



King County

Water and Land Resources Division

Environmental Laboratory

Department of Natural Resources and Parks

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TTY Relay: 711

April 27, 2017

Phuong Truong
West Point Treatment Plant
Fort Lawton, Discovery Park
1400 Utah Street
WTP-NR-0100
Seattle, WA 98199

Dear Phuong:

Enclosed please find our report on NPDES biomonitoring tests conducted with effluent collected on April 4-9, 2017 at the West Point Treatment Plant.

Detailed findings are in the "Results" section of this report. The following table shows a summary of the results:

Chronic Toxicity Tests

Test Organism	Growth IC25 ^a % Effluent	Difference in Growth from Control (Control vs ACEC ^b)
<i>Atherinops affinis</i>	>50 ^c	Not Significantly Different
<i>Mysidopsis bahia</i>	24.6	Not Significantly Different

^a Concentration of effluent inhibiting growth by 25%

^b Acute Critical Effluent Concentration (3.6% effluent)

^c Based on the surviving number of topmelt; IC25 = 55.7 % effluent based on the initial number of topmelt

If you would like additional information, please call Francis Sweeney at 477-7117.

Sincerely,

Gary Yoshida
King County Environmental Laboratory

Enclosures

cc: Jeff Lafer: KSC-NR-0505

Fritz Grothkopp: LAB-NR-0100

**NATIONAL POLLUTANT DISCHARGE ELIMINATION SYSTEM
BIOLOGICAL MONITORING REPORT FOR THE
WEST POINT TREATMENT PLANT
FORT LAWTON, DISCOVERY PARK
1400 UTAH STREET WEST
SEATTLE, WASHINGTON 98199**

**PERMIT #WA0029181
FIRST BIENNIAL TEST 2017**

**KING COUNTY DEPARTMENT OF NATURAL RESOURCES AND PARKS
WATER AND LAND RESOURCES DIVISION
ENVIRONMENTAL LABORATORY SECTION
322 WEST EWING STREET
SEATTLE, WA 98119**

Test Numbers: 8274, 8275

Test Date: April 5, 2017

Report Date: April 27, 2017

METHODS

Sample

Three samples of secondary effluent from the West Point Treatment Plant were collected on ice by time-paced compositing over 24-hour periods on Days 0, 2 and 4 of testing. The samples were delivered to the King County Environmental Laboratory (KCEL) on Days 0, 2 and 4 in one or two (Day 4) 20-L glass jars containing approximately 17 – 18.5 liters each. Samples from the two jars (Day 4) were combined and mixed for use in testing. Due to a malfunction of one of the samplers on Day 2 (resulted in insufficient sample volume to last until Day 5) the sample normally brought in on Day 5 was brought in on Day 4. The chronic tests were initiated within 6.0 hours after sampling on Day 0. The unused samples were stored in the dark in a 4 ± 2°C refrigerator.

Day/Time of Collection		Day 0	Day 2	Day 4
		4-4-17 / 0858h to 4-5-17 / 0858h	4-6-17 / 0735h to 4-7-17 / 0715h	4-8-17 / 0841h to 4-9-17 / 0841h
Delivered to KCEL		4-5-17 / 1000h	4-7-17 / 0818h	4-9-17 / 0930h
Container		(1) 5-gal. glass carboy	(1) 5-gal. glass carboy	(2) 5-gal. glass carboy
Vol. (L):		18.5	17	17 / 17.5
At West Point Plant	pH	7.07	6.85	6.84
	Temp. °C	3.86	2.24	2.37
	TRC, mg/L*	< 0.01	< 0.01	< 0.01
At KCEL	pH**	7.27	6.98	6.91 / 6.97
	Temp. °C	--	2.7	1.9 / 2.1
	DO, mg/L**	9.5	9.6	8.7 / 8.8

*Measurement by field kit

** Before compositing (Day 4)

Chemical characteristics of the samples are listed below:

Parameter	Day 0	Day 2	Day 4	Units
Total NH ₃ -N	21.0	14.6	9.9	mg/L
Conductivity	688	540	727	µmhos/cm
Total Alkalinity	140	95	83	mg/L as CaCO ₃
Total Hardness	101	90	115	"

CONTROL WATER

The control (dilution) water used for the chronic tests with *A. affinis* and *M. bahia* was artificial seawater prepared by adding 35.7 g of Hawaiian Marine Mix (HMM) brand salts and 0.20 g of NaHCO₃ per L of Millipore Super Q deionized water (DW) and mixing until dissolved to obtain a salinity of 30‰. The artificial seawater was then aerated for ≥ 2 hours and filtered to 0.45 µm before use.

