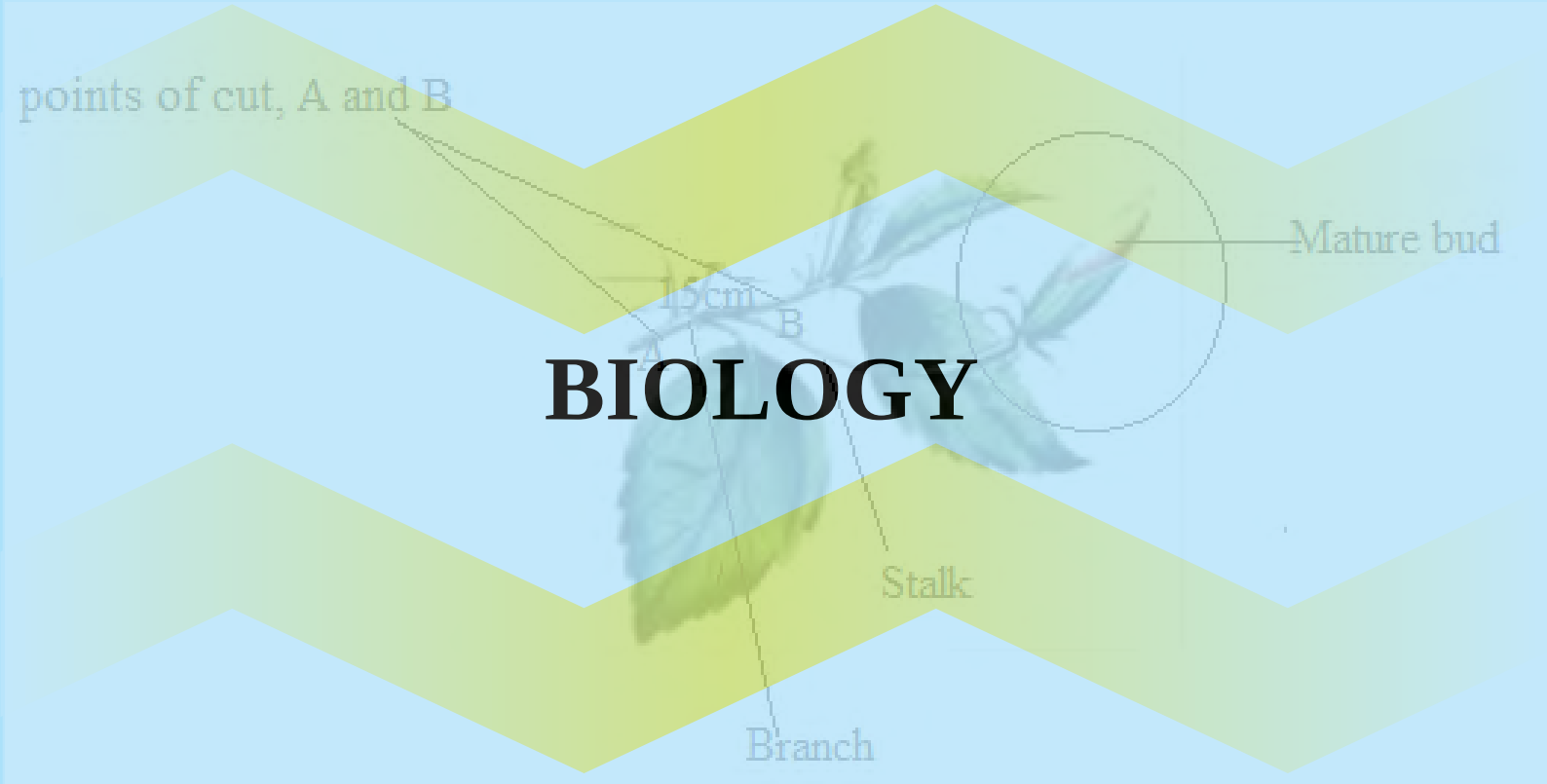
THE UNITED REPUBLIC OF TANZANIA

MINISTRY OF EDUCATION, SCIENCE AND TECHNOLOGY

NATIONAL EXAMINATIONS COUNCIL OF TANZANIA

GUIDELINES FOR PREPARING THE LABORATORY FOR PRACTICAL EXAMINATIONS AT SECONDARY AND DIPLOMA IN SECONDARY EDUCATION LEVELS



BIOLOGY

Prepared by:
The National Examinations Council of Tanzania,
P.O. Box 2624,
Dar es Salaam, Tanzania.

October 2021

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FOREWORD

The National Examinations Council of Tanzania (NECTA) administers biology practical examinations in Certificate of Secondary Education Examination (CSEE), Advanced Certificate of Secondary Education Examination (ACSEE) and Diploma in Secondary Education Examination (DSEE). The examinations aim at assessing candidates' competences in doing Biology practical at the respective education levels.

These guidelines provide important instructions on how to organise the laboratory in general, and the organisation of the laboratory for examinations in particular. It offers step by step guide notes to help the teachers and laboratory technicians in preparing and administering Biology practical examinations.

The guidelines aimed at providing the Biology teachers and laboratory technicians the basic skills needed for laboratory organisation, management and safety procedures on laboratory preparations for examinations. They also present the areas of assessment identified in the syllabi for Biology practical assessment at all levels of Secondary Education and Diploma in Secondary Education Examinations.

It is expected that, laboratory technicians and Biology teachers will use the guidelines effectively for proper and timely preparations of Biology practical examinations.

The Council extends its gratitude to all those who participated in the preparation of these guidelines.

Dr Charles E. Msonde
EXECUTIVE SECRETARY

1.0 INTRODUCTION

The National Examinations Council of Tanzania (NECTA) has prepared these guidelines for preparing the laboratory for practical examinations to cater for the needs of Biology teachers and Laboratory technicians for the ordinary and advanced certificates of secondary school education and diploma in secondary school education levels. The areas of practical assessment are in accordance with the syllabi issued by the Tanzania Institute of Education (TIE); for Ordinary Secondary Education, Advanced Secondary Education and Diploma in Secondary Education of 2010, 2009, 2009 respectively.

The guidelines highlight key issues on laboratory organisation, management and safety procedures for proper and timely preparation of the laboratory for Biology practical examinations. They also present the areas of assessment identified in the topics from Biology syllabi for ordinary, advanced and diploma levels. In addition, they present some of the apparatuses, chemicals, specimens and procedures required by Biology teachers and laboratory technicians for the preparation of the given practical examinations.

The areas of assessment presented are measurements, preparation of slides; setting up of a microscope and preparation of solutions and reagents. The guidelines further provide procedures on how to use the reagents and solutions prepared to test for the food substances present in various samples. They further include dissection of selected animals and flowers, and collection and preservation of specimens for practical examinations.

2.0 LABORATORY ORGANISATION, MANAGEMENT AND SAFETY

The organisation, management and safety of the laboratory are essential for the effective preparation of the laboratory for practical examinations.

2.1 Laboratory Organisation

Laboratory organisation is the way of handling the laboratory and its operation. It enables teachers and laboratory technicians to plan and arrange the apparatuses, chemicals and specimens properly. Laboratory organisation helps to save time and avoid confusion during preparation of laboratory for practical examinations.

It is expected that, Biology teachers and laboratory technicians are able to organise the Biology laboratory for practical examinations.

Key issues to consider while organising the laboratory

Teachers should use the checklist to identify apparatuses, specimens, chemicals, materials and other fittings, and arrange them accordingly by ensuring that:

(a) apparatuses and equipment are cleaned before they are kept in place to avoid contamination;

(b) chemicals and bench reagents are labelled and kept in a specific place;

(c) containers for preserved specimens are correctly labelled and specimens are undamaged;

(d) live specimens such as rats and cockroaches are kept in cages; frogs are kept in a well-ventilated container with little water, grasses and stones; and

(e) dissection equipment pieces are placed in boxes at a proper place to avoid damage, confusion and disturbance.

2.2 Laboratory Management

Laboratory management entails the procedure of handling of specimens, chemicals/reagents, solutions, equipment and apparatuses. It also deals with the operations of procurement, repair and maintenance of equipment in the laboratory setting.

It is expected that teachers and laboratory technicians are able to manage the Biology laboratory for preparation of practical examinations.

Key issues to consider in laboratory management

Teachers and laboratory technicians should be able to manage the chemicals, solutions, specimens, apparatuses and equipment for preparing the practical examinations in the Biology laboratory setting. For effective preparation of the laboratory for examinations, teachers and laboratory technicians should ensure the following:

(a) needs assessment is conducted by revisiting the school ledger book in order to establish the quantity of the apparatuses, specimens, chemicals and material available in the school laboratory. This will help to determine the quantity to be purchased to meet the requirements of the checklist;

(b) preserved specimens are intact and correctly labelled as prescribed by the Council in practical advance instructions;

(c) chemicals, solutions and bench reagents are freshly prepared and properly kept in labelled containers;

(d) live specimens such as rats, frogs and cockroaches are anesthetised and dissected according to practical instructions; and

(e) sections for microscopic observation are well prepared, stained and properly set on the microscope ready for observation.

2.3 Laboratory Safety

Safety in the laboratory is a state of being free from laboratory accidents or dangers. In the Biology laboratory, safety is important to both candidates and teachers.

It is expected that, teachers and laboratory technicians are able to safely handle preparation of the laboratory for practical examinations.

Key Issues to Maintaining Safety in the Laboratory

The teachers and laboratory technicians should be able to handle various apparatuses and hazardous chemicals used in the Biology laboratory to avoid accidents. In order to achieve these, adhere to the following safety considerations for effective preparation of a laboratory for practical examinations:

(a) wear a laboratory apron/coat, safety goggles, gloves when working with hot material, preserved specimens, dangerous chemicals, hot material and burners;

(b) tie back long hairs;

(c) if you have an open skin wound, be sure that it is covered with a water proof bandage;

(d) determine the location of fire extinguishers to combat fire accidents, water sources and alternative exit routes for departure in case of emergency;

(e) turn off all pieces of electrical equipment, water and gas taps when they are not in use;

(f) always wash your hands after working with live or preserved organisms and

(g) always work in a well-ventilated laboratory.

2.3.1 Safety Precautions while Preparing the Laboratory for Examinations

(a) Precautions while Working with Hot Substances or Fire

(i) When an object is removed from the heat for cooling, it should be handled with care.

(ii) Inflammable liquid bottles should neither be left open nor should they be dispensed near the flame or a hot electric element or electric motor.

(iii) Use test tube holders to handle hot laboratory equipment.

(iv) When you are heating something in a container such as a test tube, always point the open end of the container away from yourself and others.

(v) The Bunsen burner hoses should fit tightly to avoid gas leakage.

(vi) Appropriate heating apparatus should be used when heating substances, for example pyrex and borosil glass materials.

(b) Precautions while Preparing Chemicals, Reagents and Solutions

(i) Only use materials from containers that are properly labelled.

(ii) Do not add water to acid, instead, dilute the acid by adding it to water.

(iii) Wear lab coat and gloves to avoid burning with corrosive reagents.

(iv) Use fume chamber when dealing with concentrated hazardous chemicals.

(c) Precautions while Working with Electrical Equipment

(i) Make sure that the area around the electrical equipment is dry.

(ii) Never touch electrical equipment with wet hands.

(iii) The area surrounding the electrical equipment should be free from flammable materials.

(iv) Power switches should be turned off before plugging in and after using electrical appliances.

(v) Avoid use of equipment with worn out cords, damaged insulation or broken plugs.

(d) Precautions while Working with Glassware and Equipment

(i) All glassware should be cleaned before use.

(ii) If a mercury thermometer breaks, do not touch the mercury. Use a magnet enclosed in a piece of plastic sheet to collect the mercury and put it in a container. Use a pair of tongs to remove the magnet from the plastic sheet while it is inside the container to allow the mercury to drop in the container. Tightly close the container.

(iii) Be careful when handling all sharp equipment pieces such as scalpels and dissecting needles.

(iv) Never use laboratory glassware to serve food or drinks in the laboratory.

(v) All glassware should be properly stored and handled with care when used.

(e) Precautions while Preparing Live and Preserved Specimens

(i) Live specimens must be properly anaesthetized and stored in rigid leak proof containers.

(ii) Specimens for dissection should be properly mounted and supported by pins. Do not try to cut a live specimen while holding it in the air.

3.0 AREAS OF ASSESSMENT

This section presents the analysis of areas of assessment in some selected topics in the examination format for Certificate of Secondary Education Examination (CSEE), Advanced

Certificate of Secondary Education Examination (ACSEE) and Diploma in Secondary Education Examination (DSEE). The Biology practical examination will involve an assessment of different topics as shown in the Table 1.

Table 1: Areas of assessment in ordinary secondary education, advanced secondary education and diploma in secondary education syllabi

S/N	Areas of assessment			Education Levels		
	Practical Activities	**Topic**	**Subtopic**	**CSEE**	**ACSEE**	**DSEE**
1.	Measurements	• Introduction to Biology • Nutrition, • Transportation of Material in Living Things • Cytology • Basic Biology Laboratory Skills	• Scientific processes in biology • Properties of food substances • Diffusion, Osmosis and mass flow • Organic constituents of cells	√	√	√
2.	Preparation of slides and setting up of microscope	Cell Structure and Organization	The concept of cell	√	x	x
		Cytology	Cell structure and function	x	√	x
		Comparative Study of Natural Group of Organisms	Phylum • Chlorophyta • Apicomplexa • Euglenophyta • Oomycota • Ascomycota • Zygomycota	x	√	x
		Growth and Development	Primary and secondary growth in angiosperms	x	√	x
			Mitosis and growth	√	x	x
		Nutrition	Digestion in mammals	x	√	x
		Gaseous Exchange and Respiration	Gaseousexchange in mammals and plants	x	√	x
		Transportation of Material in Living Things	Transportation in plants and vertebrates	√	√	x
		Basic Biology Laboratory Skills	Plant sections	x	x	√

S/N	Areas of assessment			Education Levels		
	Practical Activities	**Topic**	**Subtopic**	**CSEE**	**ACSEE**	**DSEE**
		Coordination	Nervous coordination in human -neurons	√	x	x
			Nervous coordination in mammals	x	√	x
		Reproduction	Meiosis	√	√	x
3.	Preparation of solutions and reagents	Nutrition	Properties of food substances	√	x	x
		Cytology	Organic constituents of cell (carbohydrates, lipids, proteins and enzymes)	x	√	x
		Nutrition	Factors affecting rate of photosynthesis	x	√	x
		Basic Laboratory Skills	Preparation of common Biology chemicals and reagents	x	x	√
4.	Dissection of animals and flowers	Nutrition	Digestive system (rat)	√	x	x
		Gaseous Exchange and Respiration	Gaseous exchange in mammals	√	x	x
		Reproduction	Reproduction in mammals	√	x	x
		Comparative Study of Natural Groups of Organisms	Phylum arthropoda and Phylum chordata	x	√	x
		Basic Laboratory Skills	Dissection of small animals	x	x	√
			Plant sections	x	x	√
5.	Collection and preservation of specimen	Classification of Living Things	• Phylum chlorophyta • Kingdom fungi • Kingdom plantae • Kingdom animalia	√	x	√
		Movement	Movement of human body-human skeleton	√	x	x
		Principles of Classification	Taxonomic keys	x	√	x
		Comparative Study of Natural Groups	• Phylum chlorophyta	x	√	x

S/N	Areas of assessment			Education Levels		
	Practical Activities	**Topic**	**Subtopic**	**CSEE**	**ACSEE**	**DSEE**
		of Organisms	• Kingdom fungi • Kingdom plantae • Kingdom animalia			
		Transportation of Material in Living Things	Osmosis, diffusion and mass flow	√	x	x
		Transportation	Upward movement of water and mineral salts	x	√	x
		Growth	Growth in flowering plants	√	x	x

NB: √ - applicable; **x** - not applicabe.

3.1 Measurements

Measurements involve measuring instruments used in the Biology laboratory. The common measures in Biology are mass, length, time, temperature and pulse rate. Mass is measured by using weighing scales, length by using ruler and tape measure, time by using stop watch and clock, temperature by using thermometer and pulse rate by using stethoscope.

It is expected that, teachers and laboratory technicians are able to prepare measuring instruments for use when preparing the laboratory for practical examination for them and for the candidates in Biology practical examinations.

Key Issues to Consider while Preparing Measuring Instruments

The teachers and laboratory technicians should make sure that the measuring instruments for mass, length, time and temperature are working proparly.

Preparation of Measuring Instruments

(a) For the weighing scales such as digital and triple beam balance to measure accurate mass, the following should be observed:
- to use the digital balance, make sure that electrical installation and the ports are well functioning;
- to use the triple beam balance, make sure it is calibrated and adjusted to zero; and
- when the battery powered scales are used, the batteries should be functional.

(b) The ruler and tape measures must display the measurements.

(c) The stop watches and clocks must be functioning. Therefore, the batteries should be checked.

(d) The thermometer must display the measurements and filled with mercury, so blockage should be checked.

(e) Stethoscope must be working and should display the measurements.

3.2 Preparation of Slides and Setting up of Microscope

Slide is a small flat piece of glass on which objects are mounted for examination under microscope. Some of the microscopic objects that can be observed under the microscope are cells, thin sections from plant, animal tissues and the whole organism.

It is expected that, teachers and laboratory technicians are able to prepare stains, slides and set the microscope for observation of cells or thin sections from living organism.

Key Issues to Consider when Preparing the Slide and Setting the Microscope

The teachers and laboratory technicians should be able to do the following:

(a) prepare thin sections from plant and animal tissues;

(b) prepare stains;

(c) mount the object on the slide; and

(d) set the microscope for observation.

Apparatuses, chemicals and materials

The following are requirements for preparation of stain:
spatula, methylene blue powder, 100% ethyl alcohol, distilled water, aniline blue aniline green, backer, stirrer.

3.2.1 Preparation of Stains

The following are the procedures for preparing the stains:

(a) Preparation of methylene blue stain

To prepare 0.2% of aqueous methylene blue stain, the following are the procedures:

(i) weigh 0.2 g of methylene blue powder, then dissolve it in 75 ml of distilled water;

(ii) add distilled water in the solution to make 100 ml; and

(iii) store it in a clean labelled container ready for use as stain.

(b) Preparation of methylene green stain

To prepare 1% of alcohol methylene green stain, follow the following procedures:

(i) weigh 1 g of methylene green powder, then dissolve it in 75 ml of 95% ethyl alcohol;

(ii) add 95% ethyl alcohol in the solution to make 100 ml; and

(iii) store it in a clean labelled container ready for use as stain for small organisms.

(c) Preparation of alcohol aniline blue stain

To prepare % alcohol aniline blue stain the procedure is as follows:

(i) dissolve 1 g of aniline blue in 100 ml of 85% ethyl alcohol; and

(ii) store it in a clean labelled container ready for use as stain for cellulose.

(d) Preparation of aqueous aniline blue stain

To prepare 0.5% aqueous aniline blue stain the following are procedures:

(i) dissolve 0.5 g of aniline blue in 50 ml of distilled water;

(ii) dilute in 100 ml of distilled water and filter if necessary; and

(iii) store it in a clean labelled container ready for use as stain for fungi and algae.

3.2.2 Preparation of slides

The following apparatuses, chemicals, specimens and materials are requirements for preparation of slide:

Light microscope, plastic slides, cover slips, needles, dropper, iodine solution, methylene blue stain, beakers, petri dishes, plant tissues, animal tissues, fungi, micro organisms, surgical blade or sharp knife and toothpick.

3.2.2.1 Preparation of the slide with animal tissue

Steps for preparation of slides for animal tissues observation by using microscope are as follows:

(i) collect the thin, transparent, inner lining of animal tissue e.g. the inner lining of your cheek. You may scrape the inner surface of the cheek gently to avoid any damage or bleeding to collect some viscous transparent substance (Instead of tooth pick, you may use the uncoated end of a matchstick);

(ii) smear the transparent substance on a slide;

(iii) add a drop of water to the smear;

(iv) add a drop of methylene blue stain and leave it for about one minute;

(v) tilt the slide to let the extra stain drain off. Wash gently with water to remove the stain;

(vi) put a coverslip gently over the object with the help of a needle avoiding entry of any air bubbles. Press it gently to make the cells under the cover to slip uniformly; and

(vii) observe under 40x and 100x magnifications on the microscope and find out the structural details of an object.

3.2.2.2 Preparation of slides with plant tissues

Steps for preparation of slides for plant tissues observation by microscope are as follows:

(i) collect fresh plant specimens from the field;

(ii) wash plant specimens by slowly running water to remove soil and unwanted materials;

(iii) cut transparent sections or slices of the required tissue from the plant specimen using a surgical blade;

(iv) transfer sections into a petri dish containing 70% ethanol to kill cells and fix their histological form from being altered;

(v) transfer the perfect section on the slide (you may add a drop of Aniline blue stain for higher visibility and leave it for 2 minutes);

(vi) add a drop of methylene blue stain and leave it for 3 minutes;

(vii) take the section out of methylene blue to a petri dish containing distilled water and rinse to remove stains;

(viii) put the same section on the slide again

(ix) add drops of glycerol. For temporally mounting, add drops of 50% glycerol to prevent drying. For permanent mounting, add drops of 100% glycerol for 2 minutes to prevent drying and long-lasting;

(x) put a coverslip gently with the help of a needle to avoid entry of any air bubbles. Press it gently to make the cells under the cover slip uniform; and

(xi) set the microscope and observe under 40 x to 100x magnification.

3.2.3 Setting up the Microscope

(i) Place the microscope away from the edge of the table and rotate the objective to bring it in line with the body tube until you hear a faint clicking sound.

(ii) Look through the eye lens and adjust the mirror below to attain a good illumination of the circular area visible.

(iii) Keep the slide to be studied on the stage such that, the specimen lies just above the aperture of the stage and hold it in position by the help of the clips. The slide, the stage, and the lens should be clean.

(iv) Move the coarse adjustment knob to the cover slip of the slide.

(v) Look through the eyepiece and using the coarse adjustment knob, bring an upward movement of the body tube until the slide comes into focus. A fine adjustment knob may be used to bring in a sharp focus. Always keep both eyes open, though you may decide to use only one eye.

(vi) For focusing under the high magnification, always focus under the low power first and then switch to high power magnification. Using the fine adjustment knob, focus the object upwards, never focus downwards since the slide is very close to the lens. Never use the coarse adjustment knob during high magnification.

(vii) Leave the set-up for candidates to observe the specimen mounted on the slide.

3.3 Preparation of Solutions and Reagents

A solution is a mixture made up of water and food sample to be tested. Reagents are chemicals used to determine which substances are present in sample specimen or solution.

It is expected that, teachers and laboratory technicians are able to prepare fresh solutions, table reagents for biochemical practical examinations.

Key Issues to Consider While Preparing Solutions and Reagents

The Biology teachers and laboratory technicians should be able to prepare fresh solutions and bench reagents required for biochemical food test practical examinations. The following should be considered:

(a) all solutions and reagents are fresh; and

(b) all storage containers are well labelled.

3.3.1 Preparation of Solutions

The solutions to be prepared are glucose, sucrose, starch, protein and fat/lipid. The requirements for preparation of these solutions are:

Specimens

Food sources such as ripe fruits, carrots, onion bulbs, cassava, potatoes, eggs, sugar, coconut oil, vegetable oil and castor seeds are some of the specimens to be prepared.

Chemicals

Industrial chemicals include egg albumen powder, glucose powder, starch powder and sucrose crystals.

Apparatuses/equipment

Beam/digital balance, measuring cylinders, labelled containers spatula, funnels, graduated flasks, stirring rods, scalpel/razor blades, forceps, petri dishes, source of heat, tripod stand, and beakers.

(a) Preparation of protein solution

(i) Dissolve 10 g of egg albumen powder in a litre of distilled, rain or tap water to obtain 1% protein solution.

(ii) Stir gently and carefully to avoid foaming and obtain a homogenous solution.

Note: It takes time to get homogeneous solution, so keep on stirring.

(b) Preparation of glucose solution

(i) Dissolve about 10 g of glucose powder in a litre of distilled, rain or tap water to obtain 1% glucose solution.

(ii) Stir gently to obtain a homogenous solution.

(c) Preparation of sucrose solution

(i) Dissolve 10 g of sucrose powder in a litre of distilled, rain or tap water to obtain 1% sucrose solution.

(ii) Stir gently to obtain a homogenous solution.

(d) Preparation of starch solution

(i) Dissolve about 10 g of starch powder in a litre of distilled, rain or tap water to obtain 1% of starch solution.

(ii) Stir gently.

(iii) Boil the mixture while stirring to obtain a homogenous solution.

(iv) Leave the solution to cool at room temperature.

(e) Preparation of lipid solution

(i) Dissolve about 10 ml of vegetable oil in a litre of distilled, rain or tap water to obtain 1% lipid mixture.

(ii) Stir the mixture gently.

3.3.2 Preparation of Reagents

Reagents to be prepared are Benedict's, iodine, sodium hydroxide, copper(II) sulphate, dilute hydrochloric acid and Sudan III solutions.

Apparatuses/equipment

Apart from other fitting of the laboratory, the following are required;

Measuring scale, measuring cylinder, coated umber glass bottle, glass/plastic containers with lids to keep reagents, spatula, funnel, graduated flasks and the stirring rod.

(a) Preparation of Benedict's Solution

Chemical requirements

Sodium citrate, sodium carbonate and distilled water

Procedures

(i) Weigh 173 g of sodium citrate and 100 g of sodium carbonate and then mix the ingredients.

(ii) Put the mixture into 800 ml of distilled water, stir the solution until it dissolves.

(iii) Filter and make the volume to 850 ml by adding distilled water.

(iv) In a separate container, dissolve 17.3 g of copper (II) sulphate in 100 ml of distilled water.

(v) Slowly, while stirring constantly, add the copper (II) sulphate solution to the first solution and make the final volume to 1 litre by adding distilled water.

(vi) Store the obtained solutions in a container ready for use in testing for reducing sugar.

(b) Preparation of 1% Copper II Sulphate Solution

Chemical requirements

Copper(II) sulphate crystals and distilled water

Procedures

(i) Weigh 1 g of copper(II) sulphate crystals, dissolve it in 100 ml of distilled water.

(ii) By using a stirring rod, stir the solution until the crystals dissolve completely.

(iii) Store the obtained solution in a container ready for use.

(c) Preparation of iodine solution

Iodine crystals are not directly soluble in water. They are soluble in potassium iodide solutions.

Chemical requirements

Distilled water, potassium iodide and iodine crystals

Procedures

(i) Weigh 15 g of potassium iodide and dissolve into 125 ml of distilled water.

(ii) Weigh 3 g of iodine crystals and dissolve it into potassium iodide solution in (i).

(iii) Stir until iodine crystals dissolve completely in potassium iodide solution.

(iv) Dilute the solution and make the final volume to 1 litre by adding more distilled water.

Note: The iodine solution obtained must always be stored in dark or amber bottle to prevent photo decomposition

(d) Preparation of Sodium Hydroxide Solution

Chemical requirements

Sodium hydroxide pellets and distilled water.

Procedures

To prepare 1 M sodium hydroxide solution, the following procedures should be followed:

(i) weigh 40 g of sodium hydroxide pellets. Dissolve in 700 ml of distilled water, then stir it well;

(ii) dilute the solution and make the final volume to 1 litre by adding more distilled water; and

(iii) store it in a plastic container.

Note: Sodium hydroxide solution when stored in a glass container may form sodium silicate which hardens the stopper and become difficult to reopen when the solution is required for use.

(e) Preparation of dilute hydrochloric acid

R*equirements*

Fume chamber, laboratory coats, safety glasses or face shield, closed in shoes, concentrated HCl, glass measuring cylinder (1 L) or volumetric flask, glass stirring rod or magnetic stirrer and pre-labelled storage bottles.

Key issues to consider while preparing the dilute hydrochloric acid

The density and percentage purity are indicated on the bottle, but the molar concentration of the stock solution is not given. In order to prepare the standard

solution of the acid from the concentrated liquid, it is a requirement to understand the following:

(a) the required molarity of the solution; and

(b) the exact volume of the solution needed.

From that consideration, the volume of the concentrated acid can be calculated and diluted in distilled water to obtain the required volume.
However, the molarity of the concentrated stock solution can be calculated from the specifications given on bottle of concentrated acid by using the following formula:

$$\text{Molarity} = \frac{\%\,\text{assay} \times \rho \times 1000}{\text{Mr} \times 100} \text{ where by,}$$

% = percentage purity of a stock concentrated solution
ρ = the density of a stock solution in g/cm^3
Mr = is the molar mass of the solution

In order to obtain the volume of the concentrated stock solution which should be diluted, the dilution formula should be applied. The formula is stated as follows:

$$M_1V_1 = M_2V_2.$$

Where,

M_1 = molarity of the concentrated stock solution calculated from bottle specifications,

V_1 = the volume of the stock solution which should be drawn from bottle in order to be diluted,

M_2 = the required molarity of a solution after diluting the concentrated acid.

V_2 = the volume of the required solution to be prepared after dilution of concentrated acid.

Preparation of dilute hydrochloric acid (1 M)

In order to dilute the stock solution to obtain (1 M) dilute hydrochloric acid (HCl) the following procedures should be followed:

(i) pour the concentrated acid from bottle to the beaker and measure the required volume using a measuring cylinder (see Table 2);

(ii) measure 1000 ml of distilled water and divide the water into two beakers, each 500 ml;

(iii) pour the measured acid into one of the beakers with 500 ml of distilled water;

(iv) rinse the measuring cylinder three times with distilled water and pours in (iii); and

(v) slowly add distilled water into the contents of the beaker with acid in (iii) to make 1000 ml of 1M dilute HCl.

Note: add acid to water and **not** water to acid.

The following table presents the percentage purity and density of concentrated acid of stock solutions and the volume of concentrated acid required to be diluted in 1 litre to prepare 1 M of dilute hydrochloric acid.

Table 2: Percentage purity (assay), density and volume of concentrated acid required to prepare 1 litre of 1M HCl solution.

S/N	Percentage purity	Density (g/cm^3)	Volume of conc. Acid to be diluted to make 1L solution (cm^3)
1	31-32	1.16	99.9
2	34	1.18	90.9
3	35	1.18	88.4
4	36	1.18	85.9
5	37	1.18	83.5
6	38	1.18	81.4

(f) Preparation of Sudan III solution

Chemical requirements

Sudan III powder, ethyl alcohol 95% and distilled water.

Procedures

(i) Measure 73.5 ml of 95% of ethyl alcohol then warm it into water bath at 75 ^oC.

(ii) Measure 0.5 g of Sudan III powder and add into the solution above, then stir it to disolve completely.

(iii) Add 75^oC distilled water to just below 100 ml mark.

(iv) Stir the solution and cool it at room temperature.

(v) Dilute it up to 100 ml with distiled water.

Note: If the percentage purity of ethyl alcohol is 100%, mix 95 ml alcohol with 5 ml of water to make 95% ethyl alcohol.

(g) Preparation of Calcium Hydroxide Solution (Lime Water)

Procedures

(i) Dissolve 25 g calcium hydroxide powder in 1 litre of distilled water.

(ii) Vigorously stir the mixture for 2 minutes.

(iii) Wait for the mixture to settle and decant it to obtain clear solution.

(iv) Filter decanted solution using filter paper to obtain clear solution.

(v) Store the solution in container ready for use for detecting carbondioxide gas.

(h) Preparation of Amylase

Procedures

Procedures for preparation of 0.5% aquous amylase solution involve the following:

(i) dissolve 0.5 g of amylase in 50 ml of distilled water;

(ii) dilute it to 100 ml using distilled water; and

(iii) store the solution in a container ready for use in starch digestion.

3.3.3 Food Test

The chemical reagents prepared are used in biochemical test to identify the food substances present in the sample solutions. The reagents also are used to test the presence of starch in a leaf after photosynthesis.

3.3.3.1 Identification of Food Substances Present in the Sample Solutions

Experimental work for identification of the food substances in a given solution is shown in Table 3.

Table 3: Food Test Sample Report

Food tested	Procedure	Observation	Inference
Starch	Take 2 ml of starch solution in a test tube, then add two drops of iodine solution and shake gently.	The mixture turns into blue-black colouration	Starch present in the solution
Reducing sugar	Take 2 ml of glucose solution in a test tube, add 2 ml of Benedict's' solution then boil while shaking.	A series of colour change observed from blue, green, yellow, orange to brick red precipitate.	Reducing sugar present in the solution

Food tested	Procedure	Observation	Inference
Non reducing sugar	Take 2 ml of sucrose solution in a test tube, add 1 ml of dilute hydrochloric acid and boil the contents. After cooling add 2 ml of sodium hydroxide solution followed by 2 ml of Benedict's solution and re-boil while shaking the contents.	A series of colour change observed from blue, green, yellow, orange to brick red precipitate.	Non reducing sugar present in the solution
Protein	Take 2 ml of protein solution in a test tube, add 2 ml of sodium hydroxide solution and mix the content thoroughly. Then slowly add 2 drops of 1% copper (II) sulphate solution while shaking the mixture.	The mixture turns into purple/violet colour	Protein present in the solution
Lipid	Take 2 ml of vegetable oil solution in a test tube, add three drops of Sudan III solution then shake the content vigorously and leave to settle for 5 minutes.	A red ring layer formed at the top of the solution.	Lipid present in the solution

3.3.3.2 Identification of Starch in the Leaf

The iodine solution prepared is used in biochemical test to identify the presence of starch in the green leaves. The following procedures should be followed for preparation of green and variegated leaf for identification of starch.

Procedures

(i) Collect green leaf or variegated leaf from a photosynthesizing plant that has been exposed to the direct sunlight for 6 hours.

(ii) Prepare boiling water at 100 ^{o}C.

(iii) Immerse the leaf for about 3 minutes to kill the cells.

(iv) Remove the leaf from the water and insert it in a test tube containing less than half-filled ethanol.

(v) Plug the test tube with a piece of cotton wool.

(vi) Submerge the test tube in the boiling water and leave it to boil until the leaf loses its colour.

(vii) Remove the leaf from ethanol solution and dip it briefly into warm water to wash out ethanol and soften it.

(viii) Remove the leaf from the warm water.

(ix) Spread the decolourized leaf on the white tile and add iodine solution until the whole leaf is covered.

(x) Observe the colour change on the leaf and record your observations.

How to Test for Starch in the Leaves

Experimental work for identification of starch in a green and variegated leaf is as shown in table 4.

Table 4: Test for Starch in Green and Variegated Leaf

Test	Procedure	Observation	Inference
Test for starch in a simple green leaf	Add iodine solution until the whole leaf is covered.	The whole leaf turns into blue-black colouration.	Starch present in the simple green leaf.
Test for starch in the variegated leaf	Add iodine solution until the whole leaf is covered.	The green portion turns into blue-black colouration.	Starch present in the green portion of the leaf indicating that chlorophyll is necessary during photosynthesis.

3.4 Dissection

Dissection involves the displaying of various systems of plants and animals. It is expected that, teachers and laboratory technicians should be able to prepare and dissect animals and flowers for Biology practical examinations. They should also be able to cconduct assessment of candidates' dissections and drawings.

3.4.1 Dissection of Animals

Key Issues to Consider While Dissecting the Animals

Teachers and laboratory technicians should be able to do the following:

- prepare the dissecting tray;
- prepare specimen for dissection; and
- dissect the rats/mouse, guinea pigs, frog/toads and flowers to display various systems for candidates to observe in the examination.

(a) Preparation of the Dissecting Tray

(i) Heat some paraffin-wax to melting point.

(ii) Pour the wax in the dissecting tray.

(iii) Leave the wax to cool. The cooling process may be hastened by immersing the dissecting tray in cold water.

(b) Preparation of Animal Specimen for Dissection

Anesthetise the fresh cockroach, rats/mouse, guinea pigs and frogs before the commencement of the examination as follows:

(i) wet the cotton wool with chloroform and put it inside an air tight container;

(ii) put a live animal in the air tight container containing the wet cotton wool;

(iii) leave the animal in the air tight container for about 4 minutes to make it unconscious;

(iv) remove the animal from the air tight container by using forceps and place it in the dissecting tray; and

(v) leave the specimen for few minutes to allow evaporation of chloroform.

3.4.2 Dissection of Rat/Mouse and Guinea Pigs

The following apparatuses, specimens and materials are required for dissection:
Dissecting kit/instruments, hand lens, scalpel, microscope, needle, forceps, surgical blade, slide, fresh flowers, water, fresh rats/mouse/guinea pigs, fresh frog/toad, T-pins, pieces of cotton wool and cotton thread.

Procedures

(i) Lay the specimen with the ventral side (abdomen) facing upwards.

(ii) Pin it down on the tray through the fore and hind limbs.

(iii) By using forceps lift the skin of the abdomen and with scissors make a longitudinal slit between hind limbs.

(iv) Extend this cut straight up to the neck and make incision in the limbs.

(v) Separate and stretch the skin from the under body wall by the help of blunt forceps, scalpel and fingers.

(vi) Pin the folds of skin by placing one pin above the fore limbs, below the fore limbs and another one near the hind limbs on both sides.

(vii) Cut the body wall on either side of the midline and pin it, in order to observe different internal organs. Do not cut deeply to avoid damage of the veins and other internal organs.

(viii) Open the thoracic cavity/region by cutting through the intercostal muscles towards the neck.

(ix) Cover the dissected specimen completely with water to make clear observation.

(x) With the help of a hand lens, observe the dissected specimen and note the position and shape of the organs and systems.

(xi) Carefully remove unwanted organs and tissues to expose targeted organs or systems as per instructions.

3.4.3 Dissection of a Frog/Toad

Procedures

(i) Place the frog on the dissecting tray.

(ii) Turn the frog on its back and pin down the legs.

(iii) Look for the opening to the specimen's cloaca, located between the hind legs. Use forceps to lift the skin and use scissors to cut along the centre of the body from the cloaca to the lip.

(iv) Turn back the skin, cut towards the side at each leg and pin the skin flat.

(v) Ligature the ventral abdominal vein at two sides using thread in order to prevent bleeding. Then, cut between the ligatured points.

(vi) Using forceps lift the abdominal vein and cut alongside it to the breast bone to open up the body cavity and make insicions towards the limbs. Pin the inner skin.

(vii) Cover the frog completely with water for clear observation.

(viii) With the help of a hand lens, observe the dissected frog and note the position and shape of the organs and the system**.** You may use forceps to remove the unwanted organs to facilitate observation of the organ under investigation as per 3 Hours Practical Advance Instructions.

3.4.4 Dissection of a Flower

Identify the relevant flower by assessing the floral parts and location of the following in the whole flower:

- sepals forming the calyx;
- receptacle is the swollen part below the calyx;
- flower stalk (pedicel);
- petals that form a corolla;

- stamens with anthers and filaments; and
- pistil with stigma, style and ovary.

(a) Dissection to half flower

(i) Count the number of sepals, petals and stigma.

(ii) Cut the flower longitudinally into half and separate it into two halves.

(iii) Use the hand lens to observe the sepals, styles, petals, stamen, stigma, ovary and the ovules.

(b) Dissection to display carpel

(i) Use forceps to cut and make a small opening between the two sepals, then use hand to peel the calyx completely from the flower.

(ii) Detach the petals by pulling each one downwards towards the receptacle until the entire corolla is removed from the flower.

(iii) Use forceps to cut and make a small opening at the base of the staminal tube, then expose the ovary with your finger nails by pilling off the staminal tube of the flower around the ovary.

(iv) Use needle to cut at the base of the staminal tube, then extend the incision to the base of the stigma. Care must be taken not to damage the ovary and the style under it.

(v) Repeat the cuttings as much as possible, and then pull apart the strands to completely remove the staminal tube from the flower.

(vi) Look at the flower you will see stigma, style and ovary collectively known as the carpel.

(vii) Observe the ovary, stigma, and style of your flower with a magnifying glass.

(c) Dissection to display stamen

(i) Use forceps to cut and make a small opening between the two sepals, then use hand to peel the calyx completely from the flower.

(ii) Detach the petals by pulling each one downwards towards the receptacle until the entire corolla is removed from the flower.

(iii) Detach the filament and use the hand lens to observe anther and filament.

3.4.5 Assessment of Candidate's Dissection and Drawing

It is expected that, Biology teachers are able to conduct on-the spot assessment of each candidate's dissection and drawing in Biology practical examinations.

Key issues to consider while assessing the candidate's works

The Biology teachers should be able to make an on-the spot assessment of each candidate's dissection and drawing at least 45 minutes after the beginning of the examination. They should do the following to assess the candidate:

(i) study carefully the assessment form which shows the task of the examination question;

(ii) assess the quality of dissection according to the NECTA standards indicated in the assessment form; and

(iii) assess the quality of drawing whether it accurately represents the candidate's dissection.

Note: Drawing of the candidate who did not dissect the animal should be awarded zero mark in on-spot assessment.

3.5 Collection and Preservation of Specimens

It is expected that, teachers and laboratory technicians are able to collect, preserve and prepare the specimens for practical examinations.

Key Issues to Consider while Collecting and Preserving the Specimens

The teachers and laboratory technicians should be able to perform the following:

- prepare the preservatives.
- identify the habitat and collect the specimens for examinations; and
- preserve specimens by using appropriate preservative and make preparation for practical examinations.

Requirements

Apparatus: Scalpels, petri dish, forceps, hand-lens, a pair of scissors, knife, dropper, collecting bottles, preserving bottle, metal hooks and desiccators.
Chemicals: Insecticide, chloroform, formalin and alcohol.
Materials: Insect net/fly traps, gloves, cotton wool, thread, cello tape and plastic bags.

3.5.1 Preparation of Preservatives

Preserves which are usually used are ethyl alcohol and formaldehyde.

3.5.1.1 Preparation of 70% Ethanol (Ethyl alcohol)

Procedures

(i) Measure 70 ml of ethanol using a measuring cylinder and put it in a beaker.

(ii) Measure 30 ml of distilled water and mix it with the ethanol in the beaker.

(iii) Label the solution as 70% ethanol and is ready to be used.

3.5.1.2 Preparation of 10% Formalin (Formaldehyde)

Procedures

(i) Measure 10 ml of formalin using a measuring cylinder and put it in a beaker.

(ii) Measure 90 ml of distilled water and mix it with the formalin in the beaker.

(iii) Label the solution as 10% formalin (ready to be used).

Note: Precaution has to be taken when preparing formalin (formaldehyde). The chemical is potentially carcinogenic, so people working with it should avoid skin contact with the chemical and inhalation of its fumes. Therefore, wearing of lab coat, gas mask and gloves is very crucial when working with formalin.

3.5.2 Collection and Preservation of Spirogyra

Habitat: The specimens are abundantly available in the fresh water, neutral or slightly acidic pH ponds, lakes and slow moving streams and rivers.

Preservation

The spirogyra filaments can be preserved in the 10% formalin.

3.5.3 Collection and Preservation of Fungi Specimens

3.5.3.1 Collection of Molds (Rhizopus)

Habitat: A variety of molds are readily available on decaying organic matter.
The teachers and laboratory technicians should prepare a culture of molds or rhizopus using the following procedures:

(i) obtain a piece of bread, put it in a Petri dish and wet it with a little amount of water;

(ii) expose the prepared culture for one hour;

(iii) place the prepared in a dark warm place, for example cupboard; and

(iv) after 5 days, observe the prepared culture by using a hand lens.

3.5.3.2 Collection of Mushroom

Habitat: Mushrooms should be collected during the rainy season. Mushrooms can be found on dead and decaying materials like logs in the forest. They may also be purchased from farm, shops and supermarkets.

Preservation
Dry mushrooms in sunlight or preserve them in 70% ethanol.

3.5.3.3 Collection of Yeast

Yeast can be purchased at shops and preserved by keeping it in an air-tight container.

3.5.4 Collection and Preservation of Plant Specimens

Collect a variety of plant materials such as moss, liverwort, fern, conifers flowering plants including seedlings and parts of the plants such as leaves, flowers and roots.

General procedures
Teachers and laboratory technicians should collect, prepare and preserve a variety of plants by using the following general procedures:

(i) place the collected plants and plant parts in plastic bags;

(ii) place the plants and plant parts in wooden plant press, kept on herbarium sheets/manila papers;

(iii) tie a plant press tightly by using ropes; and

(iv) leave the plant press to dry for about one week by keeping the plant warm and well aerated.

3.5.4.1 Collection of Moss and Liverworts

Habitat: Moss should be collected during the rainy season or in damp places. Moss and liverwort can be found on rocks or trees in moist climates or in rocky river-banks. Matured mosses consist of definite sporophyte and gametophyte.

Methods and Procedures

(i) Collect organisms (mosses and liverworts) from the field, then leave them attached to the substrate on which they naturally grow.

(ii) Transport the specimens to the laboratory in plastic bags or folded in newspapers.

(iii) Transfer to terraria which can be readily prepared in the laboratory as follow:

- place 3 cm layer of coarse gravel or pebbles on the bottom of the tank;
- add a few pieces of charcoal;
- over this, spread a 2 cm layer of sand soil;
- add cover of garden loam soil about 3 cm deep;

- in this bed, sod the mosses and liverworts which have been collected from the field;
- add water to the terrarium until it is halfway up the gravel layer and the tank should be covered with grass;
- keep the tank in medium light, but when molds grow in the tank, reduce the amount of water and remove the cover until they disappear; and
- maintain the terrarium with the least amount of water needed to keep the plant alive for the examination.

3.5.4.2 Collection of the Ferns

Habitat: Ferns can be found in shady and humid environments, usually in forests. Ferns also grow in moist, shady environments like ground beds of forests. Matured ferns consist of large frond with sori underneath.

3.5.4.3 Collection of the Conifers

Coniferophyta can be found in cooler, higher climates. Specimen collected are branches, leaves and cones.

3.5.4.4 Collection of Flowering Plants

Plants can be collected as small plants or seedlings. Angiosperms are easily found in the surrounding environment. Monocotyledons are organisms like maize plants and some grasses. Dicotyledons are bean plants, groundnuts and peas.

Seedling can be prepared by the following procedure:

(i) take wet cotton wool and spread it on a large container;

(ii) put the seeds inside the cotton wool and make sure the seeds are not completely covered with water;

(iii) leave them for a week, and

(iv) make sure that the seeds are not dry.

3.5.4.5 Collection of the Plant Parts

Plants can be collected as parts, such as seeds, leaves, roots and flowers. To collect and preserve parts of the plant such as leaves, flowers, roots and stems, the following should be done:

(i) cut the branch of leaves and flowers from the plant and place it in water for hours;

(ii) to preserve flowers for some days, pick a bunch of flowers from the plant, place it in the air tight plastic bag and store in a cool place or in a refrigerator; and

(iii) fresh flowers may also be obtained from the preserved mature flower buds. The following procedures can help to obtain fresh flowers from preserved buds:

- Cut a branch with matured flower bud from the plant
- Place them inside a bundle of dry grasses and keep them in cool place. By so doing they may stay fresh for seven days
- Prepare 1% glucose solution by dissolving 10 g of glucose powder in 1000 ml of rain or tap water
- Then cut the small portion of the branch by using a pair of scissors to obtain the fresh tissue of the branch
- Immerse petioles in the 1% glucose solution one day before the commencement of the biology practical examination to obtain fresh flower.

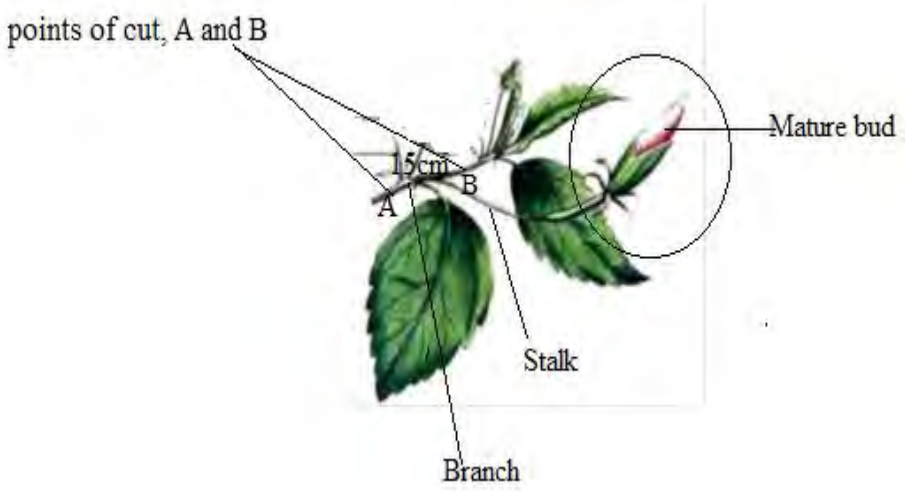

Figure 1: Mature bud of hibiscus flower

3.5.5 Collection and Preservation of Animal Specimens

3.5.5.1 Collection of Worms

Liver flukes and tapeworms can be collected when a cow, pig, goat or sheep is slaughtered and the liver or intestines get examined. There are some species that can be found in shallow tide pools along the beach. Once collected they can be preserved by keeping them in labelled air-tight containers with 10% alcohol solution.

Roundworms can be found in the stomach of fish, soil and stagnant water. They can also be found in the intestines of locally raised chickens, goats, pigs and dogs. Worms can be preserved by keeping them in labelled air-tight containers with 70% alcohol.

Earthworms can be found after rain by digging under rocks or in other damp places. Leeches can be found in rivers, paddy fields and other damp places. Live earthworms can be kept in a container with fresh soil. If they are dead, these organisms can be preserved in ethanol.

3.5.5.2 Collection of the Insect Specimens

To achieve this, the teacher should perform the following activities:

(i) sweep the vegetation with a strong insect net or use the insect killer to spray the area where insects are found;
(ii) collect the insects by using forceps and place them in a collecting bottle;
(iii) put the live insects into a bottle containing cotton wool soaked in chloroform to anaesthtize them; and
(iv) preserve the insects by using 70% ethanol.

3.5.5.3 Collection of Crustaceans

Crabs and prawns can be found in most rivers, lakes, dams, oceans and swamps. otherwise, they can be purchased in markets.

Preservation
Crustaceans can be preserved in 70% alcohol. Crustacea can also be dried for preservation purposes

3.5.5.4 Collection of Arachnids

The collection may include spiders, ticks and scorpions.
Spiders can be found in almost any environment. Scorpions can be found in dark, dry and cool areas, usually at night. Ticks are found on skins of animals such as dogs, cattle and sheep.

Preservation
Arachnida can be dried or preserved in 70% alcohol.

3.5.5.5 Collection of Chilopods and Diplopods

Chilopods includes centipedes while diplopods includes millipedes. Centipedes and milipedes can be found under rocks, in tree barks and leaf litter.

Preservation

Both can be dried or preserved in 70 alcohol.

3.5.5.6 Collection of Chordates

(a) Collection of Chondrichthyes and Osteichthyes

Chondrichthyes are catilagenous fish and osteichthyes are bony fish such as tillapia. Fish can be found in fish markets, ocean, ponds, lakes and rivers.

Preservation
Fish can be reserved in a 10% formaldehyde, dried on the sun or smoked as well.

(b) Collection of Amphibians

Amphibians include frogs and toads. These organisms can be found near rivers or ponds. Toads can also be collected at night during the rainy season. Use sweep nets to capture amphibians.

Preservation

- Make an aquarium or container with water for preservation of live specimens and provide them with small insects for food and source of water.
- For preservation of dead specimens, inject them with 10 -30% formaldehyde.

(c) Collection of Reptiles

Collection of reptiles such as lizards, snakes and chameleons can be done in rocks or in caves and inside the cracks in the walls. They can be also collected by using sweep nets, traps or fishing nets.

Preservation

Preserve the specimens by placing them in an air tight container with 10% formaldehyde solution.

(d) Collection of Birds

Birds such as chicken are kept domestically and can be easily purchased or raised. Wild birds usually live in the forest and can be captured by using bird trap nets.

Preservation

Dead specimens can be preserved by placing them in an airtight container with 10% formaldehyde solution.

(e) Collection of Mammals

Mammals can be kept as small animals such as rats/ mouse and guinea pigs. Rats can be captured overnight using a trap. Bats can be collected using sweep net during the day especially when they are asleep.

Preservation

Mammals can be preserved in a 10% formaldehyde solution.

(f) Collection of Animal Parts

Various parts of animals can be collected for biology practical examinations. The body parts from birds can be collected such as feathers and beak. Mammalian skin, bones and teeth may also be collected for biology practical examinations.

Preservation

To preserve the animal parts such bones, feathers, beaks and others the following should be done:

- Detach them from the organisms
- Dry the parts in the sun.

4.0 CONCLUSION

These guidelines have presented the information to equip teachers and laboratory technicians with knowledge and skills in preparing the Biology laboratory for CSEE, ACSEE and DSEE practical examinations. The knowledge on laboratory organisation, management and safety procedures intends to help them to have timely preparation of the laboratory for Biology practical examinations. The areas of assessment presented will assist Biology teachers and laboratory technicians to correctly prepare the measuring instruments, slides and setting up of a microscope. They will also be able to effectively prepare stains, solutions and reagents. The guidelines further provide procedures on how to use the reagents and solutions prepared to identify food substances, dissection of animals and flowers, and collection and preservation of specimens for Biology practical examinations.