1800 Cellular Chemistry - Teacher Manual

The materials provided in this kit allow up to 15 pairs of students to complete a series of experiments identifying the presence of the following molecules in samples of potato, fresh meat, and whole milk:

- carbohydrates (sugars and starches)
- proteins
- lipids

The kit also provides a brief discussion on the topics of nucleic acids, vitamins, and minerals. The material is best suited to high school biology and chemistry classes and can be completed in one to two 45 minute class periods. Students should be familiar with chemical symbols before using this kit.

Contents:

Microscope Slides (72)
Cover Slips (100)
Forceps (6)
Petri Dishes (10)
LabForms (15)
Razor Blades (15)
Pipets (2)
Chemstrip (1 container)
Filter Paper (20 disks)
Biuret Solution (2 - 30 ml bottles)
Ninhydrin Solution (2 - 30 ml bottles)
Sudan IV Solution (2 - 30 ml bottles)
Lugol's Solution (2 - 30 ml bottles)

Teacher's Manual (1)
Student Instructions Master (1)

Additional Materials Required:

Sample of whole milk or cream
5-6 oz. of fresh meat (with fat)
One potato
Microscopes (300X or better)
Access to a sink
Well ventilated area (fumehood)
Adjustable hot plate

Introduction

Most of the recent theoretical and practical advances in the field of biology have been made in the area of biochemistry. Both teaching and research have been concentrating on understanding the complex web of chemical reactions that occur in every living cell. Cell biologists especially have tried to understand the chemical interactions between the various cellular components. Great progress has been made in understanding the ways in which the cell manipulates its chemical environment to manufacture new compounds and break down existing ones.

The study of cellular chemistry depends on the recognition of certain essential groups of compounds - carbohydrates, proteins, lipids, and nucleic acids. In turn, an understanding of these compounds is based on a knowledge of their basic components - sugars, amino acids, fatty acids and glycerol, and the nucleotide bases. Study of these basic topics in cellular chemistry is a fundamental part of most modern biology courses.

This kit is designed to present the biology student with a basic discussion of these fundamental chemical groups. The student identifies the compounds with specific chemical tests, and learns something of their distribution in, and their importance to, the life of the cell.

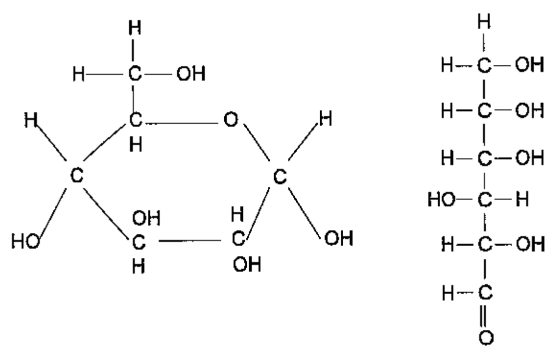
The kit is best utilized with students who have had some background in chemistry and cellular biology. The kit makes reference to several aspects of cellular structure, but a detailed background is not necessary. The students should be able to use a microscope, and should be able to handle laboratory chemicals. Discussion of cellular structure beyond that contained in this kit is recommended. The instructor may with to utilize the other kits on cell life, such as The Science Source kit #1700 on Cell Structure.

Background Information

The study of cellular chemistry is greatly simplified by understanding that most of the important chemical compounds in the cell fall into one of four groups - proteins, carbohydrates, lipids, and nucleic acids. This discussion provides an introduction to these basic groups, their characteristics, and their importance to the life of the cell.

Carbohydrates

As their name suggests, carbohydrates are composed of carbon and the elements of water: hydrogen and oxygen. The structure of glucose, which can exist either as a chain or a ring, is shown below:



For simple carbohydrates, these elements are present in the ratio $(CH_2O)_n$. The most common examples of simple carbohydrates are the simple sugars, such as glucose, $C_6H_{12}O_6$, or related compounds such as pyruvic acid, $C_3H_6O_3$.

Carbohydrates can form polymers of basic sugar units, glucose being the most common sub unit. Simple polymers are formed by joining two sugars together by dehydration synthesis, in which a molecule of water is "released". When two sugars are joined together, the resulting molecule is called a disaccharide. Many sugars may be hooked together into a straight or branched chain. Examples of these polysaccharides are starch and cellulose. The only difference between starch and cellulose is the orientation of the bonds between the sugar units. The bonds of starch are oriented in a way that is susceptible to our digestive enzymes, and those of cellulose can be digested only be specialized organisms.

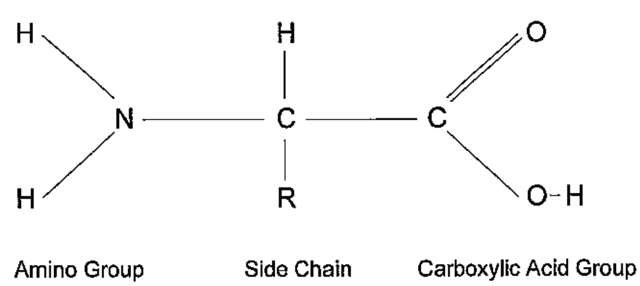
Carbohydrates are very important in energy metabolism. The processes of energy metabolism- glycolysis, the Krebs cycle, and electron transport - most often begin with glucose. Food is stored in cells in the form of *glycogen* in animals, and *starch* in plants, both of which are carbohydrates. Carbohydrates are also important in the structural makeup of the cell, particularly in cell walls of plants. Layers of cellulose molecules in these cell walls provide a tremendous degree of stability and support to the cells.

