BIOLOGY LABORATORIES

Biology teachers and their students face a wide range of potential hazards. In addition to chemical reagents, there are the hazards associated with the handling of organisms, classroom activities on the school grounds and outdoor study areas, and the containment of biological specimens. Effective control of such hazards involves both the recognition of each hazard and the development of control procedures.

Complete the Waste Disposal Form for removal of all waste (chemicals and broken glass) at the end of each semester. The Science/STEM Coordinator (Dr. Chaundra Creekmur; creekch@boe.richmond.k12.ga.us) will facilitate removal.

BIO 1: Required Materials for the High School Biology Lab

1. Broken Glass Container
2. Sharps Disposal Box
3. Biohazard Bags
4. Household Bleach
5. Fire Extinguisher
6. Spill Kit
7. First Aid Kit
8. MSDS Notebook
9. Chemical Waste Disposal Containers

BIO 2: Eye Protection

BIO 2.1 What is your obligation?

Teachers owe their students a duty of care. A teacher must reasonably address all foreseeable dangers inherent in any laboratory experiment or demonstration that will be performed in the science laboratory or classroom. A teacher must also instruct and ensure that students demonstrate the proper use of protective equipment.

An important obligation of science teachers is to provide students with appropriate eye protection. **Provision and Maintenance of PPE - 29 CFR §1910.132(d) Personal**

Protective Equipment, General Requirements Standard requires a hazard assessment to determine PPE needs and teachers must be trained in use and care of goggles.

BIO

a. The agency wants to make it clear, however, that contact lenses are not eye protection devices. If eye hazards are present, appropriate eye protection must be worn instead of, or in conjunction with, contact lenses."
b. Regulations (Preamble to Final Rules) Personal Protective Equipment for General Industry (Amended Final Rule, April 1994) Section 3- III Summary and Explanation of the Final Rule 1910.133, p. 16343.

BIO 3: Glassware

BIO 3.1 Injuries from Glassware

Glassware is the number one source of injury in the laboratory setting. More students are cut by damaged glassware and burned by heated glassware that are harmed by any other object or circumstance in the lab. To ensure the safety of students in the middle school laboratory, substitute plastic lab ware for glassware where possible. New plastics like polycarbonate (Lexan®) have been successfully used for laboratory containers. While not useful for heating, the plastic is clear and extremely hard and can be used for almost all water-soluble compounds. Beakers, flasks, graduated cylinders, and thermometers now are available in plastic. Check with your science supply company.

BIO 3.2 General Cautions

BIO 3.2.1 Broken Glass

1. Use glassware that is without defect and has smooth edges.
2. One of the most important ways to prevent glassware related injuries is to check the pieces for chips or cracks. Any damaged glassware should be disposed of in the appropriate container.
3. Glassware should have no cracks, chips, or scratches. In particular, be wary of "star cracks" that can form on the bottom of beakers and flasks. Any glassware with such cracks should be properly disposed of immediately.
4. All glass tubing should be fire-polished.

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Be careful with glassware that is "frozen." Only teachers, wearing goggles and gloves, should try to release the "frozen" glassware. If this fails, discard the glassware. Some common cases of "frozen" glassware are:

1. nested beakers that have been jammed together.
2. stoppers that cannot be removed from bottles.
3. stopcocks that cannot be moved.

BIO 3.2.3 Hot Glass

1. Use only Kimax or Pyrex brand glassware when heating substances. Common glass can break or shatter, causing serious injuries in the lab.
2. Use care when working with hot glass. Hot glass looks exactly the same as room temperature glass.
3. Do not leave hot glassware unattended, and allow ample time for the glass to cool before touching.
4. Check the temperature of the glassware by placing your hand near, but not touching, the potentially hot glass.
5. Have hot pads, thick gloves, or beaker tonnBJET2 Tf1 0 0s 12 0 612 792 reW*nQq0.000612 792 il

BIO.3.2.5 Bending.

Bending glass tubing is often necessary. Follow these procedures:

1. Place a wing-top attachment on a gas burner and heat the area of the glass to be bent while holding it with one hand on each end, rotating to ensure even heating.
2. When the glass is soft and pliable, remove it from the flame and quickly bend to the desired shape.
3. Place on insulating material until cool.

BIO.3.3 Types and Appropriate Use of Glassware

To prevent glassware related injuries always use the correct type of glass for the task you are doing. For example, a graduated cylinder should be used to measure the volume o

BIO 4: Microscope Handling

1. **DO NOT ALLOW STUDENTS WITH ACTIVE EYE INFECTIONS TO USE MICROSCOPES!**
2. Provide students with alcohol wipes to clean lenses before or after use.
3. Microscopes must be carried upright, with one hand supporting the arm of the microscope and the other hand supporting the base. Nothing else should be carried at the same time.
4. Microscope must be positioned safely on the table, NOT near the edge.
5. After plugging the microscope into the electrical outlet, the cord should be draped carefully up onto the table and never allowed to dangle dangerously to the floor.
6. The coarse adjustment must NEVER be used to focus a specimen when the 40x or oil immersion lens is in place.
7. When finished with the microscope, the cord should be carefully wrapped around the microscope before returning it to the cabinet.
8. All prepared microscope glass slides are to be returned to their appropriate slide trays; wet mount preparations are to be disposed of properly.
9. Malfunctioning microscopes should be reported to the department chairperson/laboratory safety manager.

