Gas Exchange and Respiratory Systems

In the last lab, you explored the operations of the circulatory system—the heart, vessels and blood. The circulatory system, in combination with the respiratory system, is also responsible for the transport and exchange of O2 and CO2.

In vertebrates, the respiratory pigment, hemoglobin, carries O2 from the gills or lungs to the tissues and organs and carries CO2, produced by cellular metabolism, back to the lungs. Gases are exchanged across capillary walls by diffusion as a consequence of the partial pressures of oxygen \((P O_2)\) and carbon dioxide \((P CO_2)\) in the blood and tissue cells. Both the rate of breathing and ability to exchange O2 and CO2 can be affected by external factors such as exercise, smoking, or disease.

In this laboratory, you will study the functions of gills and lungs. You will consider how hemoglobin binds O2 and CO2 and learn how your breathing rate can be affected by exercise.

STUDENT PREPARATION

Read the relevant text pages. Familiarize yourself with the information and procedures covered in this laboratory.

Key Terms

- Bronchi(oles)
- blood pressure
- fight or flight
- alveoli
- alveolar ducts
- gill arches
- trachea
- countercurrent exchange
- pulse points
- respiratory volumes
- gill lamellae
- sympathetic responses

Pre-test – bring theses answers to lab

- What physical force is responsible for air entering and leaving the lungs?
- Name a muscle that contracts to cause expansion of the chest cavity.
- Define artery.

Learning Objectives

1. Be able to describe the evolution of lungs from a gas bladder precursor
2. Compare and contrast the respiratory structures in fish versus mammals
3. Observe and label the gross anatomy of lungs and gills
4. Observe and label the microscopic anatomy of lungs and gills
5. Describe how the anatomical arrangement of capillaries and respiratory structures maximizes O2 extraction
6. Describe the respiratory volumes and measure your own
7. Understand the immediate effects of exercise on respiratory volumes

Respiratory Lab – Must have in your lab notebook

Charts

1. Respiratory volumes: VC, TV, IRV, ERV
   - After exercise: just TV
2. Respiratory rates; Before & after aerobic exercise

Diagrams
1. Lungs (labeled)
   a. Afferent & efferent blood supply & relative O₂ levels in each
   b. Microscopic anatomy of alveolar capillary beds
2. Gills (labeled)
   a. Afferent & efferent blood supply & relative O₂ levels
   b. H₂O flow & changing O₂ levels
   c. Anatomy of gill lamellae & capillary beds

Dissections
1. Pig/sheep lungs showing major anatomical features of lung
2. Perch gills showing major anatomical features & a diagram of a dissected lamellae

Slides
1. Trachea
   a. Cartilage
   b. epithelial cells & cilia
   c. smooth muscle
2. Lung tissue
   a. Bronchiole
   b. Vein/artery
   c. Alveolar ducts & alveoli

Vertebrate Respiratory System

The function of the respiratory system is to acquire O₂ that can be transported to the tissues of the body and to remove CO₂ produced by the metabolic activities of the cells. This requires a large surface area that must be protected from damage, desiccation and osmoregulatory stress. In vertebrates, there are two major respiratory surfaces: the gills and the lung/swim bladder. Accessory structures may include the skin and the cavities of the mouth and pharynx.

The first chordates were small and unprotected by scales, and gas exchange occurred across the body surface or skin. These organisms possessed slits in the pharynx (used for filter-feeding), that evolved into gills. In the earliest vertebrates, the jawless fishes, the pharynx tissue between the slits became heavily vascularized, bringing more of the blood supply into closer contact with oxygenated water, thus increasing the amount of gas exchange; the gills assumed a major role in respiration. Gas exchange through gills increased in importance as fish developed thicker protective body coverings and increased their rates of activity in response to predation.

