## STANDARD OPERATING PROCEDURE(SOP)

Treated Fecal Sludge Management & Analysis

# Fecal Sludge Laboratory



## **PURE INNOVATION, SAFE SANITATION**

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## **Standard Operating Procedure (SOP)**

Treated Fecal Sludge (Effluent) Management & Analysis

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## Preface 01

In the realms of public health engineering, managing fecal sludge effectively is crucial for community health and environmental protection. This book, **"Standard Operating Procedure (SOP):** Treated Fecal Sludge Management & Analysis (Effluent)" as a beacon of knowledge and guidance for professionals in the sector of Fecal Sludge management.

As the custodians of community health, our responsibility extends beyond the mere treatment of effluent samples. Recognizing the critical importance of a well-equipped Fecal Sludge Laboratory, this book unfolds a journey through the intricacies of laboratory setup, sample collection, preservation and advanced analytical techniques. The aim is to empower public health engineers and laboratory personnel with the knowledge and tools needed to navigate the complexities of fecal sludge management.

Within these pages, readers will discover a roadmap for establishing and maintaining a state-of-the-art laboratory, ensuring safety, precision and adherence to Bangladesh government standards for sewerage discharge (DoE guideline March 2023). From the nuances of sample collection and intricacies of cutting-edge analytical technologies, this guide

delves into every aspect of fecal sludge analysis.

Moreover, as we navigate this realm, we don't merely focus on the technicalities. The book emphasizes the broader context of community engagement, policy recommendations, and sustainable approaches. It is a holistic approach that considers the laboratory as a pivotal player in the broader ecosystem of public health and environmental preservation.

We extend our gratitude to all those who have contributed to this endeavor—researchers, practitioners, and policymakers. The shared knowledge and experiences encapsulated in this book aim to elevate the standards of fecal sludge management, not only in Cox's Bazar but as a contribution to the global effort in ensuring sanitation equity.

In closing, this book is more than a guide; it is an invitation to a collaborative journey toward better public health and environmental stewardship through ensuring fecal sludge management.

**Engineer Tushar Mohon Sadhu Khan** Chief Engineer, DPHE

## Preface 02

It is with great pleasure and a deep sense of responsibility that we present this book, "Standard Operating Procedure: Treated Fecal Sludge (Effluent) Management & Analysis." This publication is a significant milestone in our continuous efforts to address the critical issues of sanitation and environmental sustainability.

This comprehensive manual is designed to serve as an essential guide for professionals in the field of fecal sludge management (FSM), providing detailed procedures for the effective treatment, management, and analysis of treated fecal sludge effluent. The methodologies presented herein are the result of rigorous scientific research and practical field applications, aimed at ensuring the highest standards of environmental safety and public health.

We are profoundly grateful to the World Bank for their unwavering support and generous funding through the Emergency Multi-Sector Rohingya Crisis Response Project (EMCRP). This project has been instrumental in establishing our state-of-the-art Fecal Sludge Laboratory, where groundbreaking research and development in FSM are conducted. The World Bank's commitment to sustainable development improving sanitation and infrastructure has been a driving force behind our initiatives, enabling us to make significant strides in this critical area.

I extend my heartfelt appreciation to the dedicated team of researchers, scientists, and field professionals whose expertise and hard work have culminated in this publication. Their commitment to excellence and innovation is evident in every page of this book. I also wish to acknowledge the collaboration and support from our partners and stakeholders, whose contributions have been invaluable in achieving our objectives.

This book is not only a reflection of our collective achievements but also a beacon for future endeavors in fecal sludge management. It serves as a testament to what can be accomplished when vision, knowledge, and resources are combined towards a common goal of sustainable and effective sanitation solutions.

We hope that this manual (SOP) will be an indispensable resource for practitioners, policymakers, and researchers in the field of FSM, and that it will inspire further advancements and innovations in our shared mission to safeguard public health and protect our environment.

Mohammad Abdul Kaium Project Director, Emergency Multi-Sector Rohingya Crisis Response Project (EMCRP) World Bank (DPHE Part)

## Acknowledgement

The realization of this comprehensive guide on **"Standard Operating Procedure (SOP):** Treated Fecal Sludge Management & Analysis (Effluent)" is the result of collective efforts, expertise and support from various individuals and organizations. We extend our heartfelt gratitude to all those who have contributed to this endeavor.

First and foremost, we express our sincere thanks to the Embassy of Switzerland in Bangladesh for their unwavering support from September 2021 to December 2023. Their commitment and encouragement played a pivotal role in laying the foundation for the advancements in our Fecal Sludge Laboratory. Their support has been instrumental in the progress we have achieved in the initial phases of this project. Special thanks are also extended to Christopher Friedrich, Research Associate at EAWAG, Switzerland for his invaluable contributions to the lab design, feasibility study and providing training.

Furthermore, we extend our profound appreciation to the World Bank for assuming responsibility for our laboratory from January 2024 onward. Their dedication to promoting sustainable development and investing in critical infrastructure is a testament to their commitment to global public health. We look forward to the continued collaboration and the positive impact it will have on our ongoing efforts in fecal sludge management.

Our gratitude also extends to the entire team at the Department of Public Health Engineering, Cox's Bazar. The diligence, expertise and passion of our colleagues, including Md Shahidul Islam Siam (Laboratory Manager), Monuar Hosan (Laboratory Technician), Rahul, Imran and Kamruzzaman (Sample Collectors), have been the driving force behind the success of our laboratory and the creation of this guide.

We also acknowledge the invaluable contributions of researchers, practitioners and experts in the field of fecal sludge management who have shared their knowledge and experiences, enriching the content of this guide.

Last but not least, we express our appreciation to the community members in Cox's Bazar specifically WASH sector, local & iNGOs for their cooperation and understanding as we work towards improving public health and sanitation in this region.

This book stands as a collaborative achievement and it is our hope that its contents contribute to the advancement of fecal sludge management not only in our locality but also on a broader scale. Thank you to all who have been part of this journey.

> **Engineer Md. Mostafizur Rahman** Executive Engineer DPHE, Cox's Bazar

## Abbreviations

## In Alphabetical order

ABR	Anaerobic Baffled Reactor		
ADS	Anaerobic Digester system		
BD	Bangladesh		
BDRCS	Bangladesh Red Crescent Society		
BOD	Biological Oxygen Demand		
BSL	Biosafety Level		
COD	Chemical Oxygen Demand		
CF	Christopher Friedrich		
CXB	Cox's Bazar		
DEWATS	Decentralized waste water treatment system		
DoE	Department of Environment		
DPHE	Department of Public Health Engineering		
EW	Ewinur Machdar		
FDMN	Forcibly Displaced Myanmar Nationals		
FSFL	Fecal Sludge Field Laboratory		
FSL	Fecal Sludge Laboratory		
FSTP	Fecal Sludge Treatment Plant		
ICDDR, B	International Centre for Diarrhoeal Disease Research, Bangladesh		
IFRC	International Federation of Red Cross and Red Crescent Societies		
IOM	International Organization for Migration		
LSP	Lime Stabilization Pond		
m	Meter		
MRa	Md. Mostafizur Rahman		
NGO	Non-governmental organization		
iNGO	International Non-governmental organization		
ODP	Open Deviation Pond		
PPE	Personal protective equipment		
RCh	Ritthick Chowdhury		
SDC	Swiss Agency for Development and Cooperation		
SDG	Sustainable Development Goals		
SS	Settleable Solids		
SSU	Solid Separation Unit		
TN	Total Nitrogen		
TS	Total Solids		
TSS	Total Suspended solids		
UFF	Up Flow Filter		
UNHCR	United Nations High Commissioner for Refugees		
UNICEF	United Nations Children's Fund		
WASH	Water, Sanitation, and Hygiene		
WSP	Waste Stabilization Pond		

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## **Chapter 01: Executive Summary**

The Fecal Sludge Laboratory of the Department of Public Health Engineering (DPHE) in Cox's Bazar, Bangladesh, stands as a pivotal institution dedicated to addressing the pressing sanitation challenges in this densely populated and environmentally sensitive region. Its establishment is a testament to DPHE's unwavering commitment to enhancing public health, environmental sustainability and the overall wellbeing of the community. Facing significant sanitation challenges due to a growing population and its status as a renowned tourist destination, Cox's Bazar urgently required comprehensive solutions. The Fecal Sludge Laboratory was initiated to meet this need. It operates with the core objectives of ensuring quality control, driving research and innovation, and building the capacity of local personnel. Equipped with state-of-theart technology and infrastructure, it offers advanced capabilities for chemical and microbiological analysis, data management and knowledge dissemination. Since its inception, the laboratory has made significant contributions, improving sanitation practices, fostering innovation, building local capacity and enabling data-driven decision-making. With a focus on continued research, capacity building and the promotion of best practices, the Fecal Sludge Laboratory envisions a cleaner, healthier and more sustainable future, not just for Cox's Bazar but also for other regions and countries grappling with similar sanitation challenges. The DPHE is dedicated to the ongoing success of this vital institution.

## **Chapter 2: Introduction**

#### 2.1 Goals and Objectives

The "Department of Public Health Engineering (DPHE)" is the national lead agency for provision of drinking water supply, Sanitation, hygiene and waste management throughout the country. After the Rohingya influx, DPHE has done a lot of initiatives to ensure pure water supply and sanitation for the FDMN's and also established one water testing laboratory at Cox's Bazar. In continuity of these, in 2021 the DPHE also established a Fecal Sludge Laboratory (FSL) with the support of the Embassy of Switzerland in Bangladesh in Cox's Bazar. The purpose of the FSL is to test the effluent (treated fecal sludge) in Cox's Bazar district for their adherence to government standards for sewage discharge.

From January 2022 to November 2023 Fecal Sludge Laboratory Team have completed total 6 rounds and covered every FSTPs in Cox's Bazar district. During the time of reporting total 187 functional FSTPs are now in Cox's Bazar Districts. Among them 4 are in the Host community and others are in the Rohingya camps.

#### 2.2 Scope

The scope of a Fecal Sludge Laboratory operated by the Department of Public Health Engineering (DPHE) in Cox's Bazar, particularly with respect to humanitarian efforts, can be significant. Cox's Bazar is home to one of the largest refugee camps in the world, hosting hundreds of thousands of Rohingya refugees. Humanitarian organizations and government agencies are actively involved in providing essential services to this vulnerable population. A Fecal Sludge Laboratory can play a crucial role in addressing sanitation and public health challenges in such a setting. Here's how:

#### **2.2.1 Safe Sanitation and Disease Prevention**

Cox's Bazar faces significant sanitation challenges due to the high population density in the refugee camps. The laboratory can help ensure that fecal sludge management is carried out in a way that minimizes health risks and the spread of waterborne diseases.

#### 2.2.2 Data and Research

The laboratory can be a valuable resource for collecting and analyzing data on Fecal sludge quality and quantity. This information can be used to inform decision-making and improve sanitation services in the camps.

#### 2.2.3 Quality Control

A well-equipped laboratory can provide quality control for sanitation services, ensuring that emptying, transportation, and treatment of Fecal sludge are carried out to acceptable standards, thus reducing environmental contamination.

#### 2.2.4 Capacity Building

The DPHE Fecal Sludge Laboratory can serve as a training and capacity-building center for local staff and humanitarian workers. Training in Fecal sludge management is crucial for the sustainable operation of sanitation facilities.

#### 2.2.5 Innovation and Technology

The laboratory can act as a hub for testing and piloting innovative sanitation technologies, which can be crucial in humanitarian contexts where resources and infrastructure may be limited.

#### 2.2.6 Public Awareness

It can also contribute to public awareness campaigns about the importance of safe sanitation and hygiene practices, which is vital for the well-being of the refugee population.

#### 2.2.7 Coordination

The laboratory can play a coordinating role in working with various humanitarian organizations and government agencies involved in sanitation efforts, ensuring a more efficient and effective response.

#### 2.2.8 Emergency Response

In the event of emergencies or disease outbreaks, the laboratory can quickly analyze samples to assess the situation and guide rapid response efforts.

## **Chapter 3: History of the development of Fecal Sludge Laboratory, Cox's Bazar, Bangladesh**

#### 3.1 Background

Ritthick Chowdhury, as the Executive Engineer of the Department of Public Health Engineering (DPHE), is indeed the first initiative person who played a pivotal role in the establishment of the Fecal Sludge Laboratory (FSL) in Cox's Bazar. His commitment and initiative in recognizing the need for such a laboratory and actively working towards its establishment demonstrate his dedication to improving sanitation and wastewater management in the region. Under his leadership, this important facility has become a reality, contributing to better public health and environmental protection in the Cox's Bazar district.

