Environmental Toxicology Laboratory
Toxicology Centre
University of Saskatchewan

STANDARD OPERATING PROCEDURE

Management of eDNA sample: Receiving, Preservation, Storage, Documentation, Decontamination, and Disposal

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Yuwei Xie, Ph.D., Markus Hecker, Ph.D., Paul D. Jones, Ph.D. and John P. Giesy. Ph.D., FRSC, FSETAC

Supported through:
Toxicology Centre and
Department of Veterinary Biomedical Sciences

Correspondence to:
Environmental Toxicology Laboratory
Toxicology Centre
44 Campus Drive,
Saskatoon, Saskatchewan, S7N 5B3
Canada

Phone: (306) 966-5062; 966-2096
Facsimile: (306) 966-4796

APPROVAL PAGE

Revisions to an existing SOP, addition of a SOP change form, or preparation of a new SOP must be reviewed, approved, and signed by the following:

Authored By:	Yuwei Xie, Markus Hecker, Paul D. Jones and John P. Giesy	Date: 04/15/2018
Supervisor Review By:	John P. Giesy	Date: 04/30/2018
Reviewed By: (QA Coordinator)	_____	Date: _____

DEFINITIONS AND ACRONYMS

ETL	Environmental Toxicology Laboratory (University of Saskatchewan)
COC	Chain of Custody
CQA	Chemical Quality Assurance
LIMS	Laboratory Information Management System
UofS	University of Saskatchewan
DHSE	Department of Health, Safety and Environment (University of Saskatchewan)
PPE	Personal Protective Equipment
QA	Quality Assurance
QC	Quality Control
SOP	Standard Operating Procedure
STS	Sample Tracking Sheet
SAP	Sampling and Analysis Plan
eDNA	environmental DNA
GWF	Global Water Futures

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1.0 PURPOSE

This standard operating procedure (SOP) specifies the requirements for sample receipt, control, record keeping, decontamination, and disposal at the Environmental Toxicology Laboratory at the University of Saskatchewan.

2.0 SCOPE AND APPLICATION

This SOP applies to the ETL for eDNA samples supplied from the Global Water Futures (GWF) program titled "Next generation solutions to ensure healthy water resources for future generations" (eDNA project).

3.0 SAFETY CONSIDERATIONS

Safety training and medical monitoring requirements are described in the Health and Safety Plan for eDNA project of the GWF program.

In addition, there are various safety concerns regarding the receipt, storage, and disposal of sample containers at the ETL. Upon receipt, the sample containers will be monitored for breakage. If sample containers are broken, the appropriate personnel will be immediately notified and the Department of Health, Safety and Environment (DHSE) will be called in order to assess the hazard. DHSE will also be contacted in the case of chemical spills and will be responsible for the disposal of hazardous wastes.

3.1 PERSONAL PROTECTIVE EQUIPMENT

Personal protective equipment (PPE), consisting of lab coats, safety glasses, and latex gloves will be worn at all times when handling samples.

3.1 WASTE MANAGEMENT

All waste will be managed and disposed of in accordance with U of S-DHSE regulations. Waste management practices will include the control of all standards and solutions. This means that if required expired or used standards, associated solvents and other chemicals used for preservation and biological or element analysis will be disposed of in labeled waste containers and DHSE will be notified for waste pick up.

3.2 SAMPLE DECONTAMINATION

If a spill occurs in the laboratory, DHSE will be notified immediately. The area where the spill occurred will be evacuated and marked.

4.0 EQUIPMENT, MATERIALS, AND REAGENTS

The sample storage area is equipped with a locked freezer in which samples are stored as appropriate. The freezer is connected to phone alarm systems that monitor temperature and notify laboratory personnel in cases of temperature and/or power related issues. A calibrated balance is also kept in the sample storage area and is used to weigh sub-samples.

No materials or reagents are used in sample receipt.

