2006 Klamath River Blue-Green Algae Summary Report



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Table of Contents

I. Introduction	4
II. Methods	5
III. Site Selection	6
IV. Quality Assurance	9
V. Results	13
VI. Discussion	21

Figures

1 Map of monitoring locations	8
2 Results for the replicate samples containing <i>Microcystis aeruginosa</i>	10
3 Aphanizomenon flos-aquae levels, Klamath River Miles 254 to 224	17
4 Aphanizomenon flos-aquae levels, Klamath River Miles 190 to 0.5	17
5 Microcystis aeruginosa levels, Klamath River Miles 254 to 224	18
6 Microcystis aeruginosa levels, Klamath River Miles 190 to 0.5	18
7 Microcystin levels, Klamath River Miles 254 to 224	20
8 Microcystin levels, Klamath River Miles 190 to 0.5	20

Tables

1 Blank QA sample results	11
2 Phytoplankton results for water samples	14
3 Microcystin Results Klamath River Miles 254 to 0.5	19

Literature Cited	
Appendix	

I. Introduction

This report summarizes the occurrence and extent of the blue-green algae bloom on the Klamath River in 2006, excluding the Klamath Hydroelectric Project (KHP) reservoirs. The Karuk Tribe will be publishing a summary report that details the results for the algae and nutrients conditions in Copco and Iron Gate Reservoirs. YTEP, the Karuk Tribe and BOR collected water samples indicating that the water quality in the Klamath River was negatively impacted by levels of the cyanobacterium *Microcystis aeruginosa* (microcystis) and its resultant toxin, microcystin; with some levels exceeding World Health Organization (WHO) risk guidelines in all reaches of the Klamath River from downstream of Upper Klamath Lake to the Klamath River Estuary. Water quality sampling performed in 2005 and 2006 detected microcystis and microcystin in the Klamath River below Iron Gate Dam. The Karuk Tribe and YTEP were integral in collecting this data and sharing results with the local community.

Information on Microcystis aeruginosa

Microcystis aeruginosa is a type of blue-green algae which, upon death and decomposition, releases the hepatoxin microcystin, which can cause any of the following reactions in humans and/or animals: rash, irritation, conjunctivitis, nausea, vomiting, diarrhea, liver damage, tingling, numbness, paralysis, and death. Microcystin bioaccumulates in the liver, organs, and to a certain degree the muscle mass of living animals. Microcystin is not excreted by animals, and dosage over time will eventually cause liver damage, decreased liver function and increased liver size, and eventually death. Mortality in fish, domestic animals, and humans has been recorded following exposure to microcystin resultant from both single-dose events and long-term exposure.

The toxin microcystin can produce negative health effects from contact with impaired waters, from incidental nasal/oral ingestion of impaired waters, and, most seriously, from swallowing or drinking impaired waters. Microcystin can be found in the organs and muscle meat of fish who feed in impaired waters. Continued exposure to even low levels of microcystin can produce harmful cumulative response in humans and animals. Because the timing for likely bloom occurrence coincides with the annual salmon runs along the Klamath River, there is significant cause for concern; Tribal subsistence fishers not only have increased contact with impaired waters due to subsistence fishing methods, they are also more frequently visiting impaired waters than recreational or sport users.

Initial bloom characteristics are green, green-blue, yellow, or whitish waters; blooms of increasing severity can form algal mats or visible surface scum, initially in nearshore areas and backwaters or where winds cause surface detritus to accumulate, and steadily progressing to cover open areas of still or slow-moving waters. Odor and taste changes may become noticeable at any time as decomposition of organisms sets in, but should not be used as an indicator for cellular mortality.

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 microcystis/microcystin concentrations in recreational waters:

	Microcystis cells/milliliter	Microcystin micrograms/liter
Low Risk:	20,000	4
Moderate Risk:	100,000	20
Severe Risk	10,000,000 or 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.

II. <u>Methods</u>

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 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 the same water mixture into sample bottles used for algal identification and enumeration and testing for microcystin.

The filled churn was used to fill the plastic sample bottles for determination of algal species and microcystin concentrations. The sample bottle for determination of algal species contained Lugol's preservative and the toxin sample water 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. TG090106).

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 a replicate split sample and a blank sample were 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. 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 in White Salmon, WA 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.). 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 mailed on ice overnight to CyanoHAB Services at Wright State University in Dayton, OH 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 CyanoHAB Services employs and measures total microcystin.

Samples collected in October were analyzed by the California Animal Health & Food Safety Laboratory at UC Davis due to the lack of availability to CyanoHab Services. All water samples submitted to the UC Davis lab were analyzed for four microcystin variants. The microcystin variants measured include microcystin-LR, LA, YR and RR using liquid chromatography/mass spectrometry (LC/MS).

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 in bold indicate established sampling locations for the collection of water samples for nutrient analysis and phytoplankton speciation and enumeration from May through

October. Algae samples were collected at these sites and sites not in bold once the algae bloom was documented in the Klamath River below Iron Gage Dam (RM 190) to determine the extent and occurrence of the algae bloom in the Klamath River. The Lower Estuary Surface site was chosen because of concern that *Microcystis aeruginosa* bloom may increase once it reached the estuarine habitat.

Three river sites (KBL, KBK and KBB) were also added to the sampling network in the Klamath River above Iron Gate and Copco Reservoirs to compare conditions above and below PacifiCorp's Hydroelectric Project. BOR personnel coordinated with the Karuk Tribe and YTEP to collect samples at these locations on the same day the Tribes collected their samples.

The four major tributary sites were sampled for algae speciation on a monthly basis. In order to save resources increased sampling did not occur in the major tributaries because the routine algae speciation showed that no toxicogenic cyanobacteria were present. However, one toxin sampling event did occur in the tributaries to determine if microcystin was present in the tributaries.

The BOR, Karuk Tribe and YTEP collected water samples for toxin and speciation analysis at the following mainstem Klamath River locations (river miles are approximate):

- KBL- Klamath River Below Link Dam RM 254
- KBK- Klamath River Below Keno Dam RM 233
- KBB- Klamath River Below JC Boyle Dam RM 224
- IG- Klamath River Below Iron Gate Dam RM 190
- SV- Klamath River at Seiad Valley Gage RM 128.5
- OR Klamath River at Orleans Gage RM 59
- WE Klamath River at Weitchpec (upstream of Trinity River) RM 43.5
- KBW Klamath River below Weitchpec RM 42.5
- BC Klamath River Above Blue Creek RM 16.5
- TG Klamath River at Turwar Boat Ramp RM 6
- LES Lower Estuary Surface RM 0.5

The Karuk Tribe and YTEP collected water samples for toxin and speciation analysis at the following major tributary locations:

- SH Shasta River near mouth RM 0.5
- SC Scott River near mouth RM 1.5
- SA Salmon River near mouth RM 1.0
- TR Trinity River near mouth (above Klamath River confluence) RM 0.5



Figure 1. Map of monitoring locations, 2006.

IV. Quality Assurance

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 the collection and analysis of field replicates and blank samples. 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 samples were replicates.

Rinsate blank samples collected from May through July were collected after collection of all other samples at the QA/QC site. The churn was rinsed three times with distilled water before being filled with distilled water. The rinsate blank bottle sets were collected in the same way as other samples, except that distilled water was used in place of stream water. Blank samples collected from August through October involved pouring distilled water straight into the sample containers. True blank samples were collected to determine whether or not Wright State would detect low levels of microcystin in distilled water. All of the blank samples were disguised and sent with the other samples collected for analysis of algae identification and enumeration and microcystin.

The Klamath River Blue-Green Algae Workgroup sent numerous split samples to compare USEPA Region IX Lab's ELISA results. Only toxin results from Wright State are presented in this report because the USEPA final results are not available at this time. The USEPA ELISA and the UC Davis LC/MS results will be reported in a future publication distributed by the Klamath River BGA workgroup.