On 16 April 2019 Christopher Friedrich (CF), Research Assistant, Sandec, EAWAG, Switzerland met with Ritthick Chowdhury (RCh), the Executive Engineer of DPHE CXB and gave an introduction to RCh about the FSFL. RCh said that handing over the laboratory to DPHE is the way to go because any technical project of this scale related to public health should be run by a governmental agency like DPHE. RCh states that DPHE has many water laboratories but to his knowledge, they do not have an FSL, which is why this FSL could become a flagship laboratory for DPHE. RCh said that in about six months from now (Note: By approximately September 2019), there would be a new stored constructed on a DPHE building, which will be used by SDC and DPHE.

RCh also reported to CF that Ewinur Machdar (EW) has visited his office and talked to him previously about organizing a visit to the FSFL of IFRC for him and he intends to visit the FSFL soon. He emphasized that in his opinion, the transfer of the FSFL from IFRC to DPHE would be a very good idea and that he would support this transfer. RCh points out the important role DPHE played from the beginning of the emergency on and that DPHE sent 300 people from all over Bangladesh to support the operation in CXB. CF puts forward the question, if DPHE would be able to house the laboratory and estimates the space requirements of the FSL for being about 10m (Meter) by 10m, with at least two different rooms, so that one room would solely be used for microbiological analysis and another room solely for physicochemical analysis. RCh says there would be space for the FSL next to a water laboratory of DPHE and that multiple rooms for different analyses would be available. The rooms also have a connection to the water supply and the sewage network. RCh says the water laboratory of DPHE is currently performing analysis of around twenty different parameters and that he plans to increase the capacity of the water laboratory.

At last in meetings held on the **3rd** and **24th of July 2019**, **Ritthick Chowdhury, the Executive Engineer** of DPHE in Cox's Bazar district, at first confirmed DPHE's interest in establishing a permanent Fecal sludge laboratory. **Laurence West**, who was the co-chair of the sanitation technical working group of the WASH Sector in Cox's Bazar, verbally confirmed the urgent need for such a laboratory to provide essential analyses for the treatment plants in the camp.

Switzerland Embassy of Bangladesh took this opportunity and joined with DPHE as a developing partner for establishing a permanent Fecal sludge laboratory in Cox's Bazar district. After that 31 July 2019 regarding this issue a feasibility Study was done from the side of EAWAG lead by Christopher Friedrich, this feasibility study was covered Facilities, training, Human resource, Analysis capacity, Budget, Challenges and Timeline etc.

On September, 2021 DPHE & SDC by the help of NGO Shushilan requite 3 persons in the laboratory (One Laboratory Manager and two Laboratory Technicians) and on October requite 3 Sample collectors for Fecal Sludge Laboratory of DPHE. First 4 months were used for lab setup, documentation preparation and training purposes. The FSL team had started their operation in Rohingya community from 01 January 2022. The main purpose of the FSL team is collect effluent (Treated liquid from Fecal Sludge) samples from FSTPs and analyze them so that they are maintaining Government standard (DoE guideline 2023) for sewerage discharge.

In 29 August 2022, a new Executive Engineer Md. Mostafizur Rahman took the responsibility of Cox's Bazar, DPHE. Upon assuming the role of Executive Engineer Md. Mostafizur Rahman ushered in a new era of transformative leadership, significantly shaping the trajectory of the Fecal Sludge Laboratory's (FSL) growth and impact. His strategic initiatives and visionary decisions have left an indelible mark on the landscape of public health engineering in Cox's Bazar. Here some key significant contributions of him-

#### <u>Pioneering Bangladesh's First Omni Processor</u>

During his time DPHE spearheaded the introduction of Bangladesh's first Omni Processor (3<sup>rd</sup> in the world) for fecal sludge management. This groundbreaking initiative showcased a commitment to cutting-edge technology and positioned the DPHE as a national leader in innovative waste treatment solutions.

#### International Training for FSL Personnel

Recognizing the global dimensions of fecal sludge management, Md. Mostafizur Rahman arranged international training programs for FSL personnel. This initiative aimed to expose the team to international best practices, fostering a culture of continuous learning and ensuring the laboratory's alignment with global standards.

#### <u>Robust Capacity Building Strategies</u>

Understanding the pivotal role of a skilled workforce, Md. Mostafizur Rahman (MRa) implemented comprehensive capacity-building strategies. These initiatives included training programs, workshops and skill enhancement sessions for FSL personnel, empowering them with the knowledge and expertise needed for precise fecal sludge analysis.

#### Dynamic Collaboration with NGOs

MRa actively engaged in dynamic collaborations with local & International NGOs. These partnerships brought additional resources, expertise and community outreach capabilities to the FSL, amplifying its impact on the ground and fostering a collaborative approach to sanitation and public health.

#### <u>Strategic Project Extensions for Long-term Sustainability</u>

A forward-thinking leader, MRa implemented strategic project extensions to ensure the long-term sustainability of the FSL. This involved securing additional funding, expanding ongoing projects and developing a roadmap for sustained growth and impact in fecal sludge management.

#### <u>Recognition in Swiss Newspapers</u>

The transformative initiatives led by Md. Mostafizur Rahman at the Fecal Sludge Laboratory garnered international attention, leading to coverage in Swiss newspapers "One World" in 3 languages (English, French, and German). The acknowledgment in the Swiss media underscored the global significance of the laboratory's advancements under his leadership, showcasing its impact beyond national borders.

#### **3.2 Beneficiary from DPHE-FSL**

The Fecal Sludge Laboratory (FSL) established by the Department of Public Health Engineering (DPHE) in Cox's Bazar serves several beneficiaries and contributes to various aspects of public health and environmental protection. Here are some of the key beneficiaries of the FSL in Cox's Bazar:

#### **3.2.1 Local Communities**

The primary beneficiaries are the local communities, including the Rohingya refugee population in the Cox's Bazar district. The FSL ensures that fecal sludge and wastewater from treatment plants meet the required standards for safe disposal, minimizing the risk of waterborne diseases and improving overall sanitation in the area.

#### **3.2.2 Department of Public Health Engineering (DPHE)**

DPHE itself benefits from the laboratory as it helps them monitor and manage sanitation and wastewater treatment facilities more effectively, ensuring that they comply with governmental standards and guidelines.

#### 3.2.3 Government Authorities

The government agencies responsible for environmental protection, such as the Department of Environment (DoE), benefit from the laboratory's data and analyses in monitoring and enforcing regulations related to sewerage discharge and environmental protection.

#### **3.2.4 Non-Governmental Organizations (NGOs)**

NGOs working in the Cox's Bazar region use the laboratory's services to support their sanitation and wastewater management programs and projects, ensuring that these initiatives are in line with best practices and guidelines.

#### **3.2.5 International Organizations**

International organizations, such as the UNCEF, UNHCR, IOM and other global humanitarian entities involved in the Cox's Bazar are collaborating with and getting benefits from the FSL to ensure proper sanitation and environmental protection in the refugee camps by getting effluent analysis data time to time.

#### **3.2.6 Environmental Health Professionals**

Environmental health professionals, researchers, and scientists use the laboratory's data for research and to develop strategies for improving sanitation and wastewater treatment in similar contexts.

#### **3.2.7 Public Health and Safety**

The laboratory's work ultimately benefits public health and safety by reducing the risk of waterborne diseases, minimizing environmental pollution, and promoting overall sanitation in the region.

#### 3.3 Importance of FSL in Cox's Bazar

The importance of a Fecal Sludge Testing Laboratory can't be disregarded, especially given the current situation of the Rohingya refugee crisis in the area. Just now total 187 operational FSTPs are situated in Cox's Bazar District (as of November, 2023). After starting sample collection from January 2021, FSL team have felled the following reasons why Fecal Sludge Laboratory is essential-

#### **3.3.1 Public Health**

A Fecal sludge testing laboratory can help ensure the safety of the community by identifying harmful pathogens and bacteria that can cause diseases. This is especially important in overcrowded areas such as refugee camps, where the risk of infection is much higher.

#### **3.3.2 Environmental Protection**

Without proper disposal and treatment, fecal sludge can contaminate the surrounding environment, including soil and water bodies. Testing can help identify harmful levels of contaminants, enabling authorities to monitor the quality of the discharged sludge and ensure that it is being treated appropriately.

#### **3.3.3 Better Management**

Testing can help identify the composition and quality of fecal sludge, enabling better management and disposal options. This can help reduce the risk of contamination and ensure safe disposal.

#### **3.3.4 Planning and Design**

A Fecal sludge testing laboratory would provide crucial information for the planning and design of appropriate fecal sludge management systems. This information can be used to inform the development of new systems or the improvement of existing ones.

#### 3.3.5 Research and Development

Just now total 12 types treatment technology's FSTPs are present in Rohingya camps. It is an opportunity for researchers to study the composition of fecal sludge and the effectiveness of different treatment processes. This research can help to improve the efficiency and effectiveness of Fecal sludge management systems.

#### **3.3.6 Compliance**

Like many countries also in Bangladesh, regulations require that Fecal sludge is tested before disposal to ensure that it meets certain DoE guideline. Having a testing laboratory can help ensure that these regulations are being met.

#### 3.3.7 Capacity building

Finally, the laboratory can serve as a capacity-building tool, providing training and education to the community on proper sanitation and wastewater management practices.

## **Chapter 04: Laboratory Setup**

The Fecal sludge laboratory has an area of approximately 96 m<sup>2</sup> laboratory space, including a changing room and a storage area. An additional 12 m<sup>2</sup> of office space is provided in another room outside of the laboratory. This laboratory is situated in the new building next to the existing Sub Assistant Engineer's office building in Mohajer Para, Cox's Bazar Sadar, Cox's Bazar.



## **Chapter 05: Laboratory Safety**

#### **5.1 Biosafety**

Laboratory Biosafety Level 2 (BSL-2) is a crucial aspect for the Fecal Sludge Laboratory and DPHE is strongly committed to maintaining BSL-2 standards to ensure the safe and effective operation of their laboratory.\_BSL-2 encompasses a comprehensive set of safety measures and protocols, meticulously designed to safeguard laboratory personnel, the surrounding environment and the public from potential biological hazards. These precautions are especially vital when handling various samples, including fecal sludge and wastewater.

#### Key Features of BSL-2 in the DPHE Fecal Sludge Laboratory

#### Personnel Training

Personnel working in the laboratory are trained in the proper handling of biological materials, the correct use of personal protective equipment (PPE) and the recognition of potential hazards.

#### <u>Access Control</u>

Access to the laboratory is restricted to authorized personnel only. Entry and exit procedures are established to ensure that only trained and authorized individuals can enter the facility.

#### <u>Personal Protective Equipment (PPE)</u>

Laboratory personnel are provided with and required to wear appropriate PPE, such as gloves, lab coats and eye protection to prevent exposure to biological agents.

#### Engineering Controls

The laboratory is equipped with specific engineering controls such as biosafety cabinets to prevent the release of potentially hazardous materials into the environment. These controls help contain and control the materials being worked on.

#### Decontamination Procedures

The laboratory has established procedures for safely decontaminating equipment and work surfaces. This includes regular cleaning and disinfection to minimize the risk of contamination.

#### Waste Management

Proper waste disposal procedures are in place to ensure the safe removal of contaminated materials such as laboratory waste and biological samples.

#### <u>Emergency Response</u>

The laboratory has established protocols for handling spills or accidents including the use of appropriate disinfectants and notifying appropriate personnel.

#### Training and Documentation

Training records, laboratory procedures and safety manuals are maintained and all personnel are aware of the protocols in place for safe laboratory operation.

#### Monitoring and Oversight

The laboratory operates under the oversight of designated safety officers once in a year who ensure that BSL-2 standards are consistently met.

#### **5.2 Waste Management**

Here's an overview of the waste management practices at the FSL-DPHE.

#### **5.2.1 Segregation and Collection**

The laboratory follows strict protocols for the segregation of different types of waste generated during its operations. This includes the separation of general waste, hazardous waste and biohazard waste. Each type of waste is collected separately in designated containers.

#### **5.2.2 Hazardous Waste Handling**

Hazardous waste, which may include chemical reagents, contaminated materials or laboratory equipment that may have come into contact with potentially infectious agents is managed with utmost care. Such waste is appropriately labeled and stored in clearly marked containers to prevent accidental exposure. Beside these, the reagents vial which contain strong acid and heavy metals are make concrete block and dispose this to the landfill.

#### **5.2.3 Biohazard Waste Disposal**

Given the BSL-2 classification, the laboratory deals with fecal sludge that potentially contains biological agents or pathogens. Biohazard waste, including used personal protective equipment (PPE) contaminated samples and other biohazardous materials are treated in a way that ensures safe disposal. This often involves autoclaving or other approved sterilization methods to deactivate any potential pathogens before final disposal.

#### 5.2.4 General Waste Management

Non-hazardous, non-biohazard waste is disposed of following standard waste disposal regulations. This includes general laboratory waste like paper, plastic and non-contaminated materials. Recycling and proper waste disposal practices are typically employed to minimize the environmental impact.

#### 5.2.5 Documentation and Record-Keeping

Comprehensive records are maintained for all waste generated, including the types and quantities of waste, treatment methods employed, and the final disposal process. This documentation is important for regulatory compliance and transparency.

#### **5.2.6 Training and Awareness**

Personnel working in the FSL receive training on proper waste management procedures, safety protocols and the importance of adhering to BSL-2 guidelines. This training ensures that all staff members are aware of and follow best practices in waste handling.

#### **5.2.7 Regulatory Compliance**

The laboratory operates in compliance with local and national regulations governing the handling and disposal of hazardous and biohazardous waste. It also adheres to any international guidelines or standards related to biosafety and waste management.

#### **5.2.8 Continuous Improvement**

Waste management procedures are regularly reviewed and updated to reflect the latest best practices and safety guidelines. The laboratory is committed to staying current with advancements in biosafety and waste management.

## **Chapter 06: Sample Analysis Objectives and Processes**

Just now total 12 types of Treatment technologies are available in the Cox's Bazar district. Technology wise effluent sample collection area is different but maximum cases treatment plants have polishing pond from where effluent sample is collect. Sample collector team of laboratory goes to field and collect samples physically. Then they store the sample in the cooling box and send the samples in the laboratory within 2 to 3 Hours. After receiving samples in the laboratory analysis start and then report forwarding.

Environmental and Public health will be achieved through Pathogen reduction, maintaining oxygen demand, suspended solids and nutrients of effluent. As per DoE Guideline of Bangladesh, for effluent important parameters are pH, Temperature, Biological oxygen demand (BOD), Chemical Oxygen Demand (COD), Total Nitrogen, Nitrate, Phosphate, Total Suspended Solid (TSS) and Total coliform.

#### 6.1 Sample Collection from FSTPs

#### 6.1.1 Sample collection & Measuring Equipment

- ✓ Sampling Device
- ✓ Measuring Mug
- ✓ Sample Collection container
- ✓ Ice Box
- ✓ Ice
- ✓ Hand gloves
- ✓ Mask
- ✓ Apron
- ✓ Gum boot
- ✓ Safety goggles
- ✓ Tissue
- ✓ Disinfectant solution
- ✓ Clean water
- ✓ Soap
- ✓ Garbage Bag
- ✓ Multimeter (pH, Electric Conductivity, Temperature)
- ✓ Handheld GPS tracker
- ✓ Label for sample container leveling

#### 6.1.2 Collect information of sampling location

As per direction of Laboratory Manager, at first sample collector team go to FSTPs for sample collection. Before sampling they fill up a form where include the details information about the FSTPs.

#### **Details Information About Sample**

Sample ID: DPHE-FSL-S-00

Sample Type:

Sample Collection Date:

Sample Collected By:

Organization				
FSM ID				
Technology		Camp No		
Block		Sub Block		
CDS	Latitude			
GPS	Longitude			
WASH AFA	IOM	UNICEF UNHC	R IFRC	Others
FSM's In charge				
Contact Number				
Email				
	Sai	mple's Test Report		
рН		Conductivity		
Temperature		Capacity		m3/week

Note (About FSTP):

- 1. 2.
- 3.
- 4.

#### **Data Collection Sheet for Effluent**

#### 6.1.3 Sample Collection container type

Then they collect samples in the "Whirl Pak" or HDPE Autoclavable container. Use containers that are suitable for the specific analysis and that are made of materials that do not react with or contaminate the sample).

#### 6.1.4 Sample quantity and field analysis

Normally 01 to 1.5 liters sample taken from each FSTPs. pH, Conductivity and temperature are measure in the field and in front of FSTP in charge by the sample collectors during the time of sampling.

#### 6.1.5 Sampling point technology wise

S.N.	Treatment Technology	Sample Type	Collection Point	Collection process
1	Anaerobic Lagoons	Liquid	Polishing Pond	Grab/Composite
2	Lime Stabilization Pond (LSP)	Liquid	Infiltration	Grab/Composite
3	Up Flow Filter (UFF)	Liquid	Polishing Pond	Grab/Composite
4	Anaerobic Baffled Reactor (ABR)	Liquid	Polishing Pond	Grab/Composite
5	Open Deviation Pond (ODP)	Liquid	Infiltration	Grab/Composite
6	Waste Stabilization Pond (WSP)	Liquid	Infiltration	Grab/Composite
7	Solid Separation Unit (SSU)	Liquid	Infiltration	Grab/Composite
8	Decentralized waste water treatment system (DEWATS)	Liquid	Infiltration	Grab/Composite
9	Anaerobic Digester system (ADS)	Liquid	Polishing Pond	Grab/Composite
10	Geo tube Lime	Liquid	Infiltration	Grab/Composite
11	Biological Process (Planted/ Unplanted Drying Bed)	Liquid	Polishing Pond	Grab/Composite
12	Omni Processor	Liquid	Polishing Pond	Grab/Composite

#### Table 01: Treatment Technology wise effluent sample collection point



Lagoon Lime Treatment with dewatering bed









WSP (Waste Stabilization Pond)





#### 6.1.6 Labeling

Properly label each sample with relevant information such as the date and time of collection, location, analysis results and any other specific details required by the laboratory.

#### **6.1.7 Temperature Control**

After that they preserve this sample in the cooling box where temperature is maintained between  $4^{\circ}$ C to  $10^{\circ}$ C to minimize bacterial growth and changes in composition. After collecting all samples from field sampling team send the samples to the laboratory as soon as possible.

#### 6.2 Sample Receive in the Laboratory

After coming sample collector team from field, one lab technician collects the samples from them and entry all information in a log book. Another lab technician immediately starts pathogen testing (Total Coliform and E. coli). After that they do the testing of others parameters.

#### **6.3 Report Publish**

After completing analysis of all parameters, Lab technician prepare "Test Report". After that Laboratory manager check and finalize the report. Then report sent to respective In charge of DPHE and from their analysis report circulates to the respective organizations.



Sample Analysis Process (FSL, DPHE, CXB)

## **Chapter 07: Methodology**

#### 7.1 Sample Collection

The effluent samples are collected from Rohingya Camps along with host community. Sample collection was started from 01 January 2022 and up to 31 December 2023, DPHE-FSL team have covered total 217 FSTPs among which 4 are situated in the host community. Report Based on December 2023, total number of functional FSTPs in Cox's Bazar is 167.

#### 7.2 Sampling Location

Effluent samples are collected from Rohingya camps along with host community.

#### 7.3 Testing Method

Treated Fecal Sludge i.e. effluent were evaluated for various parameters including physical properties (pH, Temperature, Electrical Conductivity, Total Suspended Solids, Total Solids, Settleable Solids, Oil & Grease), Chemical Properties (BOD, COD, Total Nitrogen, Nitrate, Phosphate). The microbiological parameters included Total coliform and E coli.

#### 7.3.1 Spectrophotometric

By Spectrophotometric method Total Nitrogen (TN), Nitrate, Phosphate, Chemical Oxygen demand (COD) is determined. For this determination DR 3900 spectrophotometer and HT200S HACH reactor is use. Before sample analysis every time samples are diluted as per requirement to prevent false high readings associated with turbid solutions.

#### 7.3.2 Electrode

pH, Electrical conductivity (Ec) and Temperature were measured by Electrical conductivity meter or multimeter.

#### 7.3.3 BOD System

For measurement of Biological Oxygen demand (BOD) Lovibond BD 600 series are use. Standard for BOD measurement is at 20<sup>o</sup>C for 5 days (As per DoE guideline of BD). During the time of testing BOD range is measure depending on the value of COD.

#### 7.3.4 Oven Drying

By oven drying method Total suspended solids (TSS) and Total Solids (TS) quantity is measure. Here maintaining temperature is 103 to 105<sup>o</sup>C for one hour.

#### 7.3.5 APHA Method

By this process "Oil & Grease" amount is measure in fecal sludge.

#### 7.3.6 Colony Forming Unit

Compact Dry Ec plate is use for measuring Total Coliform and E coli. Here maintaining temperature is 37<sup>o</sup>C for 24 hours.

#### 7.3.7 AMBIC Method

The Ammonium bicarbonate (AmBic) method- Developed by Hawksworth and Archer (2010) and modified by Archer C. (2012) - Pollution Research Group, University of KwaZulu- Natal, Durban, South Africa is use for checking Helminth Eggs.

## Chapter 08: Standard Operating Procedure (SOP) for Sample collection

#### 8.1 SOP for Sampling's preparation (PPE & Self Hygiene)

Sampling procedure in field label is divided into 3 steps. They are-

- a. Preparation Before sampling.
- b. Process during sampling
- c. After sampling

#### 8.1.1 Preparation before Sampling

Before sampling start wear all personal protective equipment (PPE) in the following sequence.

Before sampling start, take off all type's ornaments, watch, bracelet, ear ring, mobile, pen etc.



Wear Gumboot

Ciean the hands properly by hand Wear Nitrile free Hand sanitizer. Gloves.









Wear body suit or Apron.



Wear Mask.

6





Wear Long hand gloves up to hand wrist.



#### 8.1.2 During Sampling

• It is strictly prohibited for the sampler to touch any of the sanitary paraphernalia (i.e. bleach, alcohol, tissues and hand sanitizers) or any other equipment rather than the sampling equipment.



• When wearing gloves, do not touch the mask, glasses, face or bring the hands near the head area.



• Equipment that had come into contact with the waste material is required to be rinsed with water and bleached after each sampling.



• No smoking, drinking or eating is allowed during the time of sampling, especially while wearing PPE.



• The sampling area must be cleaned before leaving the site. No trash should be left behind.



• The Sample collection box should be labeled with the Biological Hazard sign.



#### 8.1.3 After Sampling

Take off all PPE by following sequence-



#### **8.2 SOP for Sample Collection**

- At first were proper PPE.
- Select sample collection point
- Labeling the sample collection bag.
- Insert sampling rod to the sample collection area.
- Collect sample as such way that there is no any contamination with human body.
- Pour the sample in a beaker.
- Pour the sample from beaker to sample bag.
- Lock the bag containing sample.
- Clean all apparatus by clean water.
- Disinfectant all apparatus by spraying 65 to 70% alcohol (Isopropyl or Ethanol).
- After collecting sample immediately store them in the cooling box and transfer them to the laboratory within short period of time.


## **Chapter 09: Standard Operation Procedure (SOP) for Lab Entrance** (**Biosafety Level 2**)

Step	Details	Picture
a	Enter in the Changing Room	
b	Put on Dedicated Lab Shoe	
с	Keep Mobile in the Designated Place	
d	Collect Handwash from Soap Dispenser without touching Hand	

Step	Details	Picture
e	Wash Hand at least 20 seconds	
f	Collect Lab Coat	
g	Wear Lab Coat	
h	Enter in the main laboratory	

# **Chapter 10: Standard Operating Procedure (SOP) for Sample Analysis**

## 10.1 SOP for testing "pH & Electric Conductivity (Ec)

DPHE		Quality Management System		
DPHE-FSL-QMS-SOP-T-001		Title: Standard Operating Procedure (Testing)		
Issue No.	Issue Date	Effective Date Rev. No. Rev. Da		Rev. Date
01	01/12/2021		01	01.12.2021

#### ✤ Instruments:

- ✓ pH & Conductivity meter
- ✓ Beaker
- ✓ Autoclave

#### Personal Protective Equipment (PPE):

- ✓ Gloves
- ✓ Goggles
- ✓ Mask
- ✓ Lab Coat (Body fitting)
- ✓ Safety Shoe

#### \* <u>Reagent:</u>

- ✓ Buffer Solution pH 4 and 7.
- ✓ Buffer Solution 1413 µS/cm.
- ✓ Distill water
- ✓ KCl Solution 3M

#### ✤ <u>Procedure:</u>

- a. At first check the pH with a buffer solution. If it comes in range then continue measuring of sample's pH, if bad calibrate the pH device.
- b. For pH calibration selects "pH CALIBRATION".
- c. Then select 'Start Calibration".
- d. Then calibrate it with buffer solution 4 and 7.
- e. Clean the electrode of pH by distills water.
- f. Similarly, for conductivity check the conductivity by buffer solution. If result ok then continues, otherwise calibrate the devise.
- g. For conductivity calibration selects "EC Calibration".
- h. Select "Start Calibration".
- i. Now calibrate it with standard  $1413 \,\mu S$  solution.
- j. Clean the electrode of conductivity by distills water.

