

STANDARD OPERATING PROCEDURE FOR SAMPLE COLLECTION AND IDENTIFICATION OF HARMFUL ALGAL BLOOMS



WATER QUALITY

State of Utah
Department of Environmental Quality
Division of Water Quality

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Foreword

Utah Division of Water Quality (DWQ) Standard Operating Procedures (SOPs) are adapted from published methods, or developed by in-house technical experts. This document is intended primarily for internal DWQ use. This SOP should not replace any official published methods.

Any reference within this document to specific equipment, manufacturers, or supplies is only for descriptive purposes and does not constitute an endorsement of a particular product or service by DWQ. Additionally, any distribution of this SOP does not constitute an endorsement of a particular procedure or method.

Although DWQ will follow this SOP in most instances, there may be instances in which DWQ will use an alternative methodology, procedure, or process.

The methodology detailed below is the protocol followed by DWQ's monitoring staff and verified by DWQ's Quality Assurance officer.

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Revision Page

Date	Revision #	Summary of Changes	Sections	Other Comments
2019	4	Combined multiple version of the HAB SOP	All	Combined and updated phytoplankton collection SOP with HAB sampling SOP, and added language about toxin analysis.
3/10/2020	5	Updated language, grammar, and structure	All	Clarified and revised sentence structure and grammar throughout the entire document.
6/15/2020	5.1	Added QA/QC section	11	Increased and clarified the QA/QC protocols for HABs.
4/26/22	5.2	Updated contact information	1.0	
4/26/22	5.2	Updated sample storage and preservative usage	8.0, 9.0	
4/26/252	5.2	Clarified instances for sample collection and replicate collection	9.0, 11.0	
4/26/22	5.2	Updated bloom report form	14.0	

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1.0 SCOPE AND APPLICABILITY

This document presents the Utah Division of Water Quality's (DWQ) Standard Operating Procedure (SOP) for collecting water samples during harmful algal blooms (HABs). HABs can occur when certain cyanobacteria, a type of phytoplankton, become abundant enough to change the visual and physical nature of the waterbody. A HAB is defined as an aggregation or accumulation of either toxic or potentially-toxic cyanobacteria that poses a reasonable exposure risk to the public. Although technically inaccurate, the terms "algae" and "algal" are commonly used to refer to both algae and cyanobacteria. Most water protection agencies have adopted the term "harmful algal bloom" to describe these events, and for consistency, DWQ will use the same terminology.

This SOP is followed by all DWQ monitors and is recommended as the procedure for DWQ cooperators or local health department (LHD) staff performing HAB sampling in lakes, reservoirs, rivers, or streams. Any deviations from this procedure should be documented on the DWQ field forms (**Appendix 1**) prior to sample submission to the lab.

HAB samples collected utilizing this protocol may be used to determine the following:

1. Phytoplankton taxa identification and abundance.
2. Concentrations of cyanotoxins: anatoxin-a, cylindrospermopsins, microcystins, and nodularins (results derived from ELISA-CAAS, ELISA test strips, and LC-MS/MS test procedures).
3. Cyanotoxin gene detection/relative concentration (qPCR).

The goal of HAB sampling is to provide results that may be used:

1. To provide LHDs with information to evaluate potential recreational health risks.
2. To provide public water systems with information to evaluate potential risks to drinking water supplies.
3. To record bloom conditions (for DWQ) for use in evaluating water quality across the state.

If sampling as a cooperator:

Prior to any sample collection, contact the DWQ HAB Coordinator Hannah Bonner (hbonner@utah.gov, 385.258.6057) or Ben Holcomb (bholcomb@utah.gov, 801.536.4373). DWQ will coordinate efforts to identify sample types, quantity, and locations appropriate to HABs on a case by case basis. Additionally, the DWQ coordinator will determine if adequate funding is available to cover HAB sampling costs until completion of the bloom.

Cooperators should not assume that DWQ will pay for phytoplankton or cyanotoxin samples without prior notice and coordination.