Phytoplankton

QA replicate samples indicate that the phytoplankton speciation and enumeration results are acceptable. Not all of the phytoplankton replicate samples contained *Microcystis aeruginosa* (MSAE). Samples collected in May, June and July did not contain MSAE, therefore, no relative percent differences were calculated for these months.

Although the common algae in each replicate change rank for samples collected between May and July, 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. For example, the TSI of replicates were 40.4/42.8 in May, 50.2/79.8 in June and 45.8/42.7 in July. This is fairly close, considering the wide range of all the other samples. 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.

Samples collected in August, September and October did contain MSAE and RPD's were calculated. All relative percent differences (RPD's) for the samples containing MSAE exceeded 25%, which is the RPD employed by Kann and Corum when validating phytoplankton and toxin results in the recent technical memorandum titled "Summary of 2005 Toxic Microcystis aeruginosa Trends in Copco and Iron Gate Reservoirs on the

Klamath River, CA". However, in all three cases the primary and replicate results were above or below the action level agreed upon by the Klamath River BGA Workgroup¹ of 40,000 cells/ml (see figure 2 below). Therefore, the quality of these results has met the primary sampling objective of notifying the public when the MSAE levels exceed the action level. It is expected that the higher the cell counts are the greater the variability in counts between replicates. It is possible that two replicates could fall on opposite sides of the action level threshold, but the probability is very low that a high count would occur by chance when the true population size is low or visa versa.



Figure 2. Results for the replicate samples containing Microcystis aeruginosa, 2006.

QA blank samples also indicate that the phytoplankton speciation and enumeration results are acceptable. None of the phytoplankton rinsate or true blank samples contained MSAE or *Aphanizomenon flos-aquae* (APF9). Rinsate blank sample results contained very few species of algae in May (4), June (1) and July (2). True blank samples collected in August, September and October contained no species of algae. Therefore, it is most likely that the rinsate blank results from May through July were a product of cross contamination of the sampling equipment and not cross contamination at the laboratory because no species were found in the true blank samples. It is not believed that cross contamination between sites influences algal speciation and enumeration results because the stream sample will overwhelm any minute presence of algal species that could be

¹ It should be noted that while Siskiyou County is a participant in the Klamath River BGA Workgroup they have not endorsed the reporting limit of 8 ug/L. However, Humboldt County Public Health Department, Del Norte County Public Health Department and CA Department of Health Services have endorsed the action level which is consistent with the State of Oregon's action level.

present after the churn is rinsed three times with distilled water and with stream water at the new sampling site.

Microcystin

QA replicate samples indicate that the toxin results are acceptable. Not all of the toxin replicate samples contained total microcystin. One replicate sample event in the Klamath River on 8/23/06 that was analyzed using ELISA technology did contain total microcystin, that RPD was 28.1%. This RPD slightly exceeds 25%. However, both results for the 8/23/06 sample event were below the posting action level agreed upon by the Klamath River BGA Workgroup of 8 μ g/L. Replicate samples collected on 9/20/06 using ELISA technology were both >1 ug/L and no RPD was calculated. Therefore, the quality of these results has met the primary sampling objective of notifying the public when microcystin exceeds the action level.

Blank QA sample results indicate reason for concern when considering the precision of low levels of microcystin results reported by Wright State using the ELISA method. Two true blank QA samples that were submitted blind to Wright State were reported as containing low levels of microcystin (see Table 1, below). The blank sample that was submitted to Wright State on 8/23/06 was greater than 2 times the lab's stated reporting limit of 0.00175 μ g/L. All four tributary sites tested positive for presence of microcystin and were below 10 times the blank concentration of 0.02 μ g/L. Only one out of ten Klamath River sampling sites tested positive for presence of microcystin that was below 0.2 μ g/L, the remaining nine sites were greater than 2.0 μ g/L. These low level toxin results reported for the tributaries are of concern because no samples collected over two years of sampling these streams have contained the presence of toxigenic cynobacteria.

The blank sample that was submitted to Wright State on 9/20/06 was less than 2 X the lab's stated reporting limit of 0.00175 μ g/L. No samples were collected in tributary sampling sites. All of the mainstem sampling sites tested positive for low levels of microcystin ranging from 0.02 to 0.21 μ g/L. While these low levels of microcystin are plausible, it is difficult to say if these results are true or are an artifact of the matrix interferences associated with the ELISA method.

Parameter	Date sampled	Sample Crew	Blank Sample Result (ppb)	Reporting Limit (ppb)	2 X Reporting Limit (ppb)	Blank < 2x Reporting Limit (Pass/Fail)	Laboratory	10 X Blank concentration
total microcystin	8/23/2006	Karuk	0.02	0.00175	0.0035	FAIL	Wright State Lab	0.2
total microcystin	9/20/2006	YTEP	0.002	0.00175	0.0035	PASS	Wright State Lab	0.02

Table 1. Blank QA sample results for analysis of water samples for total microcystin or microcystin variants in the Klamath River, 2006.

As stated by Wright State in their data reports "The limit of quantification (LOQ) for microcystin on the ELISA plate is 0.175 μ g/L. However, because water samples were concentrated 100 X during preparative steps, the levels of microcystin in the sample itself can be reported down to 0.00175 μ g/L (LOQ/concentration factor). The limit of detection (LOD) for microcystin on the ELISA plate is 0.147 μ g/L. Again, because samples were concentrated 100 X, it is possible to detect microcystin in the sample itself as low as 0.00147 μ g/mL (LOD/concentration factor)."

However, Wright State has flagged low level results with the following disclaimer "Concentration increases matrix interferences and may lead to a reduction of assay sensitivity. Microcystin levels of 0.00175 μ g/L can be calculated and are sometimes reported. However, matrix interferences may reduce the degree of accuracy possible and limit a reliable estimate at such low values."

YTEP is not confident with the equation that Wright State has employed in determining their limit of quantification, which is interpreted as their reporting limit. YTEP feels that the ELISA method is useful as an initial screening tool to determine if microcystin levels in the Klamath River are above or below a posting action level of 8 μ g/L. It may not, however, be appropriate for a more quantitative analysis and regression of toxin fluxes and other variables. Therefore, all results below 1 μ g/L are reported as non-detect or <1 μ g/L in this report to include a margin of error that can be associated with the ELISA method at low levels.

This decision was also based on concurrence from multiple labs that have experience in analyzing water samples for presence of total microcystin or microcystin variants. Discrepancies among sample data can occur from sample preservation and the lab's preparation of the sample before it is analyzed. For example, Wright State University lab personnel communicated with the Klamath River BGA Workgroup that it prepared samples differently depending on the level of algae present. Furthermore, Elizabeth Tor from UC Davis Animal Health Lab indicated that she measured different levels of microcystin variants when she prepared samples using different freezing times and cell lysing techniques such as sonication. Andrew Lincoff from USEPA Region IX Laboratory in Richmond, CA has also hypothesized that microcystin may be lost by adhesion to plastics and may be increased by samples remaining frozen for a longer period of time before analysis occurs. Plastic containers are commonly used by field sampling crews to collect and store samples, mainly because they can resist breakage during freezing and transport of the sample water.