- k. Now pH+Ec measuring device is ready for use.
- 1. Pour sample in the sample beaker.
- m. Sink electrode of pH and conductivity into the sample.
- n. Select "pH+Ec MEASURE" in the machine.
- o. Select "Start measurement".
- p. If necessary put the name of sample.
- q. After that measuring have started.
- r. After establishing, take the reading from the machine.
- s. Not the data in the log book.
- t. Now get out the electrodes from the sample.
- u. Clean both electrode by Distill water and dry them by a clean tissue paper.
- v. Sink the pH electrode into the Potassium Chloride (KCl) solution.

#### ✤ Calibration's Image:







\* <u>Testing's Image:</u>



sensION"+ MM374	sensION"+ MM374		
Identification Sample? dphg 3	MEASURING by stability pH 7.32 26.3°C 829 µS/cm @25°C	Harris	
(HACH)	00:44		
(0,0)	HACH		

#### ✤ <u>Cleaning Procedure:</u>

- ✓ After finishing laboratory work thoroughly wipe down surfaces properly by tissue paper.
- $\checkmark$  Then wipe the surface by disinfectant solution so that there are no any germs in the surface.
- $\checkmark$  Clean all the apparatus using for testing by liquid soap.
- ✓ Now autoclave all the apparatus and after finishing autoclaving keep the apparatus in their dedicated places.

#### ✤ <u>Waste Management:</u>

- a. After finishing testing keep all waste in the dustbin as per following ways-
- ✓ <u>Red Dustbin:</u>

Highly Hazardous, Biohazard and Toxic elements.

✓ <u>Yellow Dustbin:</u>

Paper & Glass bottle (After Autoclave).

✓ <u>Blue Dustbin:</u>

Plastic wrappers and non-biodegradable wastes (After Autoclave).



b. Keeping wastage will be collected by the municipality department to treat these as per government standard of Bangladesh.

## **10.2 SOP for testing Temperature (T)**

DPHE		Quali	ty Management System	1
DPHE-FSL-QMS-SOP-T-002		Title: Standard Operating Procedure (Testing)		
Issue No.	Issue Date	Effective Date	Rev. No.	Rev. Date
01	01/12/2021		01	01.12.2021

#### ✤ <u>Instrument:</u>

- ✓ Beaker
- ✓ Thermometer

#### ✤ <u>Personal Protective Equipment (PPE):</u>

- ✓ Gloves
- ✓ Goggles
- ✓ Mask
- ✓ Lab Coat (Body fitting)
- ✓ Safety Shoe

#### **\*** <u>Procedure (For Dilute Solution):</u>

- a. Perform a pre-use calibration of the temperature probe.
- b. Collect the sample in the HDPE container.
- c. Pour samples into a beaker.
- d. Immerse the temperature probe into the waste water sample, ensuring that the sensing element is fully submerged.
- e. Allow the temperature reading to stabilize, typically within 30 seconds to 1 minute.
- f. Record the temperature reading in degrees Celsius or Fahrenheit, as per the project requirements.
- g. If precision is crucial, take multiple measurements at different locations within the waste water sample.
- h. Record all measurements and calculate the average temperature.



### \* <u>Cleaning Procedure:</u>

a. After completing the temperature measurement, carefully remove the temperature probe from the waste water sample, taking care not to spill any residue.

- b. Rinse the temperature probe with clean water to remove any visible contaminants or debris.
- c. Immerse the temperature probe in a mild disinfectant solution for a recommended contact time (as specified by the disinfectant manufacturer).
- d. Gently agitate the probe in the disinfectant solution to ensure thorough coverage.
- e. Rinse the temperature probe with distilled water to remove any remaining disinfectant residue.
- f. Wipe the temperature probe dry with a clean, lint-free cloth.
- g. Pay special attention to the sensing element
- h. Store the temperature probe in a clean and dry environment to prevent contamination before the next use.
- i. If applicable, store the probe in its protective case to avoid damage.

#### \* <u>Waste Management:</u>

- ✓ Dispose of the wastewater samples in accordance with local regulations and environmental guidelines.
- ✓ Do not discharge untreated or unapproved wastewater directly into the environment, storm water drains or surface water bodies.

## **10.3 SOP for testing Biological Oxygen Demand (BOD)**

DPHE		Quali	ty Management System	1
DPHE-FSL-QMS-SOP-T-003		Title: Standard Operating Procedure (Testing)		
Issue No.	Issue Date	Effective Date	Rev. No.	Rev. Date
01	01/12/2021		01	01.12.2021

#### ✤ Instruments:

- ✓ BOD measuring system.
- ✓ Incubator.
- ✓ Pipette
- ✓ Balance
- ✓ Autoclave

#### ✤ <u>Personal Protective Equipment (PPE):</u>

- ✓ Gloves
- ✓ Goggles
- ✓ Mask
- ✓ Lab Coat (Body fitting)
- ✓ Safety Shoe

### \* <u>Reagent:</u>

- ✓ Nitrification inhibitor
- ✓ Potassium Hydroxide Solution (45%)
- ✓ Diluted Hydrochloric acid (1 M)
- ✓ Sodium Hydroxide solution (1 M)

#### \* <u>Selection of Sample Volume:</u>

- ✓ The BOD value of the sample to be expected determines the volume used. A BOD measurement range (without dilution of the sample) from 0 4000 mg/L arises from this.
- ✓ BOD range will be determined by seeing the value of COD (BOD value is 80% of COD). In that case measure the COD at first before measuring BOD.
- $\checkmark$  If the value range becomes more than 4000 then dilute stock solution as per dilution factor.

BOD Range (mg/L)	Sample Volume (ml)	Nitrification inhibitor ATH dosage (Drops)
0-40	428	10
0-80	360	10
0-200	244	5
0-400	157	5
0-800	94	3
0-2000	56	3
0-4000	21.7	1

#### \* <u>Preparation of Water Sample:</u>

- a. After dilution at first test the pH value of the waste water sample. Ideal pH value is 6.5 to 7.5.
- b. If pH value is too high or too low then it will be neutralized by diluted hydrochloric acid or diluted sodium hydroxide solution.
- c. Measure exact necessary sample quantity (From above chart) with the appropriate overflow volumetric flask and add to the test bottle.
- d. Specified sample volumes must be filled very precisely. Otherwise major measurement errors can occur.
- e. Add nitrification inhibitor drop wise in the test bottle.
- f. Add a clean magnetic stir bar to the test bottle and fill the seal cup with 3-4 drops of 45% Potassium Hydroxide Solution (For binding of the CO<sub>2</sub>). Here should be aware that no contact of sample with 45% KOH solution.
- g. Then place the seal up in the test bottle.

#### **\* Procedure for BOD measuring system:**

- a. At first give connection of power line and magnetic plate.
- b. Start measurement by pressing "Power Button".
- c. Instrument menu appears after initial booting.
- d. Use "up & down" arrow keys to navigate through the menu.
- e. Select "Start Press Series".
- f. Then check the sensor, sample volume and measurement duration of every BOD bottles.
- g. Red light indicates that the sensor head is well connected to the unit.
- h. After completing setting press "function F2" to start experiment and keep it inside incubator (At 20°C for 5 Days).
- i. Enter "Display Test Series" which will provide time to time test result along with graph.
- j. Sample should be measure in triplicates and in different ranges.





#### **♦** Instruction for evaluation of results:

- a. The BOD measurement values must always be higher than on the preceding day.
- b. The values don't increase linearly. The increase is always smaller than on the preceding day.
- c. If the values increase linearly the sample has a higher BOD value than was to be expected when the sample was prepared.
- d. If the BOD measurement values should suddenly rise drastically during the measurement, it may be an indication of nitrification.
- e. If the BOD measurement values fall during the measurement, the system may have become leaky.





#### \* <u>Cleaning Procedure:</u>

- a. After finishing laboratory work thoroughly wipe down surfaces properly by tissue paper.
- b. Then wipe the surface by disinfectant solution so that there are no any germs in the surface.
- c. Clean all the apparatus using for testing by liquid soap.
- d. Now autoclave all the apparatus and after finishing autoclaving keep the apparatus in their dedicated places.

#### \* <u>Calibration Procedure:</u>

Every 4 months or after completing one round (Sample collection) BOD system should be calibrated. The following procedure should be follow-

At first place a tablet (BOD CM1) in the BOD bottle, start the measurement process.

- $\checkmark$  Read off the BOD value after 5 days.
- $\checkmark$  Compare the getting value with the defined value.
- $\checkmark$  If this value is within the quoted tolerance, this means that the BOD measuring system is functioning correctly.

#### ✤ <u>Waste Management:</u>

1. After finishing testing keep all waste in the dustbin as per following ways-

#### ✓ <u>Red Dustbin:</u>

Highly Hazardous, Biohazard and Toxic elements.

#### ✓ <u>Yellow Dustbin:</u>

Paper & Glass bottles (After Autoclave).

#### ✓ <u>Blue Dustbin:</u>

Plastic wrappers and non-biodegradable wastes (After Autoclave).



2. Keeping wastage will be collected by the municipality department to treat these as per government standard of Bangladesh.

## **10.4 SOP for testing Total Suspended Solids (TSS)**

DPHE		Quality Management System		
DPHE-FSL-QMS-SOP-T-004		Title: Standard Operating Procedure (Testing)		
Issue No.	Issue Date	Effective Date	Rev. No.	Rev. Date
01	01/12/2021		01	01.12.2021

#### ✤ <u>Instrument:</u>

- ✓ Beaker
- ✓ Weighing Scale (Close Door)
- ✓ Glass fiber filter
- ✓ Forcep
- ✓ Filtration
- ✓ Oven
- ✓ Autoclave

#### Personal Protective Equipment (PPE):

- ✓ Gloves
- ✓ Goggles
- ✓ Mask
- ✓ Lab Coat (Body fitting)
- ✓ Safety Shoe

#### \* <u>Reagents:</u>

✓ Distill water

#### **\*** <u>Procedure (For Dilute Solution):</u>

- a. Collect the sample in the HDPE container.
- b. Weight the aluminum pan in the weight scale and press "Tare" to make its weight zero.
- c. Now take weight of the filter paper and recode it.
- d. Take the sample into the stomacher bag and homogenize it in the stomacher.
- e. Connect the vacuum pump to the side arm of our vacuum flask.
- f. Place a filter paper in the filter holder, wet it with a small amount of reagent water to seat it, and secure the funnel to the base.
- g. Filter as much sample as is possible. While the sample is filtering, record the pan ID and initial weight from the label on the pan. Record the total sample volume filtered.
- h. After completing filtering use distill water to wash the container and again filter it so that no residuals remain into the container.
- i. Place the filter back in its pan and place in a drying oven set at 105°C for at least one hour.
- j. Remove filters/pans from the oven and place in a desiccator until they reach room temperature.
- k. Weigh each filter on a balance and record the weight. (Do not include the pan in this step).
- 1. Calculate the result with the following equation.

## **\*** <u>Procedure (For Heavy Thick Sample):</u>

- a. Weight the aluminum pan in the weight scale.
- b. Take a certain amount of sample (Measuring by measuring cylinder in ml) in the aluminum pan and weight it. By deducting above value from this value, we will get the sample's weight.
- c. After taking weight keep the sample along with aluminum pan in a drying oven set at 105°C for at least one hour.
- d. After one hour getting out the pan from oven and keep it in the desiccator until they reach room temperature.
- e. Now weight the dried sample and calculate the result by the following samples.

#### ✤ <u>Calculation:</u>

$$TSS\left(\frac{mg}{L}\right) = \frac{Final wt(g) - Initial wt(g) * 1000000}{sample Volume(ml)}$$





#### \* <u>Cleaning Procedure:</u>

- ✓ After finishing laboratory work thoroughly wipe down surfaces properly by tissue paper.
- $\checkmark$  Then wipe the surface by disinfectant solution so that there are no any germs in the surface.
- $\checkmark$  Clean all the apparatus using for testing by liquid soap.
- ✓ Now autoclave all the apparatus and after finishing autoclaving keep the apparatus in their dedicated places.