Carbohydrates are also found combined with proteins in compounds called *glyco-proteins*. The study of this fascinating group of molecules has begun only recently. Already, however, glycoproteins are known to be involved in cell recognition, many enzymatic reactions, control of disease organisms, and the control of cellular growth and division.

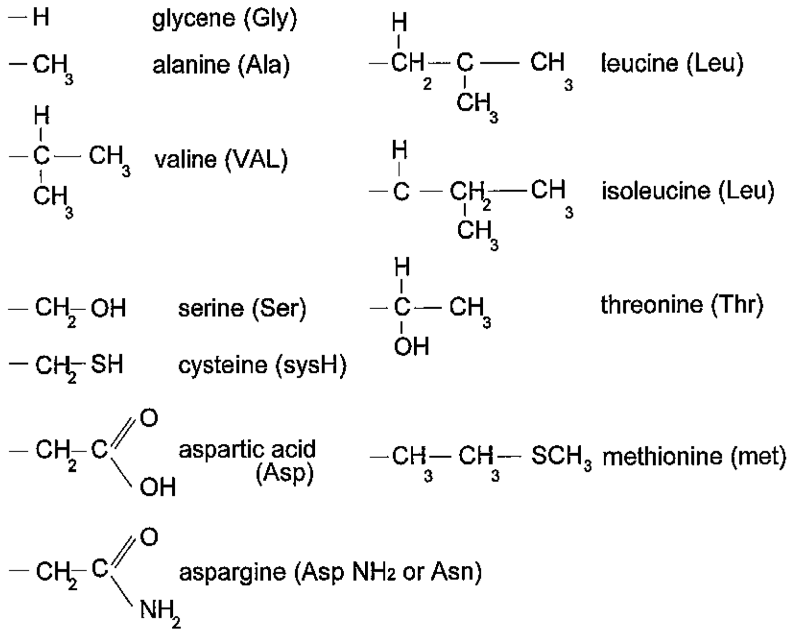
Proteins

In many ways, proteins are the most important molecules in the life of the cell. All enzymes are proteins. Enzymes catalyze most of the important chemical reactions in the cell, and are the principle link in cell activity. Beside their role as enzymes, proteins are important as structural components of cells, such as membranes and microtubules; extracellular components (e.g. collagen fibers); muscle activity; antigen/antibody reactions, etc.

Proteins are polymers of amino acids. All amino acids have the following basic structure:

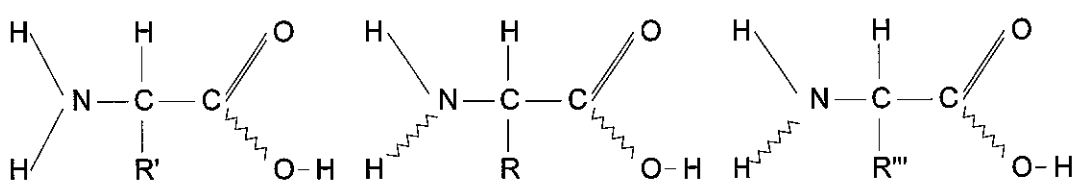


Some examples of R groups are:



The R group distinguishes one amino acid from another, it being the only variable feature in amino acid structure. There are about 20 amino acids which are important to man, and about another 10 which occur in various forms of bacteria, plants, and other animals.

During protein synthesis, the different amino acids are linked together. The process of bonding is *dehydration synthesis*, similar to carbohydrate synthesis. The bond formed is called a *peptide* bond:



The sequence of amino acids is called the *primary structure* of a protein. A difference in only one amino acid is enough to completely change the overall properties of the protein. Sickle-cell anemia is caused by only one misplaced amino acid in hemoglobin. The ^^^ bonds are drawn as such to show where water (H_2O) is released when amino acids come together to form a protein. After water is lost the carboxyl carbon (C= O) binds with the amino group of the next amino acid, forming a peptide.

The C=O, or carboxyl groups, and the N-H, or amino groups on a chain of amino acids attract each other. In regions where the R groups are not large enough to prevent it, these attractions, called *hydrogen bonds*, twist the chain into a corkscrew shape. This helical structure is referred to as the *secondary structure* of a protein. Several regions within a single chain may show the helix configuration.