Prior to any sample collection, DWQ will coordinate with the sample testing lab to ensure:

1. The lab is prepared for an additional workload
2. The appropriate samples are collected
3. The correct level of analysis is conducted
4. The expected reporting time is specified

DWQ's methodology for HAB sampling is based on a targeted, high concentration sample. These methods are not to be used to quantify or generalize abundance of cyanobacteria cells (cells/mL) or concentrations of cyanotoxins ($\mu\text{g/L}$) in the waterbody as a whole. Instead, these methods are designed to reasonably quantify the highest potential for exposure at high recreation sites.

2.0 SUMMARY OF METHOD

The primary purpose of this method is to characterize the nature of the bloom in the context of plausible exposure pathways, especially blooms with potential to harm people and animals. Therefore, sampling activities should target areas where there is a reasonable maximum risk of human-cyanotoxin interaction and exposure.

The "Reasonable Maximum" area on a waterbody is the area with the highest visible bloom accumulation at the location with the highest recreational potential.

The justification for this technique is to report on the highest potential for human exposure. This technique, along with the consideration that blooms are highly influenced by wind and current, allows DWQ to make recommendations based on the reasonable maximum location that are protective of human health and the environment. Prior to sampling, explicit site locations can be found in a project-specific SAP, where a site list identifies public access locations such as beaches, piers, shoreline access, etc. Therefore, the site conditions on the day of the sampling will be the determining factor for the reasonable maximum location.

Samples should be collected in areas of the lake where there is evidence of a potential bloom at the time of sample collection. There are two sampling techniques DWQ uses to collect samples based on the type of bloom. If the bloom is accumulating on the surface of the waterbody, a surface scum sample is collected to target the top 1-2 inches of the water column. If the bloom is concentrated in the water column, DWQ uses a composite sampling technique. Generally, when a surface sample is collected, a composite sample is also collected to better capture the extent of the bloom. However, cooperators should verify with DWQ to ensure that there is funding for both sample types.

In conjunction with water sampling, a field form is also filled out. This field form includes visual estimates, documented by taking photographs and GPS coordinates, weather conditions, and important tracking sample information. This information is requested when filling out the DWQ's field form (**Appendix 1**). Routine collection performed by DWQ should also be accompanied by multi-parameter sonde (in-situ) conditions. Consult DWQ's multi-parameter sonde SOP for further information.

This SOP details the procedure for collecting HAB samples in general. For specific information regarding response style sampling, please contact the program coordinator.

3.0 DEFINITIONS

DWQ Division of Water Quality

Field Blank	Performed after equipment decontamination from a previous site. Transfer containers and sampling bucket are filled ½ full with site or DI water, agitated, and emptied three times. This process is called a triple rinse. Triple-rinsing is an EPA verified procedure proven to remove and/or dilute contamination. It is important that every piece of reusable equipment that encounters site water is rinsed.
Field Replicate	Two composite samples taken from adjacent undisturbed locations. Replicates are collected and preserved, stored, handled, and analyzed identically to other samples.
HAB	Harmful algal bloom
L	Liter
LHD	Local Health Department
m	meter(s)
mL	milliliter(s)
Reasonable Max	The area with the highest visible bloom accumulation combined with the area with the highest recreational potential (inherent in DWQ’s site list).
Site ID	DWQ’s unique code for naming waterbody locations
SOP	Standard Operating Procedures
UDEQ	Utah Department of Environmental Quality

4.0 HEALTH AND SAFETY WARNINGS

Algal blooms may contain toxin-producing cyanobacteria. Samplers should wear elbow/shoulder length gloves, eye protection (such as goggles), and waders/boots during sampling.

Do not ingest water or allow the water to come into contact with exposed skin. Avoid inhaling spray caused by boats, wind or other water surface disturbances. If these conditions are present, wearing a face shield may reduce the risk of inhaling large droplets.