#### ✤ <u>Waste Management:</u>

- 1. After finishing testing keep all waste in the dustbin as per following ways-
- ✓ <u>Red Dustbin:</u>

Highly Hazardous, Biohazard and Toxic elements.

✓ <u>Yellow Dustbin:</u>

Paper & Glass bottle (After Autoclave).

#### ✓ <u>Blue Dustbin:</u>

Plastic wrappers and non-biodegradable wastes (After Autoclave).



2. Keeping wastage will be collected by the municipality department to treat these as per government standard of Bangladesh.

## **10.5 SOP for testing Total Solids (TS)**

DPHE		Quali	ty Management System	1
DPHE-FSL-QMS-SOP-T-005		Title: Standard Operating Procedure (Testing)		
Issue No.	Issue Date	Effective Date Rev. No. Rev. Da		Rev. Date
01	01/12/2021		01	01.12.2021

#### ✤ <u>Instrument:</u>

- i. Beaker
- j. Weighing Scale (Close Door)
- k. Crucible or Aluminum pan
- l. Oven
- m. Autoclave

#### ✤ <u>Personal Protective Equipment (PPE):</u>

- ✓ Gloves
- ✓ Goggles
- ✓ Mask
- ✓ Lab Coat (Body fitting)
- ✓ Safety Shoe

#### **\*** <u>Procedure (For Dilute Solution):</u>

- a. Collect the sample in the HDPE container.
- b. Weight the aluminum pan in the weight scale and press "Tare" to make its weight zero.
- c. Take the sample into the stomacher bag and homogenize it in the stomacher.
- d. Pour 1 to 3 gm wet samples on the pan and record the weight.
- e. Place the wet sample into the oven and set at 105°C for at least one hour.
- f. Remove pans from the oven and place in a desiccator until they reach room temperature.
- g. Weigh dry sample on a balance and record the weight. (Do not include the pan in this step).
- h. Calculate the result with the following equation

#### ✤ <u>Calculation:</u>

$$TS(\%) = \frac{Final \ sample \ wt(g) - Initial \ sample \ wt(g) * 100}{sample \ Volume \ (ml)}$$



## \* <u>Cleaning Procedure:</u>

- ✓ After finishing laboratory work thoroughly wipe down surfaces properly by tissue paper.
- $\checkmark$  Then wipe the surface by disinfectant solution so that there are no any germs in the surface.
- $\checkmark$  Clean all the apparatus using for testing by liquid soap.
- ✓ Now autoclave all the apparatus and after finishing autoclaving keep the apparatus in their dedicated places.

#### \* <u>Waste Management:</u>

- 1. After finishing testing keep all waste in the dustbin as per following ways-
- ✓ <u>Red Dustbin:</u>

Highly Hazardous, Biohazard and Toxic elements.

 ✓ <u>Yellow Dustbin:</u> Paper & Glass bottle (After Autoclave).

## ✓ <u>Blue Dustbin:</u>

Plastic wrappers and non-biodegradable wastes (After Autoclave).



1. Keeping wastage will be collected by the municipality department to treat these as per government standard of Bangladesh.

## **10.6 SOP for testing Settleable Solids (SS)**

DPHE		Quality Management System		
DPHE-FSL-QMS-SOP-T-006		Title: Standard Operating Procedure (Testing)		
Issue No.	Issue Date	Effective Date	Rev. No.	Rev. Date
01	01/12/2021		01	01.12.2021

#### ✤ <u>Instrument:</u>

- ✓ Imhoff Cone
- ✓ Imhoff Cone Stand
- ✓ Timer
- ✓ Autoclave

#### Personal Protective Equipment (PPE):

- ✓ Gloves
- ✓ Goggles
- ✓ Mask
- ✓ Lab Coat (Body fitting)
- ✓ Safety Shoe

#### ✤ <u>Procedure:</u>

- a. After collecting sample from cooling box or refrigerator allow the sample to become warm at room temperature.
- b. Thoroughly mix 01 liter sample in a bottle by shaking hand.
- c. Decant 01 Liter sample in the Imhoff Cone.
- d. Allow the sample to settle for 45 minutes.
- e. The solids will start to settle down.
- f. After 45 minutes turn the cone forward and beck to remove material on the slopped side of the cone.
- g. Allow the solid another 15 minutes for settle.
- h. After 01 Hour (45 minutes + 15 minutes) read the amount of Settleable solids at the bottom of the cone.
- i. Result will be recorded as ml/L

#### Test procedure





 Fill an Imhoff cone to the 1-L mark with a mixed sample.

 Do not move for
Turn the cone forward and back to remove materials on the sloped side of the cone.





 Do not move for
minutes to let the sample become stable.

5. Read the graduated scale on the Imhoff cone at the top of the solids layer as mL/L settleable matter.

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#### \* <u>Cleaning Procedure:</u>

- ✓ After finishing laboratory work thoroughly wipe down surfaces properly by tissue paper.
- $\checkmark$  Then wipe the surface by disinfectant solution so that there are no any germs in the surface.
- $\checkmark$  Clean all the apparatus using for testing by liquid soap.
- ✓ Now autoclave all the apparatus and after finishing autoclaving keep the apparatus in their dedicated places.

#### ✤ <u>Waste Management:</u>

2. After finishing testing keep all waste in the dustbin as per following ways-

#### ✓ <u>Red Dustbin:</u>

Highly Hazardous, Biohazard and Toxic elements.

#### ✓ <u>Yellow Dustbin:</u>

Paper & Glass bottle (After Autoclave).

✓ <u>Blue Dustbin:</u>

Plastic wrappers and non-biodegradable wastes (After Autoclave).



3. Keeping wastage will be collected by the municipality department to treat these as per government standard of Bangladesh.

## 10.7 SOP for testing "Oil & Grease"

DPHE		Quality Management System		
DPHE-FSL-QMS-SOP-T-007		Title: Standard Operating Procedure (Testing)		
Issue No.	Issue Date	Effective Date	Rev. No.	Rev. Date
01	01/11/2023		0	01.11.2023

#### ✤ <u>Instrument:</u>

- n. Beaker
- o. Weighing Scale (Close Door)
- p. Crucible or Aluminum pan
- q. Separating Funnel
- r. Measuring Cylinder
- s. Pipette
- t. Oven
- u. Autoclave

#### \* <u>Reagent:</u>

- v. Sulphuric Acid (0.2 N)
- w. Petroleum Ether

#### Personal Protective Equipment (PPE):

- ✓ Gloves
- ✓ Goggles
- ✓ Mask
- ✓ Lab Coat (Body fitting)
- ✓ Safety Shoe

#### **\*** <u>Procedure (For Dilute Solution):</u>

- a. Collect the sample in the HDPE container.
- b. Take 25ml Samples in the separating funnel.
- c. Add 01 ml Sulphuric acid to remove suspended solids.
- d. Add 5 ml petroleum ether into the solution.
- e. Shake the separating funnel properly and open the lid to come out gas.
- f. Now keep the separating funnel 15 to 20 minutes.
- g. After a while we will able to see two layers (Upper side oil & Grease layer and bottom side aqueous layer).
- h. Now using the stopper of funnel, we will separate the aqueous part.
- i. We will keep the funnel 10 minutes more to separate extra water from the mixture.
- j. Again separate the aqueous layer.
- k. Weight a dry crucible or aluminum pan (W<sub>1</sub>).
- 1. Now collect the "Oil & Grease" sample in the crucible or aluminum pan.
- m. Weight the "Oil & Grease" with pan (W<sub>2</sub>).
- n. Now dry the samples in the oven for 2 hours at  $105^{\circ}$ C.

- o. After drying keep the sample in the desiccator and let it come at room temperature.
- p. Now again weight and calculate as per following equation.

## \* <u>Calculation:</u>

 $\textit{Oil & Grease} \ (\frac{mg}{L}) = \frac{\textit{Final sample wt} \ (g) - \textit{Initial sample wt} \ (g) * 1000000}{\textit{sample Volume (ml)}}$ 





## \* <u>Cleaning Procedure:</u>

- ✓ After finishing laboratory work thoroughly wipe down surfaces properly by tissue paper.
- $\checkmark$  Then wipe the surface by disinfectant solution so that there are no any germs in the surface.
- ✓ Clean all the apparatus using for testing by liquid soap.
- ✓ Now autoclave all the apparatus and after finishing autoclaving keep the apparatus in their dedicated places.

## ✤ <u>Waste Management:</u>

- 4. After finishing testing keep all waste in the dustbin as per following ways-
- ✓ <u>Red Dustbin:</u> Highly Hazardous, Biohazard and Toxic elements.
- Yellow Dustbin: Paper & Glass bottle (After Autoclave).
- ✓ <u>Blue Dustbin:</u> Plastic wrappers and non-biodegradable wastes (After Autoclave).



2. Keeping wastage will be collected by the municipality department to treat these as per government standard of Bangladesh.

## **10.8 SOP for testing Chemical Oxygen Demand (COD)**

DPHE		Quality Management System		
DPHE-FSL-QMS-SOP-T-008		Title: Standard Operating Procedure (Testing)		
Issue No.	Issue Date	Effective Date	Rev. No.	Rev. Date
01	01/12/2021		01	01.12.2021

#### ✤ Instruments:

- ✓ Reactor
- ✓ Spectrophotometer (HACH DR 3900)
- ✓ Pipette
- ✓ Autoclave

#### ✤ <u>Personal Protective Equipment (PPE):</u>

- ✓ Gloves
- ✓ Goggles
- ✓ Mask
- ✓ Lab Coat (Body fitting)
- ✓ Safety Shoe

#### \* <u>Reagent:</u>

- ✓ LCI400
- ✓ LCK 014
- ✓ TNT 822
- ✓ TNT 825

#### **Procedure (Sample Preparation) for range LCI 400/TNT 822/TNT825:**

- a. At first take a Vial.
- b. Shake well for mixing chemical 'A" & "B".
- c. Add 2 ml sample in the vial.
- d. Close the lid tightly.
- e. Shake properly.
- f. Heat the vial for 2 Hours at  $148^{\circ}$ C.
- g. After heating allow the vial to come at room temperature.
- h. Before vial come to room temperature shake the vial twice whine it is still hot (Approx.  $60^{\circ}$ C).

#### ✤ Procedure (Sample Preparation) for LCK 014:

- a. At first take a Vial.
- b. Shake well for mixing chemical 'A" & "B".
- c. Add 0.5 ml sample in the vial.
- d. Close the lid tightly.
- e. Shake properly.
- f. Heat the vial for 2 Hours at 148°C.

- g. After heating allow the vial to come at room temperature.
- h. Before vial come to room temperature shake the vial twice whine it is still hot (Approx.  $60^{\circ}$ C).

#### **\*** <u>Procedure (Spectrophotometer):</u>

- a. Switch "ON" the spectrophotometer.
- b. Now clean the vial's outside properly by tissue and places it in the hole of spectrophotometer.
- c. The Spectrophotometer will automatically take the reading.
- d. The result is shown in the display.



#### \* Cleaning Procedure:

- ✓ After finishing laboratory work thoroughly wipe down surfaces properly by tissue paper.
- $\checkmark$  Then wipe the surface by disinfectant solution so that there are no any germs in the surface.
- $\checkmark$  Clean all the apparatus using for testing by liquid soap.
- ✓ Now autoclave all the apparatus and after finishing autoclaving keep the apparatus in their dedicated places.

#### ✤ <u>Waste Management:</u>

After measuring give cross sign into the vial and keep it in the packet.



- ✓ If possible returned the used vial back to the supplier, otherwise destroy it as per SOP of Waste management (As per Government guideline).
- $\checkmark$  If recycling not possible, in that case put them in the landfill by making a concrete block.



- $\checkmark$  After finishing testing keep all waste in the dustbin as per following ways-
- ✓ <u>Red Dustbin:</u>

Highly Hazardous, Biohazard and Toxic elements.

- ✓ <u>Yellow Dustbin:</u> Paper & Glass bottles (After Autoclave).
  ✓ Phone Dustbin:
- ✓ <u>Blue Dustbin:</u> Plastic wrappers and non-biodegradable wastes (After Autoclave).



✓ Keeping wastage will be collected by the municipality department to treat these as per government standard of Bangladesh.

