

FINAL
2009 Klamath River
Blue-Green Algae Summary
Report



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I. Introduction

This report summarizes the presence of toxicogenic cyanobacteria in the Klamath River within the Yurok Indian Reservation (YIR) boundaries in 2009. The Yurok Tribe Environmental Program (YTEP) and the Karuk Tribe collaborated to monitor water quality conditions from downstream of Iron Gate Dam to the Klamath River Estuary. The Karuk Tribe will be publishing a report that summarizes the conditions from Orleans to just downstream of Iron Gate Dam. Results from water samples collected in 2009 indicated that the water quality in the Klamath River was negatively impacted by levels of the cyanobacterium *Microcystis aeruginosa* (MSAE) and its resultant toxin, microcystin. MSAE and microcystin levels within the YIR boundaries did not exceed the State of California's recommended thresholds for recreational waters in 2009. However, MSAE and microcystin levels upstream of the YIR did exceed the State of CA's recommended thresholds for recreational waters in the Klamath River, Iron Gate Reservoir and Copco Lake.

Cyanobacteria, also known as blue-green algae (BGA), are commonly found in many freshwater systems across the world. The species of concern here are known as toxigenic species, since they have the potential to produce chemicals that are toxic to humans and animals. In general, the toxins produced by these algae can be divided into two groups, those which can cause liver damage (hepatotoxins) and those which can damage the central nervous system (neurotoxins), although other health effects are possible.

At least 46 species of cyanobacteria have been shown to be toxic to vertebrates (Chorus & Bartrum, 1999). Some of the more common toxin-producing genera include *Microcystis*, *Anabaena*, *Aphanizomenon*, *Lyngbya*, *Nodularia*, *Planktothrix*, *Nostoc* and *Cylindrospermopsis*. It should be noted that cyanobacteria likely produce toxins that have not been characterized.

Microcystis aeruginosa is a type of blue-green algae which releases the liver toxin microcystin when it dies and decomposes. Microcystin can cause rashes, skin irritation, conjunctivitis, nausea, vomiting, diarrhea, liver damage, tingling, numbness, paralysis, and death in humans and animals. Microcystin causes the most damage when it builds up in the liver; it also accumulates in other organs and in the muscle tissue of humans and animals. Microcystin is not excreted by humans and animals, so the dose can increase over time. When a large enough dose accumulates liver damage, increased liver size and death can result. Mortality in fish, domestic animals, and humans has been recorded following exposure to microcystin from a single-dose and from long-term exposure.

Exposures Pathways

The primary exposure pathway of concern for exposure to cyanotoxins is through ingestion of water. Skin irritation can result from exposure to the algae itself, however the cyanotoxins are not likely to cross the skin barrier and enter the bloodstream. Inhalation of microcystin is possible, especially during activities such as water skiing or splashing, where contaminated water is aerosolized.

Ingestion of contaminated water can occur through both incidental and intentional pathways. Incidental ingestion most commonly occurs during recreation especially in turbid or discolored lakes. The risk of incidental ingestion of the toxin is particularly high for children playing in

near-shore areas where algal scum tends to accumulate. Because of their small body size, children are at greater risk from exposure—it takes a smaller dose to make them sick than it does to sicken an adult. Exposure levels can be broadly defined as high, moderate and low based on recreational activity (Table 1).

Table 1. Exposure level of recreational activity (modified from Queensland Health, 2001).

Level of Exposure	Recreational Activity
High	Swimming, diving, water skiing
Moderate	Canoeing, sailing, rowing
Low to none	Fishing, pleasure cruising, picnicking, hiking

At this time, there is insufficient information to determine the risk of consuming fish caught in waters with toxigenic cyanobacteria. Studies have shown that toxins mainly accumulate in the liver and other internal organs of fish, although microcystin has been detected in the fillet (Vasconcelos, 1999; de Magalhães et al., 2001). However, Fetcho 2006 reports that no microcystin was detected in salmon or steelhead filets collected from fish sampled at Weitchpec during the 2005 *Microcystis* bloom. At a minimum, the internal organs and skin should be removed and discarded prior to cooking fillets. Shellfish have been shown to accumulate cyanotoxins in edible tissue (Vasconcelos, 1999). It is recommended that people call the Department of Human Services for more information on fish consumption while advisories are in effect.

Detrimental Environmental Effects of Microcystis aeruginosa

In addition to causing many known, well-documented human and animal health effects, microcystins can have a detrimental effect on the food chain by limiting growth of beneficial phytoplankton species, discouraging zooplankton feeding and population growth, decreasing total dissolved oxygen in the water column, and ultimately lowering success of fish and other large organisms.

“Even low microcystins concentration at the base of the food web poses a threat to the upper food web because microcystins may bioaccumulate.” The impact of *Microcystis* species on the quantity and quality of phytoplankton biomass available to the food web may be a greater threat to the food web than toxicity. Blooms can reduce growth of other phytoplankton species because of their buoyancy and ability to block light further down the water column, and their relative ability to out-compete species which cannot tolerate high light and temperatures at the surface. Dissolved microcystin in the water may also inhibit feeding by zooplankton (De Mott et al., 1991). In addition, high biomass produced by blooms and the associated decomposition can eventually impact fishery production through influence on dissolved oxygen concentration (Lehman et al., 2005).

Microcystin Toxin Information

WHO has established minimum tolerance levels for recreational contact with microcystin. Because of the time it takes to analyze water samples for the presence of microcystin, WHO recommends the use of cell counts per milliliter of water as a crude surrogate for concentrations of microcystin. However, because the toxin is released as the organism decomposes, the risk

from microcystin presence in waters is at its greatest after the bloom has initially begun to decompose and increases until well after the last cells are observed in samples.

WHO has set the following thresholds for MSAE/microcystin concentrations in recreational waters:

	<u>Microcystis cells/milliliter</u>	<u>Microcystin micrograms/liter (µg/L)</u>
Low Risk:	20,000	4
Moderate Risk:	100,000	20
Severe Risk	10,000,000 <i>or</i> visible scum	200

The consumption limit for microcystin is set as 0.04 micrograms per kilogram of bodyweight per day. However, because even the consumption of relatively low doses of microcystin over time will damage the liver of animals, continued consumption of known contaminated food sources is not recommended.

The State of CA has set thresholds for posting waterbodies to minimize impacts to recreational users in a document titled “Cyanobacteria in California Recreational Water Bodies Providing Voluntary Guidance about Harmful Algal Blooms, Their Monitoring, and Public Notification”.

The State of CA’s has set the following thresholds for posting recreational waters:

- Scums present containing toxigenic* species
- MSAE or *Planktothrix* \geq 40,000 cells/ml
- Population of potentially toxigenic BGA species \geq 100,000 cells/ml
- Concentration of microcystin \geq 8 ppb

*Potentially toxic blue-green algae that have been detected in California include those of the genera *Anabaena*, *Microcystis*, *Aphanizomenon*, and *Gloeotrichia*. Additional bluegreen algae that are known to be potentially toxic may be added to this list.

II. Methods

YTEP follows methods as specified in the USEPA approved “Lower Klamath River Nutrient, Periphyton, Phytoplankton and Algal Toxin Sampling Analysis Plan (SAP)” for samples collected for baseline monitoring purposes. At each sample site, sample water was collected with a pre-rinsed churn splitter as specified in the grab sample protocol located in Appendix B. The 14 Liter churn was rinsed three times with distilled water followed by three rinses with site river water. Samples were drawn in a moving portion of the river in an attempt to collect water samples to represent the river as a whole. The churn splitter allowed for distribution of a homogenous water mixture into sample bottles used for algal identification and enumeration and testing for microcystin.

YTEP follows methods for the collection of water samples and freshwater mussel tissue samples collected for public health purposes as specified in the Standard Operating Procedures Environmental Sampling of Cyanobacteria for Cell Enumeration, Identification and Toxin Analysis which was developed for the 2009 AIP Interim Measure 12 Water Quality Monitoring

Activities in the Klamath River which was prepared by the Klamath River Blue Green Algae Working Group.

At each sampling location, samplers conducted an initial visual survey of the public access area to identify where surface grab samples would be collected to represent a reasonable maximum exposure at that public access location (referred to hereafter as the RME location – see Section 2.3). Because cyanobacteria can accumulate and dissipate rapidly, depending on sun and wind conditions, a location having a greater presence of cyanobacteria should be identified within each designated public access area, where the public is likely to come into contact with cyanotoxins.

The sampler waded to where the sample was collected, and that sample was collected before other work was done at that location to minimize collection from a disturbed water column. Care was taken to avoid collecting particulates that are re-suspended as the result of accessing the sampling location. Using a glass wide mouth jar a grab sample was collected from the upper 10 cm of the water column.

The sample bottle for identification and enumeration of algal species contained Lugol's preservative and the toxin sample was preserved by freezing the bottle. Both of these samples were drawn from the same churn of water because they are complementary to one another. All samples were labeled with the following information: date, time, sampler, sample site, study name. The sample ID was comprised of a two or three digit site ID and the date (e.g. TG090108).

If a sampling crew member identified an area along the river that had scum lines, an additional sample was collected at this site. The sample was labeled appropriately and photographs of the sample area were taken. Additional quality control measures were included in the sampling. At one site per trip a replicate split sample was sent to the laboratory to assess laboratory performance and to gain improved confidence in the data.

Environmental information was also recorded at the time water samples were collected. The data included water temperature, pH, specific conductance, dissolved oxygen and other observational notes. Water samples were also collected to be analyzed for the concentration of nutrient analytes and sent to Aquatic Research Inc. in Seattle, Washington (WA). Chain-of-custody (COC) sheets were also filled out to document the handling of the samples from the time of collection to the time of laboratory analysis. This is a standard procedure for handling samples.

Water samples that were collected for algae speciation and enumeration were mailed overnight to Aquatic Analysts for analysis. Microscope slides are prepared at the laboratory from each sample by filtering an appropriate aliquot of the sample through a 0.45 micrometer membrane filter (APHA Standard Methods, 1992, 10200.D.2; McNabb, 1960). A section is cut out and placed on a glass slide with immersion oil added to make the filter transparent, followed by placing a cover slip on top, with nail polish applied to the periphery for permanency. Most algae are identified by cross-referencing several taxonomic sources.

Algal units (defined as discrete particles - either cells, colonies, or filaments) are counted along a measured transect of the microscope slide with a Zeiss standard microscope (1000X, phase contrast). Algal units are measured accurately to 0.1 mm with a stage micrometer. The algal densities are calculated from the area observed (transect length times diameter of field of view), the effective filter area, and the volume of sample filtered. Only those algae that were believed to be alive at the time of collection (intact chloroplast) are counted. A minimum of 100 algal units are counted. (Standard Methods, 1992, 10200.F.2.c.). If toxic cyanobacteria are present in the 100 algal units count the taxonomist then counts 4 times that area but only for the toxic species. Average biovolume estimates of each species are obtained from calculations of microscopic measurements of each alga. The number of cells per colony, or the length of a filament, are recorded during sample analysis to arrive at biovolume per unit-alga. Average biovolumes for algae are stored in a computer, and measurements are verified for each sample analyzed.

Water samples that were collected for microcystin processing were stored in glass containers and mailed on ice overnight to USEPA Region 9 lab in Richmond, California for analysis using the enzyme linked immunosorbent assay (ELISA) method. These methods have been adapted to a commercial ELISA kit (Microcystin Plate Kit, EP-022) that is produced by Envirologix, Inc. (Portland Maine), which USEPA Region 9 lab in Richmond, CA employs and measures total microcystin. Additional water and freshwater mussel tissue samples were submitted to the California Department of Fish and Game's Fish and Wildlife Water Pollution Control Laboratory in Rancho Cordova for the analysis of microcystin variants and anatoxin-a using liquid chromatography dual mass spectrometry (LC-MS/MS).

YTEP's real-time continuous water quality monitoring equipment on the Klamath River have phycocyanin probes that are designed to detect the presence of accessory pigment known to occur in *Microcystis aeruginosa* and other cyanobacteria. YTEP operates these data sondes according to the manufacturer's recommendations. Once the presence of MSAE was detected at sampling sites on the YIR additional samples were collected at the datasonde locations in the Klamath River to develop a relationship between the phycocyanin probe readings and blue-green algae laboratory cell counts.

III. Site Selection

In general, the various sampling locations were chosen in order to represent the average ambient water conditions throughout the water column. The sites listed below indicate established sampling locations for the collection of water samples for nutrient analysis and phytoplankton speciation and enumeration from May through October on a biweekly interval.

YTEP collected water samples for toxin and algae speciation analysis at the following mainstem Klamath River locations (river miles are approximate):

- **WE - Klamath River at Weitchpec (upstream of Trinity River) – RM 43.5**
- **TC – Klamath River Above Tully Creek (downstream of Trinity River)– RM 38.5**
- **TG - Klamath River at Turwar Boat Ramp – RM 6**
- **LES - Lower Estuary Surface – RM 0.5**

YTEP collected water samples for toxin and speciation analysis at the following major tributary location:

- **TR - Trinity River near mouth (above Klamath River confluence) – RM 0.5**

YTEP's datasondes on the Klamath River are close to the routine water sampling sites for Klamath River at Weitchpec and Klamath River at Turwar Boat Ramp, see figure 1. The Klamath River Above Tully Creek datasonde location is at the same location that routine water sampling site, see figure 1. The samples collected after datasonde maintenance occurred are denoted with a DCP (data collection platform) acronym to aid the sampling crew in tracking sample results.

YTEP collected water samples for toxin and speciation analysis at the following mainstem Klamath River datasonde locations (river miles are approximate):

- **WEDCP - Klamath River at Weitchpec (upstream of Trinity River) – RM 43.7**
- **TCDCP – Klamath River Above Tully Creek (downstream of Trinity River) – RM 38.5**
- **KATDCP – Klamath River above Turwar – RM 8**

YTEP collected freshwater mussels and water samples as part of the public health sampling funded under the AIP in the Klamath River above Starwein Riffle and in the Trinity River near the mouth (above Klamath River confluence), see figure 1. These sampling sites were selected due to the high quantity of freshwater mussels being present and of the appropriate size needed for tissue analysis.

YTEP collected water and freshwater mussel samples for toxin and speciation analysis at the following mainstem Klamath and Trinity River locations (river miles are approximate):

- **KASR – Klamath River Above Starwein Riffle – RM 10**
- **TR - Trinity River near mouth (above Klamath River confluence) – RM 0.5**

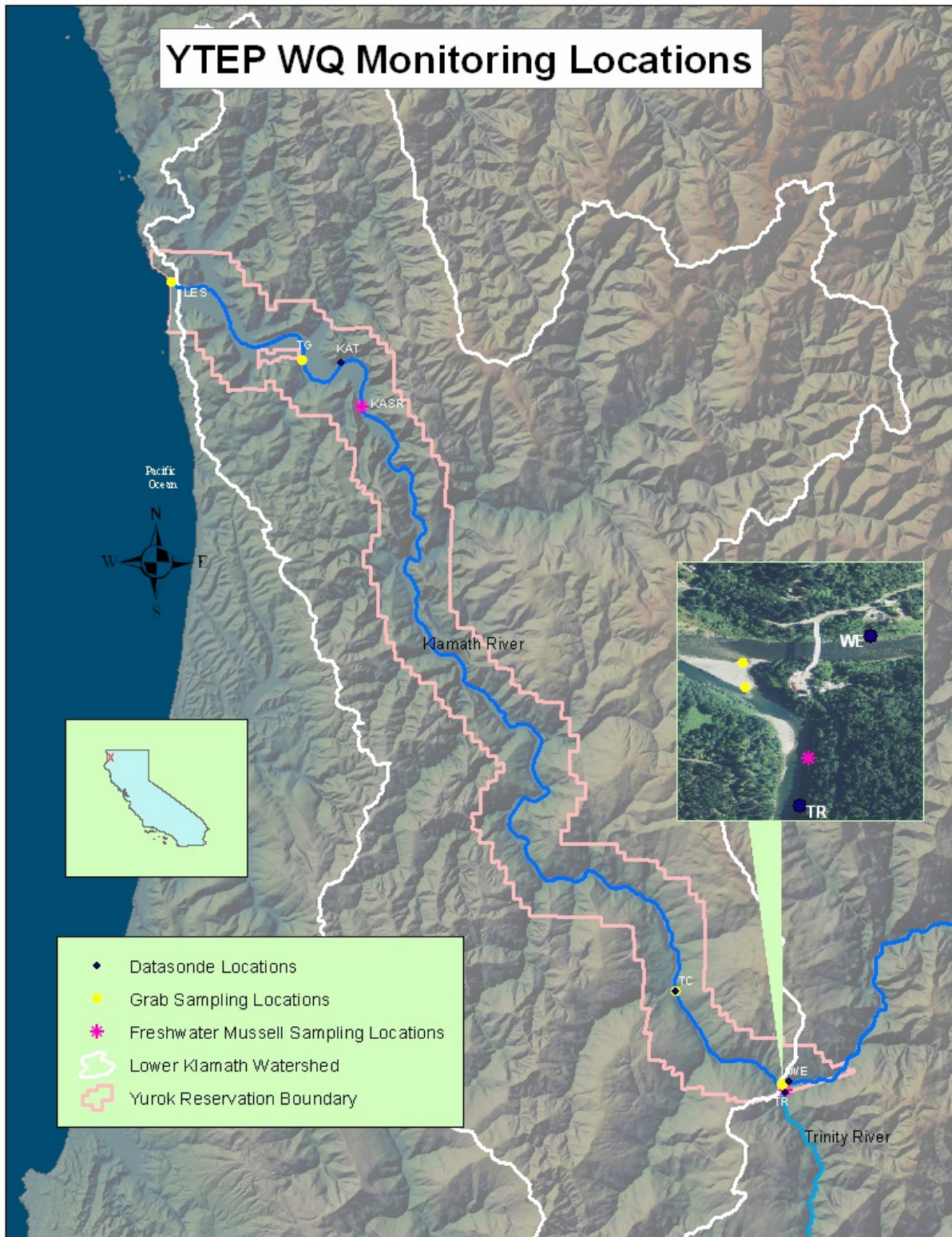


Figure 1. Map of phytoplankton and microcystin surface water and mussel tissue monitoring locations, 2009.