V. <u>Results:</u> <u>Phytoplankton</u>

-1 alige T_{i} = 1 invertibility of the warde saturation of the first of the first of the first of the first of the start -1 of the T_{i}	Table 2.	Phytoplankton	results for w	ater samples c	ollected in the	Klamath River and	mouths of Major	Tributaries May	- October 2006
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Site		Total	Total	1						•)		2		APF9	MSAE
ID	Date	Density	Biovolume	Sp1	Sp1%	Sp2	Sp2%	Sp3	Sp3%	Sp4	Sp4%	Sp5	Sp5%	#Spp	cells/ml	cells/ml
IG	5/23/2006	574	176,348	STAM	10.1	CXER	10.1	RDMN	10.1	STHN	5.8	NZFR	5.8	31	0	0
SV	5/23/2006	1,420	503,309	NZDS	18.8	ACMN	17.5	NVCV	6.3	DTTN	6.3	NZFR	5.0	27	0	0
OR	5/23/2006	1,012	359,736	NZDS	22.9	NZPC	11.4	ACMN	7.1	NZFR	4.3	NVCV	4.3	28	0	0
WE	5/23/2006	693	316,093	NZDS	16.4	DTTN	9.0	ACMN	7.5	GFAN	7.5	STHN	6.0	29	0	0
KBW	5/23/2006	834	270,313	NZDS	21.3	ACMN	16.4	DTTN	13.1	NVCV	6.6	GFAN	4.9	23	0	0
TG	5/23/2006	676	321,913	NZDS	12.7	ACMN	9.9	DTVL	7.0	NZFR	5.6	DTTN	5.6	30	0	0
SH	5/23/2006	2,041	599,783	COPC	25.6	ACMN	9.3	GFAN	9.3	NVCV	8.1	ACLC	7.0	28	0	0
SC	5/23/2006	337	120,438	ACMN	28.3	GFAN	10.9	DTTN	8.7	NVCR	4.3	CMAF	4.3	20	0	0
SA	5/23/2006	116	60,439	ACMN	16.7	GFAN	16.7	DTVL	8.3	NZDS	8.3	HNAR	8.3	15	0	0
TR	5/23/2006	844	377,551	DTTN	21.8	ACMN	10.9	GFAN	9.1	HNAR	7.3	NZDS	5.5	23	0	0
IG	6/20/2006	1,591	1,671,411	MLGR	38.8	STHN	20.4	STBN	12.6	RDMN	8.7	CXER	5.8	14	0	0
SV	6/20/2006	2,045	1,046,014	MLGR	15.9	STHN	9.3	ACMN	9.3	NVCV	6.5	COPC	6.5	28	0	0
OR	6/20/2006	1,966	979,200	STHN	13.8	STBN	11.9	NZDS	10.1	MLGR	9.2	ACMN	7.3	31	0	0
WE	6/20/2006	1,679	742,553	STBN	25.5	STHN	13.7	NZDS	10.8	ACMN	5.9	MLGR	3.9	32	0	0
KBW	6/20/2006	1,522	618,393	NZDS	16.9	STHN	12.0	DTTN	9.6	ACMN	4.8	NZFR	4.8	32	0	0
TG	6/20/2006	1,223	616,339	STHN	18.9	STBN	12.2	NZDS	10.0	DTTN	7.8	ACMN	5.6	26	0	0
SH	6/20/2006	978	321,291	COPC	43.1	NVCV	5.6	RHCU	5.6	NVML	4.2	NZFR	4.2	26	0	0
SC	6/20/2006	1,428	517,789	ACMN	50.5	CMMN	10.3	CMAF	10.3	CMSN	4.7	GFOM	2.8	20	0	0
SA	6/20/2006	163	58,871	ACMN	31.8	DTTN	18.2	GFAN	11.4	FRVA	9.1	DTHM	6.8	13	0	0
TR	6/20/2006	355	154,737	DTTN	50.0	ACMN	8.0	COPC	6.0	GFVT	4.0	NZDS	4.0	18	0	0
IG	7/18/2006	363	151,902	RDMN	29.2	MSAE	19.4	APF9	15.3	CXER	11.1	KMXX	8.3	12	832	7,063
SV	7/18/2006	1,246	255,370	ACMN	15.5	NZIN	13.6	NZPL	11.8	NZFR	10.9	COPC	8.2	25	0	0
OR	7/18/2006	1,406	442,670	NZPL	24.5	COPC	10.4	NZFR	7.5	NVCV	7.5	CMAF	6.6	27	0	0
WE	7/18/2006	1,437	423,894	NZPL	18.3	RDMN	12.8	COPC	11.9	KMXX	9.2	CMAF	8.3	24	0	0
KBW	7/18/2006	1,413	674,916	DTTN	19.2	NZPL	16.2	CMAF	10.1	KMXX	6.1	ACMN	6.1	30	0	0
ТG	7/18/2006	1,106	341,469	NZPL	24.4	DTTN	15.4	CCXX	6.4	STBN	6.4	AKFL	3.8	30	0	0
LES	7/18/2006	1,138	351,631	NZPL	30.0	DTTN	16.9	STBN	10.0	CMAF	4.6	CCXX	3.8	27	0	0
SH	7/18/2006	560	204,750	COPC	62.4	NZFR	7.1	GFAN	4.7	RHCU	4.7	CXER	3.5	15	0	0
SC	7/18/2006	2,451	661,931	ACMN	68.0	CMAF	8.8	COPC	5.6	CMSN	4.8	CMMN	4.0	13	0	0
SA	7/18/2006	252	212,101	DTTN	28.7	EPSX	27.8	ACMN	12.0	GFAN	6.5	GFSB	4.6	20	0	0
TR	7/18/2006	786	433,331	DTTN	73.6	EPSX	8.5	SNUL	3.8	ACMN	0.9	FRLP	0.9	18	0	0

APF9 = Aphanizomenon flos-aquaeMSAE = Microcystis aeruginosa

Site	× /	Total	Total		1							<i>.</i>			APF9	MSAE
ID	Date	Density	Biovolume	Sp1	Sp1%	Sp2	Sp2%	Sp3	Sp3%	Sp4	Sp4%	Sp5	Sp5%	#Spp	cells/ml	cells/ml
IG	7/26/2006	1,429	1,299,790	APF9	55.6	MSAE	24.4	NZPL	8.1	RDMN	3.7	COPC	2.2	12	14,292	35,985
SV	7/26/2006	1,295	391,942	COPC	27.7	NZFR	12.5	ACMN	8.0	ACLN	6.3	NVCV	6.3	22	0	0
OR	7/26/2006	1,604	596,741	NZPL	17.3	COPC	16.5	NZFR	15.7	RHCU	6.3	CMAF	5.5	24	0	0
WE	7/26/2006	2,301	976,574	NZPL	29.7	COPC	9.9	CMAF	8.9	AKFL	7.9	NZFR	6.9	30	0	0
BC	7/26/2006	1,817	653,402	NZPL	26.7	SLMN	13.8	CMAF	6.0	NZFR	6.0	EPSX	5.2	28	0	0
TG	7/26/2006	1,762	1,220,325	NZPL	31.0	DTTN	8.0	AKFL	7.0	COPC	6.0	CCMG	6.0	30	0	0
LES	7/26/2006	1,240	593,391	NZPL	25.3	CMAF	14.1	AKFL	9.1	DTTN	6.1	NVGR	4.0	26	0	0
KBL	8/7/2006	10,874	18,609,262	APF9	96.8	RDMN	0.9	COPC	0.5	AKFL	0.5	ACMN	0.5	7	294,653	0
KBK	8/7/2006	2,105	2,016,150	APF9	62.9	CXER	12.1	MSAE	9.3	RDMN	4.3	NZFR	1.4	16	26,459	21,107
KBB	8/7/2006	1,394	722,979	APF9	15.2	MSAE	10.9	NZPL	7.6	RDMN	6.5	COPC	6.5	31	2,546	24,247
IG	8/7/2006	2,202	2,009,933	APF9	68.5	NZPL	16.9	MSAE	10.5	CXER	0.8	NZPL	0.8	8	27,167	24,929
WE	8/7/2006	3,937	2,032,367	APF9	27.2	NZPL	13.6	SCQD	8.7	MSAE	7.8	COPC	7.8	22	12,842	30,576
TG	8/7/2006	2,192	940,617	NZPL	18.1	SCQD	14.3	EPSX	13.3	MSAE	9.5	RDMN	6.7	27	209	4,802
LES	8/7/2006	964	301,900	NZPL	26.0	RDMN	17.0	AKFL	13.0	SCQD	9.0	EPSX	7.0	27	96	289
KBL	8/23/2006	14,401	24,823,911	APF9	96.1	MKXX	1.8	MSAE	0.7	RDMN	0.7	AKFL	0.4	6	387,450	41,000
KBK	8/23/2006	5,733	5,924,189	APF9	77.3	CXER	7.2	STHN	6.1	AKFL	2.2	RDMN	1.7	12	88,680	665
KBB	8/23/2006	1,279	653,400	APF9	30.4	NZPL	8.9	NZAM	7.6	NZFR	3.8	GFAN	3.8	29	5,828	4,856
IG	8/23/2006	1,176	738,050	APF9	33.1	NZPL	26.2	MSAE	16.6	COPC	6.9	NZAM	4.8	13	5,451	28,423
SV	8/23/2006	1,844	934,024	COPC	29.2	MSAE	20	NZPL	13.8	NVCR	4.6	ACMN	3.8	25	142	41,299
OR	8/23/2006	1,821	898,256	COPC	23	MSAE	17.5	NZPL	15.9	RHCU	4.8	CMAF	4.8	25	0	31,801
WE	8/23/2006	1,720	674,350	MSAE	18.6	COPC	16.1	NZPL	15.3	NZFR	7.6	SCQD	5.9	26	0	32,069
KBW	8/23/2006	1,447	671,512	NZPL	18.4	COPC	17.5	MSAE	10.7	EPSX	9.7	SCAC	5.8	27	0	15,455
TG	8/23/2006	2,374	1,408,287	EPSX	22.5	NZPL	14.2	COPC	10.8	MSAE	9.2	SCQD	8.3	29	396	21,759
LES	8/23/2006	608	365,488	NZPL	13.4	EPSX	12.2	SCQD	11.0	COPC	9.8	NZAC	6.1	29	223	1,246
SH	8/23/2006	371	136,591	COPC	28.0	RHCU	16.0	EPSX	9.3	NZFR	8.0	GFAN	6.7	22	0	0
SC	8/23/2006	4,165	3,468,910	CMAF	30.8	SCQD	23.3	ACLN	12.0	SNUL	7.5	ACMN	5.3	17	0	0
SA	8/23/2006	218	71,171	COPC	25.4	GFAN	22.4	ACMN	20.9	GFSB	4.5	NVCV	4.5	15	0	0
TR	8/23/2006	544	280,841	COPC	6.5	CMAF	6.5	ACMN	5.4	DTTN	4.3	EPSX	4.3	18	0	0

Table 2 (contd.) Phytoplankton results for water samples collected in the Klamath River and mouths of major tributaries May - October 2006.

APF9 = Aphanizomenon flos-aquae MSAE = Microcystis aeruginosa

Site	× /	Total	Total		1							<i>.</i>			APF9	MSAE
ID	Date	Density	Biovolume	Sp1	Sp1%	Sp2	Sp2%	Sp3	Sp3%	Sp4	Sp4%	Sp5	Sp5%	#Spp	cells/ml	cells/ml
KBL	9/6/2006	50,882	76,659,881	APF9	99.4	NVPO	0.3	NZPL	0.3					3	1,214,231	0
KBK	9/6/2006	4,817	2,973,601	CXER	32.7	RDMN	17.3	APF9	15.5	AKFL	9.1	SCQD	6.4	16	29,030	0
KBB	9/6/2006	2,784	1,133,299	CXER	30.6	APF9	17.4	RDMN	9.9	STHN	8.3	SCQD	6.6	22	5,799	0
IG	9/6/2006	472	208,702	RDMN	31.7	CXER	25.7	COPC	11.9	MSAE	7.9	GFSB	3.0	18	0	3,735
SV	9/6/2006	1,108	623,106	COPC	46.6	MSAE	6.9	NZAM	6.9	NVCR	5.2	SCQD	3.4	25	0	9,555
OR	9/6/2006	825	466,168	COPC	25.2	EPSX	20.3	NZFR	5.7	SCQD	4.9	MSAE	4.1	28	0	3,356
WE	9/6/2006	1,033	724,865	EPSX	24.1	COPC	10.7	SCQD	10.7	RDMN	6.3	NZPL	5.4	33	0	1,845
KBW	9/6/2006	805	363,770	COPC	17.2	EPSX	16.1	RDMN	12.9	RHCU	7.5	SCQD	6.5	25	0	0
TG	9/6/2006	1,797	856,405	EPSX	13.3	RDMN	11.2	KMXX	9.2	SCQD	8.2	COPC	6.1	28	0	0
LES	9/6/2006	707	194,783	RDMN	30.9	KMXX	13.8	EPSX	9.6	DTTN	8.5	AKFL	6.4	19	0	0
KBL	9/20/2006	29,403	39,938,924	APF9	97.7	RDMN	1.5	CXER	0.4	STAM	0.4			4	632,284	0
KBK	9/20/2006	4,961	1,779,810	RDMN	32.2	CXER	23.1	CCMG	14.0	APF9	9.9	STHN	5.8	18	10,332	1,230
KBB	9/20/2006	550	183,337	CXER	8.8	APF9	7.4	CCMG	7.4	COPC	5.9	FRCV	5.9	29	566	0
IG	9/20/2006	498	2,389,673	FRCR	25	RDMN	22.0	CXER	12.0	MSAE	8.0	NZAM	6.0	23	0	3,982
SV	9/20/2006	723	428,501	COPC	35.1	SCQD	13.2	RDMN	9.6	NVCR	6.1	CMAF	5.3	24	0	190
OR	9/20/2006	1,007	648,054	EPSX	26.6	COPC	14.7	DTTN	10.1	SCQD	5.5	GFSB	5.5	30	0	0
WE	9/20/2006	956	654,488	EPSX	17.7	COPC	16.7	DTTN	13.5	SCQD	8.3	SNMZ	6.3	23	0	0
KBW	9/20/2006	720	485,405	EPSX	27.4	COPC	21.1	DTTN	14.7	CMAF	5.3	ACMN	3.2	23	0	0
TG	9/20/2006	1,563	1,035,460	DTTN	26.5	EPSX	15.9	COPC	8.8	SCQD	7.1	SNUL	6.2	26	0	0
LES	9/20/2006	363	247,650	DTTN	28.2	EPSX	28.2	COPC	7.8	SNUL	6.8	ACMN	3.9	21	0	35
SH	9/20/2006	524	205,158	COPC	38.0	RHCU	14.0	EPSX	6.0	NVCV	6.0	GFAN	6.0	26	0	0
SC	9/20/2006	2191	797,615	SCQD	61.8	ACMN	18.6	CMAF	8.8	NVCR	2.0	SNUL	2.0	12	0	0
SA	9/20/2006	121	55,189	COPC	21.2	ACMN	15.2	GFAN	12.1	NVXX	6.1	EPSX	6.1	16	0	0
TR	9/20/2006	318	193,560	COPC	33.3	DTTN	20.7	EPSX	10.3	CMAF	8.0	ACMN	6.9	19	0	0
KBL	10/4/2006	21.258	22.643.958	APF9	99.4	RDMN	0.6							2	359,391	0
KBK	10/4/2006	5.125	2.510.396	APF9	37.5	RDMN	22.5	STHN	15.0	CXER	13.3	KFXX	3.3	11	30,750	0
KBB	10/4/2006	1.311	225.847	RDMN	37.3	STHN	19.6	CXER	7.8	CCMG	6.9	AKFL	4.9	21	642	0
IG	10/4/2006	551	964.740	RDMN	25.5	NVCV	11.3	FRCR	11.3	MLVR	5.7	NZAM	4.7	27	0	0
SV	10/4/2006	1.658	1.775.768	COPC	26.0	SNUL	10.0	RDMN	9.0	RHCU	9.0	NZFR	8.0	20	0	0
OR	10/4/2006	1,791	1,589,543	EPSX	30.6	COPC	18.5	DTTN	9.3	SCQD	4.6	FRCR	4.6	28	0	0
WE	10/4/2006	1,648	1,231,461	EPSX	22.8	DTTN	18.4	RDMN	10.5	SCQD	9.6	COPC	7.9	24	0	0
KBW	10/4/2006	1,171	748,107	EPSX	24.1	DTTN	20.4	COPC	9.3	CMSN	5.6	ACMN	0.7	21	0	0
ΤG	10/4/2006	1,965	991,073	DTTN	22.6	EPSX	13.9	SCQD	11.3	COPC	9.6	RDMN	7.8	25	0	0
LES	10/4/2006	532	134,719	RDMN	47.2	DTTN	19.4	EPSX	6.5	COPC	6.5	AKFL	2.8	21	0	0

Table 2 (contd.) Phytoplankton results for water samples collected in the Klamath River and mouths of major tributaries May - October 2006.

