your arm extended. Focus on the pin, and gradually move the pin closer to your eye. When the image of the pin becomes blurry, have your laboratory partner measure the distance from the pin to your eye. Repeat for the other eye.

Near point accommodation - right eye _____ cm

Near point accommodation - left eye _____ cm

b. What do your results tell you about the elasticity of your lens, and it's ability to change shape?

4. Visual acuity

a. The Snellen eye chart will be used to measure your visual acuity or sharpness of vision in each eye. The line on the chart that reads 20/20, means that a person with emmetropia (what does this term mean?) can read that line clearly while standing 20 feet away from the chart.

Visual acuity - right eye_____

Visual acuity - left eye_____

b. If you wear glasses, perform this test twice - once with your glasses and once without.

c. If your visual acuity is measured as 20/50, what does this mean? Emmetropia or myopia? What do the numbers "20" and "50" mean?

d. If your visual acuity is measured as 20/15, what does this mean?

5. Color Blindness

Observe the two figures on p. 645 of your textbook. What numbers appear in the center of the circles? Do you show any indications of color blindness?

6. Astigmatism

View the figure below separately for each eye. If all of the lines are equally dark and distinct, there is not astigmatism. If some are less dark than the others or are blurred, then some degree of astigmatism is present. Record your results.

Right eye - normal or astigmatism?

Left eye - normal or astigmatism?

FIGURE HERE

7. Positioning of Rods and Cones

This experiment illustrates the relative positioning of the photoreceptors. As you know, the rods should be most peripheral, with the blue cones, followed by the red, then green cones being closest to the central fovea.

a. The subject should stand facing the chalkboard, about one foot away. The subject should close the left eye and stare at a small circle drawn on the board, directly in front of the subject's right eye.

b. While the subject continues to stare at the small circle, move a white paper disc from about 2 feet away, along the board, and into the field of vision. As soon as the subject can recognize the circle's presence AND the fact that it is white in color, place an "X" on the board. Repeat this mapping at about 6 different angles.

c. Repeat the experiment with the red, green, and blue paper discs, but make the "X" in red, green, and blue chalk, respectively.

d. Connect the "X"s with the appropriate color chalk.

e. Does your map show the white circle, representing rods, as being the most peripheral? Is the blue next, followed by red? Is the green circle the innermost? Compare your results to the normal. Account for any variations.

8. Depth perception

To illustrate that our two eyes see slightly different fields of view, perform the following exercise.

a. Hold a pencil vertically, at an arm's length, directly in front of your right eye. Close the left eye. Now hold another pencil directly beneath it and move the lower pencil, in the same plane, towards your face.

b. Now open the left eye and observe that the pencils are NOT in the same plane, as observed by the right eye.

To illustrate that this binocular vision allows our brain to provide us with information about depth perception, perform the following exercise.

a. A laboratory partner should hold an empty test tube at about one arm's length from the subject's eyes. The subject should try to quickly insert a pencil into the test tube while both eyes are open.

How easy was this to perform? How accurate were you?

b. Repeat the experiment with one eye closed.

How easy was this to perform? How accurate were you with only one eye?

Hearing

9. Auditory Acuity

The subject should pack one ear with cotton and close their eyes. The laboratory partner should take a ticking clock and slowly move it away from the unpacked ear until the subject can no longer hear it. Record the distance at which the sound can no longer be heard. Repeat with the other ear.

Right ear _____

Left ear _____

10. Sound Localization

The subject should close both eyes. The ticking clock is then placed at different locations such as on top of his/her head, to the left, right, front, and back. The subject should point to the direction that the sound is coming from. Could the subject localize the sound in all positions? In which position was sound localization more difficult?

11. Range of Hearing

Obtain three different tuning forks - one with a high frequency (4096 cps), one mid-range (1024 cps), and one with a low frequency (100 cps). Strike the tuning fork with a rubber reflex hammer. Can the subject hear all three tuning forks? Assuming that they were struck with equal force, which was heard the best? Which was heard the least well?