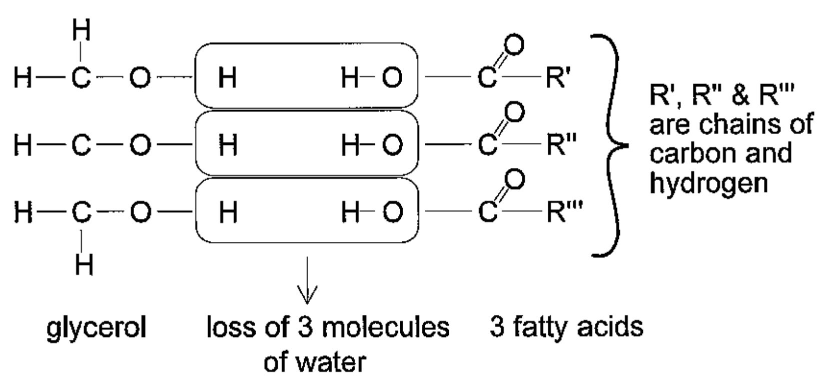
Other attractions occur in a chain of amino acids. The amino acid cysteine is an example. Sulfur atoms in two separate cysteine molecules, often spaced widely on the chain, or even on separate chains, bond to each other to form a *disulfide* bridge. This bridge pulls the two sections of the chain or chains together. In addition, the R groups of various other amino acids may hydrogen-bond to each other. The sum total of these interactions pull the protein into its overall shape, or *tertiary structure*. Some proteins, including many enzymes, have many interactions between R groups and are *globular* in shape. Others, like collagen, have few tertiary-level bonds between the R groups, and are linear or rod shaped.

The overall shape of a protein is related to its function. Enzymes in particular have an intricate relationship between function and structure. An enzyme must be able to bind onto a specific molecule. The region of the enzyme that attaches to this specific molecule is called the *active site*. Once the molecule is in the active sight, the enzyme must catalyze a reaction involving one or more of its bonds. The formation of the final product must somehow trigger the enzyme to release the new molecule. Active sites of enzymes are possibly the most complex level of protein structure.

Lipids

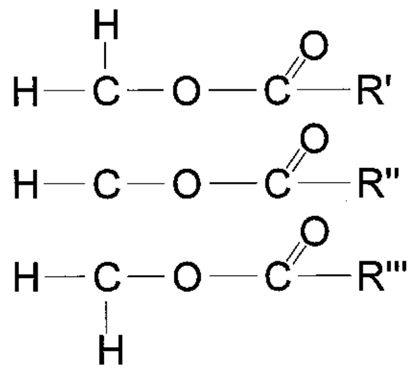
The lipids are a complex group of compounds. All lipids have fairly substantial hydrophobic, or *non-polar*, regions. The lipids include the fats, phospholipids, steroids, and a few other types of molecules. Lipids are important as energy sources for the cell, and as components of cellular membranes. The steroids are important as hormones and cellular control agents.

The most commonly known lipids are the *fats*. Fats are composed of a single molecule of glycerol and three fatty acid molecules, joined together:



In the above dehydration synthesis, three molecules of water are formed by the Hydrogens of the glycerol and the H-O of the fatty acids. These are lost, leaving the fat molecule:

(When fatty acids are needed by the organism, 3 molecules of water are added and an enzyme catalized reaction causes the break-up of the fat molecule, leaving the original reactants).



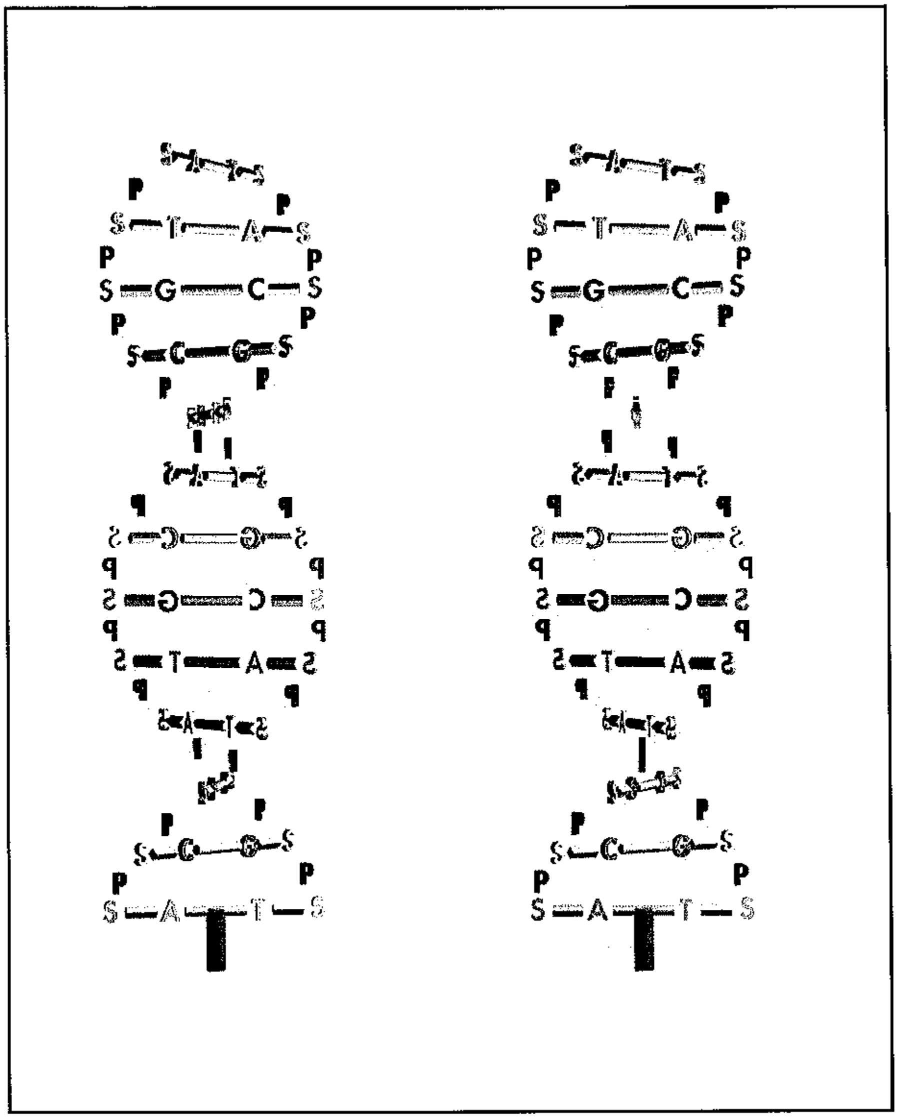
The fatty acid hydrocarbon chains may have only single bonds, in which case they are said to be *saturated*. If there are any double bonds between carbon atoms in the chain, the fatty acid, and the fat which it forms, they are called *unsaturated*. (Saturated fatty acids, characteristic of animal fats, are usually converted into cholesterol in the body, while the unsaturated fats found in plant oils tend to be metabolized in other ways.) The carbon chains may range from 3 or 4 carbons up to 20 or so. Each fatty acid molecule in a fat may be different.