APF9 = Aphanizomenon flos-aquae MSAE = Microcystis aeruginosa

Site		Total	Total												APF9	MSAE
ID	Date	Density	Biovolume	Sp1	Sp1%	Sp2	Sp2%	Sp3	Sp3%	Sp4	Sp4%	Sp5	Sp5%	#Spp	cells/ml	cells/ml
KBL	10/19/2006	13,066	12,937,996	APF9	91.4	RDMN	4.1	STAM	2.0	FRCN	1.0	NVXX	0.5	7	202,950	0
KBK	10/19/2006	1,915	1,275,326	APF9	55.6	RDMN	31.5	NZDS	5.6	NZPL	2.4	NZAM	1.6	9	19,183	0
KBB	10/19/2006	951	528,631	APF9	73.1	RDMN	0.4	FRCN	1.5	NZAM	0.7	SCQD	0.8	24	6,136	9,204
IG	10/18/2006	496	1,413,048	RDMN	25.7	FRCR	16.2	AKFL	16.2	NZAM	6.7	CXER	6.7	16	47	0
SV	10/18/2006	1,359	1,649,337	COPC	16.5	NZFR	14.3	NVCV	12.1	FRCR	8.8	NZAM	6.6	26	0	0
OR	10/18/2006	1,879	1,374,009	DTTN	16.0	NZFR	14.0	EPSX	13.0	COPC	11.0	SNUL	7.0	25	0	0
WE	10/18/2006	1,905	1,471,038	DTTN	25.5	SNUL	12.2	COPC	10.2	EPSX	8.2	NVCR	6.1	28	0	0
KBW	10/18/2006	1,769	1,344,104	DTTN	30.0	EPSX	15.0	SNUL	14.0	COPC	8.0	NZFR	6.0	23	0	0
TG	10/18/2006	1,397	776,087	DTTN	28.7	EPSX	7.9	COPC	7.9	NZFR	7.9	SCQD	6.9	25	0	0
LES	10/18/2006	413	244,57	DTTN	42.1	RDMN	12.6	SNUL	7.4	COPC	7.4	EPSX	6.3	22	0	0
SH	10/18/2006	1,037	381,713	COPC	21.9	RHCU	11.4	NZFR	7.9	GFAN	7.9	NZDS	6.1	26	0	0
SC	10/18/2006	2,197	1,045,501	SCQD	57.0	ACMN	14.9	CMAF	7.9	SNUL	6.1	DTTN	3.5	12	0	0
SA	10/18/2006	338	125,977	DTTN	49.5	ACMN	12.4	COPC	7.2	NZPC	5.2	SNUL	3.1	22	0	0

Table 2 (contd.) Phytoplankton results for water samples collected in the Klamath River and mouths of major tributaries May - October 2006.

APF9 = Aphanizomenon flos-aquae MSAE = Microcystis aeruginosa



Figure 3. *Aphanizomenon flos-aquae* levels for water samples collected in the Klamath River from RM 254 to RM 224, August through October 2006.



Figure 4. *Aphanizomenon flos-aquae* levels for water samples collected in the Klamath River from RM 190 to RM 0.5, May through October 2006.



Figure 5. *Microcystis aeruginosa* levels for water samples collected in the Klamath River from RM 254 to RM 224, August through October 2006.



Figure 6. *Microcystis aeruginosa* levels for water samples collected in the Klamath River from RM 190 to RM 0.5, May through October 2006.

Microcystin

Table 3. Microcystin results for water samples collected in the Klamath River from RM 254 to RM 0.5, July to September 2006.

TOXINS						
Total Microcystin		Date				
units: µg/L	Site	7/26/2006	8/7/2006	8/23/2006	9/6/2006	9/20/2006
Wright State ELISA reporting limit: 1µg/L	KBL	DNS	3.3	4.1	2.7	<1
	KBK	DNS	2.2	<1	<1	<1
	KBB	DNS	<1	<1	<1	<1
	IG	3.4	3.0	9.2	<1	<1
	SV	DNS	6.7	7.3	<1	<1
	OR	<1	4.1	4.6	<1	<1
	WE	<1	4.8	7.1	<1	<1
	KBW	DNS	DNS	2.8	<1	<1
	BC	<1	DNS	DNS	DNS	DNS
	TG	<1	1.3	2.7	<1	<1
	LES	<1	<1	2.0	<1	<1
	SH	DNS	DNS	<1	DNS	DNS
	SC	DNS	DNS	<1	DNS	DNS
	SA	DNS	DNS	<1	DNS	DNS
	TR	DNS	DNS	<1	DNS	DNS

DNS = Did Not Sample

* Samples analysis from July to September was performed by Wright State University using ELISA to measure total microcystin



Figure 7. Microcystin levels for water samples collected in the Klamath River from RM 254 to RM 224, August through September 2006.



Figure 8. Microcystin levels for water samples collected in the Klamath River from RM 190 to RM 0.5, July to September 2006.

VI. Discussion:

Phytoplankton

<u>Aphanizomenon flos-aquae</u>

River Reach RM 254 to RM 224

Aphanizomenon flos-aquae a nitrogen fixing blue-green algae was present in 18 of 18 samples collected in the Klamath River reach downstream of Link Dam from RM 254 to RM 224 (KBL, KBK,KBB). Summary information of all algae species identified and enumerated in this river reach is presented in Appendix A. *Aphanizomenon flos-aquae* significantly dominated all of the samples collected at the above mentioned sites with an average percent density of 53.8 per sample. The highest density of *Aphanizomenon flos-aquae* occurred at the Klamath River Below Link Dam (KBL) on September 6, 2006 and was measured at 1,214,231 cells/ml.

River Reach RM 190 to RM 0.5

Aphanizomenon flos-aquae was present in 13 of 69 samples collected in the Klamath River reach downstream of Iron Gate Dam from RM 190 to RM 0.5 (IG,SV,OR,WE,KBW,BC,TG,LES). Summary information of all algae species identified and enumerated in this river reach is presented in Appendix A. Aphanizomenon flosaquae ranked as the ninth most dominant species when looking at the average percent density (3.1%). Aphanizomenon flos-aquae was detected in this river reach beginning on July 18th, 2006 at the IG sampling site. Aphanizomenon flos-aquae continued to be present in the Klamath River at multiple sampling sites downstream of Iron Gate Dam to the Lower Klamath River Estuary until September 6th, 2006. Aphanizomenon flos-aquae was then detected again at very low levels on October 18th, 2006 at the IG sampling site. The highest density of Aphanizomenon flos-aquae occurred in this river reach at the IG sampling site on August 7th, 2006 and was measured at 27,167 cells/ml.

Mouths of Major Tributaries

Aphanizomenon flos-aquae was not present in the phytoplankton samples collected in the four major tributaries that were sampled from May through October, 2006. Summary information of all algae species identified and enumerated in the mouths of major tributaries is presented in Appendix A.

