



Ethiopian TVET– System



Water Supply and Sanitation Supervision

Level-IV

Based on Feb; 2017 G.C. Occupational Standard

Module Title: Performing Advanced Water Quality Laboratory Test

TTLM Code: EIS WSS4 TTLM 0620v1

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This module includes the following Learning Guides:

LG 53: Prepare samples for testing

LG Code: EIS WSS4 M04LO1-LG-53

LG 54: Prepare for laboratory testing

LG Code: EIS WSS4 M04LO2-LG-54

LG 55: Perform tests on samples

LG Code: EIS WSS4 M04LO3-LG-55

LG 56: Finalize work

LG Code: EIS WSS4 M04LO4-LG-56

Page 1 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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This learning guide is developed to provide you the necessary information regarding the following **content coverage** and topics:

- Identifying samples to be collected
- Compiling types, location and time of samples
- Maintaining integrity of samples
- Assessing and reporting abnormal characteristics
- Transcribing label, register and sample preparation records
- Identifying samples, test methods, equipments and safety

This guide will also assist you to attain the learning outcome stated in the cover page.

Specifically, **upon completion of this Learning Guide, you will be able to:**

- Identify samples to be collected
- Collect samples to ensure that types, locations and times complied with requirements.
- Maintain Integrity of samples during sampling
- Asses and report abnormal characteristics; and apply incident management principles.
- Label, register and record laboratory samples preparation ensuring that the required information is transcribed accurately and legibly.
- Identify samples to be tested, identify appropriate test method, equipment and safety requirements

Learning Instructions:

1. Read the specific objectives of this Learning Guide.
2. Follow the instructions described below
3. Read the information written in the “Information Sheets 1-6”. Try to understand what are being discussed.
4. Accomplish the “Self-checks 1,2,3,4, 5 and 6” in each information sheets on pages 6,11, 16,19,21and 23.

5. Ask from your teacher the key to correction (key answers) or you can request your teacher to correct your work. (You are to get the key answer only after you finished answering the Self-checks).

If you earned a satisfactory evaluation proceed to the next Learning Guide.

Page 3 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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Information Sheet-1

Identifying Samples to be Collected

1.1. Introduction

Water samples are collected and analyzed to determine the chemical, physical and bacteriological composition of a water body, and its suitability for domestic, industrial, and agricultural uses. Water samples are also analyzed to aid in understanding geochemical and hydrologic relationships in natural systems and to evaluate the influence of man's activities on these systems.

In order to get the most accurate results, samples need to be collected properly. While every lab may have their own set of sampling standard operating procedures (SOP), there are some common best practices for collecting samples.

Samples should be collected using equipment and procedures appropriate to the matrix, parameters and sampling objective. The volume of the sample collected must be sufficient to perform the analysis requested. Samples must be stored in the proper types of containers and preserved in a manner appropriate to the analysis to be performed.

1.2. Sample Collection

Before any sampling is undertaken it is important to discuss the reason for sampling with the laboratory that will analyze the sample. Usually the sample containers are specific to the tests. Therefore, it is important to get the right container for the test being done. The correct sample container for each type of analysis will be provided by the laboratory. For microbiological tests, the sample bottle/container needs to be a wide-mouth vessel of at least 200 mL capacity and with a screw cap. For general chemical tests, a sample of 1–2 litres is usually required.

Samples for testing for micro-organisms in a drinking-water supply must be collected in containers that are pre-sterilized so that there are no living organisms in the bottles. Then any 'micro-organisms' found in the samples must have come from the water being tested (or from contamination while the sample was being taken). For some tests the sample bottles may contain substances required to stabilize or preserve the sample. Sample bottles for microbiological tests may contain sodium thiosulphate to neutralize any chlorine in the sample. For tests for some chemical contaminants, preservatives are included in the sample container. These will be added by the

laboratory. Whatever is being sampled for, the act of collection must be done with utmost care to reduce the possibility of contaminating the sample.

1.3. Sample Collection Bottles and Materials

Before collecting samples, make sure you have all the proper bottles, field equipment and preservatives, such as ice. There is nothing worse than being unprepared in the field, so plan ahead as much as possible. It's best to obtain sampling bottles from the lab running the analysis, as some bottle sizes and preservatives used can differ slightly. The methods that will be followed will determine the type of bottles used.

For example, samples for metals' analyses are usually collected in plastic bottles, while analyses for volatile organics and pesticides are collected in glass containers. Bottles used to collect samples for bacteria should be sterilized.

The size of the container is important to ensure you have enough sample to run the analysis needed. Container size may also be affected by regulation, as larger samples may be needed to obtain lower detection levels

Containers for the transportation of samples are best provided by the laboratory. This ensures that large enough samples are obtained for the planned analyses and that sample bottles have been properly prepared, including the addition of stabilizing preservatives when necessary. It is essential to have enough containers to hold the samples collected during a sampling expedition. Sample containers should be used only for water samples and never for the storage of chemicals or other liquids.

Glass containers are commonly used and are appropriate for samples for many analyses, but plastic containers are preferred for samples intended for certain chemical analyses or for biota or sediments. Plastic has the obvious advantage that it is less likely to break than glass.

Page 5 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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Self-Check -1

Written Test

Direction: Choose the best answer for the following questions. Use the Answer sheet provided in the next page: Each question worth one point.

1. The purpose of water sampling is:
 - A. To determine water for irrigational and recreational purposes
 - B. To determine the chemical, physical and bacteriological composition of a water body
 - C. To analyze the chemical composition of water
 - D. All
2. Which one is correct about before sampling?
 - A. Determine the sampling locations
 - B. Determine the numbers and types of samples to be collected
 - C. Apply Safety procedures
 - D. All of the above
3. Which one of these **is not** a core activity while we carry out sampling?
 - A. Use the necessary equipments for sample taking and transportation
 - B. Label sample bottles before sampling
 - C. Takes samples carefully and systematically
 - D. Use any type of sample containers for collection of samples
4. One advantages of plastic containers than that of glass containers is that-----
 - A. They are easy to handle and manage
 - B. They are not expensive
 - C. They are less likely to break than glass
 - D. They are easily cleanable

Note: Satisfactory rating - 4 points

Unsatisfactory - below 4 points

You can ask you teacher for the copy of the correct answers.

Page 6 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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Information Sheet-2	Compiling Types, Location and Time of Sample
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2.1. Introduction: Sampling is the process of taking a portion of water for analysis or other testing, e.g. drinking water to check that it complies with relevant water quality standards, or river water to check for pollutants, or bathing water to check that it is safe for bathing, or intrusive water in a building to identify its source.

2.2. Types of water samples

- **Grab sample:** A grab sample is a discrete sample which is collected at a specific location at a certain point in time. If the environmental medium varies spatially or temporally, then a single grab sample is not representative and more samples need to be collected.
- **Composite sample:** A composite sample is made by thoroughly mixing several grab samples. The whole composite may be measured or random samples from the composites may be withdrawn and measured.

A composite sample may be made up of samples taken at different locations, or at different points in time. Composite samples represent an average of several measurements and no information about the variability among the original samples is obtained. In composite sampling, volumes of material from discrete samples are physically combined into a single homogeneous sample or a single collection device is used to collect samples from multiple locations.

2.3. Sampling Strategies

Sampling strategies are specific for the site being evaluated and should be in place prior to initiating sample collection. The following three primary categories of sampling strategies are discussed in this section:

- **Judgmental Sampling:** Judgmental sampling involves sampling at locations that have the greatest chance of detecting contamination based on the lines of evidence and best professional judgment. This strategy can help the Incident Command prioritize sampling. Sampling directed by the SAP (Sampling and analysis plan) will be conducted in a stepwise fashion, prioritizing and addressing data quality objectives as data are received and evaluated. As data becomes available, it may influence decision making and future priorities, which in turn may affect the SAP.

- Probabilistic Sampling:** To be able to make a general statement (inferences) about contamination in an area of interest, samples taken in that area must be randomized. Randomized sampling is one of the underlying assumptions that must hold true in order to reach a valid and defensible inference based on the results of that sample. To the degree that the sample was not random (biased in some way), the inference will be biased and decisions made based on the information will also be biased (flawed). The area of interest is made up of an almost infinite number of possible sampling locations.

To characterize the entire area of interest every possible sample location could be sampled; however, that would be costly in both time and resources. A limited number of randomly located samples can be taken to reduce cost and still be representative of the entire population of locations within that area of interest.

- Combined judgmental and probabilistic sampling:** Most sampling is really a combination of judgmental and probabilistic sampling. Sampling within an area of interest could be labeled as solely probabilistic; however, the area of interest is selected based on judgment. (i.e., what makes this area worth sampling must be based on some evidence that the area of interest was contaminated).

2.3. Samples Site Selection

Site selection of first importance in the selection of any water-quality sampling site is the extent to which the proposed location complements the overall program of which it is a part. Water-quality data-collection programs have historically developed to meet specific and often unique requirements of water utilization or development in a particular area.

Surface water: Surface water refers to water that is found at a source on the surface of the earth, such as a river, stream, dam or lake. It is important to remember that if a water sample is collected from one of these sources, the point where the sample is collected must be as near to the point of abstraction as possible.

A program designed to obtain proper samples from flowing water bodies must consider the need for the samples taken to represent the entire flow of the stream at that point and at that time. Sampling stations should not be established at locations where mixing is incomplete, or where significant differences in water composition.

Page 8 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
--------------	--	---	---------------------------------

A sample typical of the entire flow of the stream is usually necessary. For some purposes, however, only the composition of the water at a fixed intake point is desired. Where the water composition at such a fixed intake point differs significantly from that of the entire stream, this fact should be noted in order not to mislead users of the published water data. Each sampling station should be selected, so far as possible, to fit into a comprehensive network of sampling stations to cover a State or drainage basin.

- **Lakes and reservoirs:** The selection of sites for sampling lakes and reservoirs depends on the degree of accuracy required in determining the composition and on the degree of mixing. Water circulation and movement caused by wind, temperature changes, and currents between points of inflow and outflow cause mixing. Other factors such as irregularity in shape of the body of water, difference in composition of inflow and stored water, thermal stratification, evaporation, solution or evolution of gases at the air-water interface, and activity of aquatic plant and animal life all tend to bring about heterogeneous quality.
- **Ground water:** The chemical quality of ground water at the sampling point may vary in response to changes in rate of water movement, to pump age, or to differences in rate and chemical composition of recharge from precipitation and from the surrounding area. Although concentrations of dissolved constituents in ground water from any one weir may vary widely, sometimes several fold, in general the changes take place much slower than those commonly associated with surface water.

2.4. General Sampling Procedures

Follow the procedures described below to assist in the collection of an acceptable sample and to maintain the integrity of the sample after collection.

- Prepare a Sampling and Analysis Plan (SAP) which describes the sampling locations, numbers and types of samples to be collected, and the quality control requirements of the project.
- Check with the laboratory before collecting samples to ensure that sampling equipment, preservatives, and procedures for sample collection are acceptable. It is best to obtain sampling supplies directly from the laboratory performing the analyses. Gather all equipment and supplies necessary for the project.

Page 9 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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- The acids and bases used in preservation of many types of samples are dangerous and must be handled with care.
 - ✓ Always wear gloves and eye protection when handling preservatives.
 - ✓ When opening a preservative bottle, particularly a glass ampoule, break open the ampoule away from yourself and others.
 - ✓ Have acid/base neutralization supplies (baking soda) on hand in the event of a spill.
 - ✓ If acid spills on your skin or clothing, remove the contaminated clothing and rinse the area with water.
 - ✓ Do not apply baking soda (the heat of reaction can cause burns).
- Collect samples in an area free of excessive dust, rain, snow or other sources of contamination.
- Select a cold-water faucet for sampling which is free of contaminating devices such as screens, aeration devices, hoses, purification devices or swiveled faucets. Check the faucet to be sure it is clean. If the faucet is in a state of disrepair, select another sampling location.
- Collect samples from faucets which are high enough to put a bottle underneath, generally the bath tub or kitchen sink, without contacting the mouth of the container with the faucet.
- Open the faucet and thoroughly flush. Generally, 2 to 3 minutes will suffice, however longer times may be needed, especially in the case of lead distribution lines. Typically, the water temperature will stabilize which indicates flushing is completed. Once the lines are flushed, adjust the flow so it does not splash against the walls of the bathtub, sink or other surfaces.
- Follow the collection instructions provided for the analytes of interest.
- Wear eye protection and gloves if you are handling containers with acidic/basic preservatives and when you are collecting samples.