Among the earliest jawed fishes, a saclike outpocketing (evagination) of the pharynx provided an accessory respiratory organ, the lung (Figure 1). This ancestral lung evolved into two different structural specializations among vertebrates. First, in primitive bony fishes (and their descendants that live in well-aerated waters), the gills function as the major respiratory organs, and gases in the lung are used mainly to reduce the density of the fish (like a ballast tank in a submarine). In these organisms, this structure functions as a lung/swim bladder and remains connected to the pharynx (Figure 1). In modern body fishes, this structure loses its connection to the pharynx during embryonic development and functions only as a swim bladder, not as a lung. Its contents are regulated by the circulatory system and a gland which "secretes" oxygen into the swim bladder (Figure 1).

In reptiles, there is an increasing division of the lining of the lung; in mammals, this culminates in extremely small alveoli where air comes in close contact with the vascular system (Figure 1).

Examine Figure 1. What trend can be observed in the surface area of the lung, proceeding from amphibians to primitive reptiles to mammals?
Figure 1 Evolution of swim bladders and lungs. (a) A bilobed outpocketing of the pharynx serves as an accessory respiratory organ. (c) The lung/swim bladder assumes a more dorsal position to help the fish float upright, and the lobes of the lung/swim bladder fuse. (d) The swim bladder has lost its connection to the pharynx in adult bony fishes and no longer serves as a lung, but only as a swim bladder or "ballast" organ. 

(a) The lung of an amphibian is a relatively simple vascular sac. (b) In primitive reptiles, the surface area is increased by folds of vascularized tissue. (c) Further partitioning in mammals leads to the formation of a copious number of microscopic sacs, the alveoli.

Figure 262. Diagrammatic sections through the lung of A, a frog, and B, a lizard. C, Section of a small area of a human lung, x about 50, showing its complex construction. Abbreviations: ad, an alveolar duct, smallest component of the duct system; a, the alveoli to which it leads. (A after Vialleton; B after Goodrich.)
How Fish Breathe—Gills as a Respiratory Surface

In bony fishes, a series of **gill arches** support the gills. Attached to the arches are gill **filaments** (Figure 2) with leaf-like lobes (**lamellae**) that serve as the respiratory exchange surfaces. Blood vessels pass through the gill arches and send branches into the filaments and even smaller branches into the lamellae (Figure 2). Blood flows outward from the gill arch and through the lamella, moving across each lamella in one direction; oxygenated water flowing over the gills moves in the opposite direction (Figure 2). This countercurrent flow allows more oxygen to be picked up from the water by the capillaries in the lamellae (Figure 3). Exchange of oxygen from water to blood is dependent upon diffusion. The rate of diffusion increases as the difference in the concentration of the diffusing substance on the two sides of a membrane increases. Countercurrent flow maximizes the difference between the oxygen content of the blood and that of the water at each point of contact, so the gills always pick up the maximum amount of oxygen from the water. The blood vessels going to the gills are called afferent arterioles; those that lead away from the gills are called efferent arterioles. From the gills, oxygenated blood is distributed to the body by the dorsal aorta and is returned to the heart by the cardinal and hepatic veins.

**Procedure**

1. Examine the mouth of a bony fish (perch). Feel the teeth on the upper jaw and on the roof and floor of the pharynx.
2. The pharynx wall is perforated by five pairs of gill slits. Four gill arches are present between the slits. The operculum (gill covering) extends over the entire grouping of arches and slits.
3. Place your finger into the mouth and extend it to emerge through one of the gill slits.
4. Cut through the opercular flap (Figure 2) and bend it backwards out of the way.
5. Examine the gills of the fish. Each gill arch between the gill slits has two sets of filaments. Identify the two sets of gill filaments (Figure 2).
6. Observe a prepared slide of a gill filament if one is on demonstration. Otherwise, prepare a wet-mount slide of a gill filament and observe it under the microscope.
   - **What is the advantage of the gill filaments having lamellae?**
7. Water passes into the mouth of the fish and then from the pharynx through the gill slits. It passes to the outside at the posterior edge of the opercular flap. As water passes over the gill filaments, oxygen is carried to the filament and diffuses into the blood that is flowing in the opposite direction within the capillaries.
   - **Why does a fish continuously open and close its mouth?**
   - **If water and blood flowed in the same direction, would the blood pick up as much as much oxygen as when blood and water flow in opposite directions?**
   - **Why or why not? Examine Figure 3**
   - **Why are the blood vessels going both to and from the gills called arteries, or arterioles? What type of blood does each carry?**