IV. Quality Assurance

YTEP follows methods as specified in the “Lower Klamath River Nutrient, Periphyton, Phytoplankton and Algal Toxin Sampling Analysis Plan (SAP)” approved by the USEPA in June 2008. These methods are consistent with the methods specified in the AIP SOP. Quality Assurance/Quality Control (QA/QC) of the collection, preparation and analysis of water samples for microcystin and phytoplankton speciation and enumeration was achieved by following a standard water sample collection protocol using a churn sampler and submitting samples to labs that follow strict protocol that have QA/QC measures. All field personnel that were involved in collection of water samples have been trained appropriately by the Water Division Program Manager and are properly supervised to ensure proper protocol is followed consistently throughout the monitoring season. Field crews collecting samples ensured representativeness of samples by selecting free-flowing water from established sampling locations and using a churn splitter to mix sample water once collected. All samples were transported to the appropriate laboratories following chain of custody procedures to ensure proper handling of the samples.

The collection and analysis of field replicate samples were performed during each sampling event to determine the labs’ precision of data. Since the Yurok and Karuk Tribes collaborated on this project it was necessary for each Tribe to alternate every other event to collect the QA/QC samples. YTEP collected QA/QC samples in the sampling event that was near the beginning of the month and the Karuk Tribe would collect the QA/QC samples near the end of the month. Field replicates were collected by splitting samples in the field using the churn splitter. One of the split samples was sent with its’ associated split with a different ID code for analysis of both algae identification and enumeration and microcystin so as to not alert lab staff of the fact that the samples were replicates.

Data is thoroughly reviewed once received from the laboratory. YTEP is the primary organization responsible for data review, although the professional laboratories analyzing water quality samples will also note potential problems with outliers or other anomalies in sample results. Information regarding QA/QC procedures for the laboratory is available upon request. One hundred percent of laboratory-generated data was checked on receipt by the Project Manager for consistency and acceptability, including whether replicates are within specified targets and meet data quality objectives. Any unusual values outside the range of norm will be flagged and all aspects of field data sheets, shipping handling and laboratory handling and testing will be reviewed. Outliers will be identified and removed from the dataset if deemed necessary by the QA Officer. Water temperature, conductivity, pH and dissolved oxygen are measured in the field when samples are collected and values of these hand-held measurements can be used to check field conditions at the time of sampling.

The data manager visually inspects all entered data sets to check for inconsistencies with original field or laboratory data sheets. Where inconsistencies are encountered, data will be re-entered and re-inspected until the entered data is found to be satisfactory or results will be discarded. The Project Manager will maintain field datasheets and notebooks in the event that the QA Officer needs to review any aspect of sampling for QA/QC purposes.

Phytoplankton

QA samples collected in June, July and August did not contain MSAE, therefore, no relative percent differences were calculated for these months. The common algae in each replicate did, at times, change rank for most samples collected between May and October (see table 2). However, the same species generally make up the bulk of the samples. Furthermore, the abundances (whether density, biovolume, or Trophic State Index (TSI)) were similar for the replicates. Considering the biological variability in this river the replicate results are not significantly different. Discrepancy among replicates may also reflect how well the churn was able to split particulates in samples.

The September QA replicate sample and primary samples did contain levels of MSAE and the RPD exceeded 20%. However, since both samples were both well below the posting level of 40,000 cells/ml the data was found to be acceptable since the required action would have been the same for both sample results.

Microcystin

QA sample results collected by YTEP and submitted to the lab blindly reported good precision by the laboratory and analysis method. 4 of the 5 samples submitted to the lab had RPDs under 20%, see table 3. One sample had a RPD greater than 20% however, this was due to such low level of toxins in the sample. Since both samples were both well below the posting level of 8 µg/L the data was found to be acceptable since the required action would have been the same for both sample results. All blank sample results submitted to the lab blindly were reported at less than the reporting limit of 0.18µg/L.

The QA samples collected by YTEP indicate that the toxin results are valid and acceptable.

V. Results:

Phytoplankton QA

Table 2. Phytoplankton results for the QA replicate samples collected by YTEP, 2009.

Site	Date	Total Density	Total Bio-volume	TSI	Sp1	Sp1%	Sp2	Sp2%	Sp3	Sp3%	Sp4	Sp4%	Sp5	Sp5%	#Spp	APF9 cells/ml	MSAE cells/ml	ABX9 cells/ml	Oscillatoria sp. Cells/ml
TG	5/13/09	520	151,425	36.3	DTTN	28.7	ACMN	19.8	COPC	6.9	RHCU	6.9	GFAN	6.9	21	0	0	0	0
TG Dup	5/13/09	515	212,231	38.7	ASFO	25.9	DTTN	15.3	ACMN	10.6	COPC	5.9	RHCU	5.9	23	0	0	309	0
TG	6/25/09	1,295	804,499	48.3	DTTN	42.6	EPSX	4.9	GFVT	4.9	MLGR	4.1	ACMN	4.1	29	0	0	0	0
TG Dup	6/25/09	1,525	864,872	48.8	DTTN	41.7	MLGR	7.8	COPC	7.0	EPSX	6.1	GFVT	5.2	25	0	0	0	0
TG	7/23/09	927	612,167	46.3	EPSX	25.2	NZFR	13.6	COPC	10.7	DTTN	5.8	RHCU	4.9	26	0	0	0	0
TG dup	7/23/09	738	486,845	44.7	EPSX	27.6	NZFR	11.4	COPC	11.4	DTTN	9.5	SCQD	8.6	24	0	0	0	0
TG	8/6/09	851	982,191	49.7	EPSX	37.6	COPC	17.9	SNUL	10.3	SCQD	7.7	RHCU	5.1	21	0	0	0	0
TG Dup	8/6/09	1,186	973,023	49.6	EPSX	40.2	COPC	20.5	SCQD	8.2	RHCU	5.7	FRCV	4.1	20	0	0	0	0
TG	8/20/09	1,627	707,893	47.4	RDMN	18.8	EPSX	10.9	STHN	10.9	NZPL	9.9	COPC	8.9	24	0	0	0	0
TG dup	8/20/09	1,712	776,609	48.0	COPC	12.5	RDMN	11.5	EPSX	11.5	STHN	10.6	CCMG	8.7	27	0	0	0	0
TG	9/17/09	1,565	1,726,514	53.8	EPSX	16.8	DTTN	13.7	MSAE	7.7	COPC	5.3	SNUL	5.3	34	132	2,764	0	0
TG Dup	9/17/09	2,135	1,153,132	50.9	MSAE	40.2	EPSX	10.7	SNUL	5.9	DTTN	4.7	NZFR	4.7	31	253	10,309	0	0

Key to Species Codes is located in Combined Species List located in Appendix A

APF9 = *Aphanizomenon flos-aquae* MSAE = *Microcystis aeruginosa* ABX9= *Anabaena* sp.

Microcystin QA

Table 3. Microcystin results for the QA replicate samples submitted to USEPA Region 9 lab collected by YTEP, 2009.

site	date	Depth	primary result	duplicate result	difference	RPD
TG	6/25/2009	SG	<0.18	<0.18	N/A	N/A
TG	7/23/2009	SG	0.26	0.15	0.1	53.7
TG	8/6/2009	OC	0.64	0.73	-0.1	-13.1
TG	8/20/2009	SG	0.49	0.55	-0.1	-11.5
TG	9/17/2009	SG	1.70	2.10	-0.4	-21.1

Phytoplankton

Table 4 Phytoplankton results for baseline water samples collected with a churn splitter in the Klamath River and mouth of Trinity River May - December 2009.

Station	Date	Total	Total	TSI	Sp1	Sp1%	Sp2	Sp2%	Sp3	Sp3%	Sp4	Sp4%	Sp5	Sp5%	#Spp	APF9	MSAE	ABX9	Oscillatoria
		Density	Biolume													cells/ml	cells/ml	cells/ml	sp. Cells/ml
LES	5/13/09	341	107,480	33.8	ASFO	27.8	DTTN	15.6	ACMN	11.1	RHCU	8.9	NZFR	7.8	19	0	0	0	0
TG	5/13/09	520	151,425	36.3	DTTN	28.7	ACMN	19.8	COPC	6.9	RHCU	6.9	GFAN	6.9	21	0	0	0	0
TC	5/13/09	639	400,801	43.3	DTTN	30.3	ACMN	20.2	RHCU	11.2	ASFO	7.9	COPC	6.7	20	0	0	0	0
WE	5/13/09	1,063	302,080	41.2	DTTN	38.4	ACMN	16.2	ASFO	9.1	NZDS	6.1	COPC	6.1	18	0	0	0	0
TR	5/13/09	238	96,632	33.1	DTTN	26.2	GFAN	18.5	RHCU	12.3	ACMN	7.7	CMMN	4.6	19	0	0	0	0
LES	5/28/09	1,684	804,343	48.3	ASFO	35.7	DTTN	29.5	ACMN	9.8	STHN	6.3	RHCU	3.6	18	0	0	0	0
TG	5/28/09	1,569	660,053	46.9	ASFO	39.1	DTTN	29.7	ACMN	5.5	STHN	4.7	NVCV	2.3	17	0	0	0	0
TC	5/28/09	1,786	823,456	48.4	DTTN	33.6	ASFO	28.7	ACMN	7.4	NZDS	4.1	NZFR	3.3	23	0	0	0	0
WE	5/28/09	1,727	706,266	47.3	DTTN	32.2	ASFO	29.8	ACMN	9.9	STHN	6.6	RDMN	3.3	19	0	0	0	0
TR	5/28/09	1,151	400,058	43.2	DTTN	74.6	ACMN	6.7	CMMN	3.7	GFAN	3.0	SNRM	1.5	16	0	0	0	0
LES	6/11/09	1,247	616,040	46.4	DTTN	56.1	MLGR	8.1	STHN	8.1	ACMN	5.7	RDMN	4.9	18	0	0	0	0
TG	6/11/09	1,210	585,055	46.0	DTTN	51.9	MLGR	12.3	RDMN	9.4	ACMN	4.7	NVCV	2.8	16	0	0	0	0
TC	6/11/09	1,466	757,805	47.8	DTTN	42.7	MLGR	10.3	ACMN	9.4	RHCU	6.8	COPC	6.8	18	0	0	0	0
WE	6/11/09	1,464	986,255	49.7	MLGR	20.5	DTTN	19.6	ACMN	15.2	COPC	8.0	RHCU	7.1	21	0	0	0	0
TR	6/11/09	1,077	327,813	41.8	DTTN	76.1	ACMN	5.1	GFAN	3.4	NZFR	2.6	EPSX	1.7	17	0	0	0	0
LES	6/25/09	1,377	1,077,256	50.4	DTTN	50.4	MLGR	8.0	SLMN	4.4	EPSX	4.4	FRCV	4.4	26	0	0	0	0
TG	6/25/09	1,735	1,071,125	50.3	DTTN	40.0	MLGR	8.0	GFSA	7.0	RHCU	5.0	EPSX	4.0	24	0	0	0	0
TC	6/25/09	902	478,098	44.5	DTTN	31.6	COPC	10.5	ACMN	7.4	MLGR	7.4	CMAF	6.3	17	0	0	0	0
WE	6/25/09	1,168	712,002	47.4	DTTN	18.8	COPC	13.9	MLGR	13.9	ACMN	7.9	EPSX	5.9	26	0	0	0	0
TR	6/25/09	789	445,609	44.0	DTTN	50.9	EPSX	14.3	GFAN	5.4	ACMN	5.4	GFSA	3.6	20	0	0	0	0
LES	7/9/09	236	240,012	39.6	EPSX	36.6	RDMN	30.5	CHX1	7.3	SNUL	4.9	AKFL	2.4	19	0	0	0	0
TG	7/9/09	910	549,522	45.5	CHX1	41.4	EPSX	26.7	RDMN	12.1	COPC	5.2	NVDC	1.7	18	0	0	0	0
TC	7/9/09	546	483,042	44.6	EPSX	38.1	COPC	12.7	APFA	7.6	RDMN	5.1	DTTN	5.1	23	583	1,388	0	0
WE	7/9/09	524	427,704	43.7	EPSX	44.6	COPC	10.9	CHX1	9.9	DTTN	4.0	DTVL	4.0	20	0	0	0	0
TR	7/9/09	183	165,674	36.9	COPC	25.3	EPSX	20.3	CMAF	16.5	SNUL	8.9	DTTN	8.9	16	0	0	46	0

Key to Species Codes is located in Combined Species List located in Appendix A

APF9 = *Aphanizomenon flos-aquae* MSAE = *Microcystis aeruginosa* ABX9 = *Anabaena* sp.

Table 4(contd) Phytoplankton results for baseline water samples collected with a churn splitter in the Klamath River and mouth of Trinity River May-Dec 2009.