BIO 5: Dissections

The use of preserved animal specimens in instruction should be carefully planned to provide

BIO 5.2 Student Instruction

1. **Students should be instructed in the safe use of dissection instruments.**
    a. Scalpels and dissecting instruments should be sterilized before and after experiments.
    b. Pointed dissection probes, scalpels, razor blades, scissors, and microtome knives must be used with great care, and placed in a safe position when not in use.
    c. Scalpels and other sharp instruments are only to be used to make cuts in the specimen, never as a probe or a pointer.
    d. Leave

BIO 5.4 Disposal

1. **Body parts or scraps of the specimen should NOT to be disposed of in the sink.**
   a. **Body parts and tissue specimen should be placed into resealable plastic bags then placed into red biohazard bags. The bags should be labeled with the contents and the date the waste was generated.**
   b. **Facilities and Maintenance should be contacted for pick up and disposal. Document the date pick-up was requested and the date it occurred.**
2. Containers designated for the disposal of sharps (scalpel blades, razor blades, needles; dissection pins, etc.) and containers designated for broken glass must be present in each laboratory. Never dispose of any sharp object in the regular trash containers.
3. All bio hazardous disposable glass items (i.e., slides, cover slips, Pasteur pipets, etc.) must be disposed of properly in the **Biohazard Sharps Container, NOT** the regular trash or waste bags.
4. Dispose of dissecting pins or other sharp objects in **Biohazard Sharps Container, NOT** in the Waste Container, waste bags, or regular trash.

BIO 5.5 Cleaning Dissecting Pans

1. All solid debris should be removed from the tray.
2. Dissecting pans should be washed with soapy water.
3. Dissecting pans should be sterilized with a 5:1 ratio of water and hypochlorite solution.
4. Allow the solution to sit in the pan for a minimum of 10 minutes, then rinse thoroughly and allow to air dry.

BIO 4.6 Cleaning Dissecting Tools

1. Scalpels, probes, and other related tools should be carefully wiped with a paper towel to remove solid debris.
   a. Place the paper towel into the waste bag containing discarded body parts and tissues.
2. Tools may be placed in a 5:1 ratio of water and hypochlorite solution or a similar alcohol solution to clean and sterilize them.

3. Tools should be allowed to sit in the solution for a minimum of 90 minutes; leaving them

in the solution overnight is preferable.

4. If necessary, dissecting tools may be autoclaved.

BIO 6: Experiments with Bacteria and Fungi

This section pertains primarily to the use of viruses, bacteria, and other microscopic organisms. The handling of these pathogens is treated in 29CFR 1910.1030. This publication covers definitions, exposure control, specific procedures and protocols to comply with the regulations, precautions for specific pathogens, signs, labels, training, and record keeping. Proper laboratory technique is the basis for all cautions in this section.

Essential equipment for working with microorganisms includes:

1. Sterilization equipment (autoclave, heat sterilizer, or pressure cooker) for media preparation, sterilization of glassware and equipment, and decontamination of disposable material
2. Sterile transfer equipment (micropipettes with disposable tips or sterile pipets) for safe transfer of microorganisms
3. Adequate work space and equipment to prepare media
4. Proper storage facilities, including refrigeration and incubation equipment
5. Supplies for

c. The procedure may require the use of more than one loop so that as one is being used, others are cooling.

2. When a contaminated loop is inserted into a flame for sterilization, an aerosol may be generated by the boiling and volatilization of the material before the flame can kill all pathogenic microorganisms.

BIO 6.7 Bunsen Burner Safety Guidelines

Bunsen burners present fire hazards. They produce an open flame and burn at a high temperature, and as a result, there is potential for an accident to occur. For the safety and convenience of everyone working in a laboratory, it is important that the following guidelines be observed.

1. Remove all papers, notebooks, combustible materials and excess chemicals from the area.
2. Tie-back any long hair, dangling jewelry, or loose clothing.
3. Inspect hose for cracks, holes, pinch points or any defect and ensure that the hose fits securely on the gas valve and the burner. Replace all hoses found to have a defect before using.
4. Notify others in the laboratory that the burner will be in use.
5. Have the sparker/lighter available before turning on the gas.
6. Utilize a sparker/lighter with extended nozzle to ignite the burner. Never use a match to ignite a burner.
7. Adjust the flame by turning the collar to regulate air flow and produce an appropriate flame for the experiment (typically a medium blue flame).
8. Do not leave open flames unattended and never leave the laboratory while the burner is on.
9. Shut off gas when its use is complete.
10. Allow the burner to cool before handling. Ensure that the main gas valve is off before leaving the laboratory.

BIO 6.8 Spills & First Aid

1. **A spill kit should be prepared prior to starting microbiology labs. It should include all items required to clean up a spill, including disinfectant, paper towel, gloves and plastic bags and containers for disposal.**
2. Students must report all spills to the teacher.
3. Only the teacher or laboratory safety manager should be allowed to clean up such spills.

4.

BIO 6.9 Contaminated Broken Glassware

1. Contaminated broken glassware should never be picked up directly with the hands.
2. It should be cleaned up using aids such as brush and dustpan, forceps or cotton wool swabs.
3. Follow the procedure for liqu6 owl

a. The teacher should carefully read instructions before using an autoclave or pressure cooker.

   If using a pressure cooker, make sure the safety valve is in good working order. Materials may be sterilized by using 15 pounds of pressure at 121°C for 20 minutes.

BIO 7: DNA Study

Work with deoxyribonucleic acid (DNA) is at the core of many of the hands-on activities in molecular biology and biotechnology that have been introduced into the high school biology laboratory. The study of the chemical and physical properties of DNA often involves the spooling, isolation, enzymatic digestion, gel electrophoresis, and manipulation of bacterial cells to introduce new genetic information. Many such laboratory activities can be purchased as complete kits that provide documentation and guidelines helpful to both students and teachers. These kits are especially recommended for teachers who are not familiar with standard procedures in research laboratories. Safety, as always, is a crucial part of any molecular biology experience. **Research requiring containment is prohibited by federal law.**

BIO 7.1 Electrophoresis

Electrophoresis, a technique which separates molecules based on their electrical charge, is frequently used in today's laboratories. **Be aware that ALL components of an electrophoresis gel require an MSDS and that students and other individuals must be informed of all risks prior to use.**

BIO 7.1.1 Handling Electrophoresis Chambers

Precautions to prevent electrical shock and using electrophoresis apparatus safely include:

1. Turn the power off before connecting the electrical leads.
2. Connect one lead at a time using one hand only.
3. Insure that your hands are dry while connecting the leads.
4. Keep the apparatus away from sinks or other water sources.
5. Turn off power before opening lid or reaching inside chamber.
6. Don't override safety devices.
7. Don't run electrophoresis equipment unattended.