Many of the lipids found in the membranes of cells and organelles have one of the fatty acid molecules replaced with a modified phosphate group. The resulting molecules are called *phospholipids*. Some lipids have carbohydrate side chains; these *glycolipids* are important in cell-surface phenomena, like antigen/antibody reactions. *Steroids* such as cholesterol, have a basic structure of three six-carbon rings and one five-carbon ring joined together. The difference among the many steroids is the specific side groups which are attached to the basic structure.

Nucleic Acids

The nucleic acids, *RNA* and *DNA*, provide the templates for all protein synthesis in the cell. They are responsible for genetic continuity and the expression of genetic information. The nucleic acids are polymers made up of subunits called nucleotides. Each nucleotide consists of a phosphate group, a five-carbon sugar (either *ribose* in RNA, or *deoxyribose* in DNA), and a nitrogen-containing base (cytosine, adenine, guanine, and thymine in DNA, cytosine, adenine, guanine, and uracil in RNA). These nitrogenous bases are either single or double ring molecules which have several C=O and N-H groups. Cytosine and guanine can hydrogen bend to each other, as can adenine and either thymine or uracil.

The overall structure of the RNA molecule is a long single strand. In DNA, two strands, bonded together by the hydrogen bonds between cytosine and guanine, and adenine and thymine, twisted into a double helix. The structure of DNA is shown schematically below:



Schematic representation of DNA. The components are: S(ugar), P(hosphate), A(denine), T(hymine), C(ytosine), and G(uanine). This figure represents a stereo pair that can be free-viewed. Simply relax your eyes, bring the figure up to your nose, then slowly move it away while continuing to stare off at infinity. The "central" image of the helix will appear in 3D.

DNA is found almost entirely within the nucleus, although small amounts of it are found in the mitochondria and chloroplasts. Where DNA is the "master copy", RNA is the genetic messenger, and is found throughout the cell. In the nucleus RNA is made as a copy of the DNA which can be sent out to the ribosomes in the cytoplasm of the cell. The ribosomes use the RNA as a template for joining amino acids together into proteins.

Nucleic acids are difficult to demonstrate histologically. The stains required involve extensive preparation and a considerable amount of time. For this reason, detection of nucleic acids has not been included in this kit. If you have prepared slides showing the distribution of nucleic acids within the cell, or showing the chromosome formation or division of mitosis, they would be an excellent addition to this activity.

Using The Kit In The Classroom

This kit is designed for junior or senior high school students of life sciences (and possibly health and nutrition as well). The kit presents an introduction to the important chemical components of the cell, and discusses major aspects of each compound's importance to the metabolism of the cell.

The objectives are that the student shall:

- Perform chemical tests to identify the presence of starch, glucose, amino acids, proteins, and fats, and shall demonstrate correct and safe laboratory techniques while running these tests.
- State the importance of each of the major groups of compounds discussed in the kit, including nucleic acids, vitamins, and minerals, in terms of cellular chemistry and structure.
- State the relationship between sugars and starches, and amino acids and proteins. Knowing that there are different types of sugars and different types of amino acids, the student will be able to infer the possible existence of many different types of starches and proteins.
- State the usefulness of chemical identification tests as a source of information of food analysis. Given samples of various materials, the student shall be able to determine the presence or absence of the major chemical groups discussed above.
- Observe the distribution of fat and starch within cells, using correct microscopic technique, and record his/her observations by drawing.