Station	Date	Total	Total	TSI	Sp1	Sp1%	Sp2	Sp2%	Sp3	Sp3%	Sp4	Sp4%	Sp5	Sp5%	#Spp	APF9 cells/ml	MSAE cells/ml	ABX9 cells/ml	Oscillatoria sp. Cells/ml
		Density	Biolume																
LES	7/23/09	162	159,810	36.7	EPSX	55.9	COPC	23.5	SNUL	7.4	CMSN	5.9	RHCU	2.9	8	0	0	0	0
TG	7/23/09	368	335,316	42.0	EPSX	50.0	COPC	24.0	CMSN	3.1	SNUL	3.1	AKFL	2.1	17	0	0	38	0
TC	7/23/09	416	293,171	41.0	COPC	23.7	EPSX	17.8	SCQD	11.9	SNUL	8.5	DTTN	6.8	29	0	0	0	0
WE	7/23/09	706	763,399	47.9	EPSX	38.0	COPC	21.3	SNUL	8.3	NZFR	5.6	SCQD	3.7	23	0	0	0	0
TR	7/23/09	210	122,448	34.7	COPC	50.0	CMAF	10.5	EPSX	9.3	SCQD	3.5	GFAN	3.5	19	0	0	0	0
KATDCP	7/29/09	1,095	419,461	43.6	EPSX	21.9	SLMN	19.1	COPC	7.6	CHX1	7.6	AKFL	7.6	23	0	407	0	0
TCDCP	7/29/09	898	687,358	47.1	EPSX	25.2	COPC	24.3	MSAE	7.3	SCQD	7.2	NZFR	5.4	23	1,178	0	0	0
WEDCP	7/29/09	1,413	861,016	48.8	EPSX	27.9	COPC	12.5	MSAE	6.7	NZFR	5.8	SCQD	5.8	25	2,472	0	0	0
LES	8/6/09	106	86,815	32.3	EPSX	32.9	COPC	20.0	SCQD	15.7	MSAE	7.1	SNUL	5.7	15	0	1,150	30	0
TG	8/6/09	851	982,191	49.7	EPSX	37.6	COPC	17.9	SNUL	10.3	SCQD	7.7	RHCU	5.1	21	0	0	0	0
TC	8/6/09	911	668,093	46.9	COPC	28.0	EPSX	25.2	NZFR	7.5	DTTN	6.5	SCQD	4.7	25	0	0	0	0
WE	8/6/09	618	549,559	45.5	COPC	39.0	EPSX	27.0	SCQD	8.0	RHCU	5.0	SNUL	4.0	18	19	0	0	0
TR	8/6/09	195	173,002	37.2	SNUL	26.6	COPC	13.8	GFAN	10.6	APFA	9.6	EPSX	4.3	23	429	0	0	0
KATDCP	8/11/09	912	232,154	39.3	MSAE	53.7	COPC	12.2	RDMN	5.7	SCQD	4.9	EPSX	3.3	19	0	4,896	0	0
TCDCP	8/12/09	1,411	465,065	44.3	MSAE	59.2	EPSX	13.0	COPC	7.1	AKFL	4.1	SCQD	4.1	16	167	8,352	0	0
WEDCP	8/12/09	2,208	761,709	47.9	MSAE	41.5	COPC	13.8	RDMN	10.6	EPSX	5.7	NZPL	4.9	20	359	9,156	0	0
LES	8/20/09	195	191,465	38.0	EPSX	30.0	COPC	22.0	SNUL	9.0	CMAF	8.0	MSAE	6.0	18	0	1,125	0	0
TG	8/20/09	474	394,951	43.2	EPSX	31.3	COPC	15.7	SCQD	9.6	SNUL	8.7	FRCN	7.0	22	0	0	41	0
TC	8/20/09	511	451,324	44.1	EPSX	42.3	COPC	20.7	SCQD	6.3	MLGR	5.4	CHX1	3.6	19	0	0	0	0
WE	8/20/09	861	472,617	44.4	MSAE	22.8	EPSX	18.1	RDMN	9.4	COPC	9.4	SNUL	5.5	25	0	1,967	0	0
TR	8/20/09	83	58,822	29.5	COPC	22.8	EPSX	19.3	SNUL	10.5	AKFL	5.3	NZPC	5.3	21	0	0	0	0
KATDCP	8/25/09	1,278	734,314	47.6	EPSX	25.2	MSAE	20.2	COPC	10.9	SNUL	5.0	SCQD	4.2	25	0	2,577	0	0
TCDCP	8/26/09	586	604,649	46.2	EPSX	27.5	COPC	16.5	SNUL	7.7	NZFR	6.6	NVCV	3.3	32	0	0	77	51
WEDCP	8/26/09	791	917,608	49.2	EPSX	31.6	MSAE	19.1	COPC	10.3	NZPL	4.4	SNUL	4.4	23	0	1,513	47	0

Key to Species Codes is located in Combined Species List located in Appendix A

APF9 = *Aphanizomenon flos-aquae* MSAE = *Microcystis aeruginosa* ABX9= *Anabaena sp.*

Table 4(contd) Phytoplankton results for baseline water samples collected with a churn splitter in the Klamath River and mouth of Trinity River May-Dec 2009.

Station	Date	Total		TSI	Total										#Spp	APF9 cells/ml	MSAE cells/ml	ABX9 cells/ml	Oscillatoria sp. Cells/ml
		Density	Biovolume		Sp1	Sp1%	Sp2	Sp2%	Sp3	Sp3%	Sp4	Sp4%	Sp5	Sp5%					
LES	9/3/09	145	111,959	34.1	DTTN	26.4	EPSX	18.1	SNUL	12.5	COPC	6.9	NVCR	4.2	19	40	242	0	0
TG	9/3/09	514	509,804	45.0	EPSX	36.6	SNUL	15.9	DTTN	7.3	COPC	6.1	FRCN	4.9	19	125	0	0	0
TC	9/3/09	634	429,483	43.8	MSAE	18.9	EPSX	18.0	NZFR	13.5	COPC	8.1	DTVL	6.3	25	114	1,199	0	0
WE	9/3/09	782	526,182	45.2	EPSX	25.4	MSAE	13.8	COPC	12.3	NZFR	8.5	NZPL	6.2	27	216	1,082	0	0
TR	9/3/09	126	124,762	34.9	DTTN	33.3	EPSX	15.1	COPC	11.8	SNUL	10.8	NVCR	3.2	26	0	0	0	0
KATDCP	9/8/09	1,134	638,195	46.6	DTTN	31.5	EPSX	16.3	RDMN	13.4	SNUL	4.8	DTTE	3.8	23	169	217	0	0
TCDCP	9/9/09	990	1,118,785	50.7	EPSX	18.3	MSAE	18.3	NZFR	10.6	COPC	8.6	DTTN	4.8	24	468	1,808	0	0
WEDCP	9/9/09	2,205	1,241,933	51.4	MSAE	35.9	EPSX	20.9	NZFR	10.0	SCQD	4.2	COPC	3.3	25	1,082	7,916	0	0
LES	9/17/09	560	404,745	43.3	EUPC	16.8	EPSX	15.9	DTTN	15.9	MSAE	15.0	SNUL	7.1	22	446	6,656	0	0
TG	9/17/09	1,089	854,890	48.7	MSAE	41.0	EPSX	19.4	SNUL	7.5	DTTN	4.5	DTVL	4.5	25	81	4,469	81	0
TC	9/17/09	532	547,797	45.5	EPSX	32.6	COPC	13.0	NZFR	12.0	SNUL	6.5	DTTN	4.3	24	0	0	0	0
WE	9/17/09	928	501,438	44.9	MSAE	40.7	EPSX	24.1	SNUL	5.5	COPC	4.8	NZFR	4.1	23	128	3,774	0	0
TR	9/17/09	67	59,020	29.5	EPSX	30.4	SNUL	17.4	DTTN	6.5	ABXX	4.3	SLMN	4.3	21	0	0	44	0
TCDCP	9/23/09	3,007	667,555	46.9	MSAE	65.4	NZFR	12.0	EPSX	3.4	APFA	2.4	NZPL	1.9	25	1,084	23,591	0	0
WEDCP	9/23/09	3,975	1,334,177	51.9	MSAE	72.0	NZFR	5.4	EPSX	4.7	GMAF	3.9	COPC	2.8	26	463	31,488	0	0
LES	10/1/09	646	176,567	37.4	MSAE	68.2	EPSX	11.6	COPC	4.7	AKFL	3.9	RDMN	3.9	13	0	4,410	0	0
TG	10/1/09	1,337	813,343	48.4	MSAE	62.7	EPSX	12.9	SNUL	3.5	NVCR	3.0	COPC	2.5	23	0	8,381	0	0
TC	10/1/09	1,706	404,358	43.3	MSAE	65.9	NZFR	7.2	COPC	6.7	EPSX	3.4	SNUL	2.4	21	82	11,234	0	0
WE	10/1/09	2,781	563,380	45.7	MSAE	74.8	NZFR	7.2	COPC	3.6	EPSX	2.7	NVCR	1.8	17	206	24,955	0	0
TR	10/1/09	145	128,031	35.1	DTTN	15.9	EPSX	11.6	ACMN	11.6	SNUL	8.7	NZPC	7.2	23	0	0	0	0
KATDCP	10/6/09	2,796	601,558	46.2	MSAE	74.6	EPSX	4.8	NZPL	4.0	COPC	3.6	NZFR	1.6	22	0	20,859	0	0
TCDCP	10/7/09	2,234	681,024	47.1	MSAE	54.0	COPC	11.2	NZPL	7.5	NZFR	5.6	EPSX	5.6	21	0	14,488	0	0
WEDCP	10/7/09	2,656	624,698	46.5	MSAE	63.7	NZFR	9.9	NZPL	6.6	COPC	5.2	EPSX	3.3	18	0	16,913	0	0
LES	10/8/09	2,289	333,718	41.9	MSAE	84.4	NZPL	7.2	EPSX	1.6	COPC	1.0	NVCR	0.7	17	298	23,169	0	0
LES	10/15/09	1,054	899,018	49.1	EPSX	32.0	COPC	21.0	NZFR	9.0	NVCR	6.0	DTVL	5.0	23	316	3,477	0	0
TG	10/15/09	4,067	4,210,729	60.2	EPSX	30.7	COPC	15.8	SNUL	7.9	NZFR	7.9	NVCR	6.9	22	0	0	0	0
TC	10/15/09	5,906	7,243,382	64.1	EPSX	23.6	COPC	20.0	NZFR	10.9	NVCR	5.5	NVCR	4.5	26	0	0	0	0
WE	10/15/09	8,936	7,923,093	64.8	COPC	20.6	NZFR	19.6	EPSX	15.9	ACMN	5.6	SNUL	4.7	24	0	0	0	0
TR	10/15/09	3,876	10,499,643	66.8	EPSX	31.8	COPC	20.9	RPGB	7.3	DTTN	3.6	SNUL	2.7	29	0	0	0	0

Key to Species Codes is located in Combined Species List located in Appendix A

APF9 = *Aphanizomenon flos-aquae* MSAE = *Microcystis aeruginosa* ABX9 = *Anabaena* sp.

Table 4(contd) Phytoplankton results for baseline water samples collected with a churn splitter in the Klamath River and mouth of Trinity River May-Dec 2009.

Station	Date	Total	Total	TSI	Sp1	Sp1%	Sp2	Sp2%	Sp3	Sp3%	Sp4	Sp4%	Sp5	Sp5%	#Spp	APF9	MSAE	ABX9	Oscillatoria
		Density	Bioldume													cells/ml	cells/ml	cells/ml	sp. Cells/ml
LES	10/15/09	1,054	899,018	49.1	EPSX	32.0	COPC	21.0	NZFR	9.0	NVCR	6.0	DTVL	5.0	23	316	3,477	0	0
TG	10/15/09	4,067	4,210,729	60.2	EPSX	30.7	COPC	15.8	SNUL	7.9	NZFR	7.9	NVCR	6.9	22	0	0	0	0
TC	10/15/09	5,906	7,243,382	64.1	EPSX	23.6	COPC	20.0	NZFR	10.9	NVCV	5.5	NVCR	4.5	26	0	0	0	0
WE	10/15/09	8,936	7,923,093	64.8	COPC	20.6	NZFR	19.6	EPSX	15.9	ACMN	5.6	SNUL	4.7	24	0	0	0	0
TR	10/15/09	3,876	10,499,643	66.8	EPSX	31.8	COPC	20.9	RPGB	7.3	DTTN	3.6	SNUL	2.7	29	0	0	0	0
KATDCP	10/20/09	675	424,908	43.7	EPSX	13.2	COPC	13.2	NZFR	8.8	MSAE	8.8	RDMN	7.7	28	0	593	0	0
TCDCP	10/21/09	863	472,115	44.4	COPC	25.2	NZFR	15.3	MSAE	7.2	EPSX	5.4	RHCU	5.4	25	397	622	0	0
WEDCP	10/21/09	1,435	696,449	47.2	NZFR	19.6	COPC	17.9	MSAE	11.6	EPSX	6.3	NVCR	6.3	27	0	2,165	0	0
LES	10/29/09	122	40,162	26.8	RDMN	23.3	DTTN	11.7	COPC	11.7	NZFR	8.3	NVCR	6.7	20	0	41	0	0
TG	10/29/09	632	364,235	42.6	DTTN	25.8	NZFR	16.5	COPC	11.3	EPSX	9.3	NVCR	6.2	24	0	450	0	0
TC	10/29/09	829	461,961	44.3	NZFR	25.2	COPC	20.7	DTTN	10.8	DTVL	5.4	NVCV	4.5	26	0	597	0	0
WE	10/29/09	963	427,902	43.7	NZFR	27.6	COPC	19.0	NVCR	6.7	NZPL	6.7	NVCV	4.8	26	0	183	0	0
TR	10/29/09	166	74,957	31.2	DTTN	59.3	SNUL	5.5	NVCR	5.5	ACMN	3.3	NVCV	2.2	23	0	0	0	0
LES	11/12/09	227	136,126	35.5	COPC	34.7	NZFR	13.3	EPSX	6.7	DTTN	6.7	DTVL	4.0	22	0	0	0	0
TG	11/12/09	787	454,784	44.2	NZFR	22.3	COPC	11.7	DTVL	10.7	DTTN	9.7	RHCU	6.8	29	0	0	0	0
TC	11/12/09	921	500,601	44.9	NZFR	34.0	COPC	15.5	DTVL	7.2	DTTN	5.2	EPSX	4.1	24	0	0	0	0
WE	11/12/09	1,320	546,181	45.5	NZFR	41.6	COPC	11.9	NVCV	10.9	NVTP	3.0	DTVL	3.0	22	0	0	0	0
TR	11/12/09	346	115,630	34.3	DTTN	55.8	CMMC	4.8	NZFR	3.8	AFPR	3.8	NZDS	3.8	22	0	0	0	0
LES	12/17/09	4,628	3,133,783	58.1	NZFR	21.2	DTTN	16.9	EPSX	15.3	COPC	13.6	SNUL	4.2	23	0	0	0	0
TG	12/17/09	4,857	5,324,762	61.9	DTTN	20.5	EPSX	14.3	COPC	12.5	NZFR	12.5	SNUL	6.3	24	0	0	0	0
TC	12/17/09	4,645	3,224,921	58.3	NZFR	22.3	DTTN	20.4	EPSX	10.7	COPC	8.7	DTVL	3.9	25	0	0	0	0
WE	12/17/09	2,091	1,142,916	50.8	NZFR	23.5	COPC	11.8	DTTN	11.8	NVCV	5.9	EPSX	5.9	27	0	0	0	0
TR	12/17/09	3,979	2,122,618	55.3	DTTN	40.0	EPSX	15.0	NZFR	8.3	NVCV	5.8	COPC	5.8	21	0	0	0	0

Key to Species Codes is located in Combined Species List located in Appendix A

APF9 = *Aphanizomenon flos-aquae* MSAE = *Microcystis aeruginosa* ABX9 = *Anabaena* sp.

Table 5 Phytoplankton results for public health samples collected with a wide mouth jar in the Klamath River at Weitchpec and Turwar Gage May-Oct 2009.