BIO 7.2 Electrophoresis Gels and Additives

Many of the commonly used electrophoresis gels are harmless, but the additives can be extremely hazardous.

BIO 7.2.1 Ethidium bromide

Ethidium bromide is an intercalating agent commonly used as a fluorescent tag (nucleic acid stain) in molecular biology laboratories for techniques such as agarose gel electrophoresis. It is a mutagen and should be handled with caution, even when mixed in the gel.

BIO 7.2.2. Formamide

Formamide is also used as an RNA stabilizer in gel electrophoresis by deionizing RNA. In capillary electrophoresis, it is used for stabilizing (single) strands of denatured DNA. Also known as **methanamide**, is an amide derived from formic acid. It is a clear liquid which is miscible with water and has an ammonia-like odor. Formamide is highly corrosive on contact with skin or eyes and may be deadly if ingested. Inhalation of large amounts of formamide vapor may require medical attention. It is also a teratogen.http://en.wikipedia.org/wiki/Formamide Formamide should never be handled without proper safety attire including gloves and goggles. There is a small risk of decomposition into hydrogen cyanide and water.

BIO 7.2.3 Acrylamide

In recent years polyacrylamide gels have been prepared in some school laboratories to achieve the isolation of specific molecules by electrophoretic techniques. Schools are cautioned to cease this practice because acrylamide poses a potentially serious health hazard as a neurotoxin. This substance has been classified as 2B (possibly carcinogenic to humans) by the International Agency for Research on Cancer (IARC).

Because there is a serious risk of inhalation exposure during the weighing of acrylamide powder for the preparation of gels, schools should purchase only pre-poured polyacrylamide gels from laboratory supply houses. The pre-poured gel presents less

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BIO 7.2.4 TAE buffer

TAE buffer is a buffer solution containing a mixture of Tris base, acetic acid and EDTA. In molecular biology EDTA is used in agarose electrophoresis typically for the separation of nucleic acids such as DNA and RNA. It is made up of Tris-acetate buffer, usually at pH 8.0, and EDTA, which sequesters divalent cations. It is extremely irritating to the skin, eyes, and the upper respiratory tract. It is easily absorbed through the skin and is a mutagen. It is harmful if swallowed or inhaled.

BIO 7.2.5 Stains

Bromoph92 r0.00000912 0 612 792 re15re15re15re15re15re15re15re15re15re15re15re15re15re15re15re15re

BN0 1792 ilTOC1 0TJ0 612 792 reW*n0 g0 G2 460MSDSO1 1 06(Tr)4(is)]TJETOq0.00000912 0 612 792 reW*nBT/F1 12 Tf1

BIO 7.3.2 Electrophoresis Gels and Contaminated Non-Sharp Debris, (i.e. gloves, tips, paper towels, etc.)

1. Collect electrophoresis gels and contaminated non-sharp debris screw-top pails with a clear plastic liner. The container lid should be closed securely when the container is not in use.
2. Label electrophoresis gels and contaminated non-sharp debris. Indicate on the label that it is a solid waste, and check off the appropriate constituent box on the label.
3. Contact Facilities and Maintenance for pick-up and disposal. Document the date pick-up was requested and the date it occurred.

5. Contact Facilities and Maintenance for pick-up and disposal. Document the date pick-up was requested and the date it occurred.

BIO 7.3.4.2 Record Keeping

1. Reassigned samples must be re-labeled with the new custodian's name and the date the waste was generated and stored.
2. Contact Facilities and Maintenance for pick-up and disposal. Document the date pick-up was requested and the date it occurred.

BIO 8: Disposal and Cleanup

BIO 8.1 Disinfectants

Disinfectants and antiseptics (disinfectants for use on living surfaces e.g. skin) vary in their ability to kill bacteria, viruses, fungi, spores and protozoa. Disinfectants should always be diluted and used according to the manufacturer's instructions. The Material Safety Data Sheet should also be consulted for specific protective equipment and ventilation requirements. The following types of disinfectants are suitable for use in schools.

BIO 8.1.1 Alcohols

Alcohols have good activity on bacteria, and fungi but less on viruses and poor activity on spores. 70% ethanol is rapid acting and dries quickly. 90% ethanol is good for viruses. **100% ethanol is NOT an effective disinfectant**. 60-70% Isopropyl Alcohol (Propan-2-ol) is also effective.

BIO 8.1.2 Chlorhexidine

Chlorhexidine has good activity on gram-positive bacteria but less activity on gram negative bacteria, viruses and fungi and poor activity on spores. It has low toxicity and irritancy and so is a good antiseptic. 0.5% for face - 4% for other skin. It is often combined with alcohol, which may dry the skin.

BIO 8.1.3 Sodium Hypochlorite

Household bleach has good activity on bacteria, fungi and viruses, but less activity on spores. Varying amounts of available chlorine in hypochlorite solutions are required for different purposes. They must be prepared fresh daily from the concentrated stock solution to ensure the correct level of available chlorine. 1% for spills, 0.25% for discard jars, 0.1% for cleaning benches and 0.05-0.1% for equipment and instruments.

BIO 8.1.4 Providone-Iodine

Tincture of Iodine as 10% aqueous or alcoholic solutions is also suitable as skin disinfectant but it stains.

BIO 8.1.5 Other

DO NOT USE quaternary Ammonia compounds as they are not effective disinfectants against many bacteria and viruses. Peracetic acid, aldehydes and phenolic disinfectants are considered too hazardous for use in schools.