CHRONIC TESTS

Topsmelt - *Atherinops affinis*

The topsmelt chronic toxicity test (#8274; LIMS sample #L67475-2) was conducted as outlined in Chapman *et al.* (1995). The larvae were received from Aquatic Biosystems as 9 days old. Upon receipt the temperature was 17.2 °C, with salinity of 28-30 ppt. The larvae were placed into four 1.5-L crystallizing dishes and acclimated for 24 hours at 20°C in an environmental chamber with light aeration. During acclimation, the larvae were fed 3-4 mL

live *Artemia* nauplii per dish 2 times a day. Following acclimation, the larvae were loaded directly into the test chambers with a nylon screen. At test initiation the topsmelt larvae were 10 days old.

The effluent sample was warmed to approximately 20°C and the salinity adjusted to 30 ppt by adding 35.7 g of HMM salts and 0.20 g of NaHCO₃ per liter of sample, after which the sample was diluted with HMM artificial seawater to the concentrations listed below. Five replicates of five fish each were tested at each concentration, including the HMM-only control. Test chambers were 600-mL beakers containing 200 mL of test solution.

Assignment of the larvae to the test chambers was random, as was placement of the test chambers in the environmental chamber. The test was incubated for 7 days at 20 ± 1.0°C on a 16:8 h light:dark cycle. Solutions were renewed daily (75%), and larvae were fed newly-hatched *Artemia* nauplii two times per day (1 drop per test chamber for each feeding).

Survival and water quality measurements were recorded every 24 hours at solution renewal and can be found on the photocopied pages from the laboratory notebook in the "Bench Sheets" section of this report. Temperature was measured daily by digital thermometer in one replicate of each concentration and in replicates at six shelf positions (4 outer corner + 2 center), as well as recorded at 15-minute intervals using an Onset Tidbit data logger placed in a beaker of water amongst the test beakers.

At the end of the test, surviving animals were inactivated in ice water, rinsed, placed into tared aluminum foil weigh pans (hand-made) and dried at 60°C for 15.5 hours. Each replicate (one rep across treatments) was processed by a single analyst. After the pans cooled in a desiccator, dry weight was measured to the nearest 0.01 mg to determine growth.

Test #	Start Date/ Time	End Date/ Time	Effluent Concentrations (%)	Larvae Age	# Reps/ Trtmt	# Orgs/ Rep
8274 (Topsmelt)	4-5-17/ 1500h	4-12-17/ 1505h	0, 0.55 ^a , 3.6 ^b , 12.5, 25, 50, 100	10 days	5	5

^aCCEC (Chronic Critical Effluent Concentration, mistakenly run at 0.55% effluent, should have been 0.53% effluent)

^bACEC (Acute Critical Effluent Concentration)

Mysid Shrimp - *Mysidopsis bahia*

The mysid shrimp chronic toxicity test (#8275; LIMS sample #L67475-3) was conducted as outlined in US EPA (2002). The mysid juveniles were received from Aquatic Biosystems as 7 days old and were 7 days old at test initiation. Upon receipt the temperature was 20.3 °C and the salinity was 32 ppt. The larvae were transferred to two 1.5-L crystallizing dishes and gradually brought to the test temperature of 26 ± 1°C in a water bath with light aeration. During acclimation, the larvae were fed 4 mL live *Artemia* nauplii per dish. Following acclimation, the larvae were loaded directly into the test chambers with a nylon screen.

The effluent sample was warmed to approximately 25-26°C and the salinity adjusted to 30 ppt by adding 35.7 g of HMM salts and 0.20 g of NaHCO₃ per liter of sample, after which the sample was diluted with HMM artificial seawater to the concentrations listed below. Eight replicates of five mysids each were tested at each concentration, including the HMM-only control. Test chambers were 400-mL beakers containing 250 mL of test solution.

Assignment of the larvae to the test chambers was random, as was placement of the test chambers in the environmental chamber. The test was incubated for 7 days at 26 ± 1.0°C on a 16:8 h light:dark cycle. Solutions were renewed daily (80%), and larvae were fed newly-hatched *Artemia* nauplii two times per day (2 drops per test chamber for each feeding).

Survival and water quality measurements were recorded every 24 hours at solution renewal and can be found on the photocopied pages from the laboratory notebook in the "Bench Sheets" section of this report. Temperature was measured daily by digital thermometer in one replicate of each concentration and in replicates at six positions (4

outer corner + 2 center), as well as recorded at 15-minute intervals using an Onset Tidbit data logger placed in a beaker of water amongst the test beakers.

At the end of the test, surviving animals were rinsed with iced tap water and dried at 60°C for 15.5 hours in tared aluminum weigh boats. Each replicate (one rep across treatments) was processed by a single analyst. After the pans cooled in a desiccator, dry weight was measured to the nearest 0.01 mg.

Test #	Start Date/ Time	End Date/ Time	Effluent Concentrations (%)	Mysid Age	# Reps/ Trtmt	# Orgs/ Rep
8275 (Mysid)	4-5-17/ 1435h	4-12-17/ 1320h	0, 0.55 ^a , 3.6 ^b , 12.5, 25, 50, 100	7 days	8	5 ^c

^aCCEC (Chronic Critical Effluent Concentration, mistakenly run at 0.55% effluent, should have been 0.53% effluent)

^bACEC

^c6 in rep H, 25% effluent

QUALITY CONTROL

Copper sulfate was used as a reference toxicant in chronic toxicity tests with topsmelt and *Mysidopsis*. The precision tables located at the end of this report are constructed to monitor the sensitivity of the organisms to the reference toxicant and thereby provide an indication of their overall sensitivity to other compounds.

Temperature, pH and dissolved oxygen measurements remained within acceptable limits throughout the reference toxicant test with topsmelt (#8276) and *Mysidopsis* (#8277). All tests met acceptability criteria regarding control survival and mean control weight (Chapman *et al.*, 1995; US EPA, 2002).

In addition, the LC50 (survival) and IC25 (growth) in the topsmelt and IC25 for the mysids were within the control limits of the mean ± 2SD (Chapman *et al.* 1995; US EPA, 2002).

Endpoint data for the reference toxicant tests is summarized in the following table:

Test #:	8276	8277
	Topsmelt	Mysid
Control Survival (%)	96	90
Criteria	≥ 80	≥ 80
Acceptable?	Yes	Yes
Control Growth (mg/ind)	1.311	0.263
Criteria	≥ 0.85	≥ 0.20
Acceptable?	Yes	Yes
PMSD for Growth (%)	NA	23.2
Criteria	NA	11-37
Acceptable?	NA	Yes
Survival LC50 (µg/L)	170.3	
LC50 Control Limits	66.3 – 618.8	
Growth IC25 (µg/L)	127.9	111.1
IC25 Control Limits	72.7 – 373.6	70.2 – 143.8
Acceptable?	Yes	Yes

Water Quality Measurements

Water quality parameters and methods are listed in the following table:

Parameter	Method
Water Quality Tests	APHA (1992); US EPA (1991).
Temperature	Standard Mercury Thermometer (calibrated with a certified thermometer traceable to NBS records) and Onset, Tidbit (v2) UTBI-001 Temperature Logger.
Dissolved Oxygen	YSI membrane electrode method (Method #4500-0 G, KCEL #434).
pH	Beckman 690 meter with automatic temperature compensation and Ross combination electrode (Method #4500-H; APHA 1992; KCEL #433).
Total Alkalinity	Potentiometric Method (Method #2320 B; KCEL #319).
Total Hardness	By calculation (Method #2340 B; KCEL #612).
Conductivity	Orion Model #122 Meter with 012210 conductivity cell (KCEL #435).
Total Ammonia	Phenate Method (Standard Methods SM 4500 - NH ₃ -G; KCEL #330).
Unionized Ammonia	Calculated from total ammonia, pH and ionization constants (APHA Method #417 G).
Salinity	Temperature compensated refractometer (KCEL #438)
Pesticides and PCB's	Continuous liquid extraction method (US EPA Method #608; KCEL #733)
Organic Analysis	Continuous liquid extraction method for BNA's (US EPA Method #625; KCEL #731)
Volatile Organics	Purge and trap method (US EPA Method #624; KCEL #732)
Total Metals	ICP-MS for Cd Ref. Tox. (US EPA Method #200.8; KCEL #618); ICP for Cd, Cr, Cu, Ni, Pb and Zn (US EPA Method #200.7; KCEL #612)

RESULTS

CHRONIC TESTS

Topsmelt – *Atherinops affinis*

Average weights per fish (based on the number of surviving fish) at the end of the 7-day chronic test with topsmelt (Test # 8274) are listed in the table below. Because the standard deviation for proportion alive was > 0.2 in 12.5% and 50% effluent, the mean weights per organism in the table below are based on the number of surviving topsmelt as explained in Appendix C of WA DOE WQ-R-95-80, 2016.

% Sample	GROWTH (Mean Weight per Fish in mg at 7 Days)						# Fish Tested	Survival %
	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Mean		
0	1.342	1.500	1.148	1.283	1.153	1.285	25	80
0.55 ^a	1.116	1.364	1.313	1.263	1.210	1.253	25	92
3.6 ^b	1.462	1.072	1.475	1.184	1.385	1.316	25	92
12.5	1.515	1.338	1.528	1.217	1.365	1.393	25	72
25	1.254	1.243	1.453	1.303	1.334	1.317	25	80
50	1.302	1.210	1.266	1.170	1.263	1.242	25	76
100	0	0	0	0	0	0	25	0

^aCCEC (Chronic Critical Effluent Concentration, mistakenly run at 0.55% effluent, should have been 0.53% effluent)

^bACEC

As shown in the table above, 7-day survival was 80% in the HMM control and 25% effluent concentration. Survival was 92% in the CCEC (0.55% effluent) and ACEC (3.6% effluent). Survival was 72% in 25% effluent and 76% in

50% effluent. There were no survivors in 100% effluent. Note: the CCEC was mistakenly run at 0.55% effluent rather than 0.53% effluent.