At an early time in fetal development, humans have pharyngeal clefts (like gill slits but without four gills). We can still see evidence of these—the eustachian tube, middle ear, and external ear canal are derived from the first pharyngeal cleft.

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Figure 2 (a) In fish, oxygen enters the blood by diffusion from water flowing through the gills. (b) The anatomical structure of the gills maximizes the rate of diffusion, which is proportional not only to the surface areas exposed but also to differences in concentration of the diffusing molecules. Water, carrying dissolved oxygen, flows between the gill filaments in one direction; blood flows through them in the opposite direction. Thus the blood carrying the most oxygen meets the water carrying the most oxygen, and the blood carrying the least oxygen meets the water carrying the least oxygen. The result is that the oxygen concentration of the blood in any region of the filament is less than the oxygen concentration of the water flowing over that region. In fact, the concentration gradient of oxygen between the blood in the gill filament and the water flowing over it is constant along the entire length of the filament. Thus, the transfer of oxygen to the blood by diffusion takes place across the entire surface of the filament. (c) Enlargement of a section of a gill filament showing the lamellae and the flow of blood within the capillaries.
Figure 3 (a) When blood and water flow in opposite directions (countercurrent flow), blood becomes more saturated with \( \text{O}_2 \) than it would if blood and water flowed in the same direction (b). This is because in countercurrent flow, the difference in the \( \text{O}_2 \) content of the blood and the water is kept constant. If blood and water flow in the same direction, the difference in oxygen content disappears. Since it is the difference in the oxygen content of the two liquids that drives diffusion, without a difference no further change occurs.

How Mammals Breathe—Lungs as a Respiratory Surface

In general, the lungs function as the center for gas exchange in mammals, but accessory structures such as the nasal cavity, surrounded by the sinuses, the pharynx, larynx, trachea, and the bronchus and its divisions, are also included when studying the respiratory system.

Air passing through the nasal cavity is warmed, moistened, and filtered by the mucosa of the nasal passages. Olfactory receptors in the nasal cavity provide us with a sense of smell, and the surrounding sinuses can act as resonance chambers for speech. Because the sheet of warm, moist mucosa that lines these areas is continuous, nasal infections often spread throughout these cavities and passages. The nasal passages are separated from the mouth (oral) cavity below by a partition, the palate (anteriorly, the hard palate, and posteriorly, the soft palate). In addition to entering the nasal passages, air can also enter the mouth.

Air moves from the nasal and oral cavities through the oral cavity into the pharynx, where it passes over the larynx, commonly referred to as the Adam’s apple. The larynx consists of nine cartilages. One of these, the epiglottis, forms a lid over the larynx when we swallow so that food is routed into the esophagus rather than into the trachea lying beyond the larynx. If anything other than air tries to pass, a cough reflex expels the material. The trachea is lined with a ciliated mucus-secreting epithelium. The cilia beat in unison to propel debris trapped in the mucus upwards to the oral cavity. The walls of the trachea are reinforced with C-shaped cartilage rings to provide support and flexibility.

After passing through the trachea, air moves into the right and left bronchi (singular, bronchus), which divide into smaller and smaller branches called bronchioles (Figure 4). These bronchioles are further subdivided to form alveolar ducts that end in alveolar sacs resembling clusters of grapes. The walls of the alveoli (terminal sacs) are composed of a single layer of epithelium that allows for gas exchange (by diffusion) with the web-like mass of capillaries in the lung tissue (Figure 4).