Hands should be washed thoroughly after sampling and before eating or drinking. Waders/boots should be rinsed of algal material using tap water (not lake water) before storage.

It is important that monitors also watch for and report any symptoms of exposure to cyanotoxins, which can occur immediately to several days following exposure. Potential symptoms include:

- Liver toxicity – may take hours or days for symptoms to appear in animals and humans; they include abdominal pain, diarrhea, and vomiting.
- Kidney toxicity – acute, severe gastroenteritis (including diarrhea and vomiting).

- Neurotoxicity – symptoms often appear within 15 to 20 minutes of exposure; animals may experience increased salivation, weakness, staggering, convulsions, difficulty breathing, and in severe cases, death. Humans may experience numb lips, tingling fingers and toes, or dizziness.
- Respiratory problems – runny eyes and nose, sore throat, and asthma-like symptoms.
- Skin irritation – visible rash, hives, or blisters, especially under clothing, swimsuits, or wetsuit.

If any of these symptoms occur, monitors should leave the impacted area and seek medical treatment immediately. Also, these circumstances should be documented and filed on Form 122 by your employer within 7 days.

Field personnel should take appropriate precautions when operating watercraft and working on, in, or around water. All boats should be equipped with safety equipment such as personal flotation devices (PFDs), oars, air horn, etc. Utah's Boating Laws and Rules shall be followed by all field personnel.

Field personnel should be aware that hazardous conditions potentially exist at every waterbody. If unfavorable conditions are present at the time of sampling, the sample visit is recommended to be rescheduled. If hazardous weather conditions arise during sampling, such as lightning or high winds, personnel should cease sampling and move to a safe location.

5.0 CAUTIONS

When operating a boat, hidden hazards exist underwater. Boat operators should take caution when sampling to avoid equipment damage.

Adverse sampling conditions could increase the likelihood of equipment damage. Boat operators should take extra caution when sampling under adverse conditions. If conditions are unsafe, reschedule sampling.

6.0 INTERFERENCES

Care should be taken not to include the lake bottom materials that may be disturbed and suspended if wading.

Minimize duckweed, sediment, etc. in the sample. High turbidity or dense aquatic vegetation may also interfere with sample analysis.

Samples should not be exposed to elevated temperatures during storage (i.e., do not store in a hot vehicle outside of a cooler).

7.0 PERSONNEL QUALIFICATIONS/RESPONSIBILITIES

DWQ personnel performing water sampling must be familiar with sampling techniques, safety procedures, proper handling, and record keeping. Samplers are responsible for attending refresher meetings held each spring/summer to review procedures and techniques. New staff will be trained in the field and lab by DWQ personnel.

Cooperators are required to read this SOP annually and acknowledge they have done so via a signature page (see **Appendix 2**) that will be kept on-file at DWQ along with the official hard copy of this SOP.

8.0 EQUIPMENT AND SUPPLIES

- Copy of this SOP
- Protective equipment:
 - extended gloves, safety goggles, face shield, chest/hip waders, and PFD
- One of each bottle:
 - ½ gallon plastic bottle, amber 250 mL glass bottle (Microcystin), amber 250 mL glass bottle (Anatoxin-a) with preservative, 250 mL plastic bottle (phytoplankton).
- Clean 2 gallon or greater HDPE bucket for compositing samples
- Tablet with camera
- GPS if not included with tablet
- Pencils and sharpies
- DWQ field form (**Appendix 1**)
- Cooler with ice
- Chain of Custody forms
- Gluteraldehyde, 25% preservative

9.0 PROCEDURE

Sampling for HABs has two sections: visual assessment and sample bottle collection. The visual assessment is done at every location on the waterbody and influences if and where sample bottles are collected. Collection of sample bottles occurs only at the location determined to be the “reasonable maximum” location of the HAB on the waterbody. If no bloom is observed, then no sample bottles will

be collected. *The only exception to this* is if this location has had high toxin levels and/or high cell count levels within the last two weeks. If this is the case, it is considered “response sampling” and sample bottles should be collected to verify that the bloom has dispersed. This sample should be taken from the same location as the previous sample.