This kit is intended to be used in connection with a wider discussion of cellular function and structure. In particular, the study of cellular organelles (The Science Source Kit #6800,"Organelles") and the aspects of enzyme function are recommended.

The kit may be useful in consumer science and nutrition courses. In these courses, the student is concerned with the composition of foods; the kit provides a vehicle for considering the importance of various nutrients, and particularly chemical requirements, of the cell.

Before using the kit, the student should be familiar with chemical symbols. The use of models and diagrams of chemical structure is useful in discussing the chemical nature of each type of compound. The Background Information section of this teacher's manual, and various reference and text books, are good sources.

The students should be able to handle laboratory chemicals in a safe and organized way, and they should be able to use a microscope properly. Before using the kit, you may want to have the students practice making slides and observing them under the microscope.

If you do not feel comfortable having the students use razor blades, the materials may be prepared in advance. See the next section for preparation of materials.

Preparation of the Materials

The student should have access to microscopes of 300X capability or better, with one scope for each pair of students. Check the scopes beforehand to be sure that they are in working order. Have lens tissue available for cleaning lenses and mirrors.

The students will need samples of fresh milk, meat and potato. Two or three potatoes for a class of 30 students is enough. Use fresh meat rather than meat that has been frozen, and whole milk or cream rather than skim milk. The students will need access to a sink, preferably several sinks around the room. The ninhydrin test for amino acids requires a well ventilated area, such as a fume hood (CHECK TO BE SURE IT'S WORKING), and a heat source. The best source is an adjustable hot plate turned to a very low heat. DO NOT USE AN OPEN FLAME. Alternately, you could use a heating oven for overnight use.

If the students are not going to use the razor blades, make several dozen thin slices of potato for the microscopic examination. Also, prepare several dozen small cubes of meat and fat, in separate dishes. Store both of these in the refrigerator before using. You can use semi-dull scalpels for the activities that do not require a delicate cutting procedure.

Guide To Student Activities

The introduction directs the student's attention to the importance of the study of cellular chemistry. It may be interesting to have the students look up the number of Nobel Prize Winners in medicine and biology who have been chemists and biochemists. The introduction also points out the need for careful and organized laboratory procedures. Stress this point at the beginning of the activities.

Have the supplies and materials available in a central location for general use. Be sure that students are familiar with the use of the microscope before they begin the actual experiments.

Part A deals with identifying carbohydrates. Chemstrips are a product used by diabetics to test urine for excess sugar. This test is specific for glucose. The students are given the chemical formula for glucose. Point out that many sugars, such as fructose and galactose, have the same formula but different structures (these are called isomers). Also point out that the 1:2:1 ratio of carhon:hydrogen:oxygen is characteristic of simple sugars.

Lugol's solution, which contains iodine, is used to identify the presence of starch. The iodine/starch complex is a deep blue or purple color. The student observes a sample of potato microscopically for the distribution of starch. Advise the students to sketch the cells lightly and to add detail after they have drawn rough outlines. Potato characteristically has large starch granules, which will stain blue with the Lugol's solution. Relate the large volume of starch granules to the function of the potato as a food storage area for the plant.

Beside starch granules, the student will be able to see the cell walls of the potato cells. The cell walls are composed of cellulose combined with other carbohydrates and proteins. The color of the walls will not be as deep as the color of the granules. Most text books have pictures showing the structure of the cell walls, which may be of interest to the students.

Part B deals with identifying amino acids and proteins. The Biuret test identifies the carbamyl groups ($CONH_2$) found in proteins. A positive result, which is a pink color, may be difficult to see in the case of the meat. The LabForms may be re-used after rinsing. Be sure that anyone in contact with the Biuret solution wash their hands thoroughly. You may want to refer the students to relevant reference materials for a further discussion of proteins. This kit does not discuss the chemical structure of proteins or amino acids, although you may refer to the Background Information in this Teacher's Manual.

The test for amino acids utilizes the reagent ninhydrin, which reacts with amino acids in a multi-step reaction, producing a pinkish-purple color. Since the ninhydrin is in an acetone solution, which is irritating to the skin, eyes, and lungs, the procedure should be carried out in a well ventilated area. A well circulating fume hood is the best area for this.

In the ninhydrin test, the student will see a colored shape on the filter paper identical to that of the piece of potato which was used. In addition, the student should be able to observe his own fingerprint on the paper. Incidentally, ninhydrin is used by police for "lifting" fingerprints from rough surfaces, which do not permit the use of the standard powder technique. Both the potato and the meat will give a positive test with the ninhydrin.

After adding the ninhydrin, the paper should be completely dried before heating. Heating speeds up the rate of formation of the colored compound. If an adjustable hot plate is not available, have the students leave the papers overnight in a warm location, like a heating oven or the top of a radiator. Make sure students write their names in pencil on the paper before leaving them.

Part C deals with the identification of fats. This process depends on the ability of the nonpolar dye, Sudan IV, to dissolve in fat. The dye is absorbed by the fat droplets in the milk, and will appear under the microscope as small pink spheres. Show the solubility of the Sudan IV by filling a container with about 1 cm of water, adding some dye, then adding several drops of vegetable oil and shaking. The dye migrates from the water to the oil.

Fatty tissue cells have large fat droplets which stain quite vividly with the Sudan IV. Keep the meat cold (not frozen) before doing this experiment. The cytoplasm and nucleus of the cell may appear as a ring around the fat droplet, hence the name for these cells: signet cells. Encourage the students to think of the importance of storing energy in the form of fats.

The nucleic acids and vitamins are difficult to stain with simple procedures. The student materials discuss the basic structure and importance of these molecules. Point out the relevant sections in their texts and reference books for further discussion. The students should be aware of the importance of vitamins: their role as co-enzymes is mentioned in the student materials.

The student is told to try testing additional samples of material. You may wish to orient this activity toward an analysis of various foods. Some foods which provide interesting results are: bread, butter, cheese, cereals, fruits, candy, and the powdered soy proteins being sold as a meat additive.

Further Topics For Discussion

After you complete these activities, you may want to pursue some of the following additional topics.

1 Investigate the structure of the cell, including where various compounds are made, broken down, or used within the cell. Discuss the function of organelles within the chemical life of the cell. The Science Source kits **#1700 Cell Structure,** and **#6800 Organelles,** may be useful sources for these topics.

2 Discuss the mechanisms and features of enzyme activity. Relate the wide variety of enzymes to the complex nature of proteins. Investigate the relationship between enzymes and the other major groups of compounds: digest protein samples with protease enzymes, such as trypsin. The Science Source kit **#1000 How Poisons Work** presents an investigation of enzyme characteristics.

3 Discuss the structures and metabolism of the various groups of compounds. The processes of protein synthesis, photosynthesis, the breakdown of sugars, etc, may be studied at various levels of sophistication. Have the students develop the idea of dehydration synthesis as a basic means of polymer formation.

4 Discuss diseases related to abnormalities in chemical metabolism. Diabetes, for example, results from an inability of the body to store and break down sugar. Protein deficiencies result in many serious consequences, among them failure of brain cells, which rely on protein to develop adequately. Vitamin deficiencies, and excesses, produce a variety of effects.

CELLULAR CHEMISTRY #1800 Student Instructions (Annotated)

Introduction

In many ways, biology is actually a study of the chemistry of life. Biologists now think of life as a series of chemical reactions. Most of the important discoveries in biology in the past 30 years have been made by biochemists. To fully understand the biological processes of birth, death, growth, and disease, biochemists will need to first understand the chemistry of life.

The chemical life of the cell is very complicated. It is possible, though, to define a few basic groups of compounds which are most important to the cell. In this kit you will identify these basic groups, and learn something of how they function within a cell.

Biochemists test living material and identify these chemical groups with specific chemical tests. You will be using a variety of testing methods, some of which will involve potentially dangerous laboratory chemicals. Make sure to follow all directions and to use careful laboratory procedures.

PART A - Sugars and Starches - The Carbohydrates

Work in groups of two for these activities. Obtain the following pieces of general equipment: one LabForm, one plastic dish, one razor blade, two glass slides, two cover slips, and a microscope. Before you begin, be sure you know how to focus and use the microscope correctly. Ask your teacher for assistance if you are not sure.

1a) Get a small piece of potato, and place it in the Petri dish. Make a thin slice from the potato, place it flat on the dish, and describe its appearance.

Pale yellow or white, surface is damp. Many small bumps or particles in the potato

1b) Obtain one piece of Chemstip. Touch the reagent end of the strip to a moist surface of the potato, and let the Chemstrip set for 1 to 2 minutes. Examine the color chart on the Chemstrip container, and compare with the various shades. The darker the green/blue color, the higher the concentration of a sugar called *glucose*. According to the chart, what do you conclude about the concentration of glucose in the potato?

The potato has a concentration of about 2000-3000mg/dL. (This is the concentration of sugar in the fluid, not in the solid material)

Glucose is a simple sugar made up of carbon, oxygen, and hydrogen. Oxygen and hydrogen make up water. Combining the word carbon with the Greek word for water (hydro), we get the name carbo-hydrate. All animal and plant cells contain many different kinds of sugars. Sugars are an important source of energy for the cells: sugar is often called an "energy food". Simple sugars all have the formula $(CH_2O)_n$. For example, the formula for glucose is $C_6H_{12}O_6$.

2a) If it is available, test a small piece of meat for the presence of sugar in the same way you tested the potato. Record your results.

The strip turned light green. The meat has sugar also, but not as much as the potato.

2b) Meat is actually muscle tissue of an animal. Why would sugar be important to muscle cells?

Muscles need energy to work and move. The sugar could be a source of energy for them.