## **10.9 SOP for testing Total Nitrogen**

DPHE		Quality Management System		
DPHE-FSL-QMS-SOP-T-009		Title: Standard Operating Procedure (Testing)		
Issue No.	Issue Date	Effective Date	Rev. No.	Rev. Date
01	01/12/2021		01	01.12.2021

#### ✤ Instruments:

- ✓ Reactor
- ✓ Spectrophotometer (HACH DR 3900)
- ✓ Pipette
- ✓ Autoclave

#### Personal Protective Equipment (PPE):

- ✓ Gloves
- ✓ Goggles
- ✓ Mask
- ✓ Lab Coat (Body fitting)
- ✓ Safety Shoe

#### \* <u>Reagent:</u>

- ✓ TNT 828
- ✓ TNT 826

#### **\*** <u>Procedure (Sample Preparation) for TNT 826:</u>

- a. At first take 1.3 ml sample in the empty reaction tube.
- b. Then add 1.3 ml Chemical A (Sodium Hydroxide) into the reaction tube.
- c. Add 01 pc Oxidant tablet B.
- d. Close immediately the reaction tube.
- e. Heat the reaction tube at "HT mode" for 30 minutes at 120°C.
- f. After heating allow the tube to become cool at room temperature.
- g. Shake the tube.
- h. From reaction tube take 0.5 ml sample to the vial.
- i. Add 0.2 ml "D" solution.
- j. Close the lid and shake it well.
- k. Keep the vial for 15 minutes.

#### ✤ Procedure (Sample Preparation) for TNT 828:

- a. At first take 0.2 ml sample in the empty reaction tube.
- b. Then add 2.3 ml Chemical A (Sodium Hydroxide) into the reaction tube.
- c. Add 01 pc Oxidant tablet B.
- d. Close immediately the reaction tube.
- e. Heat the reaction tube at "HT mode" for 30 minutes at 120°C.

- f. After heating allow the tube to become cool at room temperature.
- g. Shake the tube.
- h. From reaction tube take 0.5 ml sample to the vial.
- i. Add 0.2 ml "D" solution.
- j. Close the lid and shake it well.
- k. Keep the vial for 15 minutes.

#### **\*** <u>Procedure (Spectrophotometer):</u>

- a. Switch "ON" the spectrophotometer.
- b. Now clean the vial's outside properly by tissue and places it in the hole of spectrophotometer.
- c. The Spectrophotometer will automatically take the reading.
- d. The result is shown in the display.





#### \* Cleaning Procedure:

- a. After finishing laboratory work thoroughly wipe down surfaces properly by tissue paper.
- b. Then wipe the surface by disinfectant solution so that there are no any germs in the surface.
- c. Clean all the apparatus using for testing by liquid soap.
- d. Now autoclave all the apparatus and after finishing autoclaving keep the apparatus in their dedicated places.

#### ✤ <u>Waste Management:</u>

a. After measuring give a cross signs into the vial and keep it in the packet.



- b. If possible returned the used vial back to the supplier, otherwise destroy it as per SOP of Waste management (As per Government guideline).
- c. If recycling not possible, in that case put them in the landfill by making a concrete block.



- d. After finishing testing keep all waste in the dustbin as per following ways-
- ✓ <u>Red Dustbin:</u> Highly Hazardous, Biohazard and Toxic elements.

 ✓ <u>Yellow Dustbin:</u> Paper & Glass bottles (After Autoclave).

✓ <u>Blue Dustbin:</u>

Plastic wrappers and non-biodegradable wastes (After Autoclave).



e. Keeping wastage will be collected by the municipality department to treat these as per government standard of Bangladesh.

## 10.10 SOP for testing Nitrate (NO<sub>3</sub><sup>-</sup>)

DPHE		Quality Management System		
DPHE-FSL-QMS-SOP-T-010		Title: Standard Operating Procedure (Testing)		
Issue No.	Issue Date	Effective Date	Rev. No.	Rev. Date
01	01/12/2021		01	01.12.2021

#### ✤ Instruments:

- ✓ Spectrophotometer (HACH DR 3900)
- ✓ Pipette
- ✓ Autoclave

#### Personal Protective Equipment (PPE):

- ✓ Gloves
- ✓ Goggles
- ✓ Mask
- ✓ Lab Coat (Body fitting)
- ✓ Safety Shoe

#### ✤ <u>Reagent:</u>

- ✓ <u>LCK 339 or TNT 835</u>
- ✓ 0.23-13.50 mg/L NO<sub>3</sub>-N
- ✓ 01-60 mg/L NO<sub>3</sub> Or
- ✓ <u>LCK 340</u>
- ✓ 5.0-35.0 mg/L NO<sub>3</sub>-N
- ✓ 22.0-155.0 mg/L NO<sub>3</sub>

#### **Procedure (Sample Preparation) for LCK339 or TNT 835:**

- a. At first open the lid of the vial.
- b. Add slowly 1 ml sample in the vial.
- c. Add 0.2 ml Chemical "A" in the vial.
- d. Shake it well until the solutions are fully mixed
- e. Measure after approx. 15 minutes. Do not wait several minutes longer.

#### **\*** <u>Procedure (Sample Preparation) for LCK340:</u>

- a. At first open the lid of the vial.
- b. Add slowly 0.2 ml sample in the vial.
- c. Add 1 ml Chemical "A" in the vial.
- d. Shake it well until the solutions are fully mixed.
- e. Measure after approx. 15 minutes. Do not wait several minutes longer.

#### $\div$ **Procedure (Spectrophotometer):**

- a. Switch "ON" the spectrophotometer.
- b. Now clean the vial's outside properly by tissue and places it in the hole of spectrophotometer.
- c. The Spectrophotometer will automatically take the reading.
- d. The result is shown in the display.
- The result is shown in the display. NO<sub>3</sub>-N e.
- Convert readings into NO<sub>3</sub>. f.
- We can also change the unit of spectrophotometer by NO<sub>3</sub> and can get direct result. g.





#### \* Calculation:

DoE Guidelines for Nitrate in effluent: :  $50 \text{ mg/ l } \text{NO}_3^{2-1}$ Weight of 1 Mol of  $NO_3^{2-}$ = 62 g Weight of 1 Mol of Nitrogen = 14 g

#### **Conversion:**

 $50 \text{ mg}/1 \text{ NO}_3^2$  equals (50 / 62) x 14 = 11.29 mg / 1 NO\_3^2 - N

*Nitrate* 
$$\left(\frac{mg}{l}\right) = \left(\frac{Shown \, Result}{14}\right) * 62$$

#### \* <u>Cleaning Procedure:</u>

- ✓ After finishing laboratory work thoroughly wipe down surfaces properly by tissue paper.
- $\checkmark$  Then wipe the surface by disinfectant solution so that there are no any germs in the surface.
- $\checkmark$  Clean all the apparatus using for testing by liquid soap.
- ✓ Now autoclave all the apparatus and after finishing autoclaving keep the apparatus in their dedicated places.

#### ✤ <u>Waste Management:</u>

1. After measuring give a cross signs into the vial and keep it in the packet.



- 2. If possible returned the used vial back to the supplier, otherwise destroy it as per SOP of Waste management (As per Government guideline).
- 3. If recycling not possible, in that case put them in the landfill by making a concrete block.





4. After finishing testing keep all waste in the dustbin as per following ways-

#### ✓ <u>Red Dustbin:</u>

Highly Hazardous, Biohazard and Toxic elements.
# ✓ <u>Yellow Dustbin:</u>

Paper & Glass bottles (After Autoclave).

# ✓ <u>Blue Dustbin:</u>

Plastic wrappers and non-biodegradable wastes (After Autoclave).

5. Keeping wastage will be collected by the municipality department to treat these as per government standard of Bangladesh.



# **10.11 SOP for testing Phosphate (PO4<sup>3-</sup>)**

DPHE		Quality Management System		
DPHE-FSL-QMS-SOP-T-011		Title: Standard Operating Procedure (Testing)		
Issue No.	Issue Date	Effective Date	Rev. No.	Rev. Date
01	01/12/2021		01	01.12.2021

## ✤ <u>Instruments:</u>

- ✓ Reactor
- ✓ Spectrophotometer (HACH DR 3900)
- ✓ Pipette
- ✓ Autoclave

# Personal Protective Equipment (PPE):

- ✓ Gloves
- ✓ Goggles
- ✓ Mask
- ✓ Lab Coat (Body fitting)
- ✓ Safety Shoe

## \* <u>Reagent:</u>

- ✓ <u>LCK 349 or TNT 843</u>
- ✓ 0.05-1.50 mg/L PO₄-P
- ✓ 0.15-4.50 mg/L PO<sub>4</sub>
  - Or
- ✓ <u>LCK 350</u>
- ✓ 2.0-20.0 mg/L PO<sub>4</sub>-P
- ✓ 6.0-60.0 mg/L PO<sub>4</sub>

## **\*** <u>Total Phosphate testing procedure of LCK349 or TNT 843 (Sample Preparation):</u>

- a. Unscrew the lid.
- b. Add 2 ml sample in the reaction tube.
- c. Add 0.2 ml Chemical "B" in the reaction tube immediately and close with dosi Cap "C" tightly. We don't need the first dosi cap anymore.
- d. Shake it well until chemical "B" fully dissolves.
- e. Measure after approx. 10 minutes. Do not wait several minutes longer.

## **\*** <u>Total Phosphate testing procedure by LCK350 (Sample Preparation):</u>

- a. Unscrew the lid.
- b. Add 0.4 ml sample in the reaction tube.
- c. Add 0.5 ml Chemical "B" in the reaction tube immediately and close with dosi Cap "C" tightly. We don't need the first dosi cap anymore.
- d. Shake it well until chemical "B" fully dissolves.

e. Measure after approx. 10 minutes. Do not wait several minutes longer.



#### **\*** <u>Total Phosphorous testing procedure by LCK349 (Sample Preparation):</u>

- a. At first open the lid foil of the reaction tube.
- b. Unscrew the lid.
- c. Add 2 ml sample in the reaction tube.
- d. Add the lid again but upside-down.
- e. Shake well.
- f. Heat the reaction tube in "HT mode" for 15 minutes.
- g. After heating, allow the reaction tube to become cool at room temperature.
- h. Shake well again.
- i. Add 0.2 ml Chemical "B" in the reaction tube immediately and close with dosi Cap "C" tightly. We don't need the first dosi cap anymore.
- j. Shake it well until chemical "B" fully dissolves.
- k. Measure after approx. 10 minutes. Do not wait several minutes longer.

#### **\*** <u>Total Phosphorous testing procedure by LCK350 (Sample Preparation):</u>

- a. At first open the lid foil of the reaction tube.
- b. Unscrew the lid.
- c. Add 0.4 ml sample in the reaction tube.
- d. Add the lid again but upside-down.
- e. Shake well.
- f. Heat the reaction tube in "HT mode" for 15 minutes.

- g. After heating, allow the reaction tube to become cool at room temperature.
- h. Shake well again.
- i. Add 0.5 ml Chemical "B" in the reaction tube immediately and close with dosi Cap "C" tightly. We don't need the first dosi cap anymore.
- j. Shake it well until chemical "B" fully dissolves.
- k. Measure after approx. 10 minutes. Do not wait several minutes longer.



#### \* <u>Cleaning Procedure:</u>

- ✓ After finishing laboratory work thoroughly wipe down surfaces properly by tissue paper.
- $\checkmark$  Then wipe the surface by disinfectant solution so that there are no any germs in the surface.
- $\checkmark$  Clean all the apparatus using for testing by liquid soap.
- ✓ Now autoclave all the apparatus and after finishing autoclaving keep the apparatus in their dedicated place.

#### ✤ <u>Waste Management:</u>

1. After measuring give a cross signs into the vial and keep it in the packet.



- 2. If possible returned the used vial back to the supplier, otherwise destroy it as per SOP of Waste management (As per Government guideline).
- 3. If recycling not possible, in that case put them in the landfill by making a concrete block.



4. After finishing testing keep all waste in the dustbin as per following ways-

# ✓ <u>Red Dustbin:</u> Highly Hazardous, Biohazard and Toxic elements.

✓ <u>Yellow Dustbin:</u>

Paper & Glass bottles (After Autoclave).

# ✓ <u>Blue Dustbin:</u>

Plastic wrappers and non-biodegradable wastes (After Autoclave).



5. Keeping wastage will be collected by the municipality department to treat these as per government standard of Bangladesh.

# 10.12 SOP for testing Escherichia coli (E. coli) & Total Coliform

DPHE		Quality Management System		
DPHE-FSL-QMS-SOP-T-012		Title: Standard Operating Procedure (Testing)		
Issue No.	Issue Date	Effective Date	Rev. No.	Rev. Date
01	01/12/2021		01	01.12.2021

#### ✤ <u>Instruments:</u>

- ✓ Incubator
- ✓ Pipette
- ✓ Autoclave
- ✓ Weighing Machine
- ✓ Spoon

#### Personal Protective Equipment (PPE):

- ✓ Gloves
- ✓ Goggles
- ✓ Mask
- ✓ Lab Coat (Body fitting)

#### \* <u>Reagent:</u>

- ✓ Compact Dry EC
- ✓ Magnesium Chloride
- ✓ Potassium Dihydrogen Phosphate

#### \* <u>Stock Solution Preparation:</u>

- a. At first take sterilized bottle in a weight scale.
- b. Press 'Tare" and make it zero.
- c. Take 40.5 gm Magnesium Chloride (MgCl<sub>2</sub>) into the bottle.
- d. Add distill water up to 500 gm.
- e. Close the lid and shake well.
- f. Label the bottle with manufacturing date and initial.
- g. Now again take another stylize bottle in a weight scale.
- h. Press 'Tare" and make it zero.
- i. Take 17.0 gm Potassium Dihydrogen Phosphate (KH<sub>2</sub>PO<sub>4</sub>) into the bottle.
- j. Add distill water up to 500 gm.
- k. Close the lid and shake well.
- 1. Label the bottle with manufacturing date and initial.
- m. Now stylize both bottles in the autoclave (20 minutes at  $121^{\circ}$ C).
- n. After autoclave it, store them in the refrigerator.



#### \* <u>Working Solution Preparation:</u>

- a. At first take sterilized empty bottle in a weight scale.
- b. Press 'Tare" and make it zero.
- c. Shake the stock solution of Magnesium Chloride properly.
- d. Take 2.5 ml Magnesium Chloride into the empty bottle.
- e. Again shake the stock solution of Potassium Dihydrogen Phosphate.
- f. Add 0.625 ml Potassium Dihydrogen Phosphate into the bottle on weight scale.
- g. Add distill water up to 500 gm.
- h. Close the lid and shake well.
- i. Label the bottle with manufacturing date and initial.
- j. Now autoclave it.



#### **\*** <u>Dilution and sample taken Procedure:</u>

- a. At first homogenize the sample by stomacher.
- b. Label 50ml tubes for two dilution rows
- c. Place each tube on balance, tare and fill with 36 ml of working solution
- d. Pipette 4 ml of sample into first tube, close, shake well
- e. Repeat up to required step:
  - ✓ Step 01 (1:10)
  - ✓ Step 02 (1:100)
  - ✓ Step 03 (1:1000)
  - ✓ Step 04 (1:10000)
  - ✓ So on
- f. Use a new pipette tip for every step
- g. Then take Compact Dry Ec as per dilution stage. (For getting accurate result take 2 set sample)
- h. Remove cap from Compact Dry Ec, pipette 1 mL sample in the middle of the dry sheet. The sample diffuses automatically and evenly over the entire sheet to transform it into a gel within seconds.
- i. Start with the highest dilution (lowest concentration)
- j. Switch on the incubator 1hr before using it
- k. Now keep the Compact Dry Ec in the incubator for 24 hours at 37<sup>o</sup>C.
- 1. After 24 Hours count the E. coli and Total coliform manually.
- m. After use, autoclave all Stock and working solution and keep them in the refrigerator.



#### **\*** <u>Counting Procedure:</u>

- a. After 24 hours get out all compact dry Ec.
- b. Select the compact dry Ec which is suitable to count for getting accurate result.
- c. Pink-purple colonies are indicative of coliform and blue-colored colonies are indicative of Escherichia coli. Count the total number of blue colonies to obtain E. coli count. Count all pink-purple and blue-colored colonies to obtain the total coliform count.
- d. Convert to cfu/100 ml.



## \* <u>Cleaning Procedure:</u>

- ✓ After finishing laboratory work thoroughly wipe down surfaces properly by tissue paper.
- $\checkmark$  Then wipe the surface by disinfectant solution so that there are no any germs in the surface.
- $\checkmark$  Clean all the apparatus using for testing by liquid soap.
- ✓ Now autoclave all the apparatus and after finishing autoclaving keep the apparatus in their dedicated places.

#### ✤ <u>Waste Management:</u>

- 1. After finishing testing keep all waste in the dustbin as per following ways-
- ✓ <u>Red Dustbin:</u>

Highly Hazardous, Biohazard and Toxic elements.

✓ <u>Yellow Dustbin:</u>

Paper & Glass bottles (After Autoclave).

#### ✓ <u>Blue Dustbin:</u>

Plastic wrappers and non-biodegradable wastes (After Autoclave).



2. Keeping wastage will be collected by the municipality department to treat these as per government standard of Bangladesh.

# **10.13 SOP for testing Helminth Eggs**

DPHE		Quality Management System		
DPHE-FSL-QMS-SOP-T-013		Title: Standard Operating Procedure (Testing)		
Issue No.	Issue Date	Effective Date	Rev. No.	Rev. Date
01	01/12/2023		0	01.12.2023

## ✤ Instrument:

- ✓ Compound microscope with 10x and 40x objectives (and preferably, a camera)
- ✓ Bench-top centrifuge with a swing-out rotor that can spin a minimum of 8 x 15ml plastic conical test tubes (Falcon tubes) and, if possible, buckets that can also spin a minimum of 4 x 50ml Falcon tubes
- $\checkmark$  Sink with hose attached to tap for washing using high water pressure
- ✓ Top-pan balance (scale, for weights up to 200gm and accurate to 2 decimal places)
- ✓ Magnetic stirrer and bar magnets
- ✓ Vortex mixer
- ✓ Hydrometer that can measure SG between 1.2 and 1.3
- ✓ 100µm mesh stainless steel pan sieve, diameter 200mm
- ✓ 20 $\mu$ m mesh stainless steel pan sieve, diameter 200mm
- ✓ 20µm mesh stainless steel pan sieve, diameter 100mm
- ✓ Plastic test tube racks to hold the 15ml Falcon tubes (and if using 50ml tubes, one for those)
- ✓ Plastic 250ml beakers it's good to have stock of  $\pm$  20 beakers
- ✓ Plastic "hockey-stick " shaped spreaders
- ✓ Plastic 3ml Pasteur pipettes (non-sterile)
- ✓ Non-sterile gloves
- ✓ Applicator sticks
- ✓ Wooden tongue depressors
- ✓ Microscope slides (76 x 26 x 1.2mm)
- ✓ Cover glasses (22 x 40mm)

#### \* <u>Reagent:</u>

#### ✓ Physiological Saline (8.5g/L NaCl)

Dissolve 8.5 g sodium chloride in distilled or de-ionized water. Make small amounts to use up at one time or if large amounts are made, it is preferable to decant them into smaller containers and autoclave for 15 min at 121°C. Cool to room temperature and store.

#### ✓ Ammonium Bicarbonate (AmBic)

Dissolve 119gm of ammonium bicarbonate in 1L de-ionized water (use a magnetic stirrer and bar magnet) store in a glass jar.

#### ✓ Tween 80 or 7X

#### ✓ Zinc Sulphate (ZnSO<sub>4</sub>,7H<sub>2</sub>O)

Dissolve 500gm zinc sulphate in approximately 800ml de-ionized water (use magnetic stirrer and bar magnet) and adjust specific gravity using more of the chemical or water to raise or lower the specific gravity to 1.3.

#### ✓ 0.1N Sulphuric acid (H<sub>2</sub>SO<sub>4</sub>)

Add 500ml de-ionized water to a 1 liter plastic bottle, pour 3ml concentrated Sulphuric acid into a 10ml graduated cylinder, then pour the H2SO4 into the plastic bottle containing the water, recap and shake. Un-cap, add 497ml of de-ionized water to the plastic bottle, re-cap and shake.

#### Personal Protective Equipment (PPE):

- ✓ Gloves
- ✓ Goggles
- ✓ Mask
- ✓ Lab Coat (Body fitting)
- ✓ Safety Shoe

#### Procedure - VIP, UD, and thick sludge

- a. Place a 250ml plastic beaker (labeled with sample number) on top-pan balance, zero balance, weigh 10 or 20gm of sample into beaker. NOTE: IF waste material is very dry (e.g. pelletised or totally desiccated), then soak for 12 24 hours in ±80ml physiological saline to soften. Next, break up and mix sample well in the saline. Stand to sediment solids for 4 hours. Remove as much supernatant as possible without disturbing deposit, continue with next step below.
  - b. Add 50-80ml AmBic and a magnetic stirring bar, mix on magnetic stirrer for 10 minutes.
  - c. Pour this mixture over  $100\mu m$  sieve which fits on top of  $20\mu m$  sieve (wet sieves with tap H<sub>2</sub>O first).
- d. Rinse beaker with tap H<sub>2</sub>O and pour over sieves.
- e. Wash magnet over sieves and remove, wash 100µm sieve well (using "hockey-stick" spreader, or preferably, gloved hand) over 20µm filter, regularly checking bottom sieve for fluid build-up. Use same hockey stick to stir sample on 20µm sieve while holding 100µm sieve directly above so as not to lose any sample. When 20µm sieve has drained sufficiently, place the 100µm sieve back on top and continue washing. Repeat this until sample on 100µm sieve is sufficiently well washed.
- f. Separate sieves and then rinse 20µm sieve well. Use water-pressure to wash the material to one side of sieve to make collection easier.
- g. Rinse all material off 20µm filter into original rinsed-out labeled beaker.
- h. Pour beaker contents into 4 x 15ml conical test tubes labeled with sample number or if retentate is large, use 50ml labeled tubes. (The aim after the next step is to have ≤1ml deposit in a 15ml tube and ≤5ml in a 50ml tube.)

- i. Centrifuge at 3000rpm (1389g) in centrifuge with swing-out rotor for 10 minutes.
- j. Pour off supernatant, leaving deposits in test tubes.
- k. Place test tubes in rack with applicator stick in each (as a stirring rod) and pipette in ZnSO4, 3ml at a time, vortexing in between addition of the chemical, until tubes are filled to 14ml mark for 15ml tubes / 45ml mark for 50ml tubes.
- 1. Centrifuge at 2000rpm (617g) for 10 minutes.
- m. Pour supernatant flotation fluid over smaller diameter 20µm sieve. Wash remaining deposits out of test tubes and keep one aside for re-use.
- N. Wash material on sieve well with tap water and rinse it down to one side of the sieve for collection.
  Using a 3ml plastic pipette, transfer the material back into the test tube kept aside.
- o. Centrifuge at 3000rpm (1389g) for 10 minutes to obtain the final deposit.
- p. Pour off supernatant water and pipette up the deposit, place it on one or more microscope slides (but make one slide at a time so they don't stand for long periods and dry out), place a 22x40mm cover slip on top, examine and count every Ascaris egg, classifying them as viable, potentially viable or dead. Trichuris, Taenia, hookworm spp. eggs can also be counted and assessed simply as potentially viable or dead.

#### **Procedure – water samples**

a. If the water is effluent from a waste-water treatment plant and is fairly clean with low suspended solids, then it is preferable to use a large sample of 5 - 10lt, measured out using a 1lt measuring cylinder.

#### <u>NOTE 1:</u>

IF sample is black water with a high concentration of solids, then use amounts of 250 - 1000ml. The sample should be measured out and then stood for 4 hours or overnight to sediment the solids. Then, remove the supernatant fluid and treat as in second step above of: 7. Procedure – VIP, UD, and thick sludge

#### **NOTE 2:**

IF sample is fatty, then measure out selected sample size (from 250 - 1000ml), pour into plastic beaker large enough to contain the sample with at least 5-10cm above the sample, so that it does not spill when mixing on magnetic stirrer. Add 1ml per liter of neat Tween 80 or 7X directly into the sample (so as to make a  $\pm 0.1\%$  solution). Mix well using magnetic stirrer and magnet in beaker for 20 minutes. Then proceed as for next step below.

- b. The measured sample is poured slowly through a 100µm sieve which fits on top of a 20µm sieve and is well washed, checking bottom sieve for fluid build-up.
- c. Separate sieves and then rinse 20µm sieve well and wash material to one side of sieve for collection.
- d. Rinse all material off 20µm filter into 2 4 x 15ml conical test tubes.
- e. Centrifuge at 3000rpm (1389g) in centrifuge with swing-out rotor for 10 minutes.
- f. Pour off supernatant and retain deposits left in 2 4 x 15ml test tubes.
- g. Place test tubes in rack with applicator stick in each (as a stirring rod) and pipette in ZnSO4, 3ml at a time, vortexing in between addition of the chemical, until tubes are filled to 14ml mark.
- h. Centrifuge at 2000rpm (617g) for 10 minutes.
- i. Pour supernatant flotation fluid over smaller diameter 20µm sieve. Wash out test tubes and keep one aside for re-use.
- j. Wash material on sieve well with tap water and rinse down to one side of sieve for collection. Using a 3ml plastic pipette, transfer material back into test tube kept aside.
- k. Centrifuge at 3000rpm (1389g) for 10 minutes to obtain the final deposit.
- Pour off supernatant water and pipette up the deposit, place it on a microscope slide, place a 22x40mm cover slip on top, examine and count every Ascaris egg, classifying them as viable, potentially viable or dead. Trichuris, Taenia, hookworm spp. eggs can also be counted and assessed simply as potentially viable or dead.

#### Procedure for incubating samples for viability testing

- a. Weigh 10 or 20gm into a 250ml plastic beaker, on a top-pan balance.
- b. Add approximately 10 20 ml deionized water; 0.1N H<sub>2</sub>SO4, or 1% formalin to sample.
- c. Cover with parafilm, prick holes in parafilm to allow air into sample, or place a plastic Petri dish lid on top.
- d. Incubate for 21 28 days at 25-28°C (Check regularly to see that the sample has not dried out and add more water,  $0.1N H_2SO_4$  or 1% formalin as necessary to keep sample moist. Aerate the samples daily by swirling or vortexing carefully.)
- e. After 28 days, remove from incubator, stand for 4 hours or overnight to sediment the sample, remove the supernatant fluid, and then proceed as for step 2 onwards described above in: 7. Procedure VIP, UD, and thick sludge.



# **Calculation:**

No of 
$$eggs/L = \frac{E * 1000}{Total sample Volume taken (ml)}$$

Here,

E= the sum of the egg number that count from slides A, B, C, D E = A + B + C + D

## **Cleaning Procedure:**

- ✓ After finishing laboratory work thoroughly wipe down surfaces properly by tissue paper.
- $\checkmark$  Then wipe the surface by disinfectant solution so that there are no any germs in the surface.
- $\checkmark$  Clean all the apparatus using for testing by liquid soap.
- ✓ Now autoclave all the apparatus and after finishing autoclaving keep the apparatus in their dedicated places.

#### Waste Management:

- 5. After finishing testing keep all waste in the dustbin as per following ways-
- <u>Red Dustbin:</u>
  Highly Hazardous, Biohazard and Toxic elements.
  Vollow Dustbin:
- ✓ <u>Yellow Dustbin:</u> Paper & Glass bottle (After Autoclave).
- ✓ <u>Blue Dustbin:</u>

Plastic wrappers and non-biodegradable wastes (After Autoclave).



3. Keeping wastage will be collected by the municipality department to treat these as per government standard of Bangladesh.

# **Chapter 11: Standard Operating Procedure (SOP) for Analysis Data circulation**

DPHE		Quality Management System		
DPHE-FSL-QMS-SOP-D-01		Title: Standard Operating Procedure (Data management)		
Issue No.	<b>Issue Date</b>	Effective Date	Rev. No.	Rev. Date
01	01/12/2021		01	01.12.2021

# **Step: 01**



# **Step: 02**



# **Chapter 12: Standard Operating Procedure (SOP) for Analysis Data** reservation

DPHE		Quality Management System		
DPHE-FSL-QMS-SOP-D-02		Title: Standard Operating Procedure (Data management)		
Issue No.	Issue Date	Effective Date	Rev. No.	Rev. Date
01	01/12/2021		01	01.12.2021

**Standard Operating Procedure (Documentation)** 



# References

- Method for Fecal Sludge Analysis of EAWAG (IWA Publication)
- Training from CSE India topics on "Advance Laboratory Training on the analysis of Fecal Sludge and Biosolids" in 2024.
- Training from CSE India topics on "Basic Laboratory trainings for working professionals in Faecal Sludge management" on 2023.
- Training from CSE India topics on "Resource Recovery from Faecal Sludge (Laboratory training on the quality analysis of Co-compost/ Biochar and Faecal Sludge)" on 2022.
- "Faecal Sludge Laboratory Training" from Christopher Friedrich, Sandec, Switzerland on June 2022.
- "Laboratory Analysis Training for DPHE FSL" from SPIEZ Laboratory, Switzerland on March 2022.
- "FS Laboratory Training" from SPIEZ Laboratory, Switzerland on December 2021.
- Protocol for Fecal Sludge Testing (ITN, BUET, Bangladesh)
- The Ammonium bicarbonate (AmBic) method- Developed by Hawksworth and Archer (2010) and modified by Archer C. (2012) Pollution Research Group, University of KwaZulu-Natal, Durban, South Africa.
- DoE guideline of Bangladesh for sewerage discharge
- Effluent analysis data of FSL DPHE 1<sup>st</sup> round of 2022 (January to April 2022).
- Effluent analysis data of FSL DPHE 2<sup>nd</sup> round of 2022 (May to August 2022).
- Effluent analysis data of FSL DPHE 3<sup>rd</sup> round of 2022 (September to December 2022).
- Effluent analysis data of FSL DPHE 1<sup>st</sup> round of 2023 (January to April 2023).
- Effluent analysis data of FSL DPHE 2<sup>nd</sup> round of 2023 (May to August 2023).
- Effluent analysis data of FSL DPHE 3<sup>rd</sup> round of 2023 (September to December 2023).
- Effluent analysis data of FSL DPHE 1<sup>st</sup> round of 2024 (January to June 2024)
- Mode of operation FSL-DPHE of 2023.

# Annex

# Annex-13.1: Image of FSL-DPHE during setup



# Annex-13.2: Image of FSL-DPHE



# Annex-13.3: Image of Sample Collection in the field



Annex-13.4: Image of Switzerland Ambassador of Bangladesh visit in FSL



Parameter	Unit	The Environment Conservation Rules, 1997, Ministry of Environment and Forest. Schedule 9 – Standards for Sewage Discharge	Department of Environment Guidelinesupdate 5 March 2023, Schedule 3 – Standards for Sewage Discharge
pН	-	-	6-9
BOD (At 20 <sup>0</sup> C)	mg/L	40	30
COD	mg/L	-	125
Nitrate (NO <sub>3</sub> )	mg/L	250	50
Phosphate (PO <sub>4</sub> )	mg/L	35	15
Suspended Solids	mg/L	100	100
Temperature	°C	30	30
Total Coliform	CFU/100mL	1000	1000
Oil & Grease	mg/L	-	10

# Annex-13.5: DoE guideline for sewerage discharge 2023 (Bangladesh)

## Annex-13.6: News of DPHE-FSL in Media

#### News of EAWAG in LinkedIn

https://www.linkedin.com/posts/sandec-eawag\_sha-eawag-fecalsludge-activity-7105082240321105920-d-mB/?utm\_source=share&utm\_medium=member\_android



As part of the Swiss Agency for Development and Cooperation #SHA #Eawag backstopping mandate, Sandec's Christopher Friedrich planned and assisted in setting up a faecal sludge laboratory in Cox's Bazar, Bangladesh. The lab results inform evidence-based WASH interventions to limit the spread of water-borne and water-washed diseases in the world's largest refugee camp and Cox's Bazar district. Read more about this work in SDC's One World magazine article: https://bit.ly/3YWfnwu ffaecalsludge #refugeecamp #sanitation #sanitationsolutions #coxsbazar #bangladesh #diseaseprevention #diseasecontrol #WASH







1 comment • 4 reposts

News in Switzerland's "One world" Magazine

https://www.eine-welt.ch/en/2023/issue-1/determined-fight-to-eliminate-risk-of-fecalsludge

https://www.eda.admin.ch/dam/deza/fr/documents/publikationen/Eine-Welt/eine-welt-2023-01\_FR.pdf



# LUTTER CONTRE LE PÉRIL FÉCAL

La gestion des eaux usées, des excréments et des urines constitue un défi de taille dans les camps de réfugiés à Cox's Bazar, dont la densité de population compte parmi les plus élevées du monde. Surveiller la performance des installations d'assainissement se révèle essentielle pour éviter les maladies. La Suisse a mis en place au Bangladesh un laboratoire d'analyse des effluents liquides après le traitement des excréments.

Texte: Zélie Scholler

## **\*** News in Switzerland Embassy of Bangladesh's Facebook page

 $\label{eq:https://m.facebook.com/story.php?story_fbid=pfbid02QrVw1MqDnKfQcybrswSStbqm} V4J6q92VGKjeEajzfrCYkDbEFTX7duJM1kz8682kl&id=102571866488538&mibextid=Nif50z$ 

https://m.facebook.com/story.php?story\_fbid=pfbid031xBAV66Gb9h5aj8nvjhgrprHbT rfFCJ874UHu6mhx9EdBfwJ2QazL3nwSGfrRN3Cl&id=100064404443847&mibextid =Nif5oz

https://m.facebook.com/story.php?story\_fbid=pfbid0RB4oZ33532BjMx6BfsT6NKQj3 wZQTqcCRXZfGZ22Shv9JJnDC2qnjEGNCaQnDYW51&id=102571866488538&mib extid=Nif5oz

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Embassy of Switzerland in Bangladesh Mar 24, 2022 · 🚱

R 🗇 🔳

The Faecal Sludge Laboratory of the Department of Public Health Engineering (DPHE) in Cox's Bazar has been set up in 2021 with support from Switzerland. The staff of this laboratory is currently receiving an in-depth training from two visiting specialists of the Swiss Federal Laboratory in Spiez on sampling and analysis procedures as well as on laboratory safety protocols. Several field trips for sampling of Faecal Sludge Treatment Plants are also being conducted. These trainings will contribute to building capacity of DPHE staff who are working to ensure that the environment and public health in Cox's Bazar district are protected. #SwissinBD



Embassy of Switzerland in Bangladesh 🖉

R 🗢 🔳

Switzerland is supporting the Department of Public Health Engineering (DPHE) to set up a Faecal Sludge Laboratory (FSL) in Cox's Bazar with new equipment, materials, initial salary costs and technical training for the laboratory staff. Ambassador Nathalie Chuard has recently visited the FSL to observe the laboratory set-up that will allow DPHE to test treated wastewater and faecal sludge from treatment plants in the Cox's Bazar district. Being the first of its kind, the FSL will be an official government-monitoring laboratory that will conduct regular analyses from selected treatment plants to monitor the compliance with standards of sewage discharge and to ensure the environment and public health are protected. #SwissinBD



Embassy of Switzerland in Bangladesh January 24 at 8:30 PM · 🕲

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#### C 🗢 🔳

Switzerland is supporting the Department of Public Health Engineering (DPHE) in Cox's Bazar to set up a Faecal Sludge Laboratory (FSL) that can test treated wastewater from faecal sludge treatment plants across the district. The staff of the laboratory are receiving different online and in-person training and have also started collecting samples from the camps for lab analysis. Regular analyses of these samples will aid the DPHE to ensure that the environment and public health are protected.

More 🕦 📴 https://bit.ly/3tUmV5i 🥏

#SwissinBD #SwissHumanitarianAid



News in Twitter by Ex Switzerland Ambassador "Nathalie Chuard"





Ambassador Nathalie Chuard 🤣 @SwissAmbBD

The Laboratory is soon ready! With proper facilities & trained staff, it will monitor fecal sludge at national standards. This milestone in our partnership with #DPHE will surely contribute to better public #health & #environment in #CoxsBazar @SwissHumAidUnit



5:55 PM · 21 Oct 21 · Twitter for iPhone

## News of ITN-BUET

https://itn.buet.ac.bd/web/news/dphe-coxs-bazar-fecal-sludge-laboratory-personnel-arenow-oriented-on-basic-fecal-sludge-characterization



# DPHE Cox's Bazar Fecal Sludge Laboratory personnel are now oriented on basic Fecal Sludge characterization

UPM GmBH and its local partner Bangladesh Agricultural University (BAU) handed over field laboratory facilities used by them to characterize the fecal sludge in Rohingya camps Cox's Bazar to DPHE following the hands-on training on the operation of the equipment and sampling.

October 05, 2021



# **Faecal Sludge Laboratory** Department of Public Health Engineering, Mohajer Para, Cox's Bazar, Bangladesh Mobile: +8801305950333, Email: <u>dphefslcoxsbazar@gmail.com</u>