If there are any questions on when or where to sample, contact the HABs Program Coordinator.

Note: A sampler may also take a phytoplankton sample only at locations where the phycocyanin reading is 75-100, even if no bloom is observed and no response sampling is occurring.

9.1 Visual Assessment

Due to the varied nature of HABs, a visual assessment of the bloom should be conducted prior to sample bottle collection. The visual assessment informs whether a set of bottles should be collected. A visual assessment is done by following DWQ’s field form (**Appendix 1**). This form may also be filled out electronically.

1. Obtain the DWQ field form
2. Fill out to include:
 - a. Sampler name
 - b. Location:
 - i. Waterbody, access point, GPS location
 - c. Date/Time
 - d. Bloom observed: (y/n)
 - i. If yes: collect bottles and continue through the field form**
 1. Location in water column: (surface/water column/both)
 2. Colors: (green, blue, brown, white, etc)
 3. Appearance of bloom: Spilled paint, grass clippings, clumps, etc. (See photos in **Appendix 3**).
 4. Extent of bloom: (in ft²)
 5. Photos: (close-up/landscape)
 - ii. If no: continue through the field form**
 - e. Site Photos: (general)
 - f. Samples collected: (y/n), if yes, type (surface/composite/both)
 - g. Weather conditions: (wind direction, wind speed, cloud cover, etc)

- h. Comments: (high recreation, fish kill, increase in accumulation from last visit, etc)
3. Send to the DWQ HABs Coordinator

9.1.1 Determination for Sample Bottle Collection

Below are the three cases in which collection of sample bottles will occur:

1. If there is one location on the waterbody:
 - a. If a bloom was observed in the visual assessment, then sample bottles will be collected.
2. If there are multiple locations on a waterbody
 - a. If multiple locations have visual bloom material, sample at the highest visual accumulation location.
3. If the waterbody had high toxin levels or cell counts in the previous 2 weeks (Response).
 - a. Samples will be taken regardless of a visible bloom.

9.2 Sample Bottle Collection

9.2.1 Sample Collection Method Determination

Samples should be collected if there is a visual bloom observed during the visual assessment, or if a bloom was confirmed in this location during the previous two weeks. There are two techniques for sampling HAB bloom material: a surface scum sample and a composite sample.

Surface Scum Sample: Select this method if the bloom forms a concentrated algal mat or scum on the surface of the water.

- Targeted, sample is collected directly from the water surface
- Bottle immersion at the surface (top 1-2 inches)

Composite Sample: Select this method if the bloom is concentrated in the water column.

- Elbow-depth integrated, sample is collected from a bucket
- 3x bottle immersion from elbow depth to the surface

Note: If a surface scum sample is collected, take a composite sample as well. If both methods are being performed, two sample sets should be collected (6 bottles total). If it is only a water column bloom, a composite sample is the only sample type collected.

9.2.2 Surface Scum Sample Procedure

If the bloom contains a concentrated algal mat or scum on the surface of the water, perform a surface grab sample from the center of the bloom with all three bottles (Anatoxin-a, Microcystin, and phytoplankton)(See **Figure 1**).

1. Put on protective gear (gloves, waders, etc.)
2. Target the highest visual accumulation area
3. Tilt the bottle parallel to the water surface to capture the top 1-2 inches of the surface water scum.

Note: For thick mats, you may need to help push the material into the bottle. The precise locations of these samples may be determined by the bloom extent and water uses for that particular waterbody.

4. Immediately, these samples should be placed on wet ice in a cooler.

Note: This method, although generally used for shore sampling, can be used to collect sample bottles from a dock or a boat.

9.2.3 Composite Sample Procedures

If the bloom is concentrated in the water column, or a surface scum sample was already taken, a composite sample is collected. This procedure requires the collection of three grab samples from elbow-depth to the surface in 1/2 gallon bottles. These triplicate samples are collected 10 ft apart and composited into a bucket, mixed, and poured into the Anatoxin-a, Microcystin, and phytoplankton sample bottles (See **Figure 1**). This sample type may be collected near-shore and in open water following this procedure:

1. Put on protective gear (gloves, waders, etc.)
2. Triple rinse any reusable sampling equipment (bucket and 1/2 gallon transfer bottle).
 - a. Triple rinse by filling up the sampling container half full, agitating, and emptying the container (x3). The phytoplankton and amber bottles SHOULD NOT be rinsed.
3. Carefully wade into the waterbody until knee deep and avoid collecting sediment stirred from the bottom of the waterbody.
4. Remove the lid of the 1/2 gallon plastic transfer bottle and carefully dip the inverted bottle beneath the surface of the water to elbow depth and revert the bottle and bring to the surface, evenly sampling as much of the water column as possible.
5. Pour contents into a triple rinsed bucket.
6. Walk 10 feet in one direction (paralleling the shoreline) from the first sample collection point to grab the second subsample.
7. Pour contents into the bucket
8. Walk another 10 feet to grab the third subsample, pouring it into a bucket after each sample (see **Figure 2**).

Note: Take extra care when paralleling the shoreline to minimize disturbance of the bottom sediments (i.e. do not sample the kicked up sediment plume).

Note: This method can be altered to collect sample bottles from a dock or a boat. After rinsing equipment, lay on the dock or boat, facing the water, to collect the sample from elbow depth to the surface. Move roughly 10 ft in any direction (on the boat or dock) to collect the second and third subsamples in the same way as the initial sample.

9. Return to shore and mix the samples by agitating the bucket
10. Pour water contents from the bucket into the three subsamples: Anatonxin-a, Microcystin, and phytoplankton (See Figure 1).
11. Immediately store the samples in a cooler on wet ice or ice packs.

9.3 Sample Processing

1. For samples to be delivered to a lab, fill out the appropriate chain-of-custody form, see project-specific Sampling and Analysis Plan.
2. Samples must be kept in a cooler on wet ice, or otherwise refrigerated in dark conditions until delivery to the lab for analysis.
3. Submit cyanotoxin samples within 48 hours.
 - a. If the samples are to be stored for more than 48 hours, place the glass jars in the freezer until delivery. Be sure the jar is only about half full to ensure the jar does not break.
4. Submit phytoplankton bottles to PhycoTech, Inc. within 24 hours.
 - a. If samples are to be stored for more than 24 hours, preserve sample with glutaraldehyde, 25%. 1 mL glutaraldehyde / 100mL sample and store in a dark, cool (< 25°C) location.

10.0 DATA AND RECORDS MANAGEMENT

All data recorded in the field should be reviewed for completeness before leaving the sample site. Before delivering samples to a laboratory, ensure that all lab sheets and chain-of-custody forms have been filled out correctly and completely, and sample information is consistent with bottle labels. For cyanotoxin samples, pay particular attention that the correct sample type is noted on all lab sheets.

Phytoplankton samples will be identified to the lowest possible taxonomic level (generally species) and enumerated, as negotiated with one or more contracted laboratories. Until further notice, phytoplankton will be analyzed by PhycoTech, Inc.

Data is returned to DWQ and populated in a spreadsheet along with the cyanotoxin data. This spreadsheet is shared with cooperators and LHD's to aid in advisory decisions.

Cyanotoxin samples will be analyzed by one or more contracted laboratories using ELISA-CAAS and LC-MS/MS procedures to determine the presence and concentration of specific toxins.

All field data will be reviewed by the DWQ monitoring field lead for accuracy of site ID's and completeness. All laboratory data will be received by the HABs program coordinator and evaluated for accuracy using the available lab-QC results and field-sample data-flags.