Station	Date	Total Density	Total Biovolume	TSI	Sp1	Sp1%	Sp2	Sp2%	Sp3	Sp3%	Sp4	Sp4%	Sp5	Sp5%	#Spp	APF9 cells/ml	MSAE cells/ml	ABX9 cells/ml	Oscillatoria sp. Cells/ml
TG	6/11/09	1,641	704,155	47.3	DTTN	47.0	STHN	10.3	MLGR	7.7	ACMN	6.0	NZDS	4.3	19	0	0	0	0
WE	6/11/09	1,282	552,131	45.6	ACMN	19.1	COPC	16.4	DTTN	16.4	RHCU	5.5	GFAN	3.6	27	0	0	0	0
TG	6/25/09	1,295	804,499	48.3	DTTN	42.6	EPSX	4.9	GFVT	4.9	MLGR	4.1	ACMN	4.1	29	0	0	0	0
WE	6/25/09	587	226,147	39.1	ACMN	17.2	COPC	12.1	DTTN	10.1	RHCU	6.1	EPSX	5.1	30	0	0	0	0
TG	7/9/09	1,769	850,291	48.7	RDMN	25.5	DTTN	17.6	EPSX	16.7	COPC	4.9	CHX1	4.9	23	0	0	0	0
WE	7/9/09	14,459	14,833,655	69.3	EPSX	53.2	NZFR	18.3	COPC	5.5	DTVL	3.7	MLVR	1.8	17	0	0	0	0
TG	7/23/09	927	612,167	46.3	EPSX	25.2	NZFR	13.6	COPC	10.7	DTTN	5.8	RHCU	4.9	26	0	0	0	0
WE	7/23/09	1,903	1,659,838	53.5	EPSX	48.9	NZFR	14.6	DTTN	6.2	COPC	5.2	GFAN	3.1	20	0	316	0	0
TG	8/6/09	1,517	910,867	49.2	EPSX	16.5	RHCU	12.8	SCQD	9.2	NZFR	9.2	COPC	8.3	29	0	14,929	0	0
WE	8/6/09	3,595	3,835,843	59.5	MSAE	31.7	COPC	24.4	EPSX	20.9	SCQD	4.9	SNUL	3.5	19	0	151,624	0	0
TG	8/11/09	2,578	1,406,219	52.3	COPC	23.4	EPSX	13.1	NZFR	10.3	SNUL	6.5	AKFL	5.6	28	0	0	0	0
WE	8/12/09	2,472	1,366,124	52.1	MSAE	34.9	COPC	13.8	EPSX	13.8	SNUL	8.7	SCQD	4.4	22	251	8,618	0	0
TG	8/20/09	1,627	707,893	47.4	RDMN	18.8	EPSX	10.9	STHN	10.9	NZPL	9.9	COPC	8.9	24	0	0	0	0
WE	8/20/09	1,888	1,039,922	50.1	COPC	18.4	NZPL	16.5	EPSX	10.7	CCMG	9.7	NZPC	6.8	24	0	0	0	0
TG	8/25/09	2,148	1,731,430	53.8	EPSX	17.0	NZFR	10.0	RHCU	6.0	RDMN	6.0	CMAF	5.0	29	0	0	0	0
WE	8/26/09	810	730,391	47.6	NZPL	20.7	EPSX	14.8	ABXX	9.4	NZPC	8.9	SNUL	7.9	22	0	0	1,071	0
TG	9/3/09	1,353	795,013	48.2	EPSX	22.2	NZFR	12.7	DTTN	11.6	COPC	9.5	SNUL	6.3	30	171	194	0	0
WE	9/3/09	733	320,763	41.7	NZCP	31.4	NZPL	16.9	NZFR	7.6	NZPC	5.9	CCMG	5.1	21	466	0	0	0
TG	9/8/09	2,264	2,294,301	55.8	DTTN	28.5	EPSX	14.2	COPC	7.6	DTTE	4.7	NZFR	3.8	30	215	43	209	0
WE	9/9/09	2,585	2,593,626	56.7	EPSX	27.5	NZPL	17.0	COPC	8.9	SNUL	5.7	NZCP	4.8	27	223	167	209	0
TG	9/17/09	1,565	1,726,514	53.8	EPSX	16.8	DTTN	13.7	MSAE	7.7	COPC	5.3	SNUL	5.3	34	132	2,764	0	0
WE	9/17/09	507	282,693	40.7	EPSX	21.5	NZFR	21.5	NZPL	19.4	NZAC	6.5	NZPC	4.3	20	238	0	387	0
TG	9/23/09	3,103	1,632,598	53.4	MSAE	37.4	EPSX	10.1	DTTN	8.6	SNUL	7.2	COPC	6.5	30	223	11,610	0	0
WE	9/23/09	2,622	633,703	46.6	MSAE	61.7	NZFR	9.3	EPSX	4.1	SCQD	3.6	NVCR	2.6	24	326	16,165	272	0
TG	10/1/09	1,327	477,714	44.5	MSAE	34.2	RDMN	10.8	COPC	8.1	EPSX	7.2	DTTN	5.4	25	0	7,708	157	0
WE	10/1/09	493	723,280	47.5	EPSX	21.0	NZFR	18.1	SCQD	10.5	NZPL	7.6	ABXX	6.7	26	0	0	428	0
TG	10/15/09	3,929	6,425,793	63.3	EPSX	26.1	NZFR	20.0	DTVL	7.0	SNUL	5.2	COPC	4.3	27	0	0	0	0
WE	10/15/09	27,361	20,238,174	71.5	EPSX	35.7	COPC	27.5	RHCU	9.3	NZFR	7.1	NVCR	4.4	24	0	0	0	0

Key to Species Codes is located in Combined Species List located in Appendix A

APF9 = *Aphanizomenon flos-aquae* MSAE = *Microcystis aeruginosa* ABX9= *Anabaena* sp.

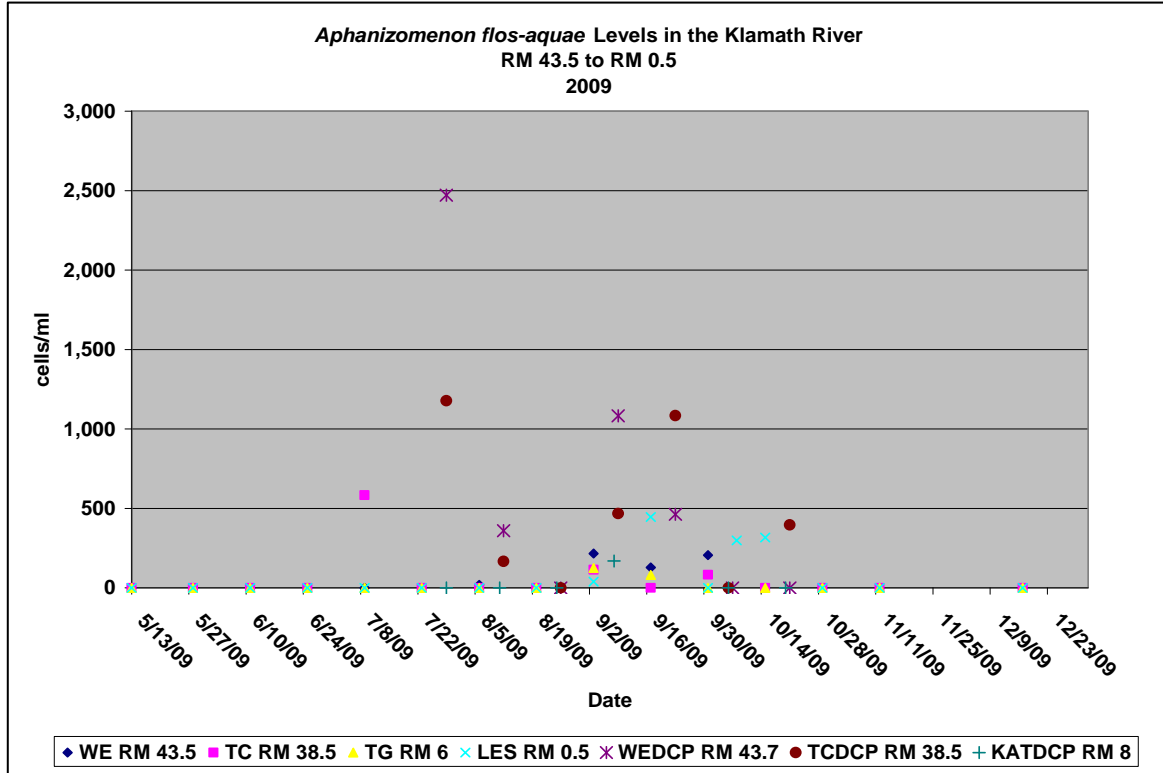


Figure 2. *Aphanizomenon flos-aquae* levels for baseline water samples collected with a churn splitter in the Klamath River from RM 43.5 to RM 0.5, May through December 2009.

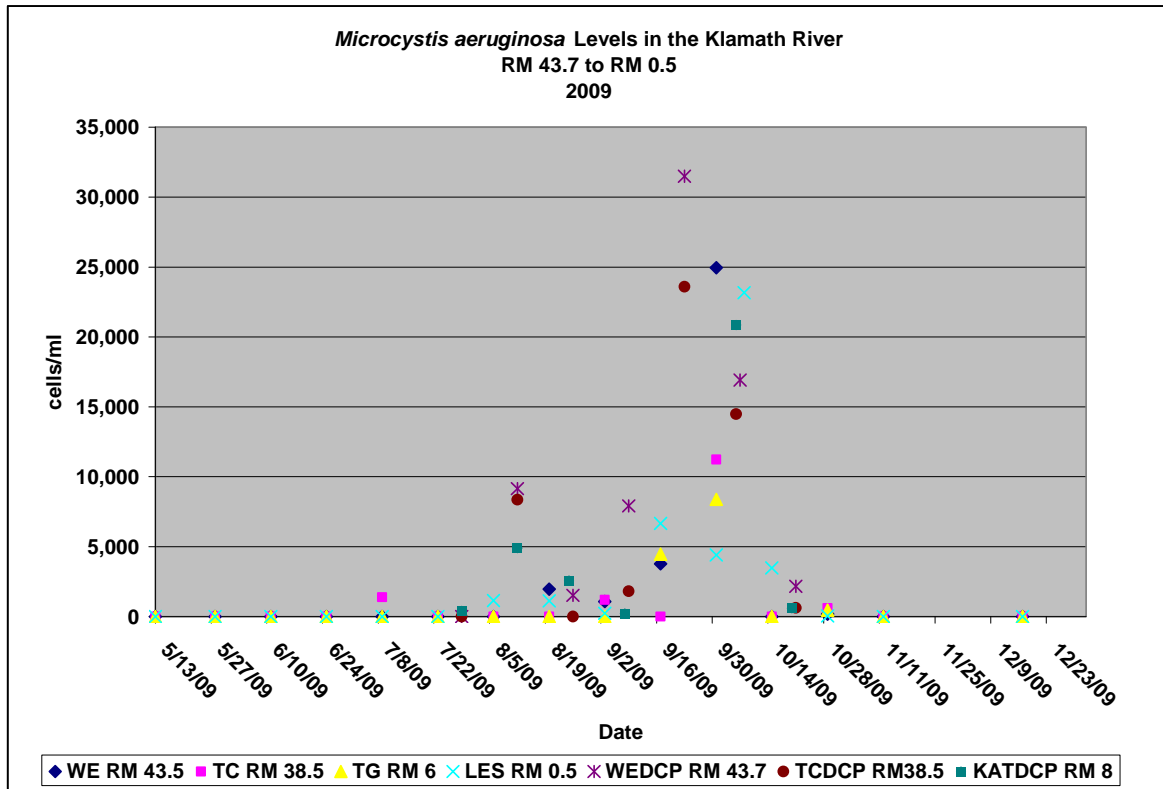


Figure 3. *Microcystis aeruginosa* levels for baseline water samples collected with a churn splitter in the Klamath River from RM 43.5 to RM 0.5, May through December 2009.

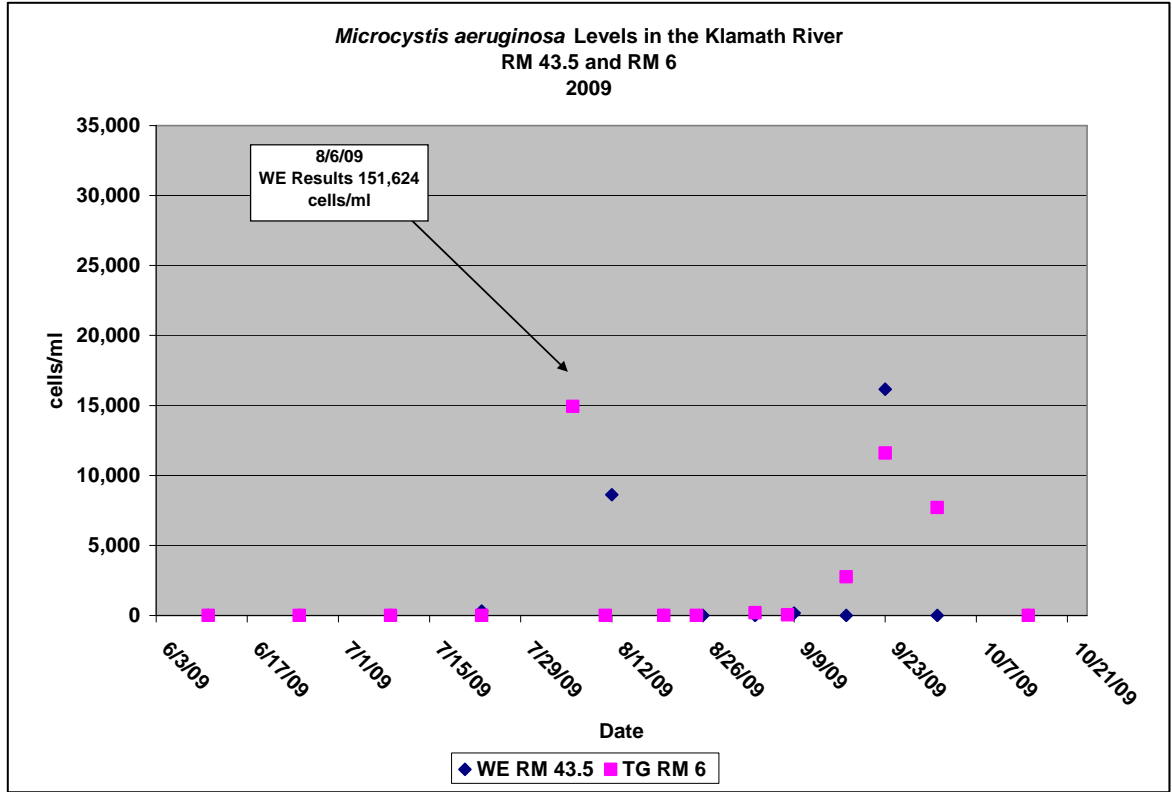


Figure 4. *Microcystis aeruginosa* levels for public health water samples collected with a wide mouth jar in the Klamath River at RM 43.5 and RM 6, May through October 2009.

Table 6. Phytoplankton results for water samples collected with a churn splitter during freshwater mussel sample collection in the Klamath River Above Starwein Riffle and Trinity River near Mouth July-Oct 2009.

Site	Date	Total		TSI	Species										#Sp	APF9 cells/ml	MSAE cells/ml	ABX9 cells/ml	Oscillatoria sp. Cells/ml
		Density	Biovolume		Sp1	Sp1%	Sp2	Sp2%	Sp3	Sp3%	Sp4	Sp4%	Sp5	Sp5%					
KASR	7/20/09	687	721,481	47.5	EPSX	60.9	COFC	9.6	NVCR	4.3	SNLL	4.3	SOCD	3.5	20	179	0	0	0
KASR	8/18/09	702	315,891	41.5	MSAE	43.7	COFC	13.4	EPSX	10.7	CHX1	6.2	SOCD	6.2	21	0	4,911	63	0
KASR	9/15/09	1,146	635,247	46.6	MSAE	39.4	EPSX	26.0	RDMN	3.9	SNLL	3.9	COFC	3.1	22	0	4,510	0	0
KASR	10/12/09	1,265	679,876	47.1	MSAE	41.9	NZPL	9.6	DTTN	8.8	EPSX	8.1	NZFR	8.1	21	0	5,300	0	0
TR	9/14/09	61	144,346	35.9	SNLL	33.3	EPSX	12.8	COFC	7.7	AKFL	7.7	DTTN	5.1	16	0	0	16	0

Key to Species Codes is located in Combined Species List located in Appendix A
 APF9 = *Aphanizomenon flos-aquae* MSAE = *Microcystis aeruginosa* ABX9 = *Anabaena sp.*

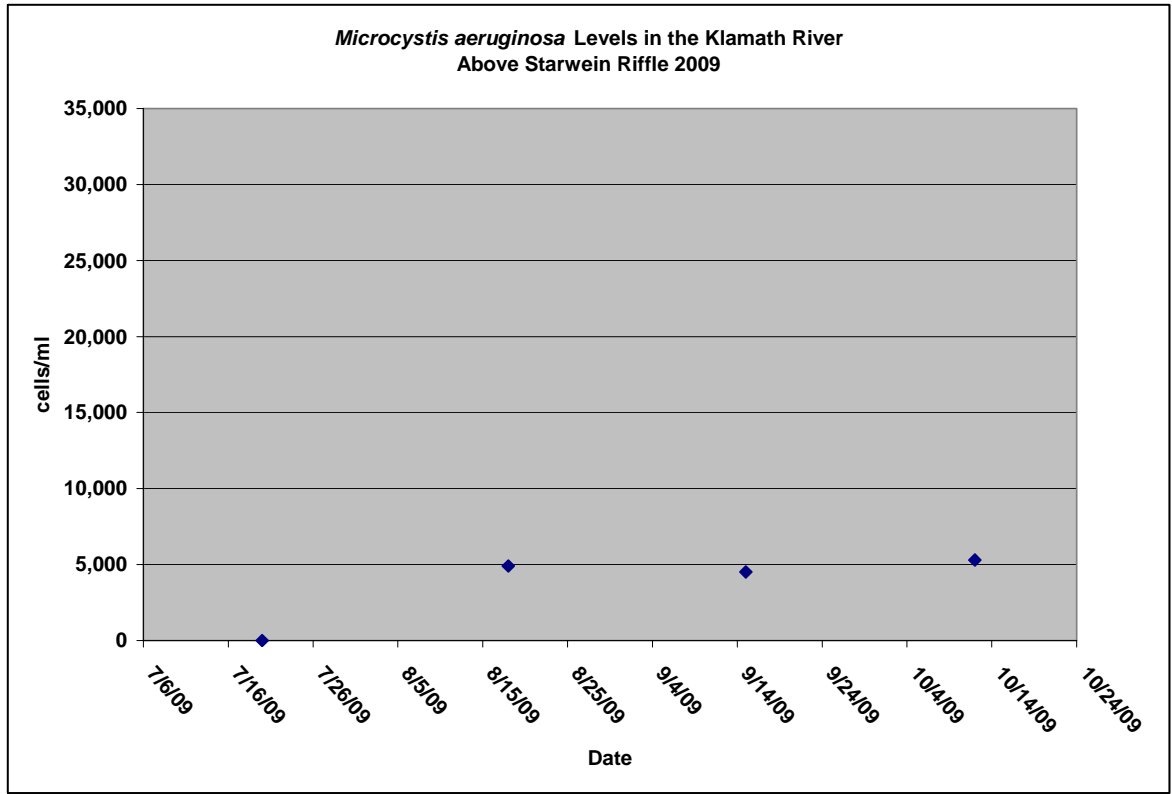


Figure 5. *Microcystis aeruginosa* levels for water samples collected with a churn splitter during freshwater mussel sample collection in the Klamath River Above Starwein Riffle July-Oct 2009.