Page 10 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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Self-Check -2**Written Test**

Direction: Choose the best answer for the following questions. Use the Answer sheet provided in the next page: Each question worth one point.

1. Water sample which is made by thoroughly mixing several samples is ----
 - A. Grab sample
 - B. Composite sample
 - C. Integrated samples
 - D. B and C

2. Which one of these is a correct water sampling requirement?
 - A. Fill out the chain of custody form with the sample collection information.
 - B. Deliver or ship samples to the laboratory to ensure that holding times are met.
 - C. Return empty preservative containers to the laboratory for proper disposal.
 - D. All

3. A sampling strategy which involves sampling at locations that have the greatest chance of detecting contamination based on the lines of evidence is known as----
 - A. Combined judgmental C. Judgmental Sampling
 - Random Sampling D. Probabilistic Sampling

4. The sample collection process should be coordinated with the laboratory
 - A. True B. False

5. Types of samples sites in which samples are normally obtained from existing drilled wells, dug (shallow) wells or springs is referred to as:
 - A. Ground sites B. Surface sites C. Tap C. Reservoir sites

Note: Satisfactory rating – 3 points**Unsatisfactory - below 3 points**

You can ask you teacher for the copy of the correct answers

Information Sheet-3	Maintaining integrity of samples
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3.1. Introduction

Sample integrity is part of Quality Assurance Project Plans. All samples should be collected with enough time for analysis within the holding time of desired analysis. Sampling requires two very important processes to be completed correctly. The first is the sampling itself, and the second is the paperwork associated with each sample. We use the terms Representative and Traceable to describe the sampling process, getting both performed correctly means that our results will be as accurate as possible. The quality of the sample and reliability of the resulting data are directly correlated to the integrity and representativeness of the sample.

3.2. Requirements of Samples integrity

- **Representative** “A sample must have the same distribution of characteristics as the body from which it is drawn. Only then, can the sample be used to draw conclusions about the greater body”. This means that if the sample is taken wrong then our results will be wrong.
- **Traceable** Traceability refers to the completeness of the information about every step from the time the sample is taken until we deliver your report. If the samples you collect get mixed up at any stage in the process then the results we deliver could be wrong.

The integrity of samples collected in the field should be preserved in order to ensure that samples are not compromised prior to sample analysis in the analytical laboratory. Preservation requirements will depend on the pathogen being analyzed, the type of sample or sample matrix, and the analytical protocol that will be used.

Some aspects of sample transport are constant across nearly all situations. Samples should be transported to the laboratory as quickly as possible to minimize pathogen degradation. There are important considerations for maintaining sample integrity when shipping samples, including sample preservation and storage.

Sample integrity elements to consider include, and are not limited to the following:

- Temperature control
- Holding times and conditions
- Preservation chemicals
- Container compatibility
- Labeling
- Volume (loss)

**Self-Check -3****Written Test**

Direction: Write **TRUE** or **FALSE** for the following questions: Each question worth one point.

1. Traceability refers to the wholeness of the information about every step from the time the sample taken until you deliver your report.
2. In keeping the integrity of samples, we have to consider only temperature and labeling of samples
3. Sampling is an often-underestimated step in the analytical process.

Note: Satisfactory rating – 3 points

Unsatisfactory - below 3 points

You can ask you teacher for the copy of the correct answers



Information Sheet-4	Assessing and Reporting Abnormal Characteristics
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4.1. Introduction: A report is a document that presents information in an organized format for a specific audience and purpose. Although summaries of reports may be delivered orally, complete reports are almost always in the form of written documents. Reports communicate information which has been compiled as a result of research and analysis of data and of issues. Reports can cover a wide range of topics, but usually focus on transmitting information with a clear purpose, to a specific audience. The scope and style of reports varies widely.

4.2. The significance of the reports

- Reports present adequate information on various aspects of the characteristics and other qualities of water samples.
- All the skills and the knowledge of the professionals are communicated through reports.
- Reports help the top line in decision making.
- A rule and balanced report also help in problem solving.
- Reports communicate the planning, policies and other matters regarding an organization to the masses. News reports play the role of ombudsman and levy checks and balances on the establishment.

4.3. Standard Elements of Report

Reports use features such as tables, graphics, pictures, voice, or specialized vocabulary in order to persuade a specific audience to undertake an action or inform the reader of the subject at hand. Some common elements of written reports include headings to indicate topics and help the reader locate relevant information quickly, and visual elements such as charts, tables and figures, which are useful for breaking up large sections of text and making complex issues more accessible.

4.4. Types of Reports

- **Daily report:** Daily reporting may sound like an annoying routine but it's probably something you need. Daily reporting is a useful method for managing both your work and personal life. It helps you keep track of your time and makes sure you only focus on important things each day.

Page 14 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
---------------	--	---	---------------------------------

- **Monthly report** serves as an important tool for communicating your achievements and ongoing projects to your employer. The report creation process can also provide you and your team with accountability as you assess your most recent performance and productivity levels
- **Annual Report:** An annual report is a comprehensive report on a company's activities throughout the preceding year. Annual reports are intended to give shareholders and other interested people information about the company's activities and financial performance.

Page 15 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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Self-Check -4**Written Test**

Direction: Choose the best answer for the following questions. Use the Answer sheet provided in the next page: Each question worth one point.

1. A comprehensive type report on a company's activities throughout the preceding year is considered as:
A. Daily B. Annual C. Monthly D. Quarterly
2. Which one of these **is not** correct about reports?
A. Present adequate information
B. Help the top line in decision making
C. Facilitate communication with stakeholders
D. Provides merely employees information
3. Elements of a report which are useful for breaking up large sections of text and making complex issues more reachable to the readers are-----
A. Tables B. Charts C. Figures D. All of these

Note: Satisfactory rating – 3 points

Unsatisfactory - below 3 points

You can ask you teacher for the copy of the correct answers

Information Sheet-5	Transcribing Label, Register and Sample Preparation Records
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5.1.Introduction: Water sampling is the process of taking a portion of water for analysis or other testing, e.g. drinking water to check that it complies with relevant water quality standards, or river water to check for pollutants, or bathing water to check that it is safe for bathing, or intrusive water in a building to identify its .

5.2. Sample Labeling

Proper labeling prevents sample misidentification and ensures the responsibility and accountability of the collector. The sample container should be labeled properly, preferably by attaching an appropriately inscribed tag or label. Alternatively, the bottle can be labeled directly with a water-proof marker. Barcode labels are also available nowadays.

Information on the sample container or the tag should include at least:

- Sample code number (identifying location)
- Date and time of sampling
- Source and type of sample
- Pre-treatment or preservation carried out on the sample
- Any special notes for the analyst
- Sampler's name

5.3. Purpose of Sample Labeling

- Reduces risk of contamination
- Reduces injury to self and others
- Maintains clean and organized working environment
- Avoids loss or damage to valuable samples
- Maintains integrity of research being conducted

5.4. Samples Preparation

Sample preparation refers to the ways in which a sample is treated prior to its analyses. Preparation is a very important step in most analytical techniques, because the techniques are often not responsive to the analyte in its in-situ form, or the results are distorted by interfering species.

Page 17 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
---------------	--	---	---------------------------------

On first impression, sample preparation may seem the most routine aspect of an analytical protocol. However, it is critical that analysts realize and remember that a measurement is only as good as the sample preparation that has preceded it. If an aliquant taken for analysis does not represent the original sample accurately, the results of this analysis are questionable. One goal of laboratory sample preparation is to provide, without sample loss, representative aliquant that are free of laboratory contamination that will be used in the next steps of the protocol.

5.5 Methods of Sample preparation

- **Dilution** is the process of decreasing the concentration of a solute in a solution, usually simply by mixing with more solvent like adding more water to a solution. To dilute a solution means to add more solvent without the addition of more solute. The resulting solution is thoroughly mixed so as to ensure that all parts of the solution are identical.

- ✓ **Solute:** this is the substance that makes up the minority of the solution, or this is the part that is dissolved. In our example of salt water, the solute is the salt.

- ✓ **Solvent:** this is the substance that makes up the majority of the solution. This is the part where the solute is dissolved. In our example of salt water, the solvent is water.

For example, if there are 10 grams of salt (the solute) dissolved in 1 litre of water (the solvent), this solution has a certain salt concentration (molarity). If one adds 1 litre of water to this solution the salt concentration is reduced. The diluted solution still contains 10 grams of salt (0.171 moles of NaCl).

Mathematically this relationship can be

shown by the equation:

$$C_1 V_1 = C_2 V_2$$

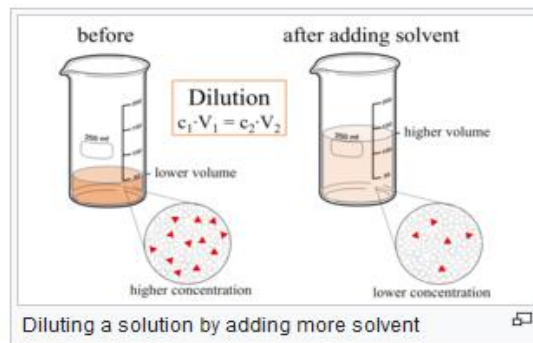
Where:

c_1 = initial concentration or molarity

c_2 = final concentration or molarity

V_1 = initial volume

V_2 = final volume



Page 18 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
---------------	--	---	---------------------------------

For Example, how much water must be added to 50ml of 1.2M HCl solution in order to produce 0.5M HCl solution?

Given: $C_1V_1 = 1.2M$; $C_2 = 0.5M$; $V_1 = 50ml$; Required: $V_2 = ?$

Using the relationship: $C_1V_1 = C_2V_2$

$$C_2 = \frac{1.2M \times 50ml}{0.5M} = 120ml.$$

120ml is the volume of the final solution. The amount of water to be added is then the difference between the volumes of the two solutions. So, the volume of water to be added is: $120 - 50 = 70ml$.

- **What is a 1% solution?**

A **one percent solution** is defined as **1 gram** of solute per **100 milliliters** final volume. For example, **1 gram** of sodium chloride, brought to a final volume of **100 ml** with distilled water, forms a **1% NaCl** solution.

- **How do I make a 1: 100 dilution?**

Example 1. For a 1:100 dilution, one part of the solution is mixed with 99 parts new solvent. Mixing **100 μ L** of a stock solution with **900 μ L** of water makes a 1:10 dilution. The final volume of the diluted sample is **1000 μ L (1 mL)**, and the concentration is **1/10** that of the original solution.

- **How to Dilute a Water Sample**

You may have to dilute your samples if there are high levels of contamination. Diluting your sample with distilled water will reduce the concentration of the contamination – making it easier to measure and obtain more accurate results.

The following table provides an example method to calculate sample dilutions for the membrane filtration method in microbiological testing. It is used to avoid too many coliform colonies growing making the sample ‘too numerous to count’ (TNTC).

Table 2. Example Sample Dilutions

Volume of sample	Volume of distilled water mixed	Volume filtered	Multiplication factor to obtain CFU per 100 ml
1 ml	99 ml	100 ml	100 x
5 ml	95 ml	100 ml	20 x
10 ml	90 ml	100 ml	10 x
50 ml	50 ml	100 ml	2 x
100 ml	0 ml	100 ml	1 x

Dilution Tips:

- Take a small sample volume with a sterile pipette
- Working with small sample volumes can reduce the accuracy of results. Moreover you need to be very careful on how you handle the sample
- If distilled water is not available, you can use boiled water (eg. clean rainwater, bottled water or spring water).
- For microbiological testing, never use chlorinated water to dilute your samples as chlorine residual will affect your test results (it will kill the bacteria you are trying to test for)

Note that: We use distilled or deionized (demineralized) water in the laboratory for dilution, preparation of reagent solutions, and final rinsing of glassware. Because; ordinary water is usually not pure. It may be contaminated by dissolved gases and by materials leached from the container in which it has been stored.

- **Neutralization/quenching:** Quenching a reaction to deactivate any un reacted reagents. Cooling the reaction mixture or adding an anti-solvent to induce precipitation, and collecting or removing the solids by filtration, decantation, or centrifugation and removal of solvents by evaporation.
 - **Decantation** is a process for the separation of mixtures of immiscible liquids or of a liquid and a solid mixture such as a suspension. An incomplete separation is witnessed during the separation of two immiscible liquids. The purpose may be either to produce a clean decant, or to remove undesired liquid from the precipitate (or other layers)
 - **Filtration:** Filtration is a process by which insoluble solids can be removed from a liquid by using a filter paper. The liquid that passes through is called the **filtrate** and the un -dissolved solid particles are called **residue**. Example: A mixture of chalk powder and water can be separated by this method
 - **Agitation:** It refers to the induced motion of a “homogenous” material in a specified way, usually in circulatory pattern inside some sort of container. It is a means whereby mixing of phases can be accomplished and by which mass and heat transfer can be enhanced between phases or with external surfaces. In its most general sense, the process of mixing is concerned with all combinations of phases like Gas, Liquid, solid. It is the heart of the chemical industry.
 - **Mixing:** It is the movement of fluids and solids to enhance a process result—is accomplished by means of an agitation source. For example, the sun is the agitation source for mixing in the earth's atmosphere. Similarly, an air compressor and/or a mechanical mixer is the agitation source in any municipal wastewater treatment plant to enhance the process results.
 - **Sub-sampling** is a procedure by which a small, representative sample is taken from a larger sample. Good sub-sampling technique becomes important when the large sample is not homogeneous.
 - **Sample digestion.** Acid digestion is a method of dissolving sample into solution, by adding acids and heating, until the complete decomposition of the matrix. Anywhere that you need to decompose a sample for release of the analyte, or for

Page 21 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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analysis of trace metals, acid digestion is much recommended. For Example: Adding HNO_3 converts metal ions into their nitrate salts, which are highly soluble.

5.5. Common Sample Preservatives

Sample Preservatives uses various preservation techniques to ensure the integrity of the test. Where acids are used the sampler must takes precautions to avoid injury to hands and eyes.

- **Sulphuric and Hydrochloric Acid**

Acid is added in small quantities in order to stop the microbial action that could remove the analyte we are looking for. The amount is small and the acid is dilute, but can still cause injury to your hands and eyes.

- **Sodium Hydroxide**

This is used in the cyanide bottles to make the sample alkaline so that cyanide is not released. The amount of used is small but can still cause injury to your hands and eyes.

- **Sodium Thiosulphate**

This chemical neutralizes the chlorine in potable water and swimming pool microbiology samples. This ensures that sterilizing effect of the chlorine does not continue during the trip to the laboratory.

- **Chilling**

Chilling a sample greatly reduces the microbial activity and is recommended for all samples taking longer than an hour to reach the laboratory. The recommended temperature is 10°C , and we have procedures in place to record the sample temperature for chilly bins arriving at the laboratory.

- **Freezing**

Freezing is a very effective way of preserving low-level nutrient samples as it almost completely stops microbial activity.

- **Others:** Some other specialized preservatives are used for tests such as Dissolved Oxygen and sulphide. These test bottles are delivered with detailed instructions

Page 22 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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Self-Check -5**Written Test**

Direction: Choose the best answer for the following questions. Use the Answer sheet provided in the next page: Each question worth one point.

1. The ways in which a sample is treated prior to its analyses;
 - A. Sample dilution
 - B. Sample preparation
 - C. Sample testing
 - D. Sample preservation
2. ----- is a method of dissolving sample into solution
 - A. Digestion
 - B. Freezing
 - C. Agitation
 - D. Sub-sampling
3. It is a means whereby mixing of phases can be accomplished is termed as:
 - A. Dilution
 - B. Agitation
 - C. Solution
 - D. Decantation
4. A chemical substance which neutralizes the chlorine in potable water and swimming pool of microbiology samples is known as-----
 - A. Sulphuric acid
 - B. Sodium sulphate
 - C. Sodium thiosulphate
 - D. Soda
5. ----- is a substance that makes up the majority of the solution in solution formation.
 - A. Solvent
 - B. Solute
 - C. Analyte
 - D. Titrate

Note: Satisfactory rating – 5 points Unsatisfactory - below 5 points

You can ask you teacher for the copy of the correct answers

Information Sheet-6	Identifying Test Methods, Equipment and Safety
----------------------------	---

6.1. Introduction: A test method is a method for a test in science or engineering, such as a physical test, chemical test, or bacteriological test. It is a definitive procedure that produces a test result. In order to ensure accurate and relevant test results, a test method should be "explicit, unambiguous, and experimentally feasible.", as well as effective and reproducible.

A test can be considered an observation or experiment that determines one or more characteristics of a given sample, product, process, or service. The purpose of testing involves a prior determination of expected observation and a comparison of that expectation to what one actually observes.

6.2. Test Methods

6.2.1. Colorimetric Methods: These tests are simple and easy to run—the basic concept is one of adding a specific reagent which reacts with the analyte being tested to create a color. The intensity of this color is then measured. You can visually compare the intensity of the color developed against a known standard.



Fig.1.Colorimeter

6.2.2. Gravimetric analysis describes a set of methods used in analytical chemistry for the quantitative determination of an analyte (the ion being analyzed) based on its mass. The principle of this type of analysis is that once an ion's mass has been determined as a unique compound, that known measurement can then be used to determine the same analyte's mass in a mixture, as long as the relative quantities of the other constituents are known.

6.2.3. Electrochemical Methods: Some parameters in water such as pH, Oxidation Reduction Potential (ORP) and Conductivity can be measured using an electrochemical meter. These are precise instruments which (in effect) measure the flow of electricity through a water sample. In addition, other electrochemical instruments can measure Dissolved Oxygen in the sample using membrane technology

6.2.4. Atomic Absorption Spectrometer (AAS) This method is used to analyze the presence of metals. Atomic Absorption Spectrometry (AAS) is based on the phenomenon that free atoms in the ground state can absorb light of a certain wavelength. Each element has its own specific absorption, meaning no other elements absorb this wavelength when light is passed through the atom in its vapour state. As this absorption of light depends on the concentration of atoms in the vapour, the concentration of the target element in the water sample can be determined.

6.2.5. Chromatography This is a separation method based on the affinity difference between two phases: stationary and mobile. A sample is injected into a column either packed or coated with the stationary phase, and separated by the mobile phase based on the difference in interaction (distribution or adsorption) between compounds and the stationary phases. Compounds with a low affinity for the stationary phase move more quickly through the column and remove earlier.

6.2.6. Titrations (Titrimetric): Titrations are simple chemical tests in which a solution or tablet of a known concentration is added slowly to a sample of unknown concentration. At a given point (called the end point), the color of the sample will rapidly change, such as from red to blue. An easy calculation will then allow determination of the concentration of the analyte.

- A typical titration begins with a beaker or Erlenmeyer flask containing a very precise amount of the analyte and a small amount of indicator (such as phenolphthalein) placed underneath a calibrated burette or chemistry pipetting syringe containing the titrant.

Small volumes of the titrant are then added to the analyte and indicator until the indicator changes color in reaction to the titrant saturation threshold, representing arrival at the endpoint of the titration. Depending on the endpoint desired, single drops or less than a single drop of the titrant can make the difference between a permanent and temporary change in the indicator. When the endpoint of the reaction is reached, the volume of reactant consumed is measured and used to calculate the concentration of analyte.

Page 25 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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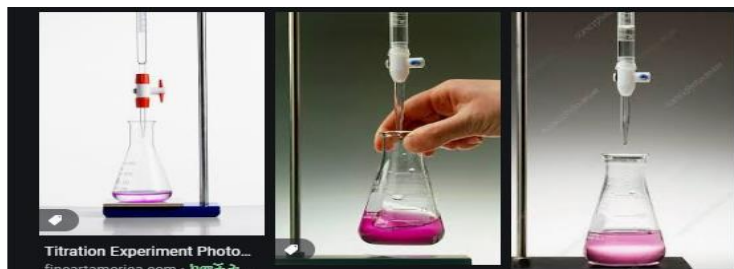


Fig 2. Photograph on Titration

❖ **Potentiometric Titration:** The end point of most titrations is detected by the use of visual indicator but the method can be inaccurate in very dilute or colored solutions. However under the same conditions, a potentiometric method for the detection of the equivalence point can yield accurate results without difficulty. The electrical apparatus required consists of a potentiometer or pH meter with a suitable indicator and reference electrode. The other apparatus consists of a burette, beaker and stirrer.

❖ **Volumetric Analysis**

Volumetric analysis is a form of quantitative analysis involving the measuring of volumes of reacting solutions; it involves the use of titrations. It's the point indicating the completion of the reaction. The volumes are measured by titration which completes the reaction between the solution and reagent. The volume and **concentration** of reagent which are used in the titration show the amount of reagent and solution.

6.3. Types of Solutions

- **Standard solution:** is a solution containing a precisely known concentration of an element or a substance. A known weight of solute is dissolved to make a specific volume. It is prepared using a standard substance, such as a primary standard. Standard solutions are used to determine the concentrations of other substances, such as solutions in titration. The concentrations of standard solutions are normally expressed in units of moles per liter.
- **A concentrated solution:** is one which contains a relatively large amount of solute per unit volume of solution.
- **A dilute solution:** is one which contains a relatively small amount of solute per unit volume of solution. The words "strong" and "weak" should not be used

Page 26 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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when referring to the concentration of a solution. Strong and weak are terms that are more properly used to describe the chemical activity of a substance.

6.4. Field Test Methods

There are different ways to test chemical contaminants in the field. The most popular are test strips, color comparators, colorimeters, and digital meters.

6.4.1. Test (Reagent) Strips

There are many different types of test strips available to measure different chemical contaminants. They are generally convenient and easy to use for technical and non-technical people; provide quick results; and are the cheapest way to do field testing.

Test strips typically have a plastic handle with a reagent area at one end. Typically, you dip the reagent area into a water sample, remove it, and compare the color of the reagent area with a color chart. Some test strips work by presence/absence of a color change at a threshold concentration.

Test strips are available to measure pH and a variety of chemicals including arsenic, chlorine and manganese.

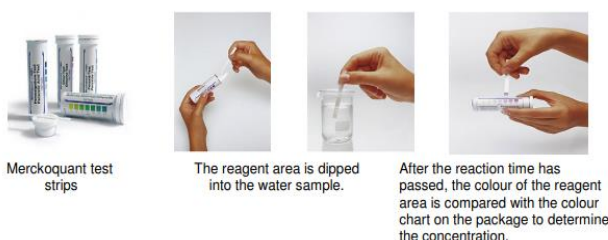


Fig 3. Test Strips

6.4.2. Color Disc Comparator

There are different types of color disc comparators that are available. The comparator is used in conjunction with a range of interchangeable color discs. These color discs are used to compare the color produced for each chemical test, against the standard test colors provided on the disc. Color discs are available for a range of chemical water testing parameters such as chlorine, fluoride, nitrate, iron and manganese. Color comparators can sometimes be more accurate than test strips, but they are more expensive, require more materials, and still require a visual determination of the chemical concentration.



Fig.4. Color comparator

6.4.3. Colorimeter and Photometer

Colorimeters and photometers use a light source to measure the chemical concentration in a water sample. Compared to test strips, they offer more accurate and repeatable results since the concentration is given as a digital reading.

As well, colorimeters and photometers can read a large variety of chemicals in a water sample as well as a wider numerical range within each parameter



Fig 5. Photo meter

6.4.4. Digital Meters

Some portable field kits include various digital meters to measure parameters like pH and EC. They are relatively easy to use and can provide more accurate measurements than other methods, such as test strips. The main disadvantages are the need for calibration and general fragility of electronic equipment.



Fig 6. Digital meter

6.5. Guidelines and Precautions for Sampling

- Collect a sample that conforms to the requirement of the sampling program and handle it carefully so that it does not deteriorate or get contaminated during its transport to the laboratory.

Page 28 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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- Before filling the container, rinse it two or three times with the water being collected.
- Representative samples of some sources can only be obtained by making composites of samples collected over a period of time or at a number of different sampling points.
- While collecting a sample from the distribution system, flush lines adequately, taking into consideration the diameter and length of the pipe to be flushed and the velocity of flow.
- Collect samples from tube-wells only after sufficient pumping (purging) to ensure that the sample represents the ground water source.
- When samples are to be collected from a river or stream, analytical results may vary with depth, flow, distance from the banks. In surface water bodies, water samples should preferably be collected at 0.2 times the depth of the water body from the top.
- Make detailed record of every sample collected (Identify each container and record information like date, time and exact location)

6.6. Safety measures in Laboratory

- All containers must be clearly labeled and read before opening. If dispensing into another container, put label along with warning.
- Minimal stocks not exceeding 500 ml of corrosive or flammable solvents only may be kept in work room. Keep rest of the volume/quantity in safe place.
- Glacial acetic acid must be regarded as a flammable solvent.
- Ether and low boiling point flammable liquids must not be kept in fridge.
- Large containers of corrosive or flammable liquids should never be put on high shelves or where they can be knocked down or fall. Also, never put liquids that react violently together closely. Never carry bottles by neck alone. Open bottles with care.
- When diluting concentrated sulphuric acid or other strong acids, it should be added to water in heat resistant vessel. Gloves and safety glasses should be used at such times.

Page 29 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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6.7. Avoidance of Hazards of Equipment

- Only trained staff should operate the equipment.
- Operating instructions should be available for each instrument.
- Check the autoclave filled with water to correct level before loading.
- If fire breaks out, nearby electrical equipment should immediately be switched off and disconnected. Take care to avoid live wires.
- When not in use, switch off and withdraw plug from socket.
- Avoid use of multi-adaptors. If necessary, to use they must be fitted with fuses.

6.8. Sampling Equipment and Supplies

Dedicated sampling equipment should be used if possible, to prevent cross-contamination. Sampling equipment should be clean and sterile whenever possible – and in good working condition.

Sampling containers should be sterile and kept in a clean environment. The sample container's inner portion should not be handled or touched by the sampler. All reusable containers must be cleaned properly and sterilized, and proven free of quantifiable target contaminant before use.

Samplers should use the necessary techniques to reduce sample contamination. After collection, samples should be placed in sealed containers to prevent contamination and to contain any sample leakage during storage and/or transport. Storage conditions, holding time (i.e., maximum time between collection and analysis), and transport procedures must also be considered

Page 30 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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Self-Check -6

Written Test

Direction: Choose the best answer for the following questions. Use the Answer sheet provided in the next page: Each question worth two points

1. In order to make a certain, truthful and relevant test results, a test method **should not** be: A. explicit B. ambiguous C. feasible D. Reproducible
2. A separation method/technique of substances based on the affinity differences between two stationary and mobile phases:
 - A. Titration B. Absorption C. Chromatography D. Colorimetric
3. An appropriate test method of measuring the pH, Oxidation Reduction Potential (ORP) and Conductivity of a sample of water is -----
 - A. Colorimetric B. Gravimetric C. Titration D. Electrometric
4. Which one of the following is a safety measure to be taken in a laboratory?
 - A. When diluting concentrated sulphuric acid or other strong acids add water to acids
 - B. When diluting concentrated sulphuric acid or other strong acids add acids to water
 - C. Carry bottles by their neck alone
 - D. All of the above
5. a type of solution containing a precisely known concentration of an element or a substance is known by-----
 - A. Standard solution B. concentrated solution C. Diluted solution A. None

Note: Satisfactory rating – 10 points

Unsatisfactory - below 10 points

You can ask you teacher for the copy of the correct answers

This learning guide is developed to provide you the necessary information regarding the following **content coverage** and topics:

- Setting up test equipments and reagents
- Conducting pre-use and safety checks
- Reporting unsafe or fault equipments
- Checking and calibrating equipments safety

This guide will also assist you to attain the learning outcome stated in the cover page.

Specifically, **upon completion of this Learning Guide, you will be able to:**

- Set up test equipment and/or reagents in accordance with the specified test method.
- Conduct pre-use and safety checks
- Identify faulty or unsafe equipment and report to appropriate personnel
- Check the calibration status of equipment and conduct calibration or report calibration requirements to appropriate personnel.

Learning Instructions:

1. Read the specific objectives of this Learning Guide.
2. Follow the instructions described below
3. Read the information written in the “Information Sheets 1- 3”. Try to understand what are being discussed.
4. Accomplish the “Self-checks 1,2, and 3” in each information sheets on pages 34,37 and 40.
5. Ask from your teacher the key to correction (key answers) or you can request your teacher to correct your work. (You are to get the key answer only after you finished answering the Self-checks).

Information Sheet-1	Setting Up Test Equipments and Reagents
----------------------------	--

1.1. Introduction: Reagents are chemical testing compounds that identify presence of chlorine, pH, alkalinity, turbidity and other metrics. Reagents must always be prepared and standardized with care and appropriate technique against reliable primary standards. They must be re-standardized or prepared fresh as often as required by their stability. Stock and working standard solutions must be checked regularly for signs of deterioration, e.g., discoloration, formation of precipitates, and concentration. Standard solutions should be properly labeled as to compound, concentration, solvent, date and prepared.



Fig 7. Laboratory equipments and reagents

1.2. Setting up reagents and chemicals

Chemical reagents, solvents, and gases are available in a wide variety of grades of purity, ranging from technical grade to various ultrapure grades. The purity of these materials required in analytical chemistry varies with the type of analysis. The parameter being measures and the sensitivity and specificity of the detection system are important factors in determining the purity of the reagents required.

Reagents must always be prepared and standardized with the utmost of care and technique against reliable primary standards. They must be re standardized or prepared fresh as often as required by their stability.

1.3. Storing and maintaining quality of reagents and solvents

Having performed the tasks of selecting, preparing, and verifying the suitability of reagents, solvents, and gases, the analyst must properly store them to prevent contamination and deterioration prior to their use. Borosilicate glass bottles with ground-glass stoppers are recommended for most standard solutions and solvents. Plastic containers such as polyethylene are recommended for alkaline solutions.

Self-Check -1**Written Test**

Direction: Write/ Say **TRUE** or **FALSE** for the following questions.

Each question worth two points

1. Reagents must always be prepared and standardized with care.
2. Stock and working standard solutions must be checked regularly for signs of deterioration.
3. Borosilicate glass bottles with ground-glass stoppers are recommended for most standard solutions and solvents
4. Plastic containers are used for reagents or solvents intended for organic analyses.
5. Solutions are chemical testing compounds that identify presence of chemicals in a given sample

Note: Satisfactory rating – 10 points

Unsatisfactory - below 10 points

You can ask you teacher for the copy of the correct answers

Information Sheet-2	Reporting Unsafe or Fault Equipments
----------------------------	---

2.1. Introduction: Report is a self-explanatory statement of facts relating to a specific subject and serves the purpose of providing information for decision making and follow up actions. It is a systematic presentation of ascertained facts about a specific event / subject. Report is a summary of findings and recommendations about a particular matter / problem. Report is for the guidance of higher authorities including company executives and directors.

Working safely with hazardous chemicals requires proper use of laboratory equipment. Maintenance and regular inspection of laboratory equipment are essential parts of this activity. Many of the accidents that occur in the laboratory can be attributed to improper use or maintenance of laboratory equipment. This chapter discusses prudent practices for handling equipment used frequently in laboratories.

2.2. Laboratory Faulty Equipment

Faulty equipment is any type of mechanical or electronic equipment that is dangerous or unsafe to use. This usually refers to equipment used in the workforce for producing or manufacturing products. Faulty equipment is responsible for several types of on the job injuries each year. Equipment can be faulty due to:

- Improper maintenance
- Errors with regards to warnings
- Incorrect instruction manuals
- Broken or old parts
- Manufacturing defects

2.3. Injuries Caused by Faulty Equipment

Faulty equipment can cause various types of injuries. These may include:

- Parts of the equipment falling onto the user
- Burns and abrasions
- Injuries to limbs or other body parts
- Toxic injuries (such as an untimely spray of chemicals or toxic materials)

Most workplaces include mechanisms to help identify machines that might be on the way to becoming dangerous. They also usually include procedures for dealing with

injuries. However, injuries can still occur due to mishaps, negligence, or other similar circumstances.

2.4. Reporting Unsafe or Broken Equipment

Implementing a simple reporting procedure in a workplace will help to obtain important information about health and safety issues in the workplace, identify problems when they arise, and address them.

Safety reporting procedures make it simpler for you and your workers to manage safety issues and prevent recurrences of incidents and injuries. They can help you understand why incidents occurred, assist you to make decisions and set priorities, and allow you to analyze trends in the workplace. These procedures may also help to identify additional hazards and risks, and will guide you in developing new risk controls.

Page 36 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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Self-Check -2**Written Test**

Direction: Choose the best answer for the following questions. Use the Answer sheet provided in the next page: Each question worth **two** points

1. A summary of findings and recommendations about a particular matter / problem is referred to as-----
A. Data B. Report C. Record D. Table
2. Implementing a simple reporting procedure in a workplace will:
A. Help to obtain important information
B. Identify problems when they arise
C. To address problems and address
D. All of these
3. One of the following is wrong about Safety reporting procedures in a work place?
A. Prevent recurrences of incidents and injuries.
B. Help you to understand why incidents occurred,
C. Hinder you to make decisions and set priorities
D. Allow you to analyze trends in the workplace

Note: Satisfactory rating – 6 points**Unsatisfactory - below 6 points**

You can ask you teacher for the copy of the correct answers

Information Sheet-3	Checking and Calibrating Equipments Safety
----------------------------	---

3.1. Introduction: Is the process of determining the accuracy of an equipment or instrument. The process involves obtaining a reading from the instrument and measuring its variation from the reading obtained from a standard instrument. Calibration of an instrument also involves adjusting its precision and accuracy so that its readings come in accordance with the established standard. **It is used to** compare the inspection, measuring, and test instruments to a recognized reference standard of known certified accuracy and precision, noting the difference and adjusting the instrument, where possible, to agree with the standard.

3.2. Calibration of equipment

Calibration of equipment is not just desirable; it is rather a necessity. All measuring instruments, whether they are used in factories, laboratories or at home, need to be calibrated on a periodic basis to ensure they are offering accurate results. However, in factories and laboratories, measurement results are usually of a critical value. Deviation from accuracy can not only affect productivity but also cause threat to the life of workers. Hence, it is imperative that instrument calibration be carried out carefully and at regular intervals.



Fig 8. Calibrating of instrument

3.3. What happens if equipment is not calibrated?

Calibration of equipment needs to be carried out on a regular basis. This is because instruments tend to deviate owing to hard operating conditions, mechanical shocks or exposure to extreme temperature or pressure. Frequency of calibration would depend on the tolerance level. When the objective of the measurement is critical calibration would need to be carried out more frequently and with great accuracy. To assure accuracy in instrument calibration, it is vital to ensure that each component of the measuring instrument is conforming to its specified standard. Regular equipment calibration carried out in a set format helps you obtain valid data and operate in a safe working environment.

3.4. Types of Calibration

Instrument calibration can be carried out on different types of instruments across sectors. Discussed here are some of most frequently performed types of calibration services.

3.4.1. Pressure Calibration: This is one of the most frequently performed types of equipment calibration. Under pressure calibration service gas and hydraulic pressure are typically measured across a variety of sectors. Various types of pressure balances and calibrators along with a number of pressure gages are used for carrying out pressure calibration.

3.4.2. Temperature Calibration: Temperature calibration is carried out in all processes where temperature readings play a critical role. Temperature calibration is carried out in a controlled environment. State-of-the-art electrical and mechanical thermometers are available that can help in the process of temperature calibration.

- **Flow Calibration:** Flow calibration services needs to be carried out on a routine basis for flow meters that check product or feedstock quality and quantity, fuel/energy quantity or function in a critical process.

Page 39 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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Self-Check -3**Written Test**

Direction: Choose the best answer for the following questions. Each question worth two points.

1. ----- is the process of determining the accuracy of an equipment or instrument.
A. Calibration B. Disinfection C. Titration D. Sterilization
2. Calibration of equipment needs to be carried out during its maintenance.
A. True B. False
3. Frequency of calibration would depend on the tolerance level.
A. True B. False
4. What will happen if equipment is not calibrated?
A. It will give reliable data
B. It yields information which deviation from accuracy
C. It can affect productivity
D. It may cause threat to the life of workers

Note: Satisfactory rating – 8 points

Unsatisfactory - below 8 points

You can ask you teacher for the copy of the correct answers

Page 40 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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Instruction Sheet-3	Learning Guide# 55: Perform Tests on Samples
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This learning guide is developed to provide you the necessary information regarding the following **content coverage** and topics:

- Preserving integrity of sample and eliminating cross contamination
- Preparing, weighing/measuring samples
- Performing tests following test methods
- Performing calculation
- Shutting down equipments
- Storing unused reagents and disposing wastes
- Cleaning and storing equipments

This guide will also assist you to attain the learning outcome stated in the cover page.

Specifically, **upon completion of this Learning Guide, you will be able to:**

- Identify samples to be collected
- Collect samples to ensure that types, locations and times complied with requirements.
- Maintain Integrity of samples during sampling
- Asses and report abnormal characteristics; and apply incident management principles.
- Label, register and record laboratory samples preparation ensuring that the required information is transcribed accurately and legibly.
- Identify samples to be tested, identify appropriate test method, equipment and safety requirements

Learning Instructions:

-
1. Read the specific objectives of this Learning Guide.
 2. Follow the instructions described below
 3. Read the information written in the “Information Sheets 1- 3”. Try to understand what are being discussed.

4. Accomplish the “Self-checks 1,2, and 3” in each information sheets on pages 46,58 and 66.
5. Ask from your teacher the key to correction (key answers) or you can request your teacher to correct your work. (You are to get the key answer only after you finished answering the Self-checks).
6. After You accomplish all self-check tests, ensure you have a formative assessment and get a satisfactory result; then proceed to the next LG.

Page 42 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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Information Sheet-1	Preserving Integrity of sample and Eliminate Cross Contamination
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1.1. Introduction: Starting from the sampling up to the time of analysis, the content of water sample can be altered due to the chemical, physical, and biological reactions it undergoes. All samples must be cooled to 4°C from the time of collection until analysis. When a preservative other than cooling is used, the preservative is generally added after the sample is collected, unless the sample container has been pre-preserved by the laboratory. If necessary, the pH must be adjusted to the appropriate level and checked with pH paper in a manner which will not contaminate the sample.

1.2. Sample Preservation Methods

1.2.1. Chemical addition: It is the most convenient preservative is a chemical which can added to a sample bottle prior to sampling. When the sample is added, the preservative disperses immediately, stabilizing the parameters of concern for long periods of time. When the preservative added interferes with other parameters being measured, additional samples for those parameters must be collected.

For example, concentrated nitric acid added for preservation of some of the metals would interfere with BOD, so an additional sample must be collected for BOD.

A common preservative used is sodium thiosulfate, a dechlorinating agent, used to stop any chlorine reactions. Sodium thiosulfate is used for bacteria analysis and volatile organics. Many laboratories will provide pre-preserved bottles wherever possible, but some analysis requires preservation in the field. Volatile organics should be further preserved in the field using hydrochloric acid, which is used to reduce any microbial activity that could reduce the contaminants while in transit to the lab.

1.2.2. pH control: pH control to preserve the sample is dependent upon chemical addition. For example; to keep metal ions in a dissolve state concentrated nitric acid is added to lower the Ph to less than 2.

1.2.3. Refrigeration: Refrigeration is a common method used in field work and has determinable effect on sample composition. Although it does not maintain integrity for all parameters, it does not interfere with any analytical methods.

1.2.4. Freezing: Freezing has been the subject of many preservation studies. It is felt by some that freezing would be a method for increasing the holding time and allowing collection of a single sample for analysis. The residue solid component (filterable and non-filterable) of the sample change with freezing and thawing.

The most common type of preservation is temperature; most analysis requires samples to be kept cool, arriving at the lab between four and six degrees Celsius.

The cooler temperature helps contaminants from breaking down during transit.

1.2.5. Holding time is another important consideration — this refers to the amount of time from collection to when the analysis begins. Regulatory samples need to meet these holding times and recognized holding times may vary from country to country. For example, most countries will recognize coliform bacteria testing as a 30-hour holding time, while some others require a stricter six hours. Keep in mind holding times when planning what time samples should be collected.

1.3. Preventing of Sample Contamination

Contamination of the sample can occur from the containers or exposure to the environment. The containers used for storage and handling are the main source of contamination. Glass containers are suitable for most samples. A variety of other materials are also available. The use of a specific material must be proven to not generate contamination,

The manner in which samples are collected has a direct impact on the quality of analytical findings. To minimize the risks of sample contamination by the sample collector and to ensure sample integrity, basic precautions must be taken to obtain a representative sample. Samples can become contaminated through careless sampling techniques. It is therefore the responsibility of the sample collector or of an artificial reservoir manager to ensure the quality of sample collection, preservation and suitable transportation of samples that are sent to a laboratory.

1.4. Precautions to Prevent Sample Contamination

- Closely follow the instructions provided by the manufacturer of the chemical test kits used at the sampling site;

Page 44 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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- Always use contaminant-free containers and where possible, prepare containers and testing equipment at sampling sites;
- Keep an accurate record of each sample collected using the correct form;
- Always use devices or instruments that have been calibrated at the required frequency (thermometer and ph-meter);
- Reagents that are used for analysis must be kept in a clean, dry, well-ventilated and dark location until use;
- Always take measurements with reagents in a suitable location. Avoid leaving reagents in prolonged sun exposure;
- Seal reagent bottles correctly after use;
- never place wet fingers on reagent bottles; this may lead to inaccurate results;
- Verify product expiry dates. Once the date indicated is past, you cannot be certain that results are reliable;
- Discard expired products, in accordance with regulations in effect.

Page 45 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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Self-Check -1	Written Test
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Direction: Choose the best answer for the following questions. Use the Answer sheet provided in the next page: Each question worth one point.

1. The main source of sample contamination comes from:
 - A. The manner in which samples are collected
 - B. The preservatives which are used
 - C. The containers used for storage and handling
 - D. All

2. Always use devices or instruments that have been calibrated at the required frequency.
 - A. True
 - B. False

3. The way in which samples are collected has a direct impact on the quality of analytical findings.
 - A. True
 - B. False

4. Always use instruments that have not been calibrated at the required frequency.
 - A. True
 - B. False

Note: Satisfactory rating – 8 points	Unsatisfactory - below 8 points
---	--

You can ask you teacher for the copy of the correct answers

Information Sheet-2	Preparing, Weighing/Measuring samples
----------------------------	--

2.1. Introduction:

Samples should be collected and analyzed by qualified individuals using the appropriate equipment and procedures. Sample preparation helps ensure that accurate and reproducible results are produced across a wide variety of water sample matrices

2.2. Sample Preparation

Sample preparation refers to the ways in which a sample is treated prior to its analyses. Preparation is a very important step in most analytical techniques, because the techniques are often not responsive to the analyte in its in-situ form, or the results are distorted by interfering species. Many samples will require some preparation before being sent to the laboratory for testing, particularly if their large sizes makes them difficult to handle or because they require special protection.

Information Sheet-3	Performing Tests Following Test Methods
----------------------------	--

3.1. Introduction: Water quality refers to the chemical, physical, biological, and radiological characteristics of water. It is a measure of the condition of water relative to the requirements of one or more biotic species, or to any human need or purpose. It is most frequently used by reference to a set of standards against which compliance, generally achieved through treatment of the water, can be assessed.

Water quality testing can be carried out in a laboratory. Performing water quality test requires facilities, trained technicians, equipment and other supporting materials.

When you pour yourself a glass of water to drink, it may appear clean, taste good, and you may expect it to be safe. However, many sources of water we assume are safe may contain dissolved minerals, organic compounds or even live organisms at harmful concentrations. Contaminated water used for drinking and cooking may affect your health. Also, high concentrations of certain minerals in your water can result in quality issues such as unpleasant taste and odors or staining of bathroom fixtures and/or laundry.

3.2. Performing Water Quality Tests

3.2.1. Color: Color in drinking water may be due to the presence of colored organic substances and certain metals such as iron, manganese and copper. This color is considered as "**apparent color**" as it is seen in the presence of suspended matter, whereas "**true color**" is derived only from dissolved inorganic and organic matters. Samples can be centrifuged and/or filtered to remove turbidity in order to measure true color.

- ✓ **True color of water** is measured after the water sample has been filtered with a 0.45-micron filter paper.
- ✓ **Apparent color:** It is what one actually sees; it is the **color** resulting from the combined effect of true **color** and any particulate matter, or turbidity. In turbid waters,

Page 48 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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the true **color** is substantially less than the **apparent color**. It is measured without filtering the water sample.

Color can be measured spectrophotometrically or using a visual comparator. In both cases, the standard unit of measurement is the Hazen unit (HU).

Levels of color above 15 TCU (True color Unit) can be detected in a glass of water by most people, although it generally does not pose a health threat. Color of water becomes noticeable to consumers at levels greater than the aesthetic objective of 5 true color units (TCU - platinum cobalt scale).

Table 3. Reporting result of color of samples

S. No.	Color Units/ Hazen units	Record to nearest
1	1 to 50	1
2	51 to 100	5
3	101 to 250	10
4	251 to 500	20

- i. **Odour and Taste:** Although taste and odour are not parameters of health concern, they are perhaps the most important characteristics of drinking water from the point of view of the user. It is next to impossible to convince people that water is safe to drink if it either tastes or smells bad. Bad taste and odour may cause people to reject the water in favor of another.

3.2.3. Alkalinity: Alkalinity is a measure of the acid-neutralizing capacity of dissolved substances in water and equals the amount of strong acid required to lower the solution from initial pH to about 4.5. Many materials may contribute to the alkalinity of water. For most practical purposes, it is due primarily to presence of salts of weak acids (mainly bicarbonate and carbonate) and hydroxide (at high pH).

The alkalinity of water is its quantitative capacity to neutralize acids. The three major forms of alkalinity ranked in order of their association with high pH values are:

- ✓ hydroxide alkalinity, [OH]

- ✓ carbonate alkalinity, $[CO_3]$ and
- ✓ bicarbonate alkalinity $[HCO_3]$

Alkalinity of waters is measured by means of titration with a standard solution of a strong acid (usually H_2SO_4) to designated pHs, and is reported in terms of equivalent $CaCO_3$.

3.2.4. Free Ammonia: Ammonia is a colorless, gaseous compound with a sharp distinctive odor. It is highly soluble in water where it exists in a molecular form associated with water and in an ionized form as NH_4^+ . The extent of association or ionization is dependent on the temperature and pH. It may also be toxic to aquatic life.. The extent of toxicity is dependent upon species and extent of dissociation. Ammonia may occur in water as a product of anaerobic decomposition of nitrogen containing compounds or from waste streams containing ammonia.

Free ammonia is that which has resulted naturally from the decay of organic matter contained in the water, and, other things being equal, shows how extensively such decomposition is going on. It is easily collected by distillation

3.2.5. Fluoride: The fluoride level in drinking water is an important parameter and has to be controlled to prevent dental and skeletal fluorosis. Almost 200 million people from 25 countries of the world are at risk of health problems for high concentration of fluoride in their drinking water.

Drinking water in some parts of Ethiopia particularly in the Rift valleys region is known to contain high levels of fluoride, which can cause dental mottling and, at high concentrations, crippling skeletal deformations.

There are few chemicals for which the contribution from drinking water to overall intake is an important factor in preventing disease. One example is the effect of fluoride in drinking water in protecting against dental caries. As per the WHO, the guideline value for fluoride content in drinking water is 1.5 mg/L (1500 μ g/L). Fluoride in drinking water can be tested using spectrophotometer and photometer.

Page 50 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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3.2.6. Iron: Iron can be naturally found in groundwater and some surface water (such as creeks, rivers and some shallow dug wells). There are areas of the world that have naturally high amounts of iron in their groundwater. Iron can also be found in drinking water that is passed through rusty steel or cast-iron pipes.

Iron can come in two forms in water: dissolved and suspended. If groundwater comes from a deep tube well, the iron may be dissolved and not visible. However, once the iron is exposed to air, it usually turns the water black or orange color. If surface water has iron in it, it will be a red-orange color from the iron that is suspended in the water.

Drinking water with high concentrations of iron will not make people sick. Iron, however, can change the color of water and it may cause people to not use it and choose another, possibly contaminated, water source instead.

In fact, iron is essential for good health because it transports oxygen in your blood.

Iron levels above 0.3 mg/L can stain water pipes and clothes during washing.

3.2.7. Manganese: Manganese can be naturally found in groundwater and surface water, and it usually occurs with iron. However, human activities may also be responsible for manganese contamination in water in some areas.