BIO 8.2 General Cleanup

All contaminated items should be decontaminated prior to reuse or disposal. Items for reuse should be immediately placed in disinfectant and soaked according to the manufacturer's instructions, prior to washing. (e.g. 25% Sodium Hypochlorite solution, soaked overnight)

BIO 8.2.1 Glassware and Sharps

1. All biohazardous dissecting pins, scalpel blades, or other such items must be disposed of in the **Red Biohazard Sharps Container**, NOT in the regular trash.
2. All bio hazardous disposable glass items (i.e., slides, cover slips, Pasteur pipets, etc.) must be disposed of properly in the **Red Biohazard Sharps Contain**

2. Once sterilized it may be poured down the sink.

BIO 8.2.4 Incubator

Following use the incubator must be thoroughly cleaned and disinfected with an appropriate disinfectant. (e.g. 0.1% sodium hypochlorite)

BIO 9: Chromatography

1. Chemical splash safety goggles and aprons should be worn.
2. Only water baths or hot plates with water baths (and not open-flame fires) should be used for chlorophyll extraction. Extraction may also be accomplished by leaving the plant material in the solvents overnight at room temperature.
3. Only Pyrex or comparable glass tubes should be used.
4. Dissolving and developing solvents give off toxic vapors. They must be stored in closed containers and the room
5. Solvents are highly flammable and must not be used near an open flame.

b. Facilities and Maintenance should be contacted for pick-up and disposal. Document the date the request was made and the date on which the waste was picked up.

BIO 12: Animals in the Classroom

BIO 12.1 Introduction

The use of live animals in the classroom can help students understand and appreciate life processes. Before bringing animals into the classroom, teachers should check the school or school system policy. It is important to select animals that are appropriate to the instructional needs and are practical to maintain. Good safety procedures should be established for the protection of students from the hazards of classroom animals as well as to ensure the humane treatment of animals.

The humane treatment of animals in research and teaching is a sensitive issue. The Council of State Science Supervisors, the National Association of Biology Teachers, the National Science Teachers Association, the Humane Society of the United States, the Animal Welfare Institute, and the National Society for Medical Research all have established guidelines and position papers supporting the safe and humane treatment of animals used for the cause of science.

The following websites offer more information on this topic:

www.enc.org/csss/index.html - Eisenhower National Clearing House

www.nabt.org - National Association of Biology Teachers

www.nsta.org - National Science Teachers Association

www.hsus.org/programs/research/animals_education.html

www.animalwelfare.com - Animal Welfare Institute

BIO 12.2 Before You Bring Animals into the Classroom

BIO 12.2.1 Permission to Keep Live Animals on Campus

You must complete a **Richmond County School System Permission to Keep Live Animals on Campus (**Appendix A) and submit it to the Curriculum and Instruction Department attn. Science Curriculum Department. It will help you think through some necessary planning measures such as animal enclosure options, how the enclosure will be cleaned, and weekend, holiday and summer care arrangements.

BIO 12.2.2 Parental Notification

You must obtain **Parental Notification Forms** (Appendix A)**.** It is not recommended that students be permitted to handle any animal(s) or be given caring or cleaning duties without prior parental/legal guardian consent.

BIO 12.2.3 Hand Washing Education

You must educate all students, paraprofessionals, and adult volunteers on proper hand washing. If anyone does handle an animal, they should wash their hands with hot soapy water for at least 60 seconds (instant hand sanitizers should only be used in addition to proper hand washing, NOT IN LIEU OF).

BIO 12.2.4 Educational Purposes

Animals in the classroom must have an educational purpose. Classroom animals should be limited to animals that are bred in captivity,

BIO 12.2.7 Special Permits

Avoid animals requiring special permits. Some animals require a written permission from the local health department, the Georgia Department of Natural Resources, and/or the United States Department of Agriculture to be kept in a classroom setting. These include venomous and nonvenomous snakes, wild turtles, certain species of frogs, wild newts and salamanders, hogs, deer, cattle, alligators, crocodiles, caimans, wild fowl, and all domestic fowl. DO NOT attempt to keep any of the animals mentioned above.

BIO 12.2.8 Animals Not Allowed in Richmond County Schools

1. Farm animals excrete *E. coli* O157:H7, *Salmonella*, *Campylobacter*, and *Cryptosporidium* intermittently and in substantial numbers; therefore these animals are not appropriate unless meticulous attention to personal hygiene can be assured.
2. Mammals at high risk for transmitting rabies (e.g., bats, raccoons, skunks, foxes, and coyotes) are not appropriate as residents in the classroom.
3. Nonpsittacine birds (any bird other than parrots, parakeets, and cockatiels).
4. Inherently dangerous animals (e.g., lions, tigers, cougars, and bears).
5. Nonhuman primates (e.g., monkeys and apes).
6. Mammals at higher risk for transmitting rabies (e.g., bats, raccoons, skunks, foxes, and coyotes).
7. Aggressive or unpredictable animals, wild or domestic.
8. Stray animals with unknown health and vaccination history.
9. Venomous or toxin-producing spiders, insects, reptiles, and amphibians.

BIO 12.3 Housing and Caring for Your Classroom Animal(s)

BIO 12.3.1 Habitats

Ensure that a proper habitat can be kept for the animal(s) (free of drafts and harsh sunlight). Also consider what type of care the animal will receive over weekends, and during school breaks (paying close attention to building heat and air conditioning status during times when school is not in session).

BIO 12.3.2 Food

Store all animal food in rigid containers with tight fitting lids to prevent access to food by pests. Also, some animals require fresh foods that may require refrigeration, or live foods. Should this be the case, make sure you have necessary equipment before bringing the animal in to the classroom. Food and water bowls should be thoroughly scrubbed and rinsed with hot soapy water.

