Jan 12, 2024 Version 1

W-1 WATER FIELD SAMPLING V.1

DOI
dx.doi.org/10.17504/protocols.io.q26g7p8m9gwz/v1

REDI-NET Consortium[1]

[1]REDI-NET Consortium

Remote Emerging Disea...

REDI-NET Consortium
University of Notre Dame, Naval Medical Research Center, Wal...

DOI: https://dx.doi.org/10.17504/protocols.io.q26g7p8m9gwz/v1

Protocol Citation: REDI-NET Consortium 2024. W-1 WATER FIELD SAMPLING. **protocols.io** **https://dx.doi.org/10.17504/protocols.io.q26g7p8m9gwz/v1**Version created by **REDI-NET Consortium**

Protocol status: Working
We use this protocol and it's working

Created: August 09, 2023

Last Modified: March 15, 2024

Protocol Integer ID: 86279

Keywords: WATER FIELD SAMPLING, SAMPLE COLLECTION, WATER STORAGE, water field sampling

Funders Acknowledgements:
USAMRAA
Grant ID: W81XWH-21-C-0001,
USAMRAA
Grant ID: W81XWH-22-C-0093
USAMRAA
Grant ID: HT9425-23-C-0059

Disclaimer

This work is supported by the US Army Medical Research and Development Command under Contract No.W81XWH-21-C-0001, W81XWH-22-C-0093 and HT9425-23-C-0059. The views, opinions and/or findings contained in this report are those of the author(s) and should not be construed as an official Department of the Army or Navy position, policy or decision unless so designated by other documentation.

Abstract

This protocol describes water field sampling.

Guidelines

OBJECTIVE

To outline steps to properly collect water samples and evaluate the risk of zoonotic disease transmission by the detection of pathogens from environmental DNA (eDNA).

SUMMARY/SCOPE

The overarching aim of the REDI-NET is to develop a collaborative laboratory network between domestic and international partnering institutions to address disease surveillance needs in order to effectively detect, predict and contain potentially emergent zoonosis. This SOP provides guidance on the Protocol for the correct sampling of water at CONUS/OCONUS sites to evaluate and predict the risk of zoonotic diseases transmission.

RESPONSIBLE PERSON

Principal Investigator, Study Coordinator, Entomology Component Lead, Managers

Note

NOTE: All study procedures must be conducted in compliance with national and local policies for prevention and control of COVID-19 infection.

MAINTENANCE OF EQUIPMENT

BEFORE EACH COLLECTION

1. Clean water collection devices.
2. Freeze and clean ice-packs.
3. Clean cool-boxes.
4. Charge all equipment(e.g., GPS unit, tablets). Make sure the tablet has enough free-space for field sampling pictures.

AFTER EACH COLLECTION

1. Clean all equipment thoroughly between sampling sites, including boots, cooler box (inside and outside), etc. Store sterile equipment separate from used equipment and samples.

QUALITY CONTROL

This SOP is reviewed by the applicable supervisor annually or as required in order to maintain its relevance.

Note

NOTE: Because ‘the edge’ of the water body may change due to precipitation changes, the edge should be defined at each month's sampling. If the water body has a "drop off" (embankment of more than 20cm), the soil and water samples should be taken where the water "meets" the soil. Aim to collect at locations where finer sediment is present, if possible.

Materials

EQUIPMENT AND MATERIALS

Note

NOTE: If product number is listed, please ensure use of this or equivalent product.

	A	B	C
	Equipment / Material	**Description**	**Mfg / Product #**
	Rubber Boots	Personal protection	Locally Sourced
	(4) Water dippers	Water collection device, labeled	BioQuip Mosquito Dipper & Handles - Product Details
	Whirl-pak bags 532 ml capacity	532 ml capacity, write on-bags, sterile	Uline, S-19793 (500/ctn)
	Writing utensils	Pen / pencils, marker pens	Locally Sourced
	Litmus paper	Litmus pH paper strips	https://www.amazon.com/s?k=Litmus+Paper+Ph+Test+Strips&ref=nb_sb_noss_2
	Depth finder	Example: wooden dowel, broom handle, or similar	Locally Sourced
	Paper towels	For drying Whirl-pak bag after sampling	Locally Sourced
	Plastic bags	Black (for used material)	Locally Sourced
	Cooler boxes	For cooling water samples during transport	Locally Sourced
	Sealable plastic bags	For sample transportation Zip-lock® slider GPS unit WGS 84 and precision of 5 decimal degrees	Locally Sourced
	Water collection forms	REDI-NET DCS W-1 Water Collection	REDI-NET Data Portal
	Tablet	For data entry and picture of the sampling sites	Locally Sourced

Water dippers

Whirl-pak bags

Troubleshooting

Safety warnings

RISKS AND PERSONAL PROTECTION

1. Know the risk associated with the study site and sampling location and take appropriate safety precautions and/or wear appropriate equipment.
2. Wear light-colored long pants and a long sleeve shirt. Wear rubber boots or hiking shoes and white socks, and tuck pant legs into socks. Be sure that clothing is sturdy and can withstand long thorns.
3. Never open a sample container until the instant you will take the sample.
4. Samples should be collected WITHOUT ENTERING THE WATER, i.e. the collector stands only on the waterhole bank or muddy pond edges. This prevents disturbance of the substrate and may limit cross-contamination.