Cyanotoxins

Table 7. Microcystin results for baseline water samples collected with a churn splitter in the Klamath River and mouth of Trinity River May-Dec 2009

Total Microcystin	Date											
	Site	5/28/09	6/11/09	6/25/09	7/9/09	7/10/09	7/23/09	8/6/09	8/20/09	8/25/09	8/26/09	9/3/09
USEPA Region 9 Lab reporting limit: 0.18 µg/L units: µg/L	WE	DNS	<0.18	DNS	<0.18	<0.18	DNS	1.2	0.89	DNS	DNS	0.75
	WEDCP	DNS	DNS	DNS	DNS	DNS	DNS	DNS	DNS	DNS	0.92	DNS
	TC	<0.18	<0.18	<0.18	<0.18	DNS	<0.18	0.98	0.43	DNS	DNS	0.51
	TCDCP	DNS	DNS	DNS	DNS	DNS	DNS	DNS	DNS	DNS	0.45	DNS
	KATDCP	DNS	DNS	DNS	DNS	DNS	DNS	DNS	DNS	DNS	0.64	DNS
	TG	<0.18	<0.18	<0.18	<0.18	DNS	<0.18	0.64	0.49	DNS	DNS	0.39
	LES	<0.18	<0.18	<0.18	<0.18	DNS	0.19	<0.18	0.34	DNS	DNS	0.39
	TR	DNS	<0.18	DNS	<0.18	DNS	DNS	<0.18	DNS	DNS	DNS	<0.18

DNS – Did Not Sample

Table 7(contd). Microcystin results for baseline water samples collected with a churn splitter in the Klamath River and mouth of Trinity River May-Dec 2009

Total Microcystin	Date										
	Site	9/8/09	9/9/09	9/14/09	9/17/09	10/1/09	10/8/09	10/15/09	10/29/09	11/12/09	12/17/09
USEPA Region 9 Lab reporting limit: 0.18 µg/L units: µg/L	WE	DNS	DNS	DNS	1.1	2.4	DNS	0.92	DNS	DNS	<0.18
	WEDCP	DNS	2.3	DNS	DNS	DNS	DNS	DNS	DNS	DNS	DNS
	TC	DNS	DNS	DNS	0.64	2.3	DNS	0.67	0.26	<0.18	<0.18
	TCDCP	DNS	1.4	DNS	DNS	DNS	DNS	DNS	DNS	DNS	DNS
	KATDCP	0.58	DNS	DNS	DNS	DNS	DNS	DNS	DNS	DNS	DNS
	TG	DNS	DNS	DNS	1.7	1.8	DNS	1.2	0.27	<0.18	<0.18
	LES	DNS	DNS	DNS	1.4	2.3	3.0	2.3	0.26	<0.18	0.23
	TR	DNS	DNS	<0.18	DNS	<0.18	DNS	DNS	DNS	DNS	DNS

DNS – Did Not Sample

Table 8. Total microcystin results for public health samples collected with a wide mouth jar in the Klamath River at Weitchpec and at Turwar Gage June-Oct 2009

Total Microcystin	Date																	
	Site	6/11/09	6/25/09	7/9/09	7/23/09	8/6/09	8/11/09	8/12/09	8/20/09	8/25/09	8/26/09	9/3/09	9/8/09	9/9/09	9/17/09	9/23/09	10/1/09	10/15/09
USEPA Region 9 Lab reporting limit: 0.18 µg/L units: µg/L	WE-SG	<0.18	<0.18	<0.18	0.30	1.7	DNS	0.72	0.22	DNS	<0.18	<0.18	DNS	<0.18	<0.18	4.4	<0.18	1.1
	TG-SG	<0.18	<0.18	<0.18	0.26	0.45	1.0	DNS	0.29	0.65	DNS	0.54	0.52	DNS	1.7	2.6	1.9	1.2

DNS – Did Not Sample

Table 9. Microcystin variants results for baseline water samples collected with a churn splitter in the Klamath River May-Nov 2009.

Microcystin Variants	Sample Date	5/28/09	6/25/09	7/10/09	7/23/09	8/6/09	8/20/09	9/3/09	9/17/09	10/1/09	10/15/09	10/29/09	10/29/09	11/12/09	11/12/09
	Sample Site	TC	TC	WE	TC	WE	TC	TC	TC	WE	TC	TC	WE	TC	WE
CA Department of Fish and Game Water Pollution Control Laboratory reporting limit 1.0 µg/L units: µg/L	MC-RR	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-Demethyl-RR*	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-LR	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-Demethyl-LR*	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-YR	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-LA	<1.0	<1.0	<1.0	<1.0	2.29	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-LW	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-LF	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0
MC-LY	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	

MC = Microcystin *Demethyl analog quantified as parent compound

Table 10. Anatoxin-a results for baseline water samples collected with a churn splitter in the Klamath River at Weitchpec and Above Tully Creek May-Nov 2009

Anatoxin-a															
CA Department of Fish and Game Water Pollution Control Laboratory reporting limit: 5.0 µg/L units: ppb (µg/L)	Sample Date	5/28/09	6/25/09	7/10/09	7/23/09	8/6/09	8/20/09	9/3/09	9/17/09	10/1/09	10/15/09	10/29/09	10/29/09	11/12/09	11/12/09
	Sample Site	TC	TC	WE	TC	WE	TC	TC	TC	WE	TC	TC	WE	TC	WE
	Anatoxin-a	<5.0	<5.0	<5.0	<5.0	<5.0	<5.0	<5.0	<5.0	<5.0	<5.0	<5.0	<5.0	<5.0	<5.0

Table 11. Microcystin results for water samples collected with a churn splitter during freshwater mussel sampling events in Klamath River Above Starwein Riffle and mouth of Trinity River July – October 2009.

Total Microcystin	Date					
USEPA Region 9 Lab reporting limit: 0.18 µg/L units: µg/L	Site	7/20/09	8/20/09	9/14/09	9/15/09	10/12/2009
	KASR -OC	0.22	0.69	DNS	1.4	1.7
	TR - OC	DNS	DNS	<0.18	DNS	DNS

DNS – Did Not Sample

Table 12. Microcystin variants results for water samples collected with a churn splitter during freshwater mussel sampling events in Klamath River Above Starwein Riffle and mouth of Trinity River July – October 2009.

Microcystin Variants	Sample Date	7/20/09	8/18/09	9/14/09	9/15/09	10/12/09
	Sample Site	KASR	KASR	TR	KASR	KASR
CA Department of Fish and Game Water Pollution Control Laboratory reporting limit 1.0 µg/L units: µg/L	MC-RR	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-Demethyl-RR*	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-LR	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-Demethyl-LR*	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-YR	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-LA	<1.0	1.05	<1.0	1.5	<1.0
	MC-LW	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-LF	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-LY	<1.0	<1.0	<1.0	<1.0	<1.0

MC = Microcystin *Demethyl analog quantified as parent compound

Table 13. Anatoxin-a results for water samples collected with a churn splitter during freshwater mussel sampling events in Klamath River Above Starwein Riffle and mouth of Trinity River July – October 2009.

Anatoxin-a		7/20/09	8/18/09	9/14/09	9/15/09	10/12/09
CA Department of Fish and Game Water Pollution Control Laboratory reporting limit: 5.0 µg/L units: µg/L	Sample Date	7/20/09	8/18/09	9/14/09	9/15/09	10/12/09
	Sample Site	KASR	KASR	TR	KASR	KASR
	Anatoxin-a	<5.0	<5.0	<5.0	<5.0	<5.0

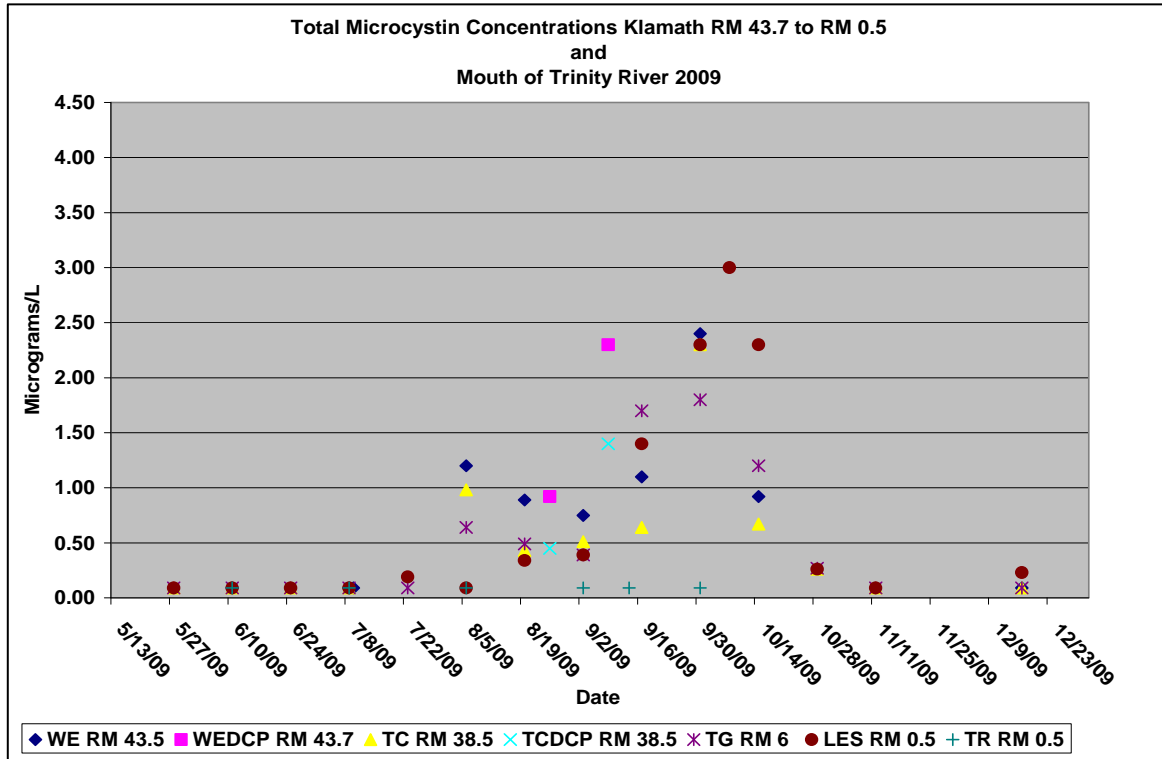


Figure 6. Microcystin results for baseline water samples collected with a churn splitter in the Klamath River and mouth of Trinity River May-Dec 2009. *sample results reported below the reporting level (0.18) were graphed at ½ the reporting limit

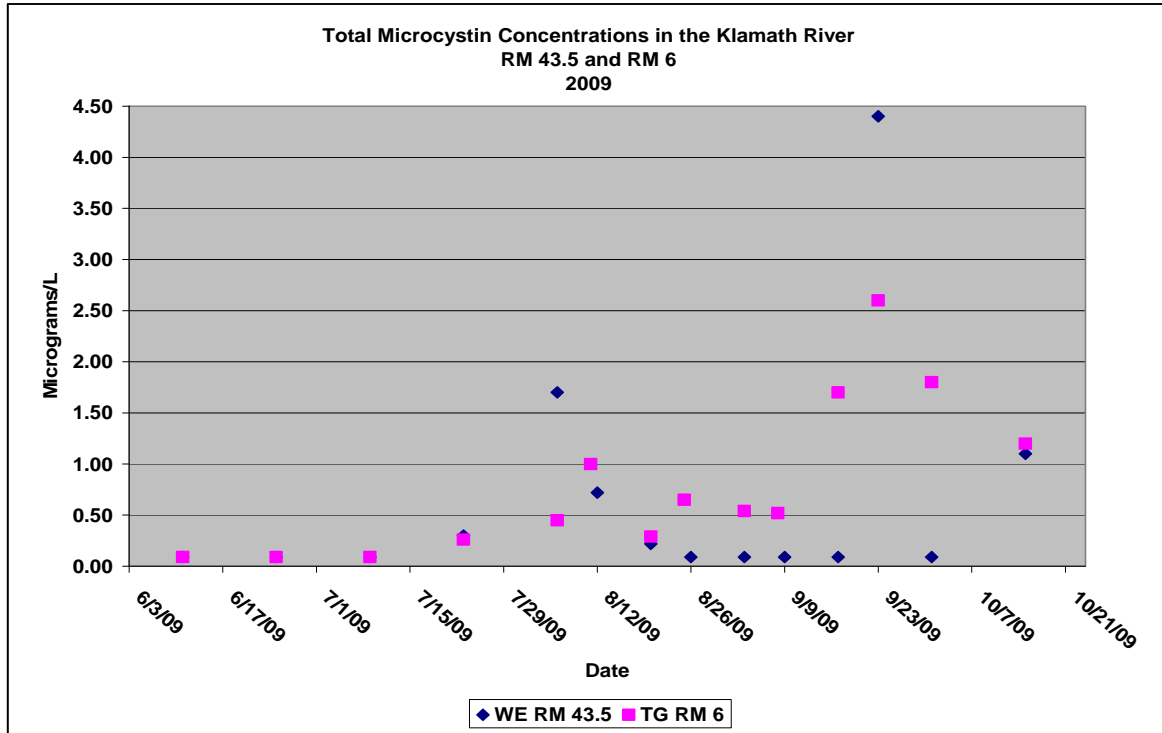


Figure 7. Microcystin results for public health samples collected with a wide mouth jar in the Klamath River at Weitchpec and at Turwar Gage June-Oct 2009. *sample results reported below the reporting level (0.18) were graphed at ½ the reporting limit.

Freshwater Mussel Tissue Cyanotoxins

Table 14. Microcystin variants results from freshwater mussel tissue sampling events in Klamath River Above Starwein Riffle and mouth of Trinity River July – October 2009.

	Sample Date	7/20/09	8/18/09	8/18/09	8/18/09	8/18/09	8/18/09	9/14/09	9/15/09	9/15/09	9/15/09	9/15/09	9/15/09
Microcystin Variants	Sample Site	KASR	KASR	KASR	KASR	KASR	KASR	TR	KASR	KASR	KASR	KASR	KASR
	Sample Number	1,2,3,4,5	1	2	3	4	5	1,2,3,4,5	1	2	3	4	5
CA Department of Fish and Game Water Pollution Control Laboratory reporting limit 1.00 ng/g MC-LY reporting limit 5.0 ng/g units: ppb (ng/g)	MC-RR	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	1.38	1.57	1.81	<1.0	1.5
	MC-Demethyl-RR*	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-LR	<1.0	3.23	3.29	11	7.07	7.17	<1.0	14.3	15.4	19.9	7.94	14.5
	MC-Demethyl-LR*	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-YR	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-LA	<1.0	5.27	6.93	34.3	13.6	16.4	<1.0	317	439	134	141	243
	MC-LW	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-LF	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-LY	<5.0	<5.0	<5.0	<5.0	<5.0	<5.0	<5.0	<5.0	<5.0	<5.0	<5.0	<5.0

Table 14 (contd). Microcystin variants results from freshwater mussel tissue sampling events in Klamath River Above Starwein Riffle and mouth of Trinity River July – October 2009.

	Sample Date	10/12/09	10/12/09	10/12/09	10/12/09	10/12/09
Microcystin Variants	Sample Site	KASR	KASR	KASR	KASR	KASR
	Sample Number	1	2	3	4	5
CA Department of Fish and Game Water Pollution Control Laboratory reporting limit 1.00 ng/g MC-LY reporting limit 5.0 ng/g units: ppb (ng/g)	MC-RR	1.8	1.97	2.24	<1.0	1.94
	MC-Demethyl-RR*	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-LR	4.64	4.21	9.24	5.32	5.33
	MC-Demethyl-LR*	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-YR	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-LA	282	234	589	247	167
	MC-LW	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-LF	<1.0	<1.0	<1.0	<1.0	<1.0
	MC-LY	<5.0	<5.0	<5.0	<5.0	<5.0

Table 15. Anatoxin-a results from freshwater mussel sampling events in Klamath River Above Starwein Riffle and mouth of Trinity River July – October 2009.