12. Weber Test

This test evaluates whether sound is heard centrally, which is normal, or lateralizes to one side or the other which is indicative of some degree of deafness due to conduction problems or hearing receptors or unilateral nerve damage.

a. Strike a tuning fork and place the handle at the top of your forehead at midline. Is the sound heard equally loud in both ears? If the answer is yes, this is a normal condition.

b. To illustrate a conduction of sound hearing loss, plug the right ear with cotton to interfere with sound wave conduction. Repeat the test. The sound should be heard more strongly in the right ear. Do your results of simulated conduction loss agree with these results?

13. Rinne Test

This test compares conduction of sound waves through bone versus through air.

a. Strike the tuning fork and place the handle on the mastoid process of the temporal bone, which is located just behind the earlobe. When the subject indicates that sound is no longer heard (i.e. no more bone conduction), place the still-vibrating tuning fork close to the ear canal. Can the subject hear the fork now, through air conduction? Record your results.

b. Repeat for the other ear. Record your results.

EAR	HEARD BY BONE CONDUCTION	STILL HEARD BY AIR CONDUCTION
Right		
Left		

b. It is normal to hear by air conduction. The hearing is normal if the tuning fork could be heard by air conduction, when it could no longer be heard by bone conduction. Do your results indicate normal hearing?

c. Repeat the test, but this time start with air conduction. Strike the tuning fork and hold it close to the subject's ear. When the subject indicates that sound is no longer heard (i.e. no more air conduction), place the handle of the tuning fork on the mastoid process. Did the sound return? If it did, there is some conductive deafness present. Repeat for the other ear.

EAR	HEARD BY AIR CONDUCTION	STILL HEARD BY BONE CONDUCTION
Right		
Left		

d. This test illustrates that we normally hear better by air or bone conduction??

Taste and Olfaction

14. Taste Bud Stimulation

The taste buds on your tongue have receptors inside of a taste pore. As food is moistened by saliva, it enters the taste pores and stimulates the receptors for taste.

a. Dry off the dorsum of the tongue with a paper towel. Place a few granules of sugar on the dry tongue. Without moving the tongue or closing the mouth, record the amount of time from when the sugar is placed on the tongue until it can be tasted as sugar.

Time to taste sugar

b. Explain why the sugar cannot be tasted immediately?

15. Mapping of Taste Zones

a. Prepare a map of the four taste zones on the tongue. Obtain a vial of sweet, sour, bitter, and salty solutions. Obtain sterile and new applicator swabs for each solution. Additionally, have a cup of water for rinsing between different solutions.

b. Dry the tongue with a paper towel. Moisten an applicator swab with one of the four solutions. Touch the swab to the tip, back, sides and center of the subject's tongue. Map where the subject can correctly taste the solution below.

c. Draw a figure of the dorsum side of the tongue. Map all four taste zones as determined by this experiment.

d. Do your results agree with the taste zones identified in your textbook?

15. Role of Olfaction in Taste

As you know from when your sense of taste diminishes if your nasal cavity is clogged with mucus from a cold, olfaction plays a definite role in the ability to taste.

a. A laboratory partner should use a clean applicator swab to apply a small amount of aromatic oil (such as peppermint, clove, butterscotch, watermelon...) to the dorsum of the subject's dried tongue WHILE THE SUBJECT"S NOSTRILS ARE CLOSED.

b. Can the subject determine the flavor?

c. Release the nostrils. Now, can the subject determine the flavor?

d. To further illustrate the importance of olfaction in taste, obtain two swabs each containing a different aromatic oil. Hold one under the subject's open nostrils, while the other is swabbed on a rinsed and dried tongue. Which flavor is noticed?

16. Olfactory Adaptation

We all are aware of an odor in a room when we first enter, but soon, we become unaware of it or "used" to it. This is olfactory adaptation.

a. Obtain a clean applicator swab. Apply one drop of aromatic oil. Hold the swab under one of the subject's nostrils, while the subject holds the other nostril closed. Record the time required for the odor to disappear, i.e. olfactory adaptation.

Time for adaptation

b. While adaptation is still occurring, introduce a swab with a different oil on it. Can the subject note the new odor? What does this indicate about the specificity of adaptation?