This gas exchange depends on differences in the concentrations of \( \text{O}_2 \) and \( \text{CO}_2 \) in the alveoli and in the lung capillaries (Figure 4). Gas concentration is expressed in terms of partial pressure, which indicates the amount of atmospheric pressure exerted by one gas in the mixture of gases that make up 1 atmosphere.

The tissue of the lungs (other than respiratory passageways and capillaries) is mostly connective tissue. The lungs are covered by a thin membrane, a pleural membrane (visceral pleura). A second pleura membrane (parietal pleura) is attached to the walls of the thorax (chest cavity). The two pleural membranes lie close to one another, and each produces a lubricating fluid that fills the space between them. Pleurisy is a condition caused by inflammation of the pleural membranes and can be quite painful.
Figure 4 The human respiratory system. Air enters through the nose or mouth and passes into the pharynx, past the larynx (voicebox), and down the trachea, bronchi, and bronchioles to the alveoli in the lungs. The alveoli, of which there are some 300 million in a pair of lungs, are the sites of gas exchange. O$_2$ and CO$_2$ diffuse into and out of the bloodstream through the capillaries surrounding the walls of the alveoli.

Objectives

- Describe the anatomy of the mammalian respiratory system.
- Explain how the respiratory and circulatory systems are related anatomically and functionally.
Figure 5. Gases are exchanged via diffusion as a consequence of the different concentrations (partial pressures) of O2 and CO2 in the alveolus and alveolar capillary. Numbers indicate partial pressure in millimeters of mercury.

Procedure

Examine the "sheep pluck" on demonstration. A sheep pluck includes the lungs and heart as well as the major blood vessels, the pulmonary artery and veins that carry blood to and from the heart. Identify the trachea and larynx. Try to feel the C-shaped cartilages in the trachea.

- What happens to these cartilages as the trachea divides into bronchi and as bronchi divide into the smaller bronchioles?
- Why are these supporting elements necessary?

Identify the lungs.

- How many lobes are present in the right lung? Left lung?

If available, place a hose attached to an air compressor into the trachea and allow air to pass into each of the lungs. Notice how the lungs inflate. Describe what you observe.

Locate the pulmonary artery and veins. In your lab notebook, diagram the passage of blood from the heart to the lungs and back to the heart. Indicate the names of the vessels and whether oxygenated or deoxygenated blood is found in each.

- What kind of blood is carried by the pulmonary artery?
- What kind of blood is carried in other arteries?
- Why is the pulmonary called an artery?

Obtain a cross section lung tissue. Observe the specimen at 40X; identify the epithelium of the alveoli and red blood cells within the capillaries embedded in the lung tissue. The lung's connective tissue is filled with elastic fibers among the cells surrounding the alveoli.

- Suggest why there is an abundance of elastic fibers in this tissue.

Look for the large nucleated white blood cells, called monocytes. These cells transform into macrophages that ingest foreign particles and fight infections.

- Why would it be advantageous for such cells to be present in the connective tissue of the lungs?

Obtain a prepared slide (cross section) of the trachea. Note the ciliated epithelium and hyaline cartilage support rings surrounding the trachea.

Use Figure 5 to explain how oxygen and carbon dioxide are exchanged between lung and capillaries.
Respiratory Pigments

Objectives

- Describe why respiratory pigments are an important component of the blood.
- Describe how differences between the oxygen concentrations in the blood and in the body tissues cause oxygen to diffuse into the tissues.

Whenever a circulatory system is responsible for the transport of oxygen, a respiratory pigment is usually present. All such pigments contain a metal ion capable of combining with oxygen; most contain iron, but some contain copper.