Anatoxin-a						
units: ppb (ng/g) CA Department of Fish and Game Water Pollution Control Laboratory reporting limit: 5.0 ng/g	Sample Date	7/20/09	8/18/09	9/14/09	9/15/09	10/12/09
	Sample Site	KASR	KASR	TR	KASR	KASR
	Sample Number	1,2,3,4,5	1,2,3,4,5	1,2,3,4,5	1,2,3,4,5	1,2,3,4,5
	Anatoxin-a	<5.0	<5.0	<5.0	<5.0	<5.0

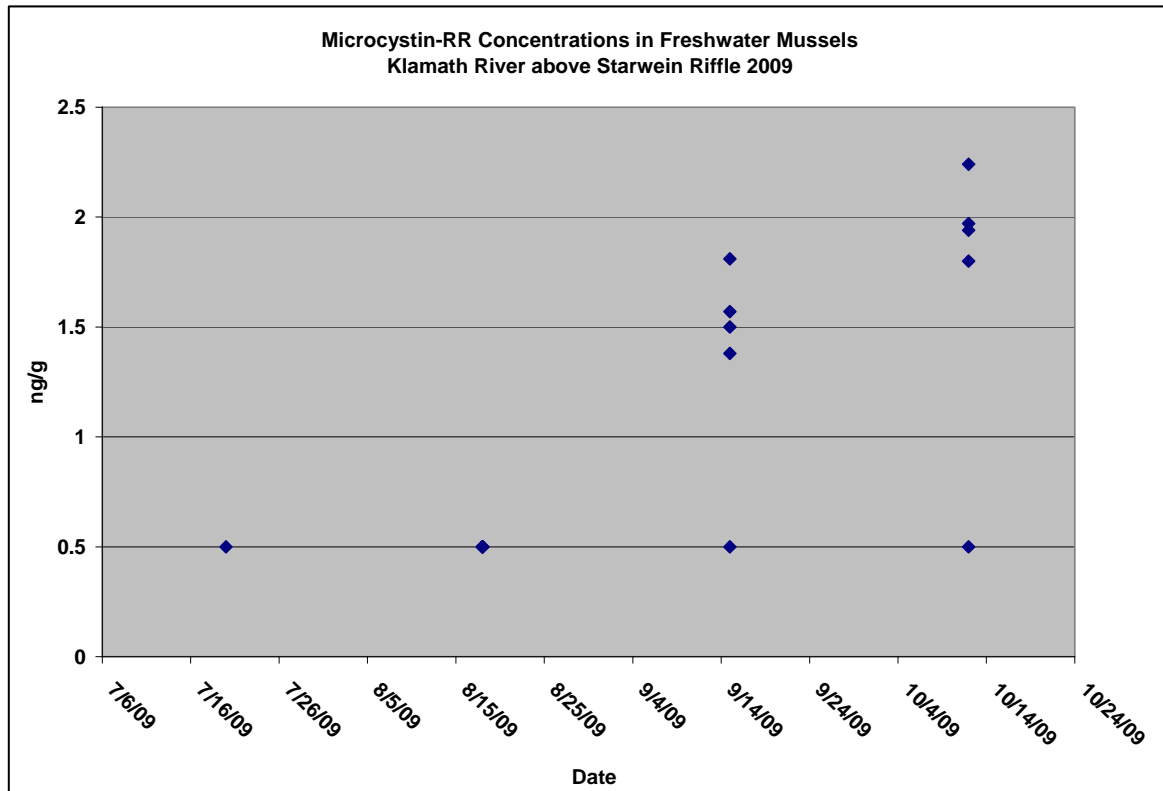


Figure 8. Microcystin-RR concentrations from freshwater mussel sampling events in Klamath River Above Starwein Riffle, July – October 2009.

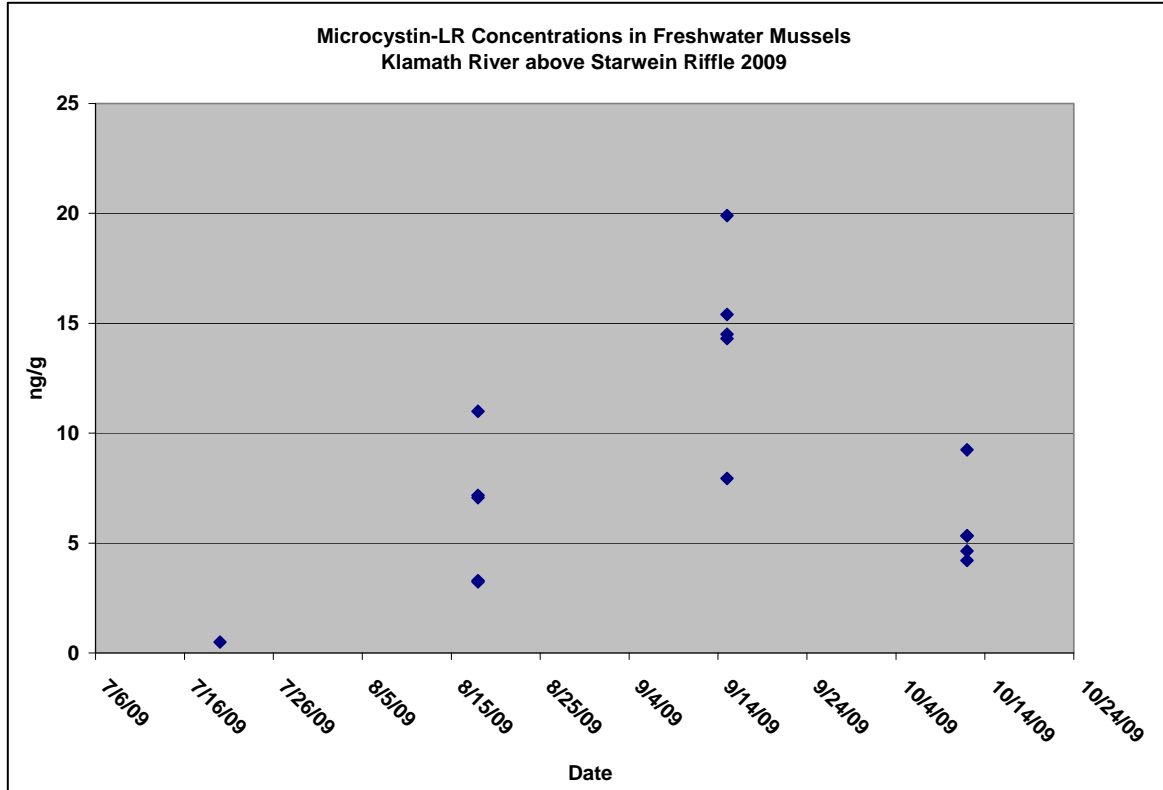


Figure 9. Microcystin-LR concentrations from freshwater mussel sampling events in Klamath River Above Starwein Riffle, July – October 2009.

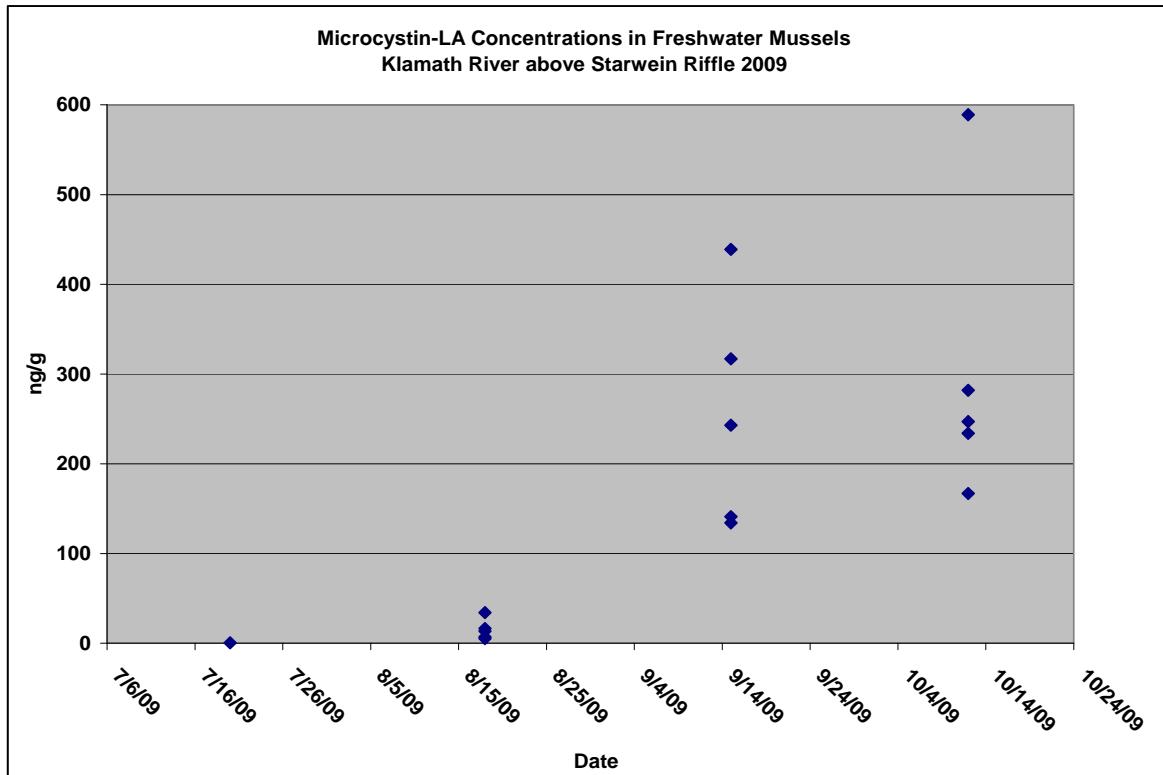


Figure 10. Microcystin-LA concentrations from freshwater mussel sampling events in Klamath River Above Starwein Riffle, July – October 2009.

Continuous Phycocyanin Probe Readings

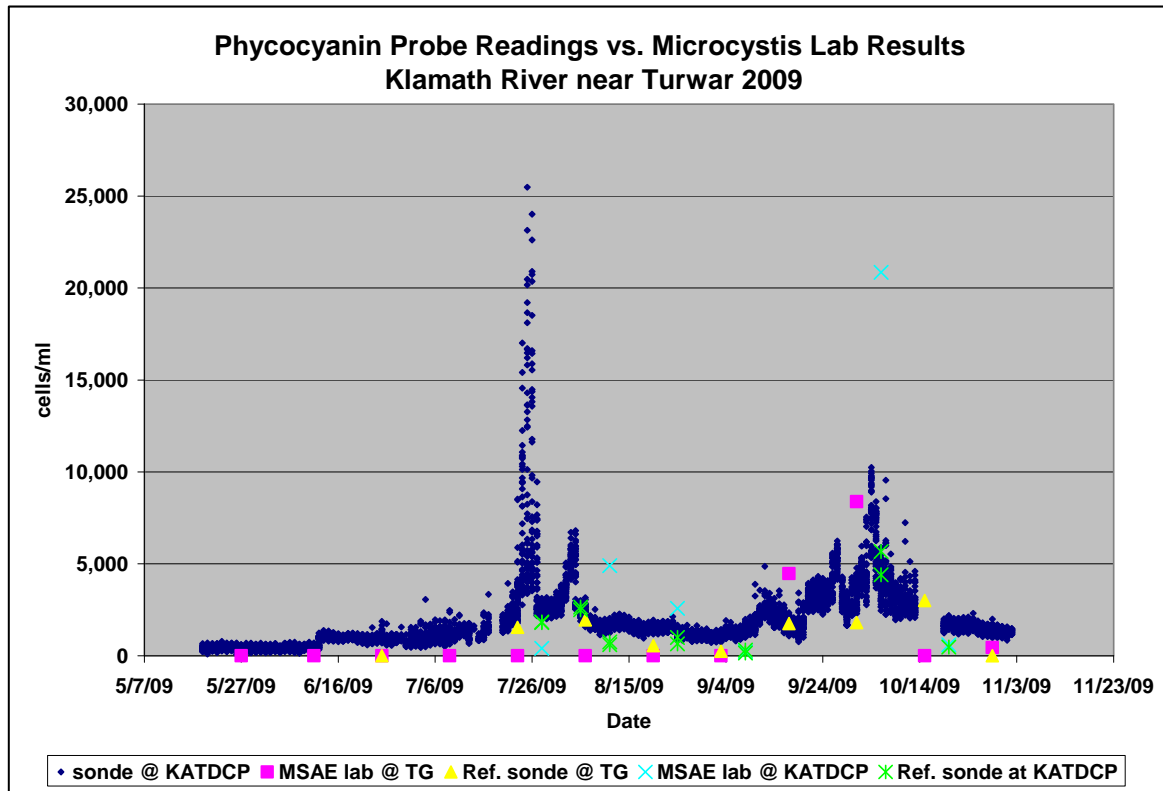


Figure 11. Continuous phycocyanin probe readings vs. microcystis lab results at Klamath River above Turwar, 2010

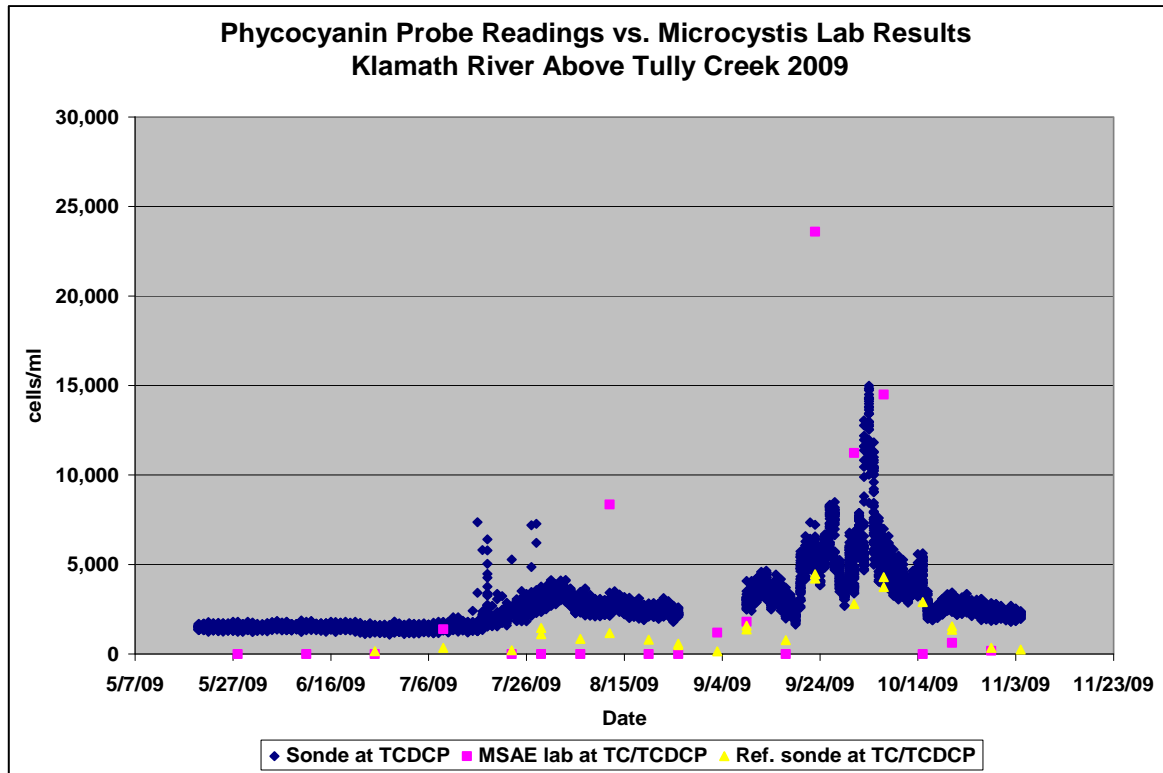


Figure 12. Continuous phycocyanin probe readings vs. microcystis lab results at Klamath River above Tully Creek, 2010