11.0 QUALITY ASSURANCE AND QUALITY CONTROL

QA/QC procedures for HAB monitoring are slightly modified from DWQ standard sample collection procedures due to the episodic nature and high spatial variability of suspected blooms. As described earlier and when required, HAB-samples will be collected from targeted areas representative of the reasonable maximum risk of human-cyanotoxin interaction and exposure, notably the interior of a surface-scum or upper water column bloom.

Since action levels for many cyanotoxins are in the low-ppb range, and since algal blooms can strongly affect the concentrations of other constituents in the waterbody (e.g. dissolved and particulate organic matter), key sample-QC concerns involve (i) detection of background contamination, and (ii) ensuring the accuracy of low (5 to 20 ug/L) cyanotoxin concentrations from diverse sample matrices. For most laboratories, DWQ requests that laboratories report analytical details equivalent to a Level II data package for all cyanotoxin results. Of primary interest to the DWQ-HAB program are results from laboratory method blanks (also referred to as 'laboratory reagent blanks') and sample matrix spikes (aka 'lab-fortified sample matrix') for each analytical batch, including any result-specific data-flags that indicate that the analysis may be outside control limits.

Because of the high sample-analysis cost and high spatiotemporal variability of suspected blooms, field replicates should be collected for approximately 5% of composite samples (note: not the surface-scum sample), and field blanks may be collected monthly or as detailed in a project-specific sampling and analysis plan (SAP). When possible, i.e. when project-resources and analytical-sampling equipment are available, additional split-samples may receive lower-level field-spikes to quantify the analytical accuracy of reported cyanotoxin concentrations and to demonstrate low-levels of matrix interference.

Field replicates should be collected from bloom sites that have a visual bloom of about 200ft² or greater to avoid sample disturbance from the initial collection. Collect a sample following the same protocol as the initial sample and make note of any differences between sample appearances.

12.0 REFERENCES

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Ohio DOH, EPA, and DNR. 2015. State of Ohio Harmful Algal Bloom Response Strategy for Recreational Waters. Available at: <http://epa.ohio.gov/portals/35/hab/HABResponseStrategy.pdf>

Oregon Health Authority. Oregon Harmful Algae Bloom Surveillance (HABS) Program. Public Health Advisory Guidelines. Available at: www.healthoregon.org/hab

Proceedings of the interagency, International Symposium on Cyanobacterial Harmful Algal Blooms (ISOC-HAB): State of the Science and Research Needs. Available at: www.cdph.ca.gov/HealthInfo/environhealth/water/Documents/BGA/ISOCHABdocument.pdf

Related DWQ SOPs

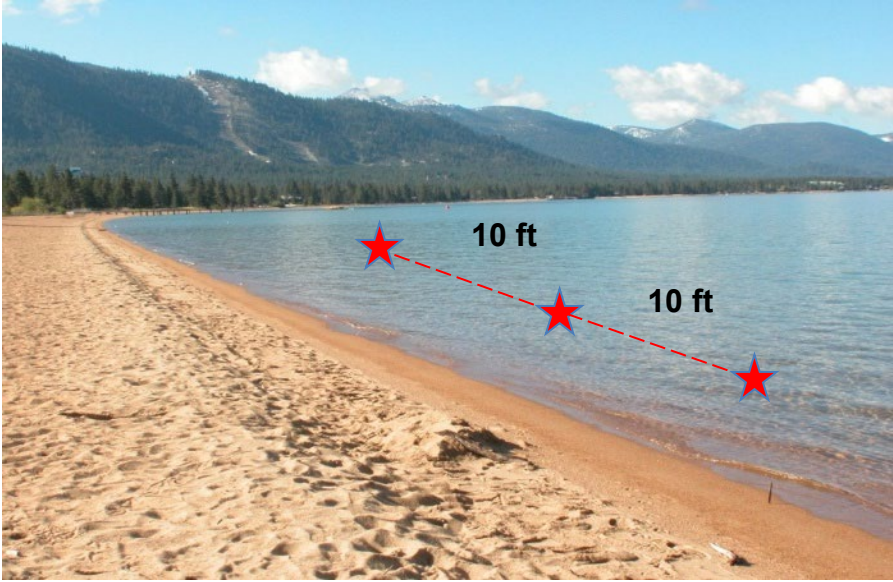
Standard Operating Procedure for Calibration, Maintenance, and Use of Multiparameter Water Quality Sondes