BIO 12.3.3 Enclosures

Animals should be housed in an enclosure constructed from a nonporous material that is easily cleanable. Cleaning of animal(s) enclosures should be done as often as necessary to keep the animal healthy, prevent odors from building up, and eliminate any unsanitary conditions. It should be noted that cleaning and disinfection may be necessary as often as daily, however it should be done weekly at a minimum.

BIO 12.3.4 Sanitation

Enclosures should be sanitized after each cleaning with a fresh bleach solution (4oz of 5.25% unscented chlorine bleach to one gallon of water) OR a quaternary ammonia solution at a dilution suggested by the manufacturer for food service uses (NEVER MIX CHEMICALS!!!). As animals can be sensitive to sanitizers, care should be taken in adequately rinsing and drying the enclosure before putting the animal back in the enclosure. Some pathogens will not be killed by the sanitizers, but may be removed by rinsing thoroughly with water (this will also remove residual amounts of sanitizers). An animal's sensitivity is not an adequate reason to avoid the use of sanitizers.

Animal enclosures must never be cleaned in plumbing fixtures used for food service, drinking water, or hand washing purposes. After cleaning the enclosure, the fixtures used

BIO 12.3.5 Security

All animal(s) enclosures should be securely covered and locked if possible. This will help protect the students and animals from one another by discouraging unsupervised handling and reducing potential of escape.

BIO 12.3.6 Aggressive Animals

It should be noted that any animal may behave aggressively, naturally aggressive species, and animals that are unusually aggressive or those displaying odd or uncharacteristic behaviors for their species should be removed immediately. Animals capable of causing substantial injury through aggressive or defensive reflexes should also be avoided (i.e. snapping turtles, venomous snakes, poisonous frogs, large birds).

BIO 12.3.7 Injured and Sick Animals

Animals that are injured or in poor health should be removed from the classroom immediately and given proper care. It should be noted, however that even animals that are or appear to be in good health can still shed potential pathogens.

BIO 12.3.8 Animal-Specific Guidelines

BIO 12.3.8.1 Invertebrates

1. Invertebrate animals are often used for observation and learning activities.
   a. Teachers should obtain manuals available from biological suppliers.
   b. These manuals are inexpensive and serve as a complete guide to maintaining and studying the organisms in the classroom.
2. If experiments are done with fruit flies, take care in quieting them and/or killing them.
3. **ETHER AND/OR TRIETHYLAMINE $(C_2H_5)_3N$ MAY NOT BE USED TO ANESTHESIZE OR EUTHANIZE ANY ORGANISM IN AN RCSS LABORATORY!**

4. Place the fruit flies in a Petri dish, gently covering them with cotton, and then invert the dish for examination under the dissecting microscope.
    a. Refrigerate culture jars and place “chilled” flies on a Petri dish over ice.
    b. Anesthetizing kits also may be used.

        FlyNap® kits containing relatively harmless components may be purchased from biological supply companies.
    c. Any anesthetic should be used in a properly ventilated room according to the supplier.

BIO 12.3.8.2 Vertebrates (Nonhuman)

1. Do not take vertebrates from the natural environment.
    a. Most municipalities prohibit the removal of vertebrates from the natural environment because doing so upsets nature’s balance and may introduce unwanted microorganisms or diseased animals into the classroom.
2. Obtain animals from a certified disease-free source such as a biological supply house or a certified breeder.
3. When studying developing chicken embryos, do not use any embryos that are more than 18 days old.
    a. Do not work with fertilized eggs.
    b. Dispose of dead embryos, which may carry pathogenic bacteria. Follow appropriate hazardous waste guidelines.
4.

BIO 13: Plants in the School

Plants can be used effectively to provide a living laboratory for high school science instruction. By providing experiential learning opportunities, science educators can help students to develop the kind of reasoned thinking that will result in responsible decision-making regarding human/ecosystem interaction. However, certain plants can trigger severe allergic reactions in the form of skin rashes and breathing difficulties in susceptible children. The following guidelines will help teachers determine how to best use plants as effective teaching tools.

BIO 13.1 Poisonous Plants and Plants with Spines

1. Teachers should confine their lesson on poisonous plants (poison ivy, poison oak or poison sumac) to pictures.
2. Cacti and other plants with spines should not be kept in the classroom. Spines can become embedded under the skin and become infected if not removed correctly.
3. When using an outdoor learning area, examine the site for the presence of poisonous plants. When visiting these sites, carefully monitor the children to keep them away from the poisonous plants.
4. Children should not put any plants or plant parts in their mouths.

BIO 13.2 Plants in the Classroom

1. Only plants that are not hazardous to children and with which you are familiar should be used.
2. Breathing spores or pollen can cause reactions in some students. Provide face masks to susceptible students as needed.
3. When using commercial seeds treat them with care because they may have been treated with toxic fungicides.
    a. Have students wear gloves when handling them. 0 0 1 126.02 191.33 Tm0 g0 G[(Ha)6(ve)4(students

6. Supervise children closely and
    a. make sure that they never place any plant or part of a plant in the mouth.
    b. make sure that they do not touch any part of their face; even 'safe' plants can have hairs, oils, and other compounds that can irritate the skin.
7. Make hand washing routine procedure after any laboratory activity even when working with plants.

BIO 13.3 School Gardens

Before you begin your school garden program, you will need to ensure that the soil, water, and working environment are safe for the students. Test the soil for contaminants, know what is in soil amendments, the water and plants, and develop rules for working in the garden. Talk with the students about these important issues, and let them help develop the rules to be used in the garden.

BIO 13.3.1 Preparation

BIO 13.3.1.1 Manure

1. Do not use fresh or unsterilized manure. All animal manure is potentially hazardous and may contain E. coli as well as other disease-causing pathogens.
2. Use only sterilized or fully composted manure.
3. Aged manure is **not** the same as composted, and can contain disease-causing organisms.
4. For more information, contact your local county health department or cooperative extension office.

BIO 13.3.1.2. Lead Contamination

1. Lead is naturally present in all soils, generally in low levels, but pollution can increase lead to harmful levels.
2. If you plan to plant an edible garden in an area that may have lead-contaminated soil, first test a soil sample for lead to determine if the soil is safe.
3. This is a critical issue for schools. Areas at risk for lead contamination include those with a history of construction before 1978, where lead may have leached

into the soil from paint or other materials, or a history of heavy exposure to traffic that at one time used fuel-containing lead.

4. To be on the safe side, it is **always** a good idea to test the soil for lead before beginning an edible garden project.
5. For information about lead testing, contact your local county health department or cooperative extension office.

BIO 13.3.2 Underground Pipes

1. Prior to developing the garden space, check with the school district or local utilities to determine if there are any underground pipes or cables that may be a potential problem.
2. If digging begins without getting an "all clear," the chance exists of running into electrical cables, water pipes, or a gas main.

BIO 13.3.3 Water

1. Make sure all water used in the garden – for watering plants, washing produce, and washing hands – is potable (drinkable) water.
2. Water for washing hands and produce should be **running** water to prevent recontamination.
   a. Some newly developed school grounds may have two separate water systems – one for potable water and one for recycled water (used for irrigation).
   b. Check with district administrators to determine if this is an issue at the school site.
   c. Then make sure only potable water is used in the school garden.
   d. For more information on recycled water, contact the **WateReuse Association.**

BIO 13.3.4 Building Materials

1. Do not use railroad ties, treated lumber, or old tires for garden boundaries, raised beds or anywhere in the garden.
   a. These items contain toxic chemicals that can leach into the soil and be absorbed by the plants.

b. Old railroad ties contain creosote, a carcinogen; treated lumber contains cyanide, a potent poison; and tires can leach petroleum products into the soil.

2. Contact your county cooperative extension office for more information.

BIO 13.4 Harmful Plants

Some plants and plant parts are poisonous. Others, such as poison ivy or stinging nettle, can irritate the skin. Teach children never to taste a plant unless an expert says it is all right to eat. Refer to the List of Hazardous Plants below for more information or contact the local poison control center.

BIO 13.4.1 Hay Fever Plants

Grasses

Ragweeds

Flowering trees, especially Alnus (Alder) and Quercus (Oaks)

Daphne

BIO 14: Greenhouse Maintenance and Operation

For schools that have greenhouses available for biology and environmental classes, the following guidelines are intended to aid in their smooth maintenance and operation. These guidelines, which supplement applicable school regulations, apply to any individual working in the greenhouse area, student or teacher.

BIO 14.1 Guidelines

The following guidelines are designed to ensure that all greenhouse components are functioning at an adequate level for optimum plant growth and at a safe level for student use.

1. Check water lines, heating system, fans, and temperature control. These are usually routine procedures that can be checked by the school maintenance staff.
2. Make sure all automatic equipment is functional and accurate.
3. Clean tools after use and store them appropriately.
4. Instruct students in the proper use of, and conduct in, the greenhouse area. It is recommended

BIO 14.2 Pesticides

1. Use organic methods of pest control when possible.
2. Make sure to maintain adequate ventilation. Ventilation is especially important when using pesticides.

BIO 15: Special Concerns

BIO 15.1 Thermometers

1. **MERCURY FILLED THERMOMETERS ARE NOT ALLOWED IN RCSS SCHOOLS.**
2. Alcohol laboratory thermometers should be used in general laboratory activities.
3. For more advanced applications, a digital laboratory thermometer may be used.
   a. Care should be taken to choose a digital thermometer that contains a changeable battery; some are not changeable.
   b. The battery is a button cell battery and may contain 5-50 mg of mercury; it should be recycled through a battery collection program.

BIO 15.2 Using Microtomes

Microtomes are commonly instruments used in laboratories to section tissues. These devices pose potential hazards to users during sectioning and/or the cleaning process. It is strongly recommended that teachers use prepared slides whenever possible.

BIO 15.2.1 Training

1. Training must be documented and provided by a knowledgeable and responsible person within the laboratory before any work is completed.
2. Standard Operation Procedures should be made available to all users and posted near the point of operation.

BIO 15.2.2 Appropriate PPE

A lab apron, chemical splash goggles, and nitrile gloves must be worn while handling tissues to be sectioned.

BIO 15.2.3 Sharpness of Blade

1. A microtome blade is extremely sharp and must be handled carefully.
2. The rotary handle of the microtome must always be set in the locked position when changing a paraffin block or the blade.
3. A new blade should be placed in the blade holder and clamped before the rotary wheel lock is released.

4. Wrist guards should be added where possible.
5. Once the blade is seated and secured the rotary wheel lock can be released and the knife and holder advanced to the specimen block.
6. If adjustments need to be made to the specimen, remove the blade from the housing.

BIO 15.2.4 Removal of the blade

1. Disposable blades must always be removed using forceps or a similar instrument.
2. Do not remove the blade holder from the microtome with a blade present or transport the housing with the blade present. .
3. Dispose of used microtome blades in the Biohazard Sharps Container.
4. The Biological sharps container must be kept adjacent to the microtome to reduce the distance that a blade would be moved
5. For microtomes with reusable blades cut resistant gloves must be used when removing and sharpeninBT/TT0 1 Tf 0.002 0 612 792 reW*nBT/F13 12 Tf1 0 0 1 168.det 556BT/TT0 1 Tf 0.002 0 0 36 611 0 0 4 (arpenin 0 0 921 0 0

BIO 16: Chemical Safety in the Biology Laboratory

All teachers should be familiar with the RCSS Chemical Management policy that addresses how