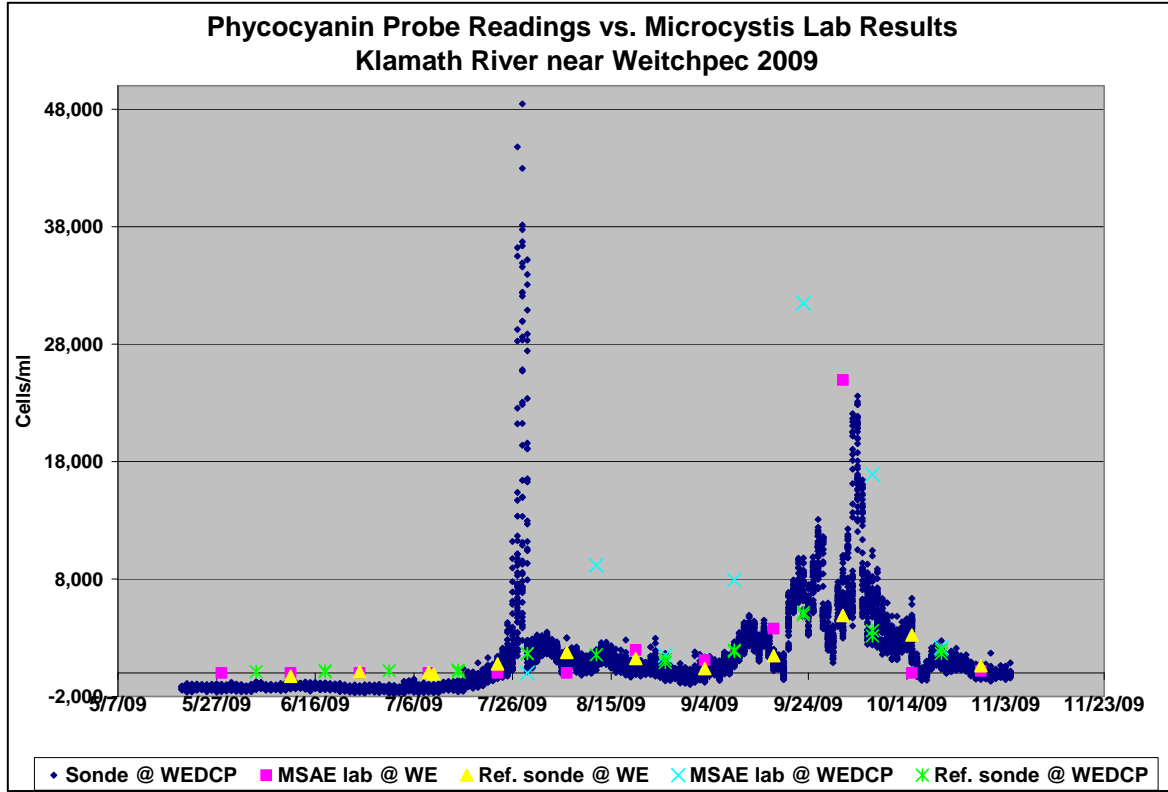


Figure 13. Continuous phycocyanin probe readings vs. microcystis lab results at Klamath River near Weitchpec, 2010

VI. Discussion: **Water Samples**

Aphanizomenon flos-aquae

Aphanizomenon flos-aquae (APF9) is a species of concern due to its ability to produce toxins and its abundance in the reservoirs managed by PacifiCorp located upstream of the YIR. To date the APF9 that is in the Klamath Basin is believed to be of a non-toxin producing strain.

However, at times when the Klamath River turns bright green APF9 is present in the water column. APF9 was found in water samples collected for baseline and public health purposes and was present in 28 out of 90 samples collected in the Klamath River reach located within the YIR from RM 44 to RM 0.5 (WE, WEDCP, TC, TCDCP, KASR, KATDCP, TG and LES) from May to December 2009. APF9 was detected in one of the 16 water samples collected at the Trinity River monitoring site in 2009. Summary information for the occurrence of APF9 in this river reach is presented in Table 4 and 5, Figure 2 and Appendix A. APF9 ranked as the 29th most dominant species out of 118 total species when looking at the average percent density (0.60%).

APF9 was first detected in the Klamath River on July 9, 2009 at the TC monitoring site. APF9 continued to be present at relatively low levels in the Klamath River at multiple sampling sites until August 20, 2009 and then was detected again on the September 3, 2009 sample event and was present at multiple sampling sites until October 21, 2009. The highest level of 2,472 cells/ml was recorded at the WEDCP monitoring site on July 29, 2009. APF9 was detected in both baseline and public health samples.

Microcystis aeruginosa

Microcystis aeruginosa (MSAE) was found in water samples collected for baseline and public health purposes and was present in 37 out of 90 samples collected in the Klamath River reach located within the YIR from RM 44 to RM 0.5 (WE, WEDCP, TC, TCDCP, KASR, KATDCP, TG and LES) from May to December 2009. MSAE was not detected in any of the 16 water samples collected at the Trinity River monitoring site. Summary information for the occurrence of MSAE in this river reach is presented in Tables 4 and 5, figures 3 and 4 and Appendix A. MSAE ranked as the 3rd most dominant species out of 118 total species when looking at the average percent density (10.63%).

MSAE was first detected in the Klamath River on July 7, 2009 at the TC sampling site. MSAE trends dipped until the August 12, 2009 sample event when they rose slightly. Further sample events indicate MSAE levels decreased slightly and then peaked on September 23, 2009. MSAE then began to decrease again and continued to be present in the Klamath River at multiple monitoring sites at low levels through October 29, 2009. MSAE was not detected in the Klamath River on the last two monitoring events of the season that took place on November 12 and December 17, 2009. The highest density of MSAE occurred at the WEDCP sampling site on September 23, 2009 and was measured at 31,488 cells/ml. MSAE did not exceed the State of California's Recommended Recreational Threshold of 40,000 cells/ml in 2009.

These results indicate that MSAE was present in the Klamath River within the YIR for over four months in 2009. Two pulses of MSAE came through the YIR with the second pulse containing the higher concentrations in the Klamath River with cell densities peaking at the end of September. Baseline and public health samples changed rank for the highest levels as the sampling season progressed. In August, the public health samples that were collected near the bank in slower moving water had higher concentrations of MSAE when compared to the baseline sampling sites. As river flows began to drop and the edge water habitats received less water the baseline sampling sites that are at locations where the water is well mixed had higher levels of MSAE, (see figures 3 and 4).

The timing of the MSAE peak is significant because of the presence of adult salmon and steelhead migrating upstream during this time period. This is also a time of increased cultural and recreational use of the Klamath River by both Tribal Members and sport fishermen.

Phycocyanin probe readings vs. *Microcystis aeruginosa* lab results

The phycocyanin probe readings had good agreement with the lab results for water samples collected at or near the datasonde locations (figures 11 -13). Each probe has a unique data signature for background conditions. Each probe reported different low level readings when cyanobacteria was not present in the Klamath River early and late in the monitoring season. For example, the phycocyanin probe reading at the Klamath River near Weitchpec sampling site background levels ranged from -700 to -1,600 cells/ml while readings at Klamath River above Tully Creek site ranged from 1,200 to 1,700 cells/ml. However, each site early and later in the season consistently had lab results that showed *Microcystis aeruginosa* and other cyanobacteria were not present in water samples. Therefore, it is the relative change from background readings that indicated the phycocyanin probe readings were detecting the presence of cyanobacteria including *Microcystis aeruginosa*

Phycocyanin probe readings increased when lab results showed *Microcystis aeruginosa* and other cyanobacteria were present. In general, lab results reported higher levels of *Microcystis aeruginosa* concentrations when compared to the phycocyanin probe readings. However, at low levels the phycocyanin probe readings were higher than what the lab results reported. YTEP's QA/QC methods included auditing the datasonde located at each site with an independent "reference" datasonde that has a phycocyanin probe. In general, the site and reference phycocyanin probe readings had good agreement. Although there was better agreement at the WEDCP monitoring site and the reference probe when compared to the KATDCP and TCDCP monitoring sites. These phycocyanin probes remain to be helpful in understanding the *Microcystis aeruginosa* dynamics in between water sampling events at different locations within the YIR.

Anabaena spp.* and *Oscillatoria spp.

Anabaena spp. and *Oscillatoria spp.* are also a species of concern due to their ability to produce toxins. *Anabaena spp.* was detected at low levels in water samples collected at the Klamath and Trinity River monitoring sites located within the YIR from RM 44 to RM 0.5 (WE, WEDCP, TC, TCDCP, KASR, KATDCP, TG, LES and TR). *Anabaena spp.* was detected in both baseline

and public health samples (see tables 4 and 5) and was present for almost three months at low levels. *Anabaena* spp. was present in 11 of 90 samples collected in the Klamath River. Public health samples had slightly higher levels when compared to the baseline samples. *Anabaena* spp. was first detected at the TR monitoring site on July 9, 2009 at low levels and was present in the YIR until October 1, 2009. The highest level of *Anabaena* spp. occurred at the WE public health sampling site on August 26, 2009 and was measured at 1,071 cells/ml. Summary information of all algae species identified and enumerated in this river reach is presented in Tables 4 and 5 and Appendix A. *Anabaena* spp. ranked as the 39th most dominant species out of 118 total species when looking at the average percent density (0.26%).

In 2009, 3 out of 16 samples collected in the Trinity River had presence of *Anabaena* spp. detected. All three detections in the Trinity River had cell concentrations below 100 cells/ml. These results are a concern due to the relatively good water quality in the Trinity River when compared to the Klamath River.

Oscillatoria spp. was detected once at the TCDCP monitoring site on August 26, 2009 and was measured at 51 cells/ml. YTEP will continue to monitor phytoplankton trends over time in the Klamath River and will respond appropriately to any increase in toxicogenic cyanobacteria species in the future.

Microcystin and Anatoxin-a

Microcystin followed a similar trend to the *Microcystis aeruginosa* concentrations in the Klamath River in 2009. Both baseline and public health sampling results indicate that microcystin was present in the Klamath River within the YIR for over four months in 2009 (see table 7 and 8, figures 6 and 7). Two pulses of microcystin were present in the Klamath River within the YIR boundaries with the second pulse containing the higher concentrations in the Klamath River with microcystin peaking on September 23rd for public health samples and on October 8th for baseline samples. Baseline and public health samples changed rank for the highest concentrations as the sampling season progressed. Anatoxin-a was not detected in any of the water samples collected within the YIR (table 10). Microcystin was not detected in any of the water samples collected in the Trinity River within the YIR (table 7).

ELISA Results

Baseline Samples

The majority of the baseline water samples submitted for microcystin analysis were analyzed by USEPA's laboratory utilizing the ELISA method. Microcystin was first detected in the Klamath River at a very low level near the reporting limit on July 23, 2009 at the LES sampling site (table 7 and figure 6). Microcystin trends rose briefly and then dipped and stayed relatively stable until September 3, 2009 and began to rise during subsequent sampling events until concentrations peaked on October 8, 2009 in the Klamath River Estuary. Further sample events indicate microcystin levels decreased until the last sampling event on December 17, 2009. The highest concentration of microcystin occurred at the LES sampling site on October 8, 2009 and was

measured at 3.0µg/L. Baseline microcystin samples did not exceed the State of California's Recommended Recreational Threshold of 8µg/L.

Public Health Samples

All of the public health water samples submitted for microcystin analysis were analyzed by USEPA's laboratory utilizing the ELISA method. The public health microcystin samples followed a similar trend as the baseline water samples. Microcystin was first detected in the Klamath River at a very low level near the reporting limit on July 23, 2009 at the WE and the TG sampling sites (Table 8 and Figure 7). Microcystin rose briefly and then dipped and stayed relatively stable until September 3, 2009 and began to rise during subsequent sampling events until concentrations peaked on September 23, 2009 at the WE sampling site. Further sample events indicate microcystin levels decreased until the last sampling event occurred on October 15, 2009. The highest concentration of microcystin occurred at the WE sampling site on September 23, 2009 and was measured at 4.4µg/L. Public Health microcystin samples did not exceed the State of California's Recommended Recreational Threshold of 8µg/L.

The timing of the MSAE peak is significant because of the presence of adult salmon and steelhead migrating upstream during this time period. This is also a time of increased cultural and recreational use of the Klamath River by both Tribal Members and sport fishermen.

LC/MS/MS Results

Additional samples were submitted to CA Fish and Game's Water Pollution Control Laboratory in Rancho Cordova, CA to cross check the microcystin results from USEPA and to determine if anatoxin-a was present in the Klamath River. These water samples were analyzed by LC/MS/MS method. Microcystin-LA was detected in one water sample that was collected for baseline monitoring purposes at the WE site at 2.29µg/L (table 9). Microcystin-LA was also detected at the KASR site when water samples were collected with a churn splitter during freshwater mussel sampling for public health purposes (table 12). Microcystin-LA was 1.05µg/L and 1.5µg/L on August 18 and September 15, respectively.

Anatoxin-a was not detected above the reporting limit of 5 µg/L for all samples that were submitted to the laboratory for testing (see tables 10 and 13).

Freshwater Mussel Tissues Samples

Freshwater mussels were collected at two locations within the YIR to determine if microcystin and anatoxin-a were present for public health purposes. 5 mussels were collected during each sampling event in the Klamath River in July 20, August 18, September 15 and October 12, 2009. 5 mussels were collected in the Trinity River near the mouth of the confluence with the Trinity River on September 14, 2009. These tissue samples were submitted to CA Fish and Game's Water Pollution Control Laboratory in Rancho Cordova, CA and analyzed by the LC/MS/MS method. Water samples were also collected with a churn splitter near the surface during the same time the freshwater mussels were collected. These water samples were analyzed for phytoplankton species identification and enumeration, total microcystin at EPA's lab via the

ELISA method and for microcystin variants and anatoxin-a at DFG's lab via the LC/MS/MS method.

Tissue and water samples collected at the Trinity River near the mouth (TR) sampling site showed negative results for microcystin variants, anatoxin-a, and *Microcystis aeruginosa*. These findings are consistent with other water samples collected in the Trinity River in 2009. However, *Anabaena spp.* has been detected in 3 samples in the Trinity River in 2009 at low levels (<100 cells/ml). Water samples collected in past years have also detected *Anabaena spp.* in one sample per year so the potential for anatoxin-a to occur in the water and tissue is possible.

Water samples collected at the Klamath River above Starwein Riffle (KASR) sampling site showed positive results for microcystin variants but varied among labs. For water samples collected at KASR *Microcystis aeruginosa* was not identified in the July sampling event but was present in water samples that were collected in August, September and October (table 6 and figure 5). Total microcystin was detected in water samples analyzed by EPA's lab show low levels in July with microcystin increasing slightly in each sample up to October (table 11). Microcystin-LA was detected slightly above the reporting limit (1µg/L) in water samples analyzed by DFG's lab in August and September but not in July or October (table 12).

Microcystin tissue results for the freshwater mussels are located in table 14 and figures 8-10. Microcystin was not detected in mussel tissue collected at the KASR sampling site during the July sampling event. During the August sampling event all five freshwater mussels had detectable levels of microcystin-LR and LA and ranged from 3.23 to 11µg/L and 5.27 to 34.3µg/L, respectively. The five freshwater mussels collected and analyzed in September had higher levels of microcystin-LR and LA and ranged from 7.94 to 19.9µg/L and 134 to 439µg/L, respectively. Freshwater mussels collected in September also had detectable levels of microcystin-RR at low levels and ranged from 1.38 to 1.81µg/L. The five freshwater mussels collected and analyzed in October showed mixed results with microcystin-LR levels slightly decreasing with results ranging from 4.21 to 9.24µg/L while microcystin-LA levels increased with results ranging from 167 to 589µg/L. Microcystin-RR levels were slightly elevated for four samples ranging from 1.8 to 2.24µg/L and one sample was below the reporting limit of 1µg/L.

A comparison of tissue concentration data relative to ambient water concentration data revealed that microcystin-LA was detected in both media, while microcystin-RR and LR were detected in tissue samples but they were not detected in water samples. In addition, the frequency of microcystin-LA detection in tissue was greater than that for water. The lack of detection or low frequency of microcystin detection in water relative to tissue samples clearly indicates that bioaccumulation mechanisms then cause a proportionally greater frequency of detection (as well as higher concentration) in freshwater mussel tissue. The importance of this finding is that even when microcystin may be below detection in the ambient water, accumulation in freshwater mussel tissue can still occur (Kann et al. 2010)

These data show that ingestion of freshwater mussels in the Klamath River system would result in microcystin doses that exceed various public health thresholds for safe consumption throughout the summer and fall (Kann et al. 2010). These are the months when traditional and subsistence use of fresh water mussels by Tribal members occurs, and at these times even one

meal could exceed safe consumption levels. It should be realized that if the use of these organisms is curtailed during these months that coincide with harvest times, their use would be effectively eliminated both from a dietary and Tribal cultural standpoint. Regardless, given that microcystin levels in ambient water that are below public health guidelines for recreational waters can result in substantial microcystin bioaccumulation in freshwater mussels, caution must be exercised when consuming these organisms in the mainstem Klamath (Kann et al. 2010)

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Appendix A

Table A-1. Combined Algae Species List for Klamath River Sites located at River Mile 44 to 0.5 (WE, TC, TG, LES, KATDCP, KASR, TCDCP, WEDCP) May to December, 2009.