Hemoglobin, the respiratory pigment present in all vertebrates (and many invertebrates), consists of four subunits, each an iron porphyrin (heme) coupled with a globin protein molecule. In solution, hemoglobin appears pink. Chlorocruorin, present in polychaete worms, also contains iron and appears green in solution. Hemerythrin, a third iron-containing pigment, is violet in color and is found in polychaete worms and the brachiopod Lingula. In mollusks and some arthropods, the copper-containing respiratory pigment hemocyanin colors blood blue.

A molecule of hemoglobin is capable of carrying four molecules of oxygen. They are added one at a time—the binding of one increases hemoglobin’s ability to bind the next. Whether oxygen combines with hemoglobin or is released from the oxygenated hemoglobin depends on the oxygen concentration (measured in terms of partial pressure of oxygen, Po) in the surrounding blood plasma. When the concentration of oxygen is high, as in the capillaries of the lung, most oxygen is bound to hemoglobin. In the body tissues where the concentration of oxygen is low, oxygen is released from the hemoglobin molecule into the plasma and diffuses into the tissues (refer to Figure 5). Carbon dioxide released from the tissues is picked up in the blood capillaries. Some CO₂ is dissolved in the plasma and some binds to the hemoglobin of the red blood cell, but most of the CO₂ picked up by red blood cells combines with water to form carbonic acid. When carbonic acid dissociates, it forms bicarbonate (HCO₃⁻) and hydrogen ions (H⁺).

- How does an increase in the concentration of H⁺ affect the pH of a solution?

Most of the HCO₃⁻ diffuses out of the red blood cells into the plasma, and some of the H⁺ ions, which could potentially reduce the pH of the blood, are bound to hemoglobin molecules within red blood cells. HCO₃⁻ can recombine with H⁺ in the plasma or in red blood cells, helping to buffer the blood. (Buffers prevent large changes in the pH of the blood by accepting H⁺ when its concentration rises above normal and giving up H⁺ when its concentration falls below normal. See Laboratory 3, Exercise E) Even though blood is buffered, increasing the CO₂ concentration in plasma does decrease the pH of the blood. As acidity increases, hemoglobin’s ability to bind oxygen decreases and the oxygenated hemoglobin gives up more oxygen to the tissues that need it.
Lung Volumes
Normal breathing usually moves about 500 ml of air into and out of the lungs. This is called **tidal volume (TV)**. After normal inspiration, you still have room for more; approximately 3,000 ml of air can be inspired forcibly. This forced inspiration is the **inspiratory reserve volume (IRV)**. You can also forcibly expel air, about 1,100 ml. This is the **expiratory reserve volume (ERV)**. Vital capacity of the lungs is calculated as follows:

\[
\text{Vital capacity} = TV + IRV + ERV
\]

Vital capacity falls within the range of about 3,000 to 5,500 ml. Variations in body size, age, and sex account for individual differences.

**Objectives**
- Determine the vital capacity of your lungs.
- Understand the relationship between tidal and reserve respiratory volumes.

**Procedure (you can also do all this with the spirometers...)**
1. Stretch a balloon several times before blowing into it.
2. Take a normal breath and then exhale into the balloon, emptying your lungs normally.
3. Hold the end of the balloon tightly so that no gas can escape. Place the balloon next to a ruler and measure its diameter in centimeters. (Placing a piece of paper or cardboard on each side of the balloon may help you to read the diameter accurately.) Use the graph in Figure 6 to convert the diameter of the balloon to liters of air. This value represents tidal volume (TV).

![Figure 6](image)

**Figure 6** Measuring lung capacity (1,000 x capacity in liters = capacity in milliliters)

Again, take a normal breath and exhale. Now place the balloon in your mouth and forcibly exhale any air that you can. Measure the balloon and convert this to volume of air. This represents the expiratory reserve volume (ERV).

\[
ERV = \text{liters.}
\]

Calculate the vital capacity of your lungs (TV + IRV + ERV).

\[
\text{Vital capacity} = \text{liters} = \text{milliliters}
\]

How does your result compare with those for 20-year-olds given in Table 7?
Table 7 Normal Vital Capacity (in milliliters) for 20-Year-Olds*