The cell contains other carbohydrates besides simple sugars. Sometimes the cell joins sugars together into chains. A very simple chain is the molecule sucrose, which is two sugars hooked together (a disacharide), a glucose molecule, and another molecule called fructose. Sucrose is the table sugar that you put in coffee or on cereal.

3 Cells can make very long chains of sugars called *starches.* Starches are one way in which the cell stores energy. A sugar molecule that is not being used by a cell must be bound into a large starch molecule so that it is not lost.

a) Place a drop of Lugol's solution on the slice of potato, and notice the reaction which occurs. Describe what you see:

The potato turns blue (or purple). Close examination will show many small blue dots floating around in the liquid.

Lugol's solution contains iodine, which will combine with starch molecules to produce a dark blue or purple color. The darker the color, the more starch that is present.

b) Use the razor blade to make a very thin slice of potato. A good technique is to slice away a wedge shaped piece of potato, and to observe the thinnest section. Use forceps to carefully place the slice of potato on a clean glass slide. Add a drop of the Lugol's solution, and place a cover slip over the slice of potato. Place the slide on the microscope stage, and focus on the slide, using first low power, then high power. Make a drawing of what you see.

c) From what you know about the test for starch, what can you say about where the starch is inside of the potato cells?

The starch is stored in granules or sacks inside the cells. This is where most of the color is.

You probably noticed that surrounding the cells was a coating which showed some of the purple or blue color. This material is the cell wall. The cell wall is composed of a special carbohydrate called *cellulose.* Cellulose is similar to starch, but is much more resistant to chemicals and breakdown.

PART B - Proteins

1) Get fresh pieces of potato and meat (both about 1 cm on a side). Using the razor blade, chop up the piece of potato in the Petri dish. Scrape the chopped potato into one tube of the LabForm, and add about 1 ml of water. Rinse the razor blade, and repeat the same procedure with the meat, putting it into a clean tube of the LabForm.

a) Get the bottle of Biuret reagent, and describe the color of the solution.

Pale blue

b) Carefully , to avoid skin contact, add 10 drops of the test solution to each of the two tubes in the LabForm. Let the mixtures sit for about 1 minute, then observe the color of the solution. Record your results.

In both tubes, the solution is pale pink There is also a bit of cloudiness to the solution.

The color change is the reaction between the Biuret solution and an important group of compounds in the cell called *proteins.* Proteins contain carbon, hydrogen, oxygen, nitrogen, and sometimes sulfur, and are essential to the life of the cell. They are a part of the cell membrane which surrounds the cell. Enzymes are complex proteins which *catalyze* (help) the necessary chemical reactions in a cell. For example, if a cell wanted to build starch to store sugar for energy, it would use an enzyme to link the sugar molecules.

2) In the same way that cellulose and starches are chains of sugar molecules, proteins are also long chains of a certain type of molecule. They are made up of smaller molecules called *amino acids.* Let's test cells for the presence of amino acids.

Get a piece of filter paper at least 1" X 1" . Handle the filter paper with the forceps. Cut a small piece of fresh potato and press one surface of the potato onto the piece of filter paper. If the piece of filter paper is large enough, carefully press your fingers on a section of it away from where the potato was pressed, as if you were making a finger print.

You will use a reagent called ninhydrin to test for the amino acids. It will react with any amino acids that are left on the paper. The ninhydrin is dissolved in acetone, which is a rather irritating solvent. *Work in a well ventilated area* for this activity. Add 4 to 5 drops of ninhydrin to the paper, spreading the drops over the area where the potato was pressed. Holding the paper with the forceps, wave it in the air to evaporate the acetone.

a) After the paper is completely dry, place it on a warm surface, such as a radiator, or on a desk in the sun. If your teacher directs, place the paper, with your name in pencil, in a heating oven overnight. Observe the paper, and record your observations.

   There is a spot of pinkish-purple color where the potato was pressed on the paper. There is also an outline of the fingerprint in the same pinkish color on the paper.

b) If you have time, blot a piece of fresh meat on a new piece of filter paper, and repeat the addition of ninhydrin, drying, and heating procedure (If you do this, remember to mark whether the paper is testing the meat or the potato). Does meat contain amino acids?

   The meat also contains amino acids, since it produced a pinkish-purple spot.

c) Why do you think it might be important for cells to have amino acids?

   The amino acids are the building blocks for new proteins. The cell might need a specific protein, and could not wait for it to just float by.

PART C - Lipids

1a) A third group of compounds contained in the cell is called *lipids.* This group, the lipids, includes substances like fats, oils, and waxes. Get a sample of whole milk, or cream, and add a drop to a clean glass slide. Add one drop of the Sudan IV dye and add a cover slip. Observe the slide under low and high power, and record what you see in a drawing below.

b) The red spots are drops of oil (lipids) scattered throughout the liquid which absorb the red dye. You can see fat stored in cells using this same procedure. Cut a very small piece of fat from a fresh piece of meat. Add a drop of Sudan IV to the sample on a glass slide. Place a cover slip on the sample, and gently squeeze the tissue by pressing with the eraser end of your pencil. Be careful not to crack the cover slip, since it is very fragile. Examine the slide under the microscope, and draw what you see below.

You should have noticed some small, round spots which had absorbed a large amount of the dye. You may be able to see that these are actually inside of the cells, and that the rest of the cell is in a very thick wall around the fat droplet. These cells store fat for use as energy when the body needs it.

There are other types of lipids beside fats: many hormones in the body are also lipids. Lipids, like proteins, are an important part of membranes surrounding the cell, and also within the cell.

PART D - Nucleic Acids

Another important group of compounds within the cell are nucleic acids, DNA and RNA. These substances control the production of protein in the cell. In this way, the nucleic acids really control the life of the cell. DNA is made up of two long chains of sugar molecules and phosphate groups, which are bonded together by small molecules called nucleotide bases that stick out from each sugar. The pattern of nucleotide bases is something like a code: the cell is able to recognize certain patterns as meaning certain proteins should be produced. The DNA is the "master copy" that carries the code from one generation of cells to the next. RNA is a temporary working copy of the DNA that the cell makes in order to produce proteins

The nucleic acids are found mostly in the nucleus of the cell. Complicated techniques are required to stain them, so these procedures are not included with this kit. Your book should have a description of their structure, and the ways in which they replicate themselves and control the activity of the cell.

PART E - Vitamins and Minerals

You probably know that vitamins are important for life. These molecules are found in very small amounts within the cell, so they are difficult to detect without complicated procedures. The vitamins usually act as an "assistant" to enzyme molecules, so they are called co-enzymes. Some vitamins are similar to proteins, and others are more similar to the lipids.

Several atoms are important to the life of the cell besides those mentioned so far. Examples are:

iron (Fe)	is found in the blood compound hemoglobin.
calcium (Ca)	important to muscle activity, bones, and teeth.
magnesium (Mg)	important for the process of photosynthesis in plant cells.
sodium (Na)	important in nerve cells and in cell membrane transport
potassium (K)	important in muscle tissue
chlorine (C)	is important in the activity of nerve cells
iodine (I)	is involved in several important hormones

Additional Work

If you have time, you may wish to test samples of various tissues and food to determine the presence of starches, sugars, amino acids, proteins, and fats.

Discussion Questions

1) A sample of cells is broken apart with a blender, then a centrifuge separates the various parts of the cell according to how heavy they are. One subset of material (in a laboratory referred to as a "fraction") is found to contain free membranes from the various parts of the cell. What groups of compounds would you expect to find in these membranes?

   Membranes contain large amounts of lipids and proteins. The proteins may be structural parts of the membrane, or enzymes which are attached to the membranes.

2) Compounds called glycoproteins are important to the membranes surrounding a cell. These molecules contain both protein and chains of sugars. A chemist gives you a sample of material which he claims is a glycoprotein. What test(s) could you perform to find out if he is correct?

   The protein should show a positive test with either the Biuret or the ninhydrin test. The ninhydrin test could be used after the protein was partially broken down into amino acids. The sugars should test positive with the Chemstrip test.

SAFETY INSTRUCTIONS: IMPORTANT

NOTICE TO TEACHERS REGARDING LABORATORY REAGENTS

Perhaps the best general rule regarding the safe handling of laboratory chemicals is to treat all of them as being potentially dangerous. This means that none of them should be taken internally, and that any external contact should be washed thoroughly. In fact, most of the chemicals provided in The Science Source kits are diluted enough that they are not hazardous. The following lists indicate appropriate antidotes for the hazardous chemicals. Check this list for the chemicals provided in the kit:

I. **Concentrated Acids & Bases** - **Do not** induce vomiting, dilute with water, then milk or egg white, call a physician immediately.

1. 25 % Acetic Acid
2. 3M Hydrochloric Acid
3. Concentrated Sulfuric Acid

II **Dilute Acids & Bases** - **Dilute** with water, then milk.

1. 1 M, 0.5M, 0.1 M Hydrochloric Acid
2. Oxalic Acid
3. Sodium Hydroxide
4. Ammonium Hydroxide

III **Miscellaneous Chemicals** - Dilute immediately with water. Induce vomiting with warm salt water, or warm baking soda solution.

1. Ammonium Chloride
2. Ammonium Oxalate
3. Barium Chloride
4. Biuret Reagent
5. Bromthymol Blue
6. Calcium Chloride
7. Ethanol (Denatured Alcohol)
8. Ferric Ammonium Sulfate
9. Hydrogen Peroxide
10. Janus Green B
11. Lead Nitrate
12. Lugol's Solution
13. Magnesium Reagent (Titan Yellow, Alcohol)
14. Mercuric Nitrate
15. Methylene Blue
16. Ninhydrin
17. Phosphorus Reagent (Ammonium Molybdate, Nitric Acid)
18. Potassium Ferricyanide
19. Potassium Permanganate
20. Silver Nitrate
21. Sodium Carbonate
22. Sodium Thiosulfate
23. Sudan IV

IV. **Organic Solvents** - **Do not** induce vomiting. Dilute with water and milk. Call a physician immediately.

Isopropyl Alcohol