Growth (based on the number of survivors) in the ACEC was not reduced relative to the control. The Chronic Statistical Power Standard (CSPS) for the ACEC was found to be 0%, which is less than the maximum allowable difference of 39%. The IC25 for growth was > 50 % effluent (using the number of surviving topsmelt). Using the initial number of topsmelt at the start of the test the IC25 was 55.7% effluent.

The unionized ammonia level in 100% effluent reached a maximum of 1.416 mg N/L during the 7-day test.

Mysid Shrimp – *Mysidopsis bahia*

Average weights per mysid larva (based on the initial number of mysids) at the end of the 7-day chronic test (Test # 8275) are listed in the following table:

% Sample	GROWTH (Mean Weight per Organism in mg at 7 Days)									#Mysids Tested	Survival %
	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Trtmt Mean		
0	0.310	0.220	0.262	0.276	0.352	0.272	0.336	0.274	0.288	40	95
0.55 ^a	0.256	0.256	0.250	0.320	0.230	0.288	0.298	0.304	0.275	40	98
3.6 ^b	0.226	0.324	0.294	0.200	0.212	0.258	0.362	0.320	0.275	40	93
12.5	0.296	0.180	0.224	0.284	0.270	0.286	0.294	0.240	0.259	40	90
25	0.220	0.238	0.236	0.196	0.220	0.286	0.136	0.183	0.214	41	95
50	0.056	0.116	0.138	0.136	0.128	0.170	0.134	0.118	0.125	40	83
100	0	0	0	0	0	0	0	0	0	40	0

^aCCEC (Chronic Critical Effluent Concentration, mistakenly run at 0.55% effluent, should have been 0.53% effluent)

^bACEC

As shown in the table above, survival was 95% in 0% (HMM control) and 25% effluent. Survival was 98% in 0.55% (CCEC) effluent and 93% in 3.6% (ACEC) effluent. Survival was 90% in 12.5% effluent and 83% in 50% effluent. There were no survivors in 100% effluent. Note: the CCEC was mistakenly run at 0.55% effluent rather than 0.53% effluent.

Growth in the ACEC was not significantly reduced relative to the control (p > 0.05; 1-tailed t-Test). The CSPS for the ACEC was found to be 4.5%, which is less than the maximum allowable difference of 39%. The IC25 for growth was 24.6% effluent.

The unionized ammonia level in 100% effluent reached a maximum of 1.391 mg N/L during the 7-day test.

QUALITY CONTROL

Salinity, pH, temperature and dissolved remained within acceptable limits throughout the chronic tests for topsmelt (Chapman *et al.*, 1995; US EPA, 2002). Salinity and pH in the mysid test remained within acceptable limits. However, the dissolved oxygen on Day 3 in 100% effluent (24 hour solution) slightly exceeded the recommended minimum of 4.0 mg/L (3.8 mg/L). In addition, the dissolved oxygen on Day 5 in 50% effluent (24 hour solution) slightly exceeded the recommended minimum of 4.0 mg/L (3.7 mg/L). At test initiation the temperature in the mysid test on Day 0 was below the target range of 26 ± 1°C.

Water quality data recorded during testing is shown on the photocopied pages from the laboratory notebook in the "Bench Sheets" section of this report. Both tests met acceptability criteria regarding control performance, including survival and growth (Chapman *et al.*, 1995; US EPA, 2002).

Effluent test and control performance results are summarized in the following table:

Test #:	8274	8275
	Topsmelt	Mysid
Control Survival (%)	80	95
Criteria	≥ 80	≥ 80
Acceptable?	Yes	Yes
Control Growth (mg/ind)	1.285*	0.288
Criteria	≥ 0.85	≥ 0.20
Acceptable?	Yes	Yes
CSPS	0	4.5
Criteria	< 39%	< 39%
Acceptable?	Yes	Yes

*Based on the number of survivors (1.022 mg/ind based on the start #)

Tested By:

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REFERENCES

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- Chapman, G., D. Denton and J. Lazorchak. 1995.** Short-term methods for estimating the chronic toxicity of effluents and receiving waters to west coast marine and estuarine organisms. 1st Edition. EPA/600/R-95-136.
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