BIO 16.2.2 Secondary Containers and Prepared Solutions

1. When a material is transferred from the original manufacturer's container to other vessels, these vessels are referred to as —secondary containers.
2. Label all containers used for storage with the following:
   a. Chemical name (as it appears on the MSDS)
   b. Name of the chemical manufacturer or person who prepared the solution
   c. Necessary handling and hazard information
   d. Concentration or purity
   e. Date prepared
   f. Expiration or —use by date

BIO 16.2.3 Containers in Immediate Use

1. These chemicals are to be used within a work shift or laboratory session.
2. Label all containers in immediate use with the following:
3. Chemical name (as it appears on the MSDS)
4. Necessary handling and hazard information

BIO 16.2.4 Chemical Waste

All containers used for chemical waste should be labeled with the following:

1. HAZARDOUS WASTE
2. Chemical name (as it appears on the MSDS)
3. Accumulation start date
4. Hazard(s) associated with the chemical waste
5. Date generated
6. ***Complete the Waste Disposal Form for removal of all waste (chemicals and broken glass) at the end of each semester. The Science/STEM Coordinator (Dr. Chaundra Creekmur; creekch@boe.richmond.k12.ga.us) will facilitate removal.***

BIO 16.3 Material Safety Data Sheets (MSDS)

1. There must be an MSDS on file for every chemical compound in use in the lab.
2. At a minimum, MSDS information should be o(‡ 0 0 1 108.02 232.7q0.00000912 0 612 792 reW*nBT/F1

3. A copy must be kept in an area that is accessible to all individuals during periods of building operations.
4. If no MSDS is available for a product because 1) the manufacturer no longer exists; or 2) the manufacturer cannot be identified from the label that material should be considered hazardous waste and disposed of in a manner consistent with federal and state regulations.

BIO 16.4 Proper Chemical Storage

Guidelines for chemical storage must follow **O.C.G.A 45-22-2, O.C.G.A. 25-2, OSHA Standard 29 CFR 1910, and NFPA 45: Standard on Fire Protection for Laboratories Using Chemicals** and **NFPA 30: Flammable and Combustible Liquids Code.**

1. Hazardous chemicals in schools should be stored in accordance with MSDS specifications
2. Chemicals should not be stored in areas that are occupied by or accessible to students, such as classrooms or restrooms; they should preferably be stored in a central, secure location.
3. Organize chemicals first by **COMPATIBILITY**—not alphabetic succession (refer to section entitled Shelf Storage Pattern). Store alphabetically within compatible groups.

BIO

BIO 16.5.3 Storage Guidelines

1. Chemicals that are stored for disposal off-site should be placed in suitable closed containers and should be clearly marked with the contents. If the chemicals are a RCRA hazardous waste, the school must ensure that they are transported offsite for proper disposal.
2. Store all waste in containers that are in good condition and are compatible with their contents. Avoid using metal containers; certain chemicals can cause the metal to corrode and the container to leak.
3. Store waste in a designated area away from normal laboratory operations and to prevent unauthorized access. Store waste bottles away from sinks and floor drains.
4. Do not completely fill waste bottles; leave several inches of space at the top of each waste container. Securely cap all waste bottles.

BIO 16.5.4 Disposal of Hazardous Waste

1. **THE USE OF SINKS FOR THE DISPOSAL OF CHEMICALS IS STRICTLY PROHIBITED!**
   a. When rinsing glassware that contained chemical, discard the first rinse volume into the appropriate waste container.
   b. Subsequent rinses can be discarded to the sink.

2. Water/air reactive wastes are restricted by waste disposal companies and must be deactivated prior to disposal.
   a. This is particularly true of materials which ignite or release gases on contact with air or water.
3. Dispose of chemically contaminated paper and disposable clothing in approved solid waste containers.
4. Do not treat hazardous waste on-site. Exception: Acids may be neutralized with sodium bicarbonate in a 50-50 ratio by weight.
5. Contact Facilities and Maintenance for pick-up and disposal. Document when pick-up was requested and when it occurred.

BIO 16.5.6 Record Keeping

1. Reassigned samples must be re-labeled with the new custodian's name and the date the waste was generated and stored.
2. A waste management log must be maintained and should indicate how and when the waste was generated, how and when it was isolated and stored, by whom it was generated and stored, and date and method in which it was disposed.

BIO 16.6 Drug-Related Items

1. **THE FOLLOWING SUBSTANCES ARE NOT ALLOWED IN RCSS LABS!**

a. **Acetaldehyde**	f. **Histamine**
b. **Adrenalin**	g. **Nicotine**
c. **Colchicine**	h. **Testosterone**
d. **Caffeine**	i. **Thiourea**
e. **Ethyl Alcohol (grain)**	j. **Tobacco**

BIO 17: Fire Hazards

Fire is a real danger in any laboratory setting, and all teachers need to be aware of how to prevent fires. In the event a fire does occur, teachers need to know how to respond appropriately. The following information is provided as guidance in preventing or combatting fires in the science laboratory.

BIO 17.1 Preventing Burns and Fires

BIO 17.1.1 When planning to heat materials or use open flames

1. instruct students on STOP DROP AND ROLL in the event clothing catches fire
2. make sure students know how to evacuate the classroom in the event of a large fire

3. know the location of the nearest fire extinguisher and make sure you know how to use it.
4. have a bucket of sand or a fire blanket nearby in the event that the nearest fire extinguisher too far outside of the classroom.

BIO 17.1.2 When heating materials

1. **DO NOT USE ALCOHOL BURNERS! T**hey are extremely hazardous. Safer alternatives to alcohol burners include candles and hot plates.
2. **DO NOT USE STERNO HEATERS!**
3. make sure that the area surrounding a heat source is clean and has no combustible materials nearby.
4. do not allow students to work with hot materials, such as very hot water.
5. do not use household glass. Use only borosilicate laboratory glassware, such as Kimax™ or Pyrex™ when heating substances.
6. do not heat common household liquids, such as alcohol or oil; these are flammable and should not be heated. Heat only water or water solutions.
7. handle all hot materials using the appropriate type of tongs or heat resistant gloves (those made of asbestos or thick silicon rubber).