13.0 FIGURES

Figure 1: HAB Sample Bottles



Figure 2: Composite Sampling Diagram



14.0 APPENDICES

Appendix 1: Bloom Report Form

Utah DWQ Cyanobacteria Bloom Report Form

Recreation Season 2022

Fill in all information, as applicable



UTAH DEPARTMENT of
ENVIRONMENTAL QUALITY
**WATER
QUALITY**

Sampler Name: _____

Contact Information (email/phone): _____

Organization: _____

Sample location: _____

Site description: _____

GPS Coordinates (latitude/longitude): _____

Sample Date: _____ Sample Time: _____

Bloom Observed? <input type="checkbox"/> Yes <input type="checkbox"/> No	Bloom Colors Observed (select all that apply): <input type="checkbox"/> Green <input type="checkbox"/> Rust <input type="checkbox"/> Blue <input type="checkbox"/> Brown <input type="checkbox"/> Red <input type="checkbox"/> White <input type="checkbox"/> Milky <input type="checkbox"/> Other: _____ <input type="checkbox"/> Purple
Estimated Bloom Extent (ft²): <input type="checkbox"/> 1-10 <input type="checkbox"/> 10-50 <input type="checkbox"/> 50-200 <input type="checkbox"/> >200	Material Present: <input type="checkbox"/> Spilled Paint <input type="checkbox"/> Grass Clippings <input type="checkbox"/> Cottage Cheese <input type="checkbox"/> Isolated Clumps <input type="checkbox"/> Decaying Bloom Material <input type="checkbox"/> Other: _____
Location in Water Column: <input type="checkbox"/> Surface Only <input type="checkbox"/> Water Column Only <input type="checkbox"/> Both Surface and Water Column	
Sample Type Collected (select all that apply): <input type="checkbox"/> Surface <input type="checkbox"/> Composite	
Additional Comments (wind speed/direction, recreational activity, nutrient inputs, etc.): 	
Please Take Pictures: <u>Landscape</u> (to show extent) & <u>Close up</u> (to show bloom material)	

Please contact Ellen Bailey ASAP to coordinate sample submission. Submissions may be limited due to priority or lab capacity. Samples need to be kept cold and in the dark.

Send Pictures and form to Ellen Bailey via email: ellenbailey@utah.gov or text 407-310-8883.

SOP Training Form

Training is required for all new samplers and Refreshers are required annually. This signature page

Trainee: Sign below to acknowledge that training on this SOP was received, understood, and all questions/concerns were addressed by the trainer.

Trainer: Sign below to acknowledge that training on this SOP was completed for the individual listed and that trainee is competent to perform the procedures described within.

Date of Training	Trainee Printed Name	Trainee Signature	Trainer Printed Name	Trainer Signature

Management Approval

Printed Name:

Signature:

Date:

Appendix 3: Cyanobacteria Identification Guide

Commonly Identified Cyanobacteria

- Types (pictured below)
 - Spilled Paint
 - Grass Clippings
 - Cottage Cheese
 - Decaying “spilled paint” bloom
 - Isolated chunks
- In general, the water will have a bright green hue and lack water clarity
- Typically accumulates along shorelines, but does persist in the open water



Spilled Paint



Grass Clippings



Cottage Cheese



Decaying bloom material



Isolated clumps

Commonly *Misidentified* Cyanobacteria

- Duckweed (bottom photo)
 - Can form into mats
 - Has small clover like leaves
- Filamentous algae
- Macrophytes