#	Algae Species	Ave %		Code
		Den	# samples	
1	<i>Epithemia sorex</i>	18.16	78	EPSX
2	<i>Diatoma tenue</i>	11.07	63	DTTN
3	<i>Microcystis aeruginosa</i>	10.63	37	MSAE
4	<i>Cocconeis placentula</i>	9.85	86	COPC
5	<i>Nitzschia frustulum</i>	4.60	76	NZFR
6	<i>Synedra ulna</i>	3.79	77	SNUL
7	<i>Rhodomonas minuta</i>	3.10	62	RDMN
8	<i>Achnanthes minutissima</i>	2.90	51	ACMN
9	<i>Scenedesmus quadricauda</i>	2.74	63	SCQD
10	<i>Asterionella formosa</i>	2.47	17	ASFO
11	<i>Rhoicosphenia curvata</i>	2.32	70	RHCU
12	<i>Nitzschia palea</i>	2.14	54	NZPL
13	<i>Chlamydomonas</i> sp.	1.77	52	CHX1
14	<i>Melosira granulata</i>	1.72	34	MLGR
15	<i>Ankistrodesmus falcatus</i>	1.47	64	AKFL
16	<i>Navicula cryptocephala veneta</i>	1.25	56	NVCV
17	<i>Stephanodiscus hantzschii</i>	1.20	35	STHN
18	<i>Gomphonema angustatum</i>	1.16	50	GFAN
19	<i>Diatoma vulgare</i>	1.13	53	DTVL
20	<i>Navicula cryptocephala</i>	1.08	53	NVCR
21	<i>Nitzschia paleacea</i>	0.93	43	NZPC
22	<i>Cymbella affinis</i>	0.91	37	CMAF
23	<i>Cymbella sinuata</i>	0.89	50	CMSN
24	<i>Cyclotella meneghiniana</i>	0.87	29	CCMG
25	<i>Gomphonema subclavatum</i>	0.75	38	GFSB
26	<i>Selenastrum minutum</i>	0.71	28	SLMN
27	<i>Nitzschia dissipata</i>	0.64	22	NZDS
28	<i>Nitzschia capitellata</i>	0.63	15	NZCP
29	<i>Aphanizomenon flos-aquae</i>	0.60	28	APF9
30	<i>Fragilaria construens venter</i>	0.59	34	FRCV
31	<i>Gomphonema ventricosum</i>	0.54	33	GFVT
32	<i>Nitzschia acicularis</i>	0.53	25	NZAC
33	<i>Cryptomonas erosa</i>	0.52	33	CXER
34	<i>Gomphoneis herculeana</i>	0.45	32	GSHR
35	<i>Navicula decussis</i>	0.35	23	NVDC
36	<i>Cymbella minuta</i>	0.34	21	CMMN
37	<i>Fragilaria construens</i>	0.33	18	FRCN
38	<i>Amphora perpusilla</i>	0.27	21	AFPR

Table A-1(contd.). Combined Algae Species List for Klamath River Sites located at River Mile 44 to 0.5 (WE, TC, TG, LES, KATDCP, KASR, TCDCP, WEDCP) May to December, 2009.

#	Algae Species	Ave %		Code
		Den	# samples	
39	Anabaena sp.	0.26	11	ABX9
40	Synedra mazamaensis	0.26	16	SNMZ
41	Navicula tripunctata	0.25	19	NVTP
42	Achnanthes lanceolata	0.25	18	ACLC
43	Tetraedron minimum	0.24	18	TEMN
44	Rhopalodia gibba	0.21	17	RPGB
45	Fragilaria vaucheriae	0.20	16	STHN
46	Gomphonema olivaceum	0.20	18	GFOM
47	Eunotia pectinalis	0.20	2	EUPC
48	Navicula sp.	0.16	15	NVXX
49	Diatoma tenue elongatum	0.14	4	DTTE
50	Navicula graciloides	0.13	11	NVGC
51	Navicula gregaria	0.11	8	NVGR
52	Scenedesmus denticulatus	0.10	9	SCDT
53	Nitzschia linearis	0.10	8	NZLN
54	Amphora ovalis	0.08	6	AFOV
55	Sphaerocystis schroeteri	0.08	7	SFSR
56	Melosira varians	0.08	6	MLVR
57	Nitzschia amphibia	0.08	6	NZAM
58	Scenedesmus abundans	0.07	7	SCAB
59	Pediastrum boryanum	0.06	6	PSBR
60	Ulothrix sp.	0.06	5	ULX9
61	Nitzschia communis	0.06	6	NZCM
62	Navicula viridula	0.06	6	NVVR
63	Gyrosigma spencerii	0.05	5	GYSP
64	Achnanthes linearis	0.05	3	ACLN
65	Nitzschia innominata	0.04	4	NZIN
66	Pediastrum tetras	0.04	3	PSTT
67	Coelastrum microporum	0.04	4	CUMC
68	Epithemia turgida	0.04	4	EPTR
69	Nitzschia microcephala	0.04	4	NZMC
70	Achnanthes clevei	0.04	3	ACCV
71	Synedra radians	0.03	2	SNRD
72	Nitzschia volcanica	0.03	3	NZVL
73	Navicula capitata	0.03	3	NVCP
74	Cymbella tumida	0.03	3	CMTM
75	Anabaena flos-aquae	0.03	3	ABFA
76	Pinnularia sp.	0.03	3	PLXX
77	Scenedesmus acuminatus	0.03	3	SCAC

Table A-1(contd.). Combined Algae Species List for Klamath River Sites located at River Mile 44 to 0.5 (WE, TC, TG, LES, KATDCP, KASR, TCDCP, WEDCP) May to December, 2009.

# Algae Species	Ave % Den	# samples	Code
78 <i>Synedra cyclopum</i>	0.03	2	SNCY
79 <i>Gomphonema</i> sp.	0.02	2	GFXX
80 <i>Scenedesmus</i> sp.	0.02	2	SCXX
81 <i>Synedra rumpens</i>	0.02	2	SNRM
82 <i>Fragilaria capucina mesolepta</i>	0.02	3	FRCM
83 <i>Cyclotella comta</i>	0.02	2	CCCM
84 <i>Stephanodiscus astraera minutula</i>	0.02	2	STAM
85 <i>Nitzschia</i> sp.	0.02	3	NZXX
86 <i>Glenodinium</i> sp.	0.02	2	GDXX
87 <i>Caloneis ventricosa minuta</i>	0.02	2	CAVM
88 <i>Fragilaria pinnata</i>	0.02	2	FRPN
89 <i>Navicula minuscula</i>	0.02	2	NVML
90 <i>Cyclotella stelligera</i>	0.02	2	CCST
91 <i>Anomoeoneis vitrea</i>	0.01	1	AOVT
92 <i>Botryococcus braunii</i>	0.01	1	BTBR
93 <i>Oscillatoria</i> sp.	0.01	1	OSXX
94 <i>Rhopalodia musculus</i>	0.01	1	RPMS
95 <i>Melosira ambigua</i>	0.01	1	MLAM
96 <i>Denticula elegans</i>	0.01	1	DNEL
97 Unidentified flagellate	0.01	1	MXFG
98 <i>Hannaea arcus</i>	0.01	1	HNAR
99 <i>Melosira italica</i>	0.01	1	MLIT
100 <i>Synedra tenera</i>	0.01	1	SNTN
101 <i>Nitzschia fruticosa</i>	0.01	1	NZFU
102 <i>Caloneis ventricosa</i>	0.01	1	CAVT
103 <i>Pediastrum duplex</i>	0.01	1	PSDP
104 <i>Achnanthes lewisiana</i>	0.01	1	ACLW
105 <i>Gomphonema clevei</i>	0.01	1	GFCL
106 <i>Tetrastrum staurogeniaforme</i>	0.01	1	TTS9
107 <i>Eunotia incisa</i>	0.01	1	EUIN
108 <i>Gomphonema tenellum</i>	0.01	2	GFTN
109 <i>Cosmarium</i> sp.	0.01	1	CSXX
110 <i>Gomphonema acuminatum</i>	0.01	1	GFAC
111 <i>Tetraedron</i> sp.	0.01	1	TEXX
112 <i>Navicula rhynchocephala</i>	0.01	1	NVRH
113 <i>Spirogyra</i> sp.	0.01	1	SPXX
114 <i>Synedra socia</i>	0.01	1	SNSC
115 <i>Melosira distans alpigena</i>	0.01	1	MLDA
116 <i>Nitzschia clausii</i>	0.01	1	NZCL
117 <i>Navicula pupula</i>	0.01	1	NVPP
118 <i>Navicula minima</i>	0.01	1	NVMN

Table A-2 Combined Algae Species List for Mouth of Trinity River Site (TR) May to December, 2009.

#	Algae Species	Ave % Den	# samples	Code
1	<i>Diatoma tenue</i>	28.77	15	DTTN
2	<i>Epithemia sorex</i>	11.87	14	EPSX
3	<i>Cocconeis placentula</i>	11.26	16	COPC
4	<i>Synedra ulna</i>	7.95	10	SNUL
5	<i>Achnanthes minutissima</i>	3.21	13	ACMN
6	<i>Gomphonema angustatum</i>	3.18	11	GFAN
7	<i>Cymbella affinis</i>	3.00	14	CMAF
8	<i>Navicula cryptocephala veneta</i>	1.97	12	NVCV
9	<i>Ankistrodesmus falcatus</i>	1.90	10	AKFL
10	<i>Rhoicosphenia curvata</i>	1.77	11	RHCU
11	<i>Nitzschia frustulum</i>	1.49	8	NZFR
12	<i>Cymbella sinuata</i>	1.41	9	CMSN
13	<i>Nitzschia paleacea</i>	1.33	8	NZPC
14	<i>Navicula cryptocephala</i>	1.03	7	NVCR
15	<i>Scenedesmus quadricauda</i>	0.95	6	SCQD
16	<i>Rhodomonas minuta</i>	0.94	7	RDMN
17	<i>Rhopalodia gibba</i>	0.93	4	RPGB
18	<i>Diatoma vulgare</i>	0.88	8	DTVL
19	<i>Selenastrum minutum</i>	0.87	6	SLMN
20	<i>Nitzschia palea</i>	0.85	6	NZPL
21	<i>Cymbella minuta</i>	0.81	6	CMMN
22	<i>Chlamydomonas</i> sp.	0.76	7	CHXX
23	<i>Gomphonema subclavatum</i>	0.75	8	GFSB
24	<i>Amphora perpusilla</i>	0.73	6	AFPR
25	<i>Cryptomonas erosa</i>	0.68	6	CXER
26	<i>Fragilaria construens venter</i>	0.64	4	FRCV
27	<i>Nitzschia acicularis</i>	0.61	6	NZAC
28	<i>Nitzschia dissipata</i>	0.60	5	NZDS
29	<i>Aphanizomenon flos-aquae</i>	0.60	1	APFA
30	<i>Epithemia turgida</i>	0.56	6	EPTR
31	<i>Navicula decussis</i>	0.54	6	NVDC
32	<i>Synedra rumpens</i>	0.52	4	SNRM
33	<i>Cymbella microcephala</i>	0.52	4	CMMC
34	<i>Anabaena</i> sp.	0.51	3	ABX9
35	<i>Achnanthes lanceolata</i>	0.48	3	ACLC
36	<i>Navicula tripunctata</i>	0.42	4	NVTP
37	<i>Diatoma tenue elongatum</i>	0.38	2	DTTE
38	<i>Fragilaria pinnata</i>	0.34	4	FRPN

Table A-2 (Contd.) Combined Algae Species List for Mouth of Trinity River Site (TR) May to October, 2009.

#	Algae Species	Ave % Den	# samples	Code
39	Caloneis ventricosa minuta	0.25	2	CAVM
40	Scenedesmus denticulatus	0.24	3	SCDT
41	Navicula pupula	0.24	4	NVPP
42	Navicula sp.	0.22	3	NVXX
43	Gomphonema olivaceum	0.20	3	GFOM
44	Gomphonema ventricosum	0.19	2	GFVT
45	Mougeotia sp.	0.17	1	MGXX
46	Caloneis ventricosa	0.16	1	CAVT
47	Gomphoneis herculeana	0.16	2	GSHR
48	Amphipleura pellucida	0.16	2	AMPL
49	Gomphonema clevei	0.15	2	GFCL
50	Fragilaria vaucheriae	0.14	2	FRVA
51	Tetraedron regulare	0.14	1	TERG
52	Fragilaria construens	0.13	2	FRCN
53	Crucigenia tetrapedia	0.13	2	CGTR
54	Fragilaria capucina mesolepta	0.12	2	FRCM
55	Pediastrum duplex	0.11	1	PSDP
56	Cosmarium sp.	0.09	1	CSXX
57	Crucigenia quadrata	0.09	1	CGQD
58	Tetraedron minimum	0.07	1	TEMN
59	Synedra radians	0.07	1	SNRD
60	Diploneis interrupta	0.07	1	DPIN
61	Nitzschia capitellata	0.07	1	NZCP
62	Tetrastrum staurogeniaforme	0.07	1	TTST
63	Navicula graciloides	0.07	1	NVGC
64	Achnanthes clevei	0.06	1	ACCV
65	Navicula minima	0.06	1	NVMN
66	Nitzschia sp.	0.06	1	NZXX
67	Spirogyra sp.	0.06	1	SPXX
68	Nitzschia communis	0.06	1	NZCM
69	Cymbella tumida	0.05	1	CMTM
70	Nitzschia linearis	0.05	1	NZLN
71	Synedra mazamaensis	0.05	1	SNMZ
72	Fragilaria crotonensis	0.05	1	FRCR

Appendix B Grab Sample Protocol

‘Grab sampling’ refers to water samples obtained by dipping a collection container into the upper layer of a body of water and collecting a water sample (USGS File Report -00213). For quality assurance/quality control (QA/QC) purposes replicate, and blank bottle sets will be prepared and collected for one site each sampling period. These additional bottle sets will be handled, prepared and filled following the same protocol used for regular bottle sets and samples. General water quality parameters will also be measured with a freshly calibrated portable multi-probe water quality instrument during grab samples and recorded onto data sheets.

Upon arrival at each site, the sampling churn will be rinsed three times with distilled water. The goal of rinsing is ‘equipment decontamination – the removal from equipment, residues from construction and machining and the removal of substances adhering to equipment from previous exposure to environmental and other media’ (USGS Open File Report 00213). After rinsing with D.I. water, the churn will be rinsed three times with stream water. The churn is then fully submerged into the stream and filled to the lid with sample water. Completely filling the churn allows for all samples to be filled from one churn; thereby minimizing differences in water properties and quality between samples.

Proper use of the churn guarantees the water is well mixed before the sample is collected. The churn should be stirred at a uniform rate by raising or lowering the splitter at approximately 9 inches per second (Bel-Art Products, 1993). This mixing must continue while the bottles are being filled. If filling is stopped for some reason, the stirring rate must be resumed before the next sample is drawn from the churn. As the volume of water in the churn decreases, the round trip frequency increases as the velocity of the churn splitter remains the same. Care must be taken to avoid breaking the surface of the water as the splitter rises toward the top of the water in the churn.

Sample bottles and chemical preservatives used were provided by associated laboratories and were considered sterile prior to field usage. Sample bottles without chemical preservatives were rinsed with stream water from the churn 2-3 times before filling with sample water. In the case of bottles that contained chemical preservatives, bottles were not rinsed before sample collection and care was taken to avoid over-spillage that would result in chemical preservative loss. Collected samples will be placed in coolers on ice or dry ice for transport to contracted laboratories for analysis.

QA/QC – Replicate bottle set

To ensure laboratory and sampling accuracy, one site every sampling period was randomly selected to receive one additional QA/QC bottle set. This bottle set contain replicate water samples. Replicate samples are obtained using the same process as regular samples. These are used to assure the laboratory maintains precision within results.

All bottle sets are then placed on ice and are transported to the associated laboratories. All grab samples were processed within 24 hours or within known laboratory holding periods.

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