Manganese can come in two forms in water: dissolved and suspended. If groundwater comes from a deep tube well, the manganese may be dissolved and not visible. In surface water, manganese can be dissolved or suspended. Water with high levels of suspended manganese usually has a black color or black flakes in it.

People need small amounts of manganese to keep healthy and food is the major source for people. However, too little or too much manganese can cause adverse health effects. High levels of manganese, however, can turn water a black color and it may cause people to not use it and choose another, possibly contaminated, water source instead.

The method used to test water for iron and manganese depends on the form of the element. If water is clear when first drawn but red or black particles appear after the water sits in a glass, dissolved (ferrous) iron/manganese is present. If the water has a

Page 51 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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red tint with particles so small, they cannot be detected nor do they settle out after a time, colloidal (ferric) iron is the problem.

3.2.8. Aluminum: The concentration of aluminum in natural (drinking) waters can vary significantly depending on various physicochemical and mineralogical factors. Aluminum levels in drinking-water vary according to the levels found in the source water and whether aluminum coagulants are used during water treatment.

Aluminum may be removed from water by means of ion exchange or coagulation/flocculation. Aluminum salts are applied in water treatment for precipitation reactions. Adding aluminum sulphate and lime to water causes aluminum hydroxide formation, which leads to settling of pollutants.

3.2.9. Hardness: Water hardness is the traditional measure of the capacity of water to react with soap, hard water requiring considerably more soap to produce lather. Hard water often produces a noticeable deposit of precipitate (e.g. insoluble metals, soaps or salts) in containers, including “bathtub ring”. It is not caused by a single substance but by a variety of dissolved polyvalent metallic ions, predominantly calcium and magnesium cations, although other cations (e.g. aluminum, barium, iron, manganese, strontium and zinc) also contribute.

Hardness is most commonly expressed as milligrams of calcium carbonate equivalent per liter. Water containing calcium carbonate at concentrations below 60 mg/l is generally considered as soft; 60–120 mg/l, moderately hard; 120–180 mg/l, hard; and more than 180 mg/l, very hard).

3.2.10. Jar Testing: The jar test attempts to duplicate in the laboratory what is occurring in the plant in relationship to detention times, mixing and settling conditions. By watching the jar test floc form and settle, the operator can get a good idea of what should happen in the plant for that chemical dose. The jar test should be used as an indication of what to expect in the water treatment plant. By closely watching the floc form in the flocculators and settle out in the sedimentation basin of the plant, the operator can also get a good indication of whether the best coagulant dosage has been

Page 52 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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chosen. It should be realized that it is almost impossible to duplicate in the jar test exactly the flow conditions that are occurring in the treatment plant.

Also, by observing the performance of the filters and by looking at the laboratory test results, the operator will gain additional information that will help make the necessary adjustments to the actual chemical feed rates.

- **Jar Test Results Evaluation**

Several factors are important in evaluating jar test results. These include:

- ✓ Rate of floc formation;
- ✓ Type of floc particles;
- ✓ Clarity of water between floc particles;
- ✓ Size of floc;
- ✓ Amount of floc formed;
- ✓ Floc settling rate;
- ✓ Clarity of water above the settled floc

3.2.11. Chemical Oxygen Demand (COD): It is an indicative measure of the amount of oxygen that can be consumed by reactions in a measured solution. It is commonly expressed in mass of oxygen consumed over volume of solution which in SI units is milligrams per liter (mg/L). A COD test can be used to easily quantify the amount of organics in water. The most common application of COD is in quantifying the amount of oxidizable pollutants found in surface water (e.g. lakes and rivers) or wastewater. COD is useful in terms of water quality by providing a metric to determine the effect an effluent will have on the receiving body, much like biochemical oxygen demand (BOD).

3.2.12. Biochemical oxygen demand (BOD) is the amount of dissolved oxygen needed (i.e. demanded) by aerobic biological organisms to break down organic material present in a given water sample at certain temperature over a specific time period. The BOD value is most commonly expressed in milligrams of oxygen consumed per liter of sample during 5 days of incubation at 20 °C and is often used as a surrogate of the degree of organic pollution of water.

Page 53 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
---------------	--	---	---------------------------------

BOD reduction is used as a gauge of the effectiveness of wastewater treatment plants. BOD of wastewater effluents is used to indicate the short-term impact on the oxygen levels of the receiving water.

BOD analysis is similar in function to chemical oxygen demand (COD) analysis, in that both measure the amount of organic compounds in water. However, COD analysis is less specific, since it measures everything that can be chemically oxidized, rather than just levels of biologically oxidized organic matter.

3.2.13. Total Organic Carbon (TOC): It is the amount of carbon found in an organic compound and is often used as a non-specific indicator of water quality or cleanliness of pharmaceutical manufacturing equipment. TOC may also refer to the amount of organic carbon in soil, or in a geological formation, particularly the source rock for a petroleum play; 2% is a rough minimum.

TOC in source waters comes from decaying natural organic matter (Humic acid, fulvic acid, amines, and urea) as well as synthetic sources

A typical analysis for total carbon (TC) measures both the total organic carbon (TOC) present and the complementing total inorganic carbon (TIC), the latter representing the amount of non-organic carbon, like carbon in carbonate minerals. Subtracting the inorganic carbon from the total carbon yields TOC. Another common variant of TOC analysis involves removing the TIC portion first and then measuring the leftover carbon.

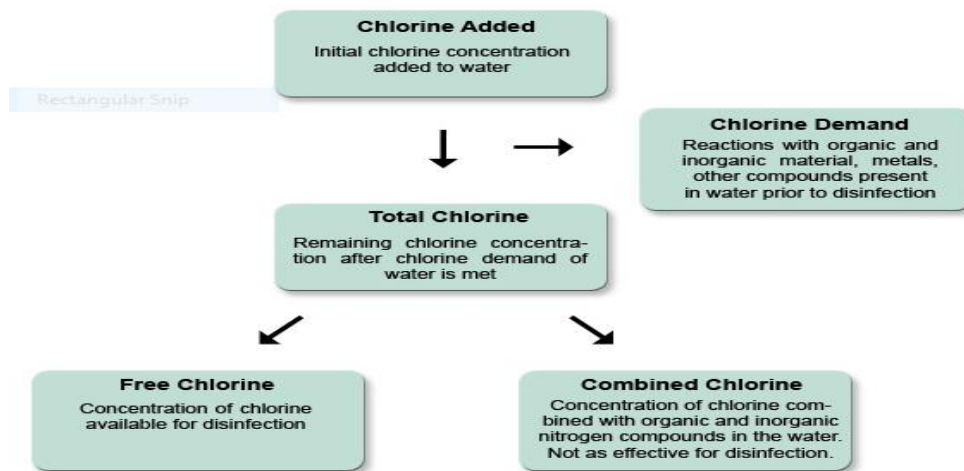
Page 54 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
---------------	--	---	---------------------------------

3.2.14. Chlorine: Chlorine is a common chemical widely used for the disinfection of water because of its strong oxidizing property and its effectiveness in preventing bacterial, viruses and protozoa infections. When chlorine is added to water, some of the chlorine reacts first with inorganic and organic materials and metals in the water and is not available for disinfection (this is called the **chlorine demand** of the water). After the chlorine demand is met, the remaining chlorine is called **total chlorine**.

Total chlorine is further divided into:

- ✓ **Combined chlorine**, which is the amount of chlorine that has reacted with inorganic (nitrates, etc.) and organic nitrogen-containing molecules (urea, etc.) to make weak disinfectants that are unavailable for disinfection and,
- ✓ **Free chlorine**, which is the chlorine that is left over and is available to inactivate disease-causing organisms; it is a measure of the potability of the water.
- ✓ Thus, total chlorine = combined chlorine + free chlorine
- ✓ When chlorine is added to drinking water, it proceeds through a series of reactions described below.

Chlorine Addition Flow Chart



Chlorine is widely used to disinfect drinking water as the final step in the water treatment process. The objective of chlorination is to add enough chlorine to leave 0.2 – 0.5 mg/L free residual chlorine after half an hour contact time.



Fig 9. Residual chlorine test

3.3. Interpreting Test Results

Evaluating what levels of contamination is acceptable, and understanding the nature of problems caused by different contaminants, are the basic considerations in interpreting the test results.

Water that has been contaminated with chemicals exceeding the national standards or WHO Guidelines for Drinking Water Quality should not be used for domestic purposes, if at all possible.

Any chemical concentration above 10% of the national standards or WHO Guidelines increases the risk for serious health effects. Therefore, appropriate local or national authorities should be notified as early as possible so that further investigation can occur, and possible solutions to the problem can be implemented.

The WHO Guidelines for Drinking Water Quality do not include some chemicals such as iron, calcium, sodium, magnesium and zinc. This is due to the fact that they pose no health risk at the levels generally found in drinking water.

3.4. What is a settle ability test?

The settle ability test is an analysis of the settling characteristics of the activated sludge mixed liquor suspended solids (MLSS). It is often referred to as “running a settle meter.” The test is normally done at the treatment plant rather than a certified laboratory. It includes five basic items:

- A clear container to hold the MLSS
- A timer or clock to track elapsed time
- A paddle or other mixing device

Page 56 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
---------------	--	---	---------------------------------

- A place to record the readings
- Operator patience, attentiveness and diligence

The idea of the test is to provide a place where the MLSS can quietly separate from the liquid water. The MLSS solids consist mostly of bacteria with some organic and inorganic debris mixed in. The debris can be finely shredded toilet paper, paper towel fibers, vegetable fibers, plastic material, seeds, insect parts — you name it.



Fig.10 Settle ability test

During the first five minutes of the settleability test, the bacteria clump together, forming large clumps (floc). These floc particles are slightly denser than water — this helps with settling and compaction. After initially clumping together, the floc begins to settle toward the bottom of the container, squeezing the clear liquid out and up toward the surface. Running a settle ability test gives the plant operator information about the settling characteristics of the biomass and the potential quality of the clarifier effluent. This article will discuss the basic settleability test and shed light upon a modification of that test called the diluted settleability test.

Page 57 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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Self-Check -2

Written Test

Direction: Choose the best answer for the following questions. Use the Answer sheet provided in the next page: Each question worth two points

1. The difference between the amounts of chlorine added to a water and the amount of chlorine residual left after a certain length of time.
 - A. Chlorine residual
 - B. Chlorine demand
 - C. Combined chlorine
 - D. A and C
2. The amount of oxidizable pollutants found in surface water/ waste water can be quantified by ----- .
 - A. BOD
 - B. COD
 - C. DO
 - D. TOC
3. Which one of the following factor **is not** important in evaluating jar test results?
 - A. Rate of floc formation
 - B. Clarity of water between floc particles
 - C. Size of floc
 - D. Apparent color observed
4. Residual Chlorine is ----- .
 - A. The amount of chlorine needed to destroy bacteria in water
 - B. Amount of chlorine which will be left in the water after all organisms have been destroyed.
 - C. Any excess chlorine above the level needed to satisfy the demand
 - D. None of the above
5. The color of water resulting from the combined effect of factual **color** and any organic and inorganic particulate matter is termed as-----
 - A. True color
 - B. Primary color
 - C. Apparent color
 - D. None
6. ----- Chlorine is added to water to:
 - A) Make its taste better
 - B) Kill/inactivate harmful microorganisms
 - C) Improve its pH
 - D) Remove turbidity

Note: Satisfactory rating – 12 points Unsatisfactory - below 12 points

You can ask you teacher for the copy of the correct answers

Information Sheet-4	Performing Calculation
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4.1. Introduction

Calculation is a deliberate process that transforms one or more inputs into one or more results. To calculate means to determine mathematically in the case of a number or amount, or in the case of an abstract problem to deduce the answer using logic, reason or common sense.

4.2. Performing Calculations

A/ How do you calculate residual chlorine in water?

The **chlorine residual** can be calculated by subtracting the amount of iodate solution used from the amount of thiosulfate solution added originally. (This gives you the amount of thiosulfate solution that reacted with the **chlorine**. The ratio of reacted thiosulfate (mls) to **chlorine** in mg/l is 1:1.

B/ How do you calculate water in a rectangular tank?

Multiply its **width (W)** by its **height (H)** and its **depth (D)**. The result is the cubic meter of space the **tank** holds. To get the answer in litre multiply the result by 1000 which gives you the number of liters of **water** the **tank** holds.