SAMPLING TEAMS

1

Note

NOTE: Suggested sampling frequency at each site is one time per month or, when not logistically possible, at minimum two times per unique season representative of the laboratory setting (e.g., early wet/late wet; early dry/late dry; rainy/dry season).

Field sampling of eDNA water samples involves two people. One person serves as the ‘sampler’ and the other person serves as a ‘helper’. The helper can look up details in these instructions when needed, keep track of samples, handle objects that are contamination risks, serve as a second set of eyes for potential contamination, and ensure safety of the sampler in potentially hazardous field conditions.

SAMPLING SITE SELECTION

2 Find two suitable sampling locations within the water body representing different ecological systems, if possible.

3 At each sampling location, soil samples should be taken from two points, one at the edge of the waterhole and the second one meter in from the edge. Sediment collection at each sampling location should be done in triplicate.

4 Because ‘the edge’ of the water body may change due to precipitation changes, the edge should be defined at each month's sampling.

5 If the water body has a "drop off" (embankment of more than 20 cm), the soil and water samples should be taken where the water "meets" the soil. Aim to collect at locations where finer sediment is present, if possible.

SAMPLE COLLECTION

6

Note

NOTES:
- Water samples collection should be performed before Sediment samples collection (REDI-NET SOP S-1 Soil Collection) at each sampling location point.
- Sampling from the edge should begin before sampling at the one meter location. The edge of the waterbody may change between collection times (depending on the rain/drought).

At each sampling location, the helper opens the bag containing the vials to have one open and ready for the sampler. The helper will open and close the bags containing the collection materials. This is to prevent potential contamination of samples.

7 The Sampler takes the appropriately labeled water dipper (for example, SL1-edge would be the dipper head for sampling location 1 at the edge) from the bag and opens the corresponding Whirl-Pak® Bag which can be placed in a bucket for easier manipulation.

Note

NOTE: A single dipper can be used for all triplicate water samples from the same sampling location (for example, SL1-edge replicate 1, 2, and 3).

8 Check the depth of the water. If the depth of the water is greater than 1 m, the water column should be gently mixed using the water dipper without disturbing bottom sediment.

Note

NOTE: Do not collect samples any deeper than 20 cm from the surface of the water when collecting at the edge of the water body.

9 Once the water column is homogenized, collect water filling the dipper, and empty the sample into the Whirl-Pak® Bag until the 250 mL sample is collected (3/4 of a 532 mL Whirl-Pak® Bag).

Note

NOTE: Remove debris from water samples as much as possible before pouring into Whirl-Pak® bags.

10 Once the water is collected, measure pH of the sample by emerging litmus paper.

11 Close the Whirl-Pak® Bag and dry it using paper towels.

12 Label the Whirl-Pak® Bag with a Sharpie or water-proof marker.

13 Place the Whirl-Pak® Bags into the cooler box filled with ice for storage.

14 Proceed with REDI-NET SOP S-1 Soil Collection at the same sampling location.

15 Repeat the water and soil sampling 3 times per sampling location point.

16 Repeat steps 8-15 at the one meter mark at the same sampling location.

17 Clean boots thoroughly between sampling location sites.

18 Repeat steps 7-17 at the second sampling location until all the samples are collected.

19 When all samples for a collection site are taken, bring the samples back to the laboratory facility for further processing according to REDI-NET SOP W-2 Water Processing (filtration should be done within 24:00:00). 1d

WATER STORAGE

20 Keep freshly collected water in labeled whirl-pak bags at 4 °C cold chain (cooler box) until return to laboratory. Water samples can be stored at 4 °C for up to 1 week.

21 Once at the laboratory, it is optimal to filter the water samples and freeze the filter disk in separate, labeled ziplock bags as soon as possible. If water samples can not be processed on the same day of collection, whole sample bags should be frozen at -80 °C . If -80 °C storage is unavailable, water bags/ filter disks should be

> stored at -20 °C until processing. Water samples can be stored at -20 °C for up to 1 month and at -80 °C for longer term storage.

Protocol references

REFERENCES

REDI-NET Overview Summary