<u>Microcystis aeruginosa</u>

River Reach RM 254 to RM 224

Microcystis aeruginosa was present in 6 of 18 samples collected in the Klamath River reach downstream of Link Dam from RM 254 to RM 224 (KBL, KBK,KBB). Summary information of all algae species identified and enumerated in this river reach is presented in Appendix A. *Microcystis aeruginosa* ranked as the eighth most dominant species when looking at the average percent density (1.5%) at the above mentioned sites. The highest density of *Microcystis aeruginosa* occurred at the Klamath River Below Link Dam (KBL) sampling site on August 23rd, 2006 and was measured at 41,000 cells/ml.

This was the single event in which the cell density exceeded the Klamath River Blue-Green Algae Workgroup posting action level of 40,000 cells/ml in this river reach.

River Reach RM 190 to RM 0.5

Microcystis aeruginosa was present in 20 of 69 samples collected in the Klamath River reach downstream of Iron Gate Dam from RM 190 to RM 0.5 (IG,SV,OR,WE,KBW,BC,TG,LES). Summary information of all algae species identified

and enumerated in this river reach is presented in Appendix A. *Microcystis aeruginosa* ranked as the tenth most dominant species when looking at the average percent density (2.9%). *Microcystis aeruginosa* was detected in this river reach beginning on July 18th, 2006 at the IG sampling site. *Microcystis aeruginosa* was present in the Klamath River at multiple sampling sites downstream of Iron Gate Dam until September 20th, 2006. The highest density of *Microcystis aeruginosa* occurred at the SV sampling site on August 23rd, 2006 and was measured at 41,299 cells/ml. This was the single event in which the cell density exceeded the posting action level of 40,000 cells/ml in this river reach. *Microcystis aeruginosa* was not present at sampling sites in this river reach in October.

Mouths of Major Tributaries

Microcystis aeruginosa was not present in the phytoplankton samples collected in the four major tributaries that were sampled from May to October, 2006. Summary information of all algae species identified and enumerated in the mouths of major tributaries is presented in Appendix A.

Microcystin

These results indicate that *Microcystis aeruginosa* was present in the Klamath River downstream of Iron Gate Dame for over two months, with cell density and microcystin levels peaking at the end of August. The timing is of significance 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 fisherman.

River Reach RM 254 to RM 224

Microcystin was present in 4 of 12 samples collected in the Klamath River reach downstream of Link Dam from RM 254 to RM 224. Microcystin was present in at least one site from August 7th to September 6th 2006. The highest reported microcystin level in this river reach occurred at the KBL sampling site on August 23rd, 2006 and was measured at $4.1\mu g/L$.

River Reach RM 190 to RM 0.5

Microcystin was present in 13 of 34 samples collected in the Klamath River reach downstream of Iron Gate Dam from RM 190 to RM 0.5. Microcystin was first detected in this river reach beginning on July 26th at the IG sampling site. On August 23rd, 2006 microcystin was present in the Klamath River downstream of Iron Gate Dam to the Lower Klamath River Estuary. The highest reported microcystin level in this river reach occurred at the IG sampling site on August 23rd, 2006 and was measured at 9.2 μ g/L.

This was the single event in which microcystin exceeded the Klamath River Blue-Green Algae Workgroup posting action level of $8 \mu g/L$ in this river reach.

<u>Mouths of Major Tributaries</u> Microcystin was not present in the samples collected in the four major tributaries that were sampled from May to September, 2006.

Literature Cited

Kann, J. and Corum, S. 2005. *Microcystis aeruginosa* Trends in Copco and Iron Gate Reservoirs, Technical Memorandum prepared for the Karuk Tribe. March, 2006.

Appendix A

Table A-1. Combined Algae Species List for	Klamath River	Sites located	at River Mile	e 254 to 224	(KBL,
KBK and KBB) August to October, 2006.					

	18 samples total		
# Algae Species	Average % Den	# samples	Code
1 Aphanizomenon flos-aquae	53.8	18	APF9
2 Rhodomonas minuta	10.5	16	RDMN
3 Cryptomonas erosa	7.9	11	CXER
4 Stephanodiscus hantzschii	3.9	10	STHN
5 Cyclotella meneghiniana	2.0	9	CCMG
6 Ankistrodesmus falcatus	1.9	11	AKFL
7 Nitzschia amphibia	1.7	11	NZAM
8 Microcystis aeruginosa	1.5	6	MSAE
9 Scenedesmus quadricauda	1.4	11	SCQD
10 Nitzschia palea	1.3	6	NZPL
11 Cocconeis placentula	1.0	7	COPC
12 Selenastrum minutum	1.0	7	SLMN
13 Fragilaria construens venter	0.9	5	FRCV
14 Gomphonema angustatum	0.9	7	GFAN
15 Stephanodiscus astraea minutula	0.8	8	STAM
16 Chlamydomonas sp.	0.7	8	CHXX
17 Asterionella formosa	0.7	6	ASFO
18 Nitzschia frustulum	0.6	7	NZFR
19 Navicula cryptocephala veneta	0.6	4	NVCV
20 Fragilaria construens	0.4	3	FRCN
21 Nitzschia dissipata	0.4	3	NZDS
22 Fragilaria vaucheria	0.3	4	FRVA
23 Navicula minima	0.3	3	NVMN
24 Achnanthes minutissima	0.3	5	ACMN
25 Cymbella minuta	0.3	5	CMMN
26 Kephyrion sp.	0.2	2	KFXX
27 Navicula sp.	0.2	5	NVXX
28 Scenedesmus acuminatus	0.2	3	SCAC
29 Fragilaria capucina mesolepta	0.2	2	FRCM
30 Synedra ulna	0.2	3	SNUL
31 Nitzschia paleacea	0.2	4	NZPC
32 Melosira granulata	0.2	2	MLGR
33 Chromulina sp.	0.2	3	KMXX
34 Rhoicosphenia curvata	0.2	2	RHCU
35 Actinastrum hantzschii	0.1	2	ATHN
36 Amphora perpusilla	0.1	2	AFPR
37 Navicula cryptocephala	0.1	2	NVCR
38 Diatoma vulgare	0.1	1	DTVL
39 Navicula pupula	0.1	3	NVPP
40 Oocystis pusilla	0.1	2	OCPU
41 Navicula minuscula	0.1	1	NVML
42 Gomphonema ventricosum	0.1	1	GFVT
43 Cyclotella pseudostelligera	0.1	2	CCPS
44 Nitzschia acicularis	0.1	2	NZAC
45 Achnanthes linearis	0.1	2	ACLN

	18 samples total		
# Algae Species	Average % Den	# samples	Code
46 Nitzschia sp.	0.1	2	NZXX
47 Synedra cyclopum	0.1	1	SNCY
48 Achnanthes sp.	0.1	1	ACXX
49 Melosira varians	0.1	1	MLVR
50 Gomphonema olivaceum	0.1	1	GFOM
51 Nitzschia communis	0.1	1	NZCM
52 Navicula seminulum	0.1	1	NVSM
53 Achnanthes hauckiana	0.1	1	ACHK
54 Nitzschia capitellata	0.1	1	NZCP
55 Epithemia sorex	0.1	1	EPSX
56 Eudorina elegans	0.1	1	EDEL
57 Amphora coffeiformes	0.1	1	AFCF
58 Amphora ovalis	0.1	1	AFOV
59 Scenedesmus abundans	0.1	1	SCAB
60 Kephyrion littorale	0.1	1	KFLT
61 Synedra rumpens	0.1	1	SNRM
62 Scenedesmus denticulatus	0.1	1	SCDT
63 Navicula rhynchocephala	0.1	1	NVRH
64 Chrysococcus rufescens	0.1	1	CYRF
65 Stephanodiscus binderanus	0.0	1	STBN
66 Tetraedron sp.	0.0	1	TEXX
67 Sphaerocystis schroeteri	0.0	1	SFSR
68 Crucigenia quadrata	0.0	1	CGQD
69 Tetraedron minimum	0.0	1	TEMN
70 Cymbella affinis	0.0	1	CMAF
71 Neidium sp.	0.0	1	NDXX
72 Gomphonema subclavatum	0.0	1	GFSB
73 Caloneis sp.	0.0	1	CAXX
74 Nitzschia linearis	0.0	1	NZLN
75 Navicula protracta	0.0	1	NVPO

 Table A-1 (contd.). Combined Algae Species List for Klamath River Sites located at River Mile 254 to 224

 (KBL, KBK and KBB) August to October, 2006.