Which can be calculated using a formula of: $V = W \times H \times D$

If the water is in a circular tank or well; its volume can be calculated by multiplying its depth by the square of its radius square and multiplying 3.14 (pi)

$$V = \pi r^2 D$$

Where $\pi = 3.14$ which is nearly equivalent to 3.14

r = Radius of the water tank or circular well.

D = Depth or height of the water level

C. Concentration of a Solution

The amount of solute present in a given amount of solution is known as concentration of the solution. There are various ways of expressing the concentration of a solution The most common units are i/ Mass percent (Percentage composition) and ii/ Molarity

- **How to calculate mass percent**

Dividing the mass of solute that you measured to its volume of solution and multiplying by 100.

Which is; **Mass percent of solute = $\frac{\text{Amount of Solute}}{\text{Amount of solution}} \times 100$**

For example, a 10% solution of sodium chloride means a solution containing 10 gram of sodium chloride in a 100 gram of the solution (Solute +Solvent).

- **How to calculate Molarity**

- ✓ Molarity is defined as the number of moles of solute present per liter of the solution.
- ✓ Molarity = Number of moles of solute/ volume of solution in liter
- ✓ Number of moles = given mass of solute/ Molar mass of solute

- **Dosage Calculations**

It is most necessary for a plant operator to know how to calculate the dosages of the various chemicals used in water treatment. It is important to be accurate when calculating dosages as too little chemical may be ineffective and too much a waste of money. In process control the exact dose of chemical must be determined through calculation for the purposes of efficient operation and economy.

- **EXAMPLE 1**

The chlorine dosage of an effluent is 15 mg/L. How many kilograms of chlorine will be required to dose a flow of 8 500 M³/day?

In this question, it will be necessary to utilize your knowledge of the metric system.

$$1 \text{ mg/L} = 1 \text{ kg/l } 1000 \text{ M}^3$$

For every 1 000 M³ of water of flow, we will need to use 15 kg chlorine.

$$\frac{15 \text{ kg Cl}_2 \times 8\,500 \text{ M}^3/\text{d}}{1000\text{M}^3} = 127.5 \text{ kg Cl}_2/\text{d}$$

Above we expressed 15 mg/L as 15 kg Cl₂ /1 000 M³ and multiplied by the flow to obtain the answer expressed as 127.5 kg/Day.

Page 60 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
---------------	--	---	---------------------------------

■ Measurement Conversions

Volume

1 gallon = 3.78 L

1 L = 0.26 gallons

1 L = 33.8 fluid oz (US)

1 L = 1,000 mL

1 mL = 0.001 L

Weight

1 kg = 1,000 g

1 g = 0.001 kg

1 g = 1,000 mg

1 mg = 0.001 g

1 mg = 1,000 μ g

1 μ g = 0.001 mg

Concentration

1 ppm = 1 mg/L = 1/1 million = 0.000001

1 ppb = 1 μ g/L = 1/1 billion = 0.000000001

To convert ppb to ppm divide by 1,000

To convert ppm to ppb multiply by 1,000

5. 1. Introduction

Each laboratory should develop a plan to protect laboratory equipment, materials, and tools from loss and to prevent hazardous conditions from developing during emergencies. All laboratory personnel should be trained in their role in shutting down equipment.

5.2. Laboratory Shutdown Checklist

- Clean glassware and store appropriately
- Turn off the lights
- Turn off plumbed natural gas
- Cancel deliveries, if possible
- Lock all lab doors
- Turn off (close) taps (water faucet)
- Store samples in an appropriate temperature and places.
- Dispose of all biological materials appropriately.
- Turn off UV light
- Be diligent in returning chemicals to their proper storage location after use.
- Label and securely cap every container.
- Move hazardous waste with completed waste tag to the proper waste storage area.
- Close sashes on chemical fume hoods.
- Check mechanical rooms
- Check water distillation units
- Shut off copy machines, printers and computers

Page 62 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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6.1. Introduction

Cleaning and waste disposal services in a laboratory requires strict adherence to applicable policies and procedures. Hazardous waste must be stored in containers (including lids) made of materials that are compatible with the waste. Hazardous waste containers must be in good condition and free of leaks or any residue on the outside of the container. Unacceptable containers include household detergent and food service containers. The best container for your hazardous waste is the original chemical container.

All potential wastes that arise from laboratory operations needs to be assessed and an appropriate disposal route selected prior to waste being generated. Waste should be collected in a suitable container and labeled.

6.2. Activities in disposing and storing of unused materials

- All waste must be appropriately packaged for the waste category. For example: corrosive waste should be stored in non-metallic containers
- Solid debris must be placed in plastic bags. Do not place chemical or other non-biohazardous material in a biohazard bag. Biohazard bags are for biohazardous material only. Any waste disposed of in these bags will be treated as such.
- All liquid waste must be stored in leak proof containers with a screw- top or other secure lid.
- Liquid dumps are intended for liquids only. Do not place glass or plastic items, such as tubes or pipettes, into solution dumps. If these items require disposal, package them separately. (Keep plastic and glass waste separate.)

Information Sheet-7	Cleaning and Storing Equipments
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7.1. Introduction

Adequate cleaning of analytical glassware is an essential procedure that determines the reliability of assays and tests carried out in laboratories, keeping the glassware free of interference from residues left by previous tests.

7.2. Cleaning Agents of Equipments

A/ Detergents: A detergent is a surfactant or a mixture of surfactants with cleansing properties in dilute solutions. These substances are usually alkylbenzene sulfonates that are similar to soap but are more soluble in hard water. These are commonly available as powders or concentrated solutions. Detergents, like soaps, work because they are amphiphilic: partly hydrophilic (polar) and partly hydrophobic (non-polar)..

- **Surfactants** are compounds that lower the surface tension (or interfacial tension) between two liquids, between a gas and a liquid, or between a liquid and a solid.

B/ Degreasers: A degreaser is a cleaner designed to remove grease, oils, cutting fluids, corrosion inhibitors, handling soils, finger prints, and other contamination common in assembly, stamping, other types of metal fabrication, refineries, motor repair, airplane hangars, and many other applications.

Many of the most effective degreasers are also the safest for the environment. These include vinegar, lemon juice, cornstarch, borax, baking soda, and castile soap. Several makers of dishwashing liquids also add natural grease-cutters, such as lemon and other citrus oils, to their formulas.

C/ Abrasives: Are materials made of grit papers which can be used for cleaning of a surface or to make it smooth.

D/ Acids: are chemicals usually liquid, which contain hydrogen ions and has a pH of less than 7. Mildly acidic cleaning products include vinegar (acetic acid) and lemon juice (citric acid). Mild cleaners made from these acids are safe for use around children and pets. Other acids are often found in household cleaning products.

7.3. Storing of Equipments

Cleaning and sanitizing equipment is essential because it helps us to be more secured, far from bacteria and other causes of illnesses. It is also important to store the equipment properly for us to avoid accidents. If the equipment is not clean, sanitized or stored properly, it can cause a lot of trouble.

Make it a habit to clean tools and equipments after each use before you return them to storage. Wipe them down with a rag or old towel and be sure they are free of dust, grease and debris before you put them into their proper places. This is also an opportunity to look for any damage or defects.

Page 65 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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Self-Check -3	Written test
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Directions: Choose the correct answer from the given options. Each question worths two points

1. Materials made of grit papers which can be used for cleaning or making surfaces smooth are known as-----

- A. Abrasives B. Degreasers C. Detergents D. Acids

2. ----- Are compounds that lower the surface tension between two liquids.

- A. Degreasers B. Detergents C. Surfactants D. Analytes

3. Chemicals usually liquids, which contain hydrogen ions and having a pH value of greater than 7

- A. Acids B. Bases C. Detergents D. Salts

4. Which one of the following is a mild acid that can use for cleaning purposes?

- A. Nitric acid B. Citric acid C. Hydrochloric acid D. A and B

Note: Satisfactory rating – 8 points	Unsatisfactory - below 8 points
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You can ask you teacher for the copy of the correct answers

Instruction Sheet-4	Learning Guide 55: Finalize Work
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This learning guide is developed to provide you the necessary information regarding the following **content coverage** and topics:

- Applying principle of chain of custody
- Recording results
- Reporting observation of further action
- Identifying atypical results
- Cleaning and restoring work area

This guide will also assist you to attain the learning outcome stated in the cover page.

Specifically, **upon completion of this Learning Guide, you will be able to:**

- Apply chain of custody principles.
- Record results.
- Identify and report ‘out of specification’ or atypical results.
- Report observations or measurements requiring further action.
- Clear and restore work area.

Learning Instructions:

-
1. Read the specific objectives of this Learning Guide.
 2. Follow the instructions described below
 3. Read the information written in the “Information Sheets 1-2”. Try to understand what are being discussed.
 4. Accomplish the “Self-checks 1-3” in each information sheets on pages 70, and 75.
 5. Ask from your teacher the key to correction (key answers) or you can request your teacher to correct your work. (You are to get the key answer only after you finished answering the Self-checks). If you earned a satisfactory evaluation proceed to “Operation sheets 1-9 on pages 76,77,78,79,80,81,and 82.and do the LAP Test on page 83”.
 6. However, if your rating is unsatisfactory, see your teacher for further instructions or go back to Learning Activity.

Page 67 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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Information Sheet-1	Applying Principle of Chain of Custody
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1.1. Introduction

Chain of custody (CoC), in legal contexts, is the chronological documentation or paper trail that records the sequence of custody, control, transfer, analysis, and disposition of physical or electronic evidence.

The chain of custody is a tracking record beginning with detailed scene notes that describe where the evidence was received or collected. Collection techniques, preservation, packaging, transportation, storage and creation of the inventory list are all part of the process used in establishing the chain of custody

1.2. Importance of The Chain of Custody

Properly designed and executed chain-of-custody forms will ensure sample integrity from collection to data reporting. This includes the ability to trace possession and handling of the sample from the time of collection through analysis and final disposition.

1.3. Objectives of Chain of Custody

The primary objective of the COC is to create an accurate written record that traces samples from the moment of collection through receipt in the laboratory, to the eventual destruction or disposal.

A chronological record of who has possessed the sample(s) and of all analyses that were performed on the samples must be maintained. In general, the following COC guidelines should be followed:

- A minimum number of people should collect and handle samples and data
- The transfer of samples and data from one person to another must be documented on the COC
- Any unusual appearance, such as the condition of the sample and sample container, should be noted on the COC
- COC forms must accompany samples and data
- Samples and data must include identification that is legible and written with permanent ink.

2. Sample Handling and Custody

A critical activity within any data collection phase involving physical samples is the handling of sample media prior to sampling, handling/transporting sample media to the field, handling samples from the field at the time of collection, storage of samples (at field or other locations), transport of samples from the field site, and the analysis of the samples.

Documentation ensuring that proper handling has occurred throughout these activities is part of the custody record, which provides a mechanism for tracking samples through sample collection, processing and analysis. Custody records document the “chain of custody”; the date and person responsible for the various sample handling steps associated with each sample. Custody records also provide a reviewable trail for quality assurance purposes and as evidence in legal proceedings.

Table 4 Chain of Custody record format

Chain of Custody Record					
Project No.		Project Title			Organization
Shipping Container No.					
Field Samplers: <i>print</i>		<i>signature</i>			
					Address
Date	Time	Site/Location	Sample Type	Sample ID	Remarks
Relinquished by (<i>print and signature</i>):			Received by (<i>print and signature</i>):		Comments

Self-Check -1	Written test
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Directions: Say **TRUE** for **FALSE** for the following statements. Each question worth two points

1. Sample planning is a tracking record beginning with detailed sight notes that explain where the evidence of sampling was received or collected.
2. Samples and data must include identification that is legible and written with permanent ink.
3. Properly designed and executed chain-of-custody forms will ensure sample integrity from collection to data reporting.
4. Any unusual appearance, such as the condition of the sample and sample container, should be noted on the chain -of -custody.

Note: Satisfactory rating – 8 points	Unsatisfactory - below 8 points
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You can ask you teacher for the copy of the correct answers

Information Sheet-2	Recording results
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2.1. Introduction

Documents and records are essential for assuring accuracy and consistency in the laboratory, and documents are the reflection of the laboratory's organization and its quality management. A well-managed laboratory will always have a strong set of documents to guide its work.