5.0 METHOD, PROCEDURES, AND REQUIREMENTS

5.1 Sample Receipt

The physical condition of coolers or other containers used for transportation, and each individual sample container, will be inspected upon arrival at the ETL. The following methods have been established for sample receiving:

A. Inspect sample coolers and samples for signs of damage upon receipt at the laboratory.

B. Attach air bill or shipping receipt to the chain of custody (COC) form.

C. Examine individual samples and record their status (frozen/ not frozen; immersed in preservation liquid, etc.) on a sample receipt form.

D. Verify that a COC form is submitted with samples, and that the COC contains all information required for analysis and reporting. Maintain custody of samples by ensuring that all dates, times, and signatures are provided on the COC forms. If the COC form is lost or all required information of the COC form is not included with samples, please contact the project manager and sampler for double check.

E. Identify and reconcile any discrepancies between the COC and sample labels following section 5.3.3 of this SOP.

F. Verify that sample containers, labeling, or other requirements are correct. Assign a unique lab identification number to each sample and log samples into the sample tracking sheet (STS). (See attached STS.) Identify any hazards or special precautions associated with the incoming samples.

G. Notify appropriate laboratory and field study personnel when samples have arrived. These individuals are to be identified in either a Work Plan or SAP.

H. Track and document the handling of samples from receipt through data reporting to final disposal. This will be accomplished by keeping all of the log forms in a binder kept in the laboratory.

5.2 Sampling Documentation

Upon arrival, the shipping receipts will be collected from the cooler and be stapled to the COC form. Samples submitted to the ETL will be accounted for by documenting their arrival and condition on COC and STS. Within the ETL, the STS will be used to monitor the samples whereabouts at all times. Fractional sample removed will be recorded on the STS. While handling samples, any anomalies or problems will be noted in bound laboratory notebooks.

5.3 Sample Storage and Preservation

Samples will be stored in freezer, fridge or locked storage room at room temperature (formalin preserved sturgeon samples) in the laboratory according to the sample type and correlated sampling SOP or proceeding SOP. This room is accessible only to lab personnel. The freezer and fridge will be set at -20 ^{o}C, and +4 ^{o}C, respectively, and the temperature will be monitored daily. The Thermo Scientific Forma -86C Ultra Freezer in Room 261, Toxicology Building, U of S, is used to store biological samples for analysis. If for any reason there is a power outage or an increase in temperature, the facility manager on call and/or other lab personnel will be immediately notified by the automated phone alarm system that will automatically call the cell phone of the person on duty. The necessary action will then be taken to ensure that sample integrity is not compromised. If samples are removed from any of the storage compartments/units for any reason, this activity will be documented on the STS form. Copies of the forms will be placed in the records archive. When samples are removed for preparation and analysis, a sample extraction form will be completed.

5.3.1 Scheduled Monitoring

All refrigerators and freezers used in the ETL will be examined frequently due to constant use and will be monitored at a minimum daily. Freezer temperatures are maintained at a nominal -20 ^{o}C. And -86C Ultra Freezer temperatures are maintained at a nominal -80 ^{o}C. If the freezer temperature rises to -15 ^{o}C, -86C Ultra Freezer temperature rises to -70 ^{o}C such that it triggers the alarm of the fridge temperature rises over +7 ^{o}C

corrective action must be taken. Actions include adjusting the thermostats, refilling liquid nitrogen, having the unit serviced, or moving the samples to another unit.

5.3.2 Sample Accountability

To ensure that all samples will be accounted for, the following guidelines will be followed:

A. The person obtaining the sample or submitting the sample to the laboratory for analysis must establish sample identity by checking against the Filed Record Sheet and COC form.

B. Integrity of sample must be maintained from collection to delivery.

C. Composition of sample must remain the same during handling and storage before analysis.

D. Evidence must exist of sample's receipt and COC record filled out, and appropriate personnel notified of the sample arrival.

E. Person preparing sample must not allow composition of sample to change or integrity to be questioned.

F. Analyst must ensure correct sample is analyzed.

G. Analyst must record all data contributing to the analysis on both local storage server and cloud server.

H. Records must be kept to trace sample from retrieval through data reporting.

I. Special storage conditions must be documented on the plan of data storage for eDNA project.

5.3.3 Label and COC Discrepancies

Discrepancies between the sample labels and COC will be noted on the COC or Sample Receipt Form. The sample manager will resolve any documentation discrepancies by contacting the personnel that submitted samples. For discrepancies impacting sample viability (i.e., improper sample temperature) where a CAR is required to be completed, the sample manager will coordinate with the sample submitter, QA, and Project Study Group representatives to determine the appropriate corrective action.