<table>
<thead>
<tr>
<th>Height (in inches)</th>
<th>Males Normal Vital Capacity</th>
<th>Females Normal Vital Capacity</th>
</tr>
</thead>
<tbody>
<tr>
<td>60</td>
<td>3,885</td>
<td>58</td>
</tr>
<tr>
<td>62</td>
<td>4,154</td>
<td>60</td>
</tr>
<tr>
<td>64</td>
<td>4,410</td>
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<td>66</td>
<td>4,675</td>
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<tr>
<td>74</td>
<td>5,736</td>
<td>72</td>
</tr>
</tbody>
</table>


How Does Smoking affect Lung Capacity?

Hemoglobin binds carbon monoxide (CO) more rapidly and strongly than it binds O₂. This is why a person can die even when O₂ is available but CO concentrations are high: hemoglobin will preferentially bind CO, which cannot be used in cellular respiration. By binding too many of the hemoglobin sites that are normally occupied by O₂, the CO in cigarette smoke reduces the blood's ability to carry O₂. In response, the body makes more red blood cells. As a result, the blood of smokers is usually thicker than that of nonsmokers, making the heart pump harder. Stress on the heart increases the incidence of heart attacks in smokers.

Smoking also affects the lungs by building up tars within the lung membranes. When enzymes in the cells begin to digest the tars, they may also digest the cell membranes themselves. This often results in the onset of emphysema, a disease characterized by deterioration of the walls of the alveoli and a loss of elasticity in the lung tissues.

- What do you think smoking does to the vital capacity of the lungs? Is there a smoker in your class or among your friends? Formulate a hypothesis about how smoking might affect lung capacity.

HYPOTHESIS:

NULL HYPOTHESIS:

What do you predict will be the outcome of measuring lung capacity in smokers and nonsmokers?

What is your independent variable?

What is your dependent variable?

Follow the procedure outlined in Respiratory Volumes, and record your results in Table 10 (provided).

- Do your results support your hypothesis or your null hypothesis?
- What do you conclude about the effects of smoking on respiration?

Note: To obtain valid data to support your conclusion, you would need to compare the lung capacities of large
numbers of smokers and nonsmokers matched for many variables, including age, height, and weight.

**Effects of Exercise on Heart and Respiratory Rates**

**Objectives**
Determine how exercise, both moderate & vigorous, affects respiratory rate.
Explain how the elevation of pulse rate is related to increasing respiratory rate.

**Procedure**

1. **Work in pairs.** Determine your partner’s sitting pulse rate.
2. **Listen closely to your partner’s respiration rate.** In Table 8, record the number of breaths per minute, and describe the depth of breathing in the "Before Exercise" column.
3. One partner should now exercise moderately by raising alternate knees to the chest for approximately 30 seconds. The other partner should exercise strenuously by running up several flights of stairs as quickly as possible.
4. In Table 8, record the pulse rate, the rate and depth of respiration, the time required for the return of normal respiration, and the time required for the return of normal pulse rate for your partner in the appropriate column (moderate or heavy exercise).
5. **Now repeat steps 3 and 4 with each partner performing the alternative type of exercise.**
6. **Record the data for your partner in Table 8.**

<table>
<thead>
<tr>
<th>Table 8</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
</tr>
<tr>
<td>Before Exercise</td>
</tr>
<tr>
<td>Pulse Rate</td>
</tr>
<tr>
<td>Respiration Rate – depth of respiration</td>
</tr>
<tr>
<td>Time to return to normal respiration</td>
</tr>
<tr>
<td>Time to return to normal pulse</td>
</tr>
</tbody>
</table>

The speed with which the heart returns to the sitting pulse rate after exercise serves as one index of circulatory efficiency. Here is an example of a normal response:

<table>
<thead>
<tr>
<th>Pulse</th>
</tr>
</thead>
<tbody>
<tr>
<td>Before exercise</td>
</tr>
<tr>
<td>Immediately after exercise</td>
</tr>
<tr>
<td>2 minutes after exercise</td>
</tr>
</tbody>
</table>

- *How is increased heart rate (measured as pulse rate) related to increased respiratory rate?*

- *As respiratory rates return to normal following exercise, what happens to the pulse rate?*

- *Can you explain why the observations you reported in a and b occur?*
Physiology of Respiration – the lung model

Breathing (pulmonary ventilation) requires the movement of air into and out of the lungs. This movement of air occurs when there is a difference between the pressure inside the body and the atmospheric pressure outside the body. The pressure inside the body in the space surrounding the lungs is changed primarily by the action of the **diaphragm**. When this dome-shaped muscle contracts, it moves downward, thereby expanding the chest cavity. The volume in the chest cavity is increased as a result. The increase in volume causes the pressure of the gas to decrease inside the chest. The pressure decrease occurs because gas molecules have more room to spread out once the volume expands. (Pressure and volume are inversely proportional. \( PV = kT \).) Gases flow from areas of high pressure to low pressure, thus air flows into the lungs when pressure inside the chest is less than it is outside the body. This movement of air into the lungs is inhalation (or inspiration). Air moves out of the lungs when the diaphragm muscle relaxes and moves upward, returning the chest cavity to the lower resting volume. Air pressure in the lungs increases because once the volume decreases, the gas molecules have less space and are crowded together. Pressure outside the body is now lower than inside the chest, so the gas molecules move out of the body. This is exhalation (or expiration).

The functional lung model (Figure 10) demonstrates that air pressure differences cause the lungs to expand and contract. The role of muscle contraction and relaxation is to expand and contract the chest cavity, which reduces or increases the pressure and allows air to move down its pressure gradient.

**Figure 9**

![Figure 9](image1.png)

**Figure 10** Lung model. Operate the lung model by pulling down on the knotted end of the large balloon. This will expand the volume inside the bottle and cause air to enter the balloon "lung." Release the knotted end of the balloon to decrease the volume of the bottle, causing air to leave the "lung."
Respiratory Volumes

Figure 10 depicts a graph of respiratory volumes. A wet spirometer will be used to measure the amount of air exhaled into the spirometer. (The spirometer does not directly measure volume of air inhaled, and you should never inhale through the spirometer tube.) On the spirometer, each number from 1 to 6 represents volume in liters and each smaller mark represents 0.1 liter or 100 milliliters. Measure the respiratory volumes in milliliters. Before beginning, set the marker (rubber disk) at 0. Each person should attach an unused cardboard tube or plastic tube (mouthpiece) to the spirometer hose before using the spirometer. Dispose of used mouthpieces in the biohazard bag. Measure each respiratory volume described below, for one group member.

**Tidal volume**: Tidal volume is the amount of air inhaled and exhaled during normal breathing (about 500 ml). Take a normal (shallow) breath and exhale normally into the spirometer. Do not force the expiration! (Note: Remember to inhale without the spirometer tube in your mouth. Exhale into the spirometer.). Do this three times, find the average, and record your results (Table 10).

**Inspiratory reserve volume**: This is the volume of air that can be forcibly inhaled following a normal inspiration (about 2,800 ml). Breathe normally several times. Take the deepest breath possible and exhale normally into the spirometer tube. (Do not forcefully exhale). Do this three times, find the average, subtract the average tidal volume, and record your results (Table 10).

Figure 11 Idealized tracing of the various respiratory volumes

<table>
<thead>
<tr>
<th>Respiratory Volume</th>
<th>Trail 1</th>
<th>Trail 2</th>
<th>Trail 3</th>
<th>Average</th>
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<tbody>
<tr>
<td>TV</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>IRV</td>
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<td></td>
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</tr>
<tr>
<td>ERV</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>VC (measured)</td>
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<tr>
<td>VC (calculated)</td>
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