The lab notebook is usually written as the experiments progress, rather than at a later date. In many laboratories, it is the original place of record of data (no copying is carried out from other notes) as well as any observations or insights.

The most important concepts in record keeping are accuracy and continuity. Laboratory records and log books are your proof of the work that you do. The more accurate information you record in them, the better you will be able to diagnose and deal with problems arising at your laboratory. Not only that, but your records and logs are legal documents and are your proof that you are doing your job properly.

2.2. Recording and Reporting Test Results

- Work safely at all times, complying with health and safety, environmental and other relevant regulations and guidelines
- Obtain and use the correct quality control documentation for the manufacturing method and product
- Record the results of the inspection activities in the appropriate format
- Record inspection and test results accurately and legibly
- Record all required details of the inspection and test activities and results
- Pass completed records on to the correct person/location
- Provide required reports on time and through the correct channels of communication
- Report any problems that you cannot solve or are outside your permitted authority to the appropriate person

2.3. Recording and Storing Results

All results must be recorded and stored in a way that allows them to be easily located again. It is recommended that records are kept on formatted sheets, either paper or computer-based. On these sheets it is important to include the target levels so that all readings can be compared to them. These values may help you identify the need to take corrective action as appropriate.

2.4. Collect Data Recording Forms

Data recording forms are used to document your samples and test results. See Table--- for an example data recording form. Many forms may be used, depending on how many samples are taken and the number of tests performed, so it is important to choose the correct forms for data entry. It is essential to collect all data forms and to verify the information and results for each day before preparing a database.

Table 5 Data Recording Form

Location: _____		Sample Taken By: _____									
Date of Sampling: _____		Sample Tested By: _____									
Sample Description	Sample ID #	Turbidity (NTU)	pH	Chlorine (mg/L)	Iron (mg/L)	Manganese (mg/L)	Phosphate (mg/L)	Nitrate (mg/L)	Flouride (mg/L)	E. coli (CFU/100 ml)	Total Coliform Fecal Coliform (CFU/100 ml)
Date of Report: _____											
Comments: _____											

Information Sheet-3	Identifying and reporting 'Out of specification'/ atypical results
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3.1. Introduction:

3.1. Terms for Identifying out of specification results

- **Out of specification (OOS):** The test results that fall outside the acceptance criteria established in SOP. (Against Specification limit).
- **Out of Trend (OOT):** The test results that may be within specification but show significant variation from the historical results.
- **OOS (out of specification)** is the comparison of one result versus predetermined specification criteria.
- **OOT (Out of Trend)** is the comparison of many historical data values versus time

Information Sheet-4	Cleaning and Restoring Work Area
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4.1. Introduction: Upon the completion of laboratory work, all tools, equipments, chemicals, laboratory wares, reagents, and personal protective equipments shall be taken from the work area and store in appropriate safe location in accordance to the correct conditions.

4.2. Cleaning: It is a method of removing of wastes and dirt from surfaces of tools, equipments and work place.

4.3. Advantages of Cleaning Work Area

- A safe working area must be provided for efficient work
- To make the work area attractive and conducive for work
- To make equipments, tools and other materials clean and to have long span to use.
- To prevent our health and the environment from pollution
- Use protective safety equipment where there is a job hazard
- Keep the work area clear of hazards, with plenty of working space for solid footing. Tools should be in good condition and used for the purpose for which they were intended.
- All areas where solvents or other compounds are used and stored must be well ventilated.
- Areas around sinks and taps should be kept clear so that chemicals spilled on one's hands or person can be washed off quickly.
- Wipe up all spills immediately.

6.5. Restoring Work Place and Disposing Waste

- ✓ After you have completed your work you need to clean your work area and put back all the materials you used back to their original place.
- ✓ Wastes that are generated when you do your analysis should be disposed properly.
- ✓ Solid wastes should be disposed in trash bins and liquid wastes in sinks.

Self-Check -2**Written test**

Directions: Say **TRUE** for **FALSE** for the following statements. Each question worth two points

1. Dilution is a method of removing of wastes and dirt from surfaces of tools, equipments and work place.
2. Work efficiency can be directly related with safe and conducive work area.
3. Wastes which are generated during your laboratory analysis should be disposed to a any near area.
4. Use protective safety equipment where there is a job hazard.

Note: Satisfactory rating – 8 points Unsatisfactory - below 8 points

You can ask you teacher for the copy of the correct answers

Page 75 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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Operation Sheet 1	pH Meter Calibration
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Procedure for pH Calibration

Step 1: Set up all equipment needed for calibration. You will need: a drip mat; clean glass beakers; a bottle of pH4 solution; a bottle of pH7 solution; distilled water; a box of tissues.

Step 2: Place a clean probe in the pH4 solution.

Step 3: Read the meter and adjust the settings until it has a pH value of 4.

Step 4: Use distilled water to rinse the probe thoroughly.

Step 5: Place the clean probe in the pH7 solution and adjust the meter settings until it has a pH value of 7.

Step 6: Use the distilled water again to rinse the probe.

Step 7: Dry the probe when you have finished calibrating. The probe is now calibrated.

Operation Sheet -2	Measurement of Color
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Directions: Measure color of sample water using spectrophotometer

Step1. Place the cell containing the blank (distilled water) in the sample compartment

Step 2. Close the sample compartment lid.

Step 3. Press “**AUTOZERO**” key to set the zero absorbance.

Step 4. Discard the distilled water and place the cell containing the **sample** in the measuring position.

Step 5. Close the sample compartment lid.

Step 5. Press “**START**” key to measure **COLOR**.

Step 6. Record the “Conc” as COLOR units for the sample

Step 7. Report the results as follows:

Page 76 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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COLOR Units	Record to nearest
1 ~ 50	1
51 ~ 100	5
101 ~ 250	10
251 ~ 500	20

Operation Sheet -3

Measurement of Total Alkalinity

Directions: Measure Total Alkalinity of a sample of water using Titration Method

Procedures:

Step 1. Place 100 ml of sample in an Erlenmeyer flask

Step 2. Add 3 drops of methyl orange indicator solution to the flask

Step 3. Titrate sample with 0.02 N H₂SO₄ (Sulphuric acid)

Step 4. Swirl constantly the flask content until the COLOR of the flask content change from yellow to red

Step 5. Record the volume of titrant used.

Step 6. Calculate the total Alkalinity of the sample

Operation Sheet -4	Measurement of Total Iron
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Directions: Measure the iron concentration of water sample using spectrophotometer

Procedures:

- Step 1. Prepare all required reagents, chemical and equipments
- Step 2. Select the test (# 265, Iron Ferrover)
- Step 3 Prepared Sample: Fill a clean square sample cell with 10 ml of sample
- Step 4 Add the contents of one FerroVer Iron Reagent Powder Pillow to the sample cell. Swirl to mix. An orange color will form, if iron is present
- Step 5. Press **TIMER>OK** (A three-minute reaction period will begin
- Step 6. Fill a second square sample cell with 10 mL of sample
- Step 7. When the timer expires, insert the blank into the cell holder with the fill line facing right. Press **ZERO**. The display will show: **0.00 mg/L Fe**
- Step 8. Place the prepared sample into the cell holder with the fill line facing right.
- Step 9. Press **READ**. Results are in mg/L Fe.

Operation Sheet -5	Fluoride Measurement
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Directions: Measure fluoride concentration of water sample using spectrophotometer (Using Spa dons reagent)

Procedures:

- Step 1. Press **STORED PROGRAMS**
- Step 2. Select the test (# 190 flouride)
- Step 3 **Prepared Sample:** Pipet 10.0 mL of sample into a dry square sample cell.
- Step 4. **Blank Preparation:** Pipet 10.0 mL of deionized water into a second dry square sample cell.
- Step 5. Carefully pipet 2.0 mL of SPADNS Reagent into each cell. Swirl to mix.
- Step 6. Press **TIMER>OK**.A one-minute reaction period will begin.
- Step 7. Insert the blank into the cell holder. Press **ZERO**. The display will show:
0.00 mg/L F–
- Step 8. Insert the prepared sample into the cell holder with the fill line facing right.
Press **READ**. Results are in mg/L F–.

Operation Sheet -6	Measurement of Chloride
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Direction: Measure the concentration of Chloride of sample water using spectrophotometer

Procedures:

Step 1 Press **STORED PROGRAMS**.

Step 2. Select the test (# chloride 70)

Step 3 **Prepared Sample:** Fill a square sample cell with 10 ml of sample

Step 4 **Blank Preparation:** Fill another square sample cell with 10 ml of deionized water.

Step 5 Pipette 0.8 ml of Mercuric Thiocyanate Solution into each sample cell

Step 6 Swirl to mix.

Step 7 Pipette 0.4 ml of Ferric Ion Solution into each sample cell.

Step 8 Swirl to mix. An orange color will develop if chloride is present.

Step 9 Press **TIMER>OK**. A two-minute reaction time will begin

Step 10. Within five minutes after the timer expires, wipe the blank and insert it into the cell holder with the fill line facing right. Press **ZERO**. The display will show: 0.0 mg/L Cl⁻

Step 11 Wipe the prepared sample and insert it into the cell holder with the fill line facing right

Step 12 Press **READ**. Results are in mg/L Cl⁻

Operation Sheet -7	Measurement of Manganese
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Directions: Perform the concentration of manganese of sample water using spectrophotometer

Procedures:

Step 1. Press **STORED PROGRAMS**.

Step 2. Select the test (# 295, Mn)

Step 3. Prepared Sample: Fill a square sample cell with 10 mL of sample.

Step 4. Add the contents of one Buffer Powder Pillow Citrate Type for Manganese.

Stopper and invert to mix.

Step 5. Add the contents of one Sodium Periodate Powder Pillow to the sample cell.

Stopper and invert to mix. A violet color will develop if manganese is present.

Step 6. Press **TIMER>OK**. A two-minute reaction period will begin.

Step 7. **Blank Preparation:** Fill a second square sample cell with 10 ml of sample.

Step 8. When the timer expires; insert the blank into the cell holder with the fill line facing right.

Step 9. Press **ZERO**. The display will show: 0.0 mg/l Mn

Step 10. within eight minutes after the timer expires; insert the sample into the cell holder with the fill line facing right.

Step 11. Press **READ**. Results are in mg/ Mn.

Operation Sheet -8

Total Hardness

Directions: Measure total hardness of sample water using Titration method

Prepare all required reagents, equipments and apparatuses

Procedures:

Step1. Take 50 ml of sample water from your sample bottle and transfer to 250 ml Erlenmeyer flask.

Step2. Add 1ml of hardness buffer solution using calibrated dropper and swirl it.

Step3. Add one Manver 2 hardness indicator powder pillow and swirl the flask to mix until the color of the mixture/solution changed from wine red to blue at the end point.

Step4. Titrate the content of the flask with 0.01M EDTA solution.

Step5. Multiply the volume of titrate you obtained by 20 to get the concentration of total hardness in milligram per liters

Operation Sheet -9

Measurement of Biochemical oxygen demand (BOD)

Directions: Measure the concentration of BOD

Procedures

Step1. Fill a DO (Dissolved oxygen) water sample bottle to over flowing with sample water

Step 2. Do not fix the samples immediately. Instead, store the full bottle in darkness at 20 degree Celsius for five days.

Step 3. After five days, test the sample for dissolved oxygen. (Follow the procedures for the dissolved oxygen test)

Step 4. Subtract the five-day result from the initial DO result to find the BOD, and record the BOD the data recording format.

Page 82 of 85	Author/copyright: Federal TVET AGENCY	Water Supply and Sanitation Supervision Level-IV	Version 1 September, 2020
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LAP Test -1**Practical Demonstration**

Name: _____

Date: _____

Time started: _____

Time finished: _____

Instruction I: Given necessary tools, equipment, lab wares, chemicals and reagents you are required to perform the following tasks within **5 hours**.

Task 1: Perform **pH** Calibration**Task 2:** Perform the color of a sample of water**Task 3:** Perform the **total alkalinity** of a sample water**Task 4:** Perform the **total iron** concentration of water sample**Task 5:** Perform the **fluoride** concentration of water sample**Task 6:** Perform the **Chloride** content of a raw water sample**Task 7:** Perform the concentration of **manganese** of a waste water sample**Task 8:** Perform the total hardness of a sample of water**Task 9:** Perform the concentration of Dissolved Oxygen of a sample of waste water

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