	· · · · ·	69 samples total		
#	Algae Species	Average % Den	# samples	Code
1	Cocconeis placentula	9.8	66	COPC
2	Rhodomonas minuta	8.4	55	RDMN
3	Diatoma tenue	7.3	45	DTTN
4	Nitzschia palea	7.3	45	NZPL
5	Epithemia sorex	6.9	42	EPSX
6	Nitzschia frustulum	3.7	59	NZFR
7	Scenedesmus quadricauda	3.3	49	SCQD
8	Achnanthes minutissima	3.2	52	ACMN
9	Aphanizomenon flos-aquae	3.1	13	APF9
10	Microcystis aeruginosa	2.9	20	MSAE
11	Nitzschia dissipata	2.7	33	NZDS
12	Cymbella affinis	2.4	50	CMAF
13	Ankistrodesmus falcatus	2.4	46	AKFL
14	Navicula cryptocephala veneta	2.1	50	NVCV
15	Synedra ulna	2.1	47	SNUL
16	Cryptomonas erosa	2.0	33	CXER
17	Stephanodiscus hantzschii	1.9	21	STHN
18	Rhoicosphenia curvata	1.9	51	RHCU
19	Navicula cryptocephala	1.5	43	NVCR
20	Stephanodiscus binderanus	1.4	15	STBN
21	Nitzschia amphibia	1.3	40	NZAM
22	Gomphonema angustatum	1.3	46	GFAN
23	Melosira granulata	1.3	16	MLGR
24	Fragilaria crotonensis	1.3	17	FRCR
25	Cymbella sinuata	1.1	40	CMSN
26	Diatoma vulgare	1.1	31	DTVL
27	Nitzschia paleacea	1.0	34	NZPC
28	Chromulina sp.	0.9	15	KMXX
29	Nitzschia innominata	0.8	28	NZIN
30	Selenastrum minutum	0.8	24	SLMN
31	Nitzschia communis	0.6	22	NZCM
32	Cyclotella meneghiniana	0.6	22	CCMG
33	Amphora perpusilla	0.5	29	AFPR
34	Achnanthes lanceolata	0.5	27	ACLC
35	Stephanodiscus astraea minutula	0.5	12	STAM
36	Gomphonema ventricosum	0.4	20	GFVT
37	Scenedesmus acuminatus	0.4	14	SCAC
38	Synedra mazamaensis	0.4	14	SNMZ
39	Navicula tripunctata	0.4	18	NVTP
40	Gomphonema subclavatum	0.4	18	GFSB
41	Nitzschia sp.	0.4	21	NZXX
42	Nitzschia acicularis	0.4	14	NZAC
43	Navicula minuscula	0.4	13	NVML
44	Achnanthes linearis	0.3	12	ACLN
45	Cyclotella stelligera	0.3	9	CCST

Table A-2. Combined Algae Species List for Klamath River Sites located at River Mile 190 to 0.5 (IG,SV,OR,WE, KBW, BC, TG, LES) May to October, 2006.

	69	samples total	
# Algae Species	Average % Den	# samples	Code
46 Gomphonema olivaceum	0.3	14	GFOM
47 Melosira varians	0.3	13	MLVR
48 Chlamydomonas sp.	0.3	15	CHXX
49 Cymbella minuta	0.3	14	CMMN
50 Fragilaria vaucheria	0.2	10	FRVA
51 Gomphoneis herculeana	0.2	13	GSHR
52 Scenedesmus denticulatus	0.2	12	SCDT
53 Navicula gregaria	0.2	6	NVGR
54 Navicula sp.	0.2	12	NVXX
55 Asterionella formosa	0.2	8	ASFO
56 Cyclotella sp.	0.2	3	CCXX
57 Cyclotella pseudostelligera	0.2	6	CCPS
58 Navicula decussis	0.2	7	NVDC
59 Melosira ambigua	0.1	6	MLAM
60 Fragilaria construens venter	0.1	9	FRCV
61 Kephyrion sp.	0.1	4	KFXX
62 Achnanthes hauckiana	0.1	7	ACHK
63 Nitzschia linearis	0.1	6	NZLN
64 Navicula minima	0.1	5	NVMN
65 Nitzschia volcanica	0.1	8	NZVL
66 Svnedra tenera	0.1	3	SNTN
67 Hannaea arcus	0.1	4	HNAR
68 Nitzschia fonticola	0.1	7	NZFT
69 Fragilaria capucina mesolepta	0.1	5	FRCM
70 Navicula viridula	0.1	3	NVVR
71 Glenodinium sp	0.1	5	GDXX
72 Diatoma hiemale mesodon	0.1	3	DTHM
73 Cryptomonas ovata	0.1	2	CXOV
74 Tetraedron minimum	0.1	4	TEMN
75 Navicula menisculus upsaliensis	0.1	5	NVMU
76 Cocconeis klamathensis	0.1	4	COKI
77 Synedra rumpens	0.1	2	SNRM
78 Scenedesmus abundans	0.1	4	SCAB
79 Gomphonema sp.	0.1	4	GFXX
80 Cymbella microcephala	0.1	3	CMMC
81 Mallomonas sp.	0.0	1	MMXX
82 Lagynion sp.	0.0	1	LGXX
83 Schroderia sp	0.0	1	SHXX
84 Nitzschia capitellata	0.0	2	NZCP
85 Stephanodiscus sp	0.0	1	STXX
86 Navicula graciloides	0.0	2	NVGC
87 Gomphonema clevei	0.0	3	GECI
88 Surirella linearis	0.0	2	SULN
89 Navicula protracta	0.0	2	NVPO
90 Coelastrum microporum	0.0	2	CLIMC
91 Fragilaria construens	0.0	2	FRCN
92 Synedra cyclopum	0.0	2	SNCY

Table A-2 (contd.). Combined Algae Species List for Klamath River Sites located at River Mile 190 to 0.5 (IG,SV,OR,WE, KBW, BC, TG, LES) May to October, 2006.

(10,5 /	, , , , , , , , , , , , , , , , , , ,	69 s	amples total	
#	Algae Species	Average % Den	# samples	Code
93	Cyclotella ocellata	0.0	2	CCOC
94	Synedra sp.	0.0	2	SNXX
95	Pediastrum tetras	0.0	2	PSTT
96	Navicula pupula	0.0	2	NVPP
97	Tetrastrum staurogeniaforme	0.0	1	TTST
98	Synedra socia	0.0	2	SNSC
99	Tetraedron regulare	0.0	2	TERG
100	Navicula mutica	0.0	2	NVMT
101	Melosira distans alpigena	0.0	1	MLDA
102	Fragilaria pinnata	0.0	1	FRPN
103	Cyclotella atomus	0.0	1	CCAT
104	Closteriopsis longissima	0.0	1	CBLG
105	Gomphonema gracile	0.0	1	GFGC
106	Oocystis pusilla	0.0	1	OCPU
107	Amphora ovalis	0.0	1	AFOV
108	Peridinium sp.	0.0	1	PRXX
109	Gomphonema truncatum	0.0	1	GFTR
110	Synedra parasitica	0.0	1	SNPR
111	Rhopalodia gibba	0.0	1	RPGB
112	Sphaerocystis schroeteri	0.0	1	SFSR
113	Eudorina elegans	0.0	1	EDEL
114	Synedra radians	0.0	1	SNRD
115	Melosira granulata angustissima	0.0	1	MLGA
116	Mougeotia sp.	0.0	1	MGXX
117	Gomphonema tenellum	0.0	1	GFTN
118	Gloeocystis sp.	0.0	1	GLXX
119	Nitzschia microcephala	0.0	1	NZMC
120	Unidentified flagellate	0.0	1	MXFG
121	Achnanthes sp.	0.0	1	ACXX
122	Navicula capitata	0.0	1	NVCP
123	Gyrosigma spencerii	0.0	1	GYSP
124	Tetraedron sp.	0.0	1	TEXX
125	Navicula anglica	0.0	1	NVAG
126	Pediastrum boryanum	0.0	1	PSBR
127	Gyrosigma sp.	0.0	1	GYXX
128	Caloneis sp.	0.0	1	CAXX

Table A-2 (contd.). Combined Algae Species List for Klamath River Sites located at River Mile 190 to 0.5 (IG,SV,OR,WE, KBW, BC, TG, LES) May to October, 2006.

	,	23	samples total	
#	Algae Species	Ave % Den	# samples	Code
1	Cocconeis placentula	16.8	20	COPC
2	Achnanthes minutissima	15.2	23	ACMN
3	Diatoma tenue	13.0	18	DTTN
4	Scenedesmus quadricauda	6.4	5	SCQD
5	Gomphonema angustatum	5.9	17	GFAN
6	Cymbella affinis	4.0	11	CMAF
7	Epithemia sorex	3.8	13	EPSX
8	Rhoicosphenia curvata	2.8	11	RHCU
9	Navicula cryptocephala veneta	2.3	14	NVCV
10	Nitzschia frustulum	2.0	12	NZER
11	Nitzschia paleacea	1.8	14	NZPC
12		1.6	12	
13	Syneora uina	1.4		SNUL
14	Cympella minuta	1.2	9	
10		1.2	10	
10	Cumballa aigusta	1.1	5	
10	Comphonoma subolavatum	1.0	9	CESE
10	Fragilaria vauchoria	1.0	9	
20	Achaanthas lingaris	0.9	5	
20	Comphonema olivaceum	0.9	9	GEOM
22	Diatoma vulgare	0.0	3 7	
23	Navicula cryptocephala	0.8	, 10	NVCB
24	Amphora perpusilla	0.0	9	AFPR
25	Navicula sp.	0.6	5	NVXX
26	Nitzschia sp.	0.6	8	NZXX
27	Navicula gregaria	0.6	8	NVGR
28	Diatoma hiemale mesodon	0.6	4	DTHM
29	Cryptomonas erosa	0.5	5	CXER
30	Navicula tripunctata	0.5	7	NVTP
31	Nitzschia acicularis	0.4	7	NZAC
32	Synedra rumpens	0.4	4	SNRM
33	Fragilaria construens venter	0.4	4	FRCV
34	Ulothrix sp.	0.3	3	ULXX
35	Nitzschia linearis	0.3	5	NZLN
36	Navicula minuscula	0.3	3	NVML
37	Gomphonema tenellum	0.3	5	GFTN
38	Ankistrodesmus falcatus	0.3	5	AKFL
39	Rhodomonas minuta	0.3	3	RDMN
40	Cyclotella meneghiniana	0.3	4	CCMG
41	Scenedesmus denticulatus	0.3	3	SCDT
42	Gomphonema ventricosum	0.3	3	GFVT
43	Nitzschia communis	0.3	3	NZCM
44	Scenedesmus abundans	0.2	4	SCAB
45	Cymbella microcephala	0.2	4	CMMC

Table A-3. Combined Algae Species List for Mouths of Major Tributary Sites (SH,SC,SA,TR) May to October, 2006.

•	23 9	samples total	
# Algae Species	Ave % Den	# samples	Code
46 Pinnularia sp.	0.2	2	PLXX
47 Nitzschia palea	0.2	5	NZPL
48 Navicula pupula	0.2	2	NVPP
49 Nitzschia recta	0.2	1	NZRC
50 Navicula decussis	0.2	3	NVDC
51 Synedra mazamaensis	0.2	3	SNMZ
52 Navicula minima	0.2	2	NVMN
53 Synedra radians	0.2	1	SNRD
54 Meridion circulare	0.2	1	MRCR
55 Navicula cascadensis	0.2	3	NVCS
56 Selenastrum minutum	0.2	3	SLMN
57 Epithemia turgida	0.1	1	EPTR
58 Nitzschia fonticola	0.1	2	NZFT
59 Caloneis sp.	0.1	2	CAXX
60 Denticula elegans	0.1	1	DNEL
61 Achnanthes lewisiana	0.1	1	ACLW
62 Achnanthes exigua	0.1	1	ACEX
63 Navicula seminulum hustedtii	0.1	1	NVSH
64 Scenedesmus acuminatus	0.1	1	SCAC
65 Navicula contenta biceps	0.1	1	NVCB
66 Fragilaria virescens	0.1	1	FRVR
67 Tabellaria fenestrata	0.1	1	TBFN
68 Synedra parasitica	0.1	1	SNPR
69 Nitzschia amphibia	0.1	2	NZAM
70 Gomphonema sp.	0.1	1	GFXX
71 Fragilaria construens	0.1	1	FRCN
72 Nitzschia capitellata	0.1	1	NZCP
73 Nitzschia volcanica	0.1	1	NZVL
74 Stephanodiscus hantzschii	0.1	1	STHN
75 Eunotia pectinalis	0.1	1	EUPC
76 Chodatella wratislawiensis	0.1	1	CDWR
77 Surirella ovata	0.1	1	SUOV
78 Glenodinium sp.	0.1	1	GDXX
79 Nitzschia microcephala	0.1	1	NZMC
80 Nitzschia innominata	0.0	1	NZIN
81 Neidium affine	0.0	1	NDAF
82 Navicula radiosa	0.0	1	NVRD
83 Amphipleura pellucida	0.0	1	AMPL
84 Sphaerocystis schroeteri	0.0	1	SFSR
85 Navicula mournei	0.0	1	NVMO
86 Rhopalodia musculus	0.0	1	RPMS
87 Melosira varians	0.0	1	MLVR
88 Fragilaria leptostauron	0.0	1	FRLP
89 Gomphoneis herculeana	0.0	1	GSHR

Table A-3 (contd.). Combined Algae Species List for Mouths of Major Tributary Sites (SH,SC,SA,TR) May to October, 2006.

·	23 samples total		
# Algae Species	Ave % Den	# samples	Code
90 Fragilaria capucina mesolepta	0.0	1	FRCM
91 Rhopalodia gibba	0.0	1	RPGB
92 Cymbella tumida	0.0	1	CMTM
93 Navicula rhynchocephala	0.0	1	NVRH
94 Scenedesmus sp.	0.0	1	SCXX
95 Chroococcus minimus	0.0	1	CKMN

Table A-3 (contd.). Combined Algae Species List for Mouths of Major Tributary Sites (SH,SC,SA,TR) May to October, 2006.

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 deionized (D.I.) 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 and Blank bottle sets

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

Blank sample bottles are utilized to assess accuracy of the analysis and verify that the sampling method or equipment does not influence the results. After collection of all other samples at the QA/QC site, the churn is rinsed three times with D.I. water before being filled with D.I. water. The blank bottle sets are collected in the same way as other samples, except using D.I. water in place of stream water. Blank samples are collected after all stream water samples are taken and act as a final rinse to decontaminate the churn.

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.

Bibliography

Bel-Art Products. Churn Sample Splitter Instructions, 37805 Series. Pequannock, NJ, 1993.

Eaton, Andrew D., Lenore S. Clesceri, and Arnold E. Greenberg., ed. Standard Methods for the Examination of Water and Wastewater. 19th Edition. Washington D.C., 1995.

Lurry,D.L. and C.M. Kolbe. Interagency field manual for the collection of Water Quality Data. USGS Publication, Open File Report 00-213.