BIO 17.1.3 When using Hot Plates

1.

6. make sure that the hotplate is both unplugged and cool before handling a hotplate. You can check to see if a hot plate is still too hot by placing a few drops of water on the surface. If the water does not evaporate, it should be cool enough to touch.

BIO 17.1.4 When using open flames

1. use only safety matches. Make sure the matches are stored in a secure place between uses.
2. closely supervise students when they use matches. Make sure students are dressed properly (baggy clothes are tucked in, long sleeves are rolled up, smocks/aprons are properly tied) and have long hair/braids tied up.
3. closely supervise students when they use candles. Make sure students are dressed properly (baggy clothes are tucked in, long sleeves are rolled up, smocks/aprons are properly tied) and have long hair/braids tied up.
4. use tea candles that are short and wide, and cannot be knocked over in normal use.
5. place all candles in a “drip pan,” such as an aluminum pie plate, that is large enough to contain the candle if it is knocked over.
6. never leave the room while a flame is lit or other heat source is in use.

BIO 17.2 In the event of a large, uncontainable fire

1. evacuate the classroom immediately.
2. locate and pull the nearest fire alarm.
3. notify public safety and/or administration about the fire. Make sure you include the location and source (chemical, paper, petroleum) of the fire.

BIO 17.3 In the event of a small, containable fire

1. identify the type of fire. The table below lists the four classes of fires and methods for extinguishing them:

Class	To Fight Fires Involving	Method to Extinguish
A	wood, paper, cloth	Use water or dry chemical extinguisher.
B	gasoline, alcohol, paint, oil, or other flammable liquids	Smother by using carbon dioxide or dry chemical extinguisher.
C	fires in live electrical equipment	Cut off power to electrical equipment. Use ABC or carbon dioxide fire extinguisher.
D	metals (Na, K, Mg, etc.)	Scoop dry sand onto fire.

2. Use the appropriate method to extinguish the fire.
3. File an incident report.

BIO 17.4 In the event a student's clothes catch fire

1. Roll the child on the floor to smother the fire.
2. Use a fire blanket if one is available.
3. Do not direct a carbon dioxide (CO_2) fire extinguisher at an individual because such extinguishers produce dry ice that can cause frostbite.
4. **DO NOT ATTEMPT TO ADMINISTER FIRST AID TO ANY BURNS THE**

BIO 18: Electrical Hazards

BIO 18.1 Burns and Shock

1. Many electrical devices become quite hot while in use.
    a. In addition, "shorted" dry cells and batteries can produce very high temperatures.
    b. Students should never grasp a recently operated device or wiring without first checking for excess heat.
2. Students must be warned of the high death potential present even when the voltage is low.
    a. The severity of an electrical shock depends primarily on the amount of current to which a person is exposed.
    b. Since the current is related to the resistance and voltage, these two factors, as well as the part of the body involved and the duration of the contact, determine the extent of injuries to the victim.
    c. If the skin is wet or the surface broken, the resistance drops off rapidly, permitting the current to flow readily through the bloodstream and body tissues.

BIO 18.2 Electrical Apparatus

BIO 18.2.1 Batteries.

1. A battery is an unregulated source of current capable of producing large currents when resistance is low.
    a. When short-circuited, connecting wires can become very hot, raising the risk of burns. Short-circuited mercury batteries may even explode.
    b. Chemical leakage from batteries is a potential hazard, especially in the case of wet cells that contain caustic chemicals such as sulfuric acid.
2. Certain types of batteries are rechargeable while others are not.
    a. Carbon-zinc and nickel-cadmium type batteries can be recharged.
    b. Do not, however, attempt to recharge a completely dead carbon-zinc battery, a leaking or corroded battery, or any battery that carries a warning against recharging.

c. Such batteries can cause damage to the charger and may explode, causing personal injury. Lead-acid batteries can be recharged but produce explosive hydrogen gas during the process.
d. They should only be recharged in a well-ventilated area with an appropriate charger.

3. Do not discard any battery in the trash.
4. Contact Facilities and Maintenance for pick-up and disposal. Document the date of the request and the date the pick-up occurred.

BIO 18.2.2 Circuit Loads

1. Most school laboratory electrical circuits have a maximum power rating of 1,500 watts (if fuses are 15 amp) or 2,000 watts (if fuses are 20 amp).
2. The total power load on a circuit should not exceed these values.
3. The total load is the sum of the power ratings of all apparatus plugged into that circuit.
4. The individual power rating is usually found printed on a plate somewhere on the apparatus.

BIO 18.2.3 Extension Cords.

1. Use extension cords only when there is no convenient way to connect equipment directly to a receptacle.
2. If an extension cord must be used, it should be checked for damage, proper grounding, and electrical capacity.
3. An extension cord should be marked with its capacity in amperes and watts and the total load should not exceed these values.
4. If the cord is unmarked, assume that it is 9 amperes or 1,125 watts.
5. If an extension cord becomes very warm to the touch, it should be disconnected and checked for proper size.
6. In general, science laboratories should be equipped with sufficient receptacles to minimize extension cord use.

BIO 18.2.4 Fuses/Circuit Breakers.

1. Replace blown equipment fuses with fuses of the same amperage.
2. Replace fuses with the equipment unplugged.
3. Failure to use the correct fuse can cause damage to equipment and overheating.
4. Frequent blowing of circuit fuses or tripping of circuit breakers usually indicates that the circuit is overloaded or a short exists.
5. Circuit breakers and fuses that are tripped or blown should be turned on or replaced only after the cause of the short or overload is removed from the circuit.

BIO 18.2.5 Grounding

1. Use grounded 3-prong plugs when available.
2. If the outlet is 2-prong, use an adapter and secure the ground wire to the cover-plate screw on the outlet.
3.