6.0 RECORDS, DOCUMENTATION, AND QC REQUIREMENTS

The primary analyst shall document any anomalies and/or deviation from the specified method in a bound, serially numbered, laboratory notebook with tear-out carbon copies. All electronic files and hardcopies will be kept at the participating laboratory.

The carbon copies from data notebooks will be removed and archived in a separate building. Copies of the COC forms, the STS, and laboratory notes will be kept in 3-ring binders in separate places at all times in case of fire or other disaster.

7.0 RESPONSIBILITIES

Project Manager — Dr. Yuwei Xie will oversee and approve all project activities, authorize necessary actions and adjustments, and act as liaison between the principal investigator and other U of S personnel and the sponsor Project Manager.

Principal Investigator — Prof. John P. Giesy will advise the Project Manager in overseeing and approving all project activities, authorize necessary actions and adjustments related to U of S activities to accomplish program QA objectives; and act as liaison between agencies, staff, and the sponsor Project Manager.

Study Team Leaders (STL) — Dr. Yuwei Xie, under the supervision of Prof. John P. Giesy, will oversee all research activities and supervise all personnel involved with the assemblage of the experimental exposure systems. The STLs will ensure that proper sample collection, preservation, storage, transport, and COC QC procedures are followed and will inform the Project QA Manager when problems occur, and will communicate and document corrective actions taken. The STLs will discuss study activities with the Project Manager.

Quality Assurance (QA) Manager — Prof. Paul D. Jones will initiate audits on work completed by project personnel. The manager will review program QA activities, quality problems, and quality-related requests. In response to experimental findings, the QA manager will approve corrective actions. The QA manager will report quality non-conformances to the Project Manager.

8.0 REFERENCES

Environmental Analytical Laboratory, Laboratory Quality Control Plan, April 1997.

APPENDIX A: FIELD RECORD SHEET

Field Record Sheet

Book No: ETL-eDNA-FRS Page No.: _____

Project:		
Sample Identification:		
Date:		Time:
Location:	Latitude:	Longitude:
Description of Sample Location/ Observations:		
Conditions:		
Principal Scientist:		
Crew Members:		
Water Depth (m):		Water Temp (°C):
Specific Conductivity (µmoh):		Air Temp (°C):
Dissolved Oxygen(mg/L):		pH:
Container Preparation/Storage:		
Samples Collected and Preserved:		
Sample Distribution:		

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APPENDIX B: EXAMPLE OF A PRE-PRINTED LABEL

University of Saskatchewan
eDNA Project -- (Subproject)
SAMPLE ID#:

Date:_____Time:_____WaterShed: _____
Sample type:_____
Preservation Method:_____
Preparing Method:_____
Sample Note:_____
Sampler initials: _____

APPENDIX C: CHAIN OF CUSTODY FORM

CHAIN OF CUSTODY FORM

Book No: ETL-eDNA-COC Page No.:_____

Project: GWF-eDNA

Samplers (signature) :

Station	Date	Time	Station Location	No. of Container	Container ID	Remarks

Relinquished By (signature)	Date	Time	Received By (signature)
Relinquished By (signature)	Date	Time	Received By (signature)
Relinquished By (signature)	Date	Time	Received By (signature)
Received for Laboratory By (signature)	Date	Time	Remarks

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APPENDIX D: SAMPLE TRACKING SHEET

SAMPLE TRACKING SHEET

Book No: ETL-eDNA-STS Page No.: _____

Sample Identification

Analyst:	
Date:	
COC received and correct?	
Subproject:	
Sampling Campaign:	
Sampling Location:	
Sample Type:	
Note about Sample:	
Preservation Method:	

SAMPLE HANDLING

Date	Analyst	Sample used	Sample remaining

Comments and Notes:

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