

Biological Analysis Report

NW-DIST-2019-11-06-01

Florida Department of Environmental Protection
Central Laboratory
2600 Blair Stone Road
Tallahassee, FL 32399-2400
DOH Accreditation E31780

Overflow Analyses Performed By:
University of West Florida
11000 University Parkway
Pensacola, FL 32514
DOH Accreditation E71969

Event Description: **Pensacola Monthly**
Request ID: **RQ-2019-11-04-53**
Customer: **NW-DIST**
Project ID: **SMP**

Send Reports to:
FL Dept. of Environmental Protection
160 Government Center
Suite 208
Pensacola, FL 32502-5794
Attn: Cheryl Bunch

For additional information please contact
Cheryl Swanson - Administrator
Jacqueline Savage - Microbiology
Diane Power - Bench Biology
Andrew Tintle - Aquatic Toxicology
Puja Jasrotia, Ph.D. - Molecular Biology
Amelia Rankin - Taxonomy (Invertebrate & Algal)
Thekkekalathil Chandrasekhar, Ph.D, QA Officer
Phone (850) 245-8177

Certified by: Puja Jasrotia, Environmental Manager

Date Certified: 10-DEC-2019 13:34



Case Narrative

Unless otherwise noted, all samples included in this report were received in accordance with protocols referenced in Chapter 62-160, Florida Administrative Code (F.A.C.). Results published in this report pertain only to the samples as submitted to, and received by the laboratory. All times in this report are adjusted to the applicable Eastern Time Zone (EST or EDT).

Results for the following analytical groups are included in this report: Chlorophyll/Grain Size/BOD, Molecular and Overflow.

Scientific notation may be used in reporting very large or small values. Values reported using scientific notation will take the form of the following example: 1.3E+03, which is equivalent to 1.3×10^3 or 1300.

Unless otherwise noted, analytical values for soil and sediment samples are reported on a dry weight basis, and analytical values for waste and tissue samples are reported on a wet weight basis.

Results for TNI accredited tests met requirements established by The NELAC Institute. A double asterisk (**) is used to indicate an analyte/matrix/method for which the laboratory is not TNI accredited by the Florida Department of Health Environmental Laboratory Certification Program or where accreditation for that field of testing is not applicable.

Any significant anomalies or deviations from established protocols are documented in Non-Conformance Reports, which, where appropriate, are included within this analytical report. Additional comments related to specific analytical tests may be included as remarks following the analytical results for each sample. Such comments and remarks are for informational purposes only and are not intended to convey judgement about the usability of the reported data.

A quality control report on the performance of the test method for the submitted samples is included. Uncertainty associated with the analytical results contained in this report can be estimated from the reported quality assurance results and from published quality control acceptance limits for each analytical test. Matrix quality control results (matrix spike recoveries and matrix sample precision) pertain only to the matrix sample tested and do not necessarily reflect test method performance for other samples.

Typical matrix quality control (QC) measurements may include matrix spike recovery, matrix spike duplicate recovery, matrix spike precision and matrix sample precision. Not all matrix QC results may be available or reportable; where they are not an explanation is provided. Typical reasons for unavailable QC results include, but are not limited to, a) insufficient matrix sample to perform some or all QC measurements; b) analyte concentration in the sample replicated was too low for a meaningful measurement of precision and c) analyte concentration in the matrix sample spiked was too high (relative to the amount of analyte spiked) for a meaningful measurement of recovery. Where matrix QC results are unavailable, other method performance metrics (e.g., LCS recovery, LCS precision, surrogate recovery) may be used to assess performance of the method. Comments explaining any missing QC measurements are not intended to convey any adverse conclusions about the quality of the reported data.

Precision is reported as relative percent difference unless otherwise noted.

Quality Control codes as defined below may be used in this report to indicate results that are associated with one or more quality control elements which did not fall within established test method criteria. Such results may be qualified as estimates using a J qualifier as required by 62-160 F.A.C. Explanations are included in the report for any results that were reported as estimates for other reasons.

QC Codes used in this report may include:

- LCS – Recovery for the batch Laboratory Control Sample (LCS) was outside existing control limits;
- MS – Recovery for the batch matrix spike (MS) was outside existing control limits;
- CCV – Recovery for a continuing calibration verification (CCV) standard was outside existing control limits;
- SUR – Recovery of a surrogate (SUR) for associated analytes was outside existing control limits;
- RPD – The precision, measured as relative percent difference (RPD), of batch replicate measurements was outside existing control limits;
- RSD – The precision, measured as relative standard deviation (RSD), of batch replicate measurements was outside existing control limits;
- SMP – Sample - used precision derived from replicate analyses of a sample;

The following data qualifiers are used, where applicable, in this report as specified in 62-160 F.A.C.

- A - Value reported is the mean of two or more determinations.
- B - Results based on colony counts outside the acceptable range.
- I - The reported value is between the laboratory method detection limit and the laboratory practical quantitation limit.
- J - Estimated value and/or the analysis did not meet established quality control criteria.
- K - Actual value is known to be less than value given.
- L - Actual value is known to be greater than value given.
- N - Presumptive evidence of presence of material.
- O - Sampled, but analysis lost or not performed.
- Q - Sample held beyond normal holding time.
- T - Value reported is less than the criterion of detection.
- U - Material was analyzed for but not detected. The reported value is the method detection limit for the sample analyzed.
- V - Analyte was detected in both sample and method blank.
- X - Too few individuals to calculate SCI value.
- Y - The laboratory analysis was from an unpreserved or improperly preserved sample. The data may not be accurate.
- Z - Colonies were too numerous to count (TNTC).

Quality control information from overflow laboratories may not be included in this report. Please refer to the associated report from the overflow laboratory for additional information.

Sample Location: Ten Mile Creek upstream of Pine Forest Road **Collection Date/Time: 11/05/2019 09:53**

Field ID: G5NW0051

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2133881	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	109		MPN/100 mL		
2133882	SOP-PCR-1.0	HF183-qPCR**	280	U	GEU/100 mL	P373742	
	SOP-PCR-1.2	GULL2-qPCR**	1.7E+04	U	TSC/100 mL	P373742	
	SOP-PCR-1.3	GFD-purified-qPCR**	170	UJ	TSC/100 mL	P374479	LCS, MS
	SOP-PCR-1.4	BacR-qPCR**	3.0E+03	U	TSC/100 mL	P373742	
	SOP-PCR-1.5	DG3-qPCR**	2.6E+03	U	TSC/100 mL	P373742	

Ref. Method and Comment:
SOP-PCR-1.0: Precision data unavailable for Analysis Batch A95935.
SOP-PCR-1.3: Quality control failure(s) observed. See QA report for details.

Sample Location: Pace Mill Creek upstream of Hwy 90 **Collection Date/Time: 11/05/2019 10:37**

Field ID: G4NW0481

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2133877	SM 10200 H (mod.)	Chlorophyll-a, Corrected	0.96	U	ug/L	P374167	
		Phaeophytin-a	1.0	U	ug/L	P374167	
		Chlorophyll-a, Uncorrected	0.70	U	ug/L	P374167	
2133879	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	63		MPN/100 mL		

Ref. Method and Comment:
SM 10200 H (mod.): Precision data is not available for at least one component due to the small amount of analyte in the QC sample. Refer to QA report for available precision data.

Sample Location: Direct Runoff - Graveyard Branch @ Tradewinds Dr. **Collection Date/Time: 11/05/2019 11:10**

Field ID: G4NW0176

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2133878	SM 10200 H (mod.)	Chlorophyll-a, Corrected	0.96	U	ug/L	P374167	
		Phaeophytin-a	1.0	U	ug/L	P374167	
		Chlorophyll-a, Uncorrected	0.70	U	ug/L	P374167	
2133880	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	820		MPN/100 mL		

Ref. Method and Comment:
SM 10200 H (mod.): Precision data is not available for at least one component due to the small amount of analyte in the QC sample. Refer to QA report for available precision data.

Quality Assurance Report Method Blank Results

Reference Method: SM 10200 H (mod.)
Batch ID: P374167

Component	Result	Code	Units
Chlorophyll-a, Corrected	0.96	U	ug/L
Chlorophyll-a, Uncorrected	0.70	U	ug/L
Phaeophytin-a	1.0	U	ug/L

Reference Method: SOP-PCR-1.0
Batch ID: P373742

Component	Result	Code	Units
-----------	--------	------	-------

Quality Assurance Report Method Blank Results

Reference Method: SOP-PCR-1.0
Batch ID: P373742

Component	Result	Code	Units
HF183-qPCR	280	U	GEU/100 mL

Reference Method: SOP-PCR-1.2
Batch ID: P373742

Component	Result	Code	Units
GULL2-qPCR	1.7E+04	U	TSC/100 mL

Reference Method: SOP-PCR-1.3
Batch ID: P374479

Component	Result	Code	Units
GFD-purified-qPCR	340	U	TSC/100 mL

Reference Method: SOP-PCR-1.4
Batch ID: P373742

Component	Result	Code	Units
BacR-qPCR	3.0E+03	U	TSC/100 mL

Reference Method: SOP-PCR-1.5
Batch ID: P373742

Component	Result	Code	Units
DG3-qPCR	2.6E+03	U	TSC/100 mL

Quality Assurance Report Laboratory Control Sample Accuracy

Reference Method: SM 10200 H (mod.)
Batch ID: P374167

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
Chlorophyll-a, Corrected	96.9		P	84 - 116

Reference Method: SOP-PCR-1.0
Batch ID: P373742

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
HF183-qPCR	123	130	P/P	50 - 175

Reference Method: SOP-PCR-1.2
Batch ID: P373742

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GULL2-qPCR	96.6	82.7	P/P	50 - 300

Reference Method: SOP-PCR-1.3
Batch ID: P374479

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
-----------	---------	---------	-----------	----------------

Quality Assurance Report Laboratory Control Sample Accuracy

Reference Method: SOP-PCR-1.3
 Batch ID: P374479

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GFD-purified-qPCR	139	49.5	P/F	50 - 175

Reference Method: SOP-PCR-1.4
 Batch ID: P373742

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
BacR-qPCR	143	173	P/P	50 - 175

Reference Method: SOP-PCR-1.5
 Batch ID: P373742

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
DG3-qPCR	112	139	P/P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0
 Batch ID: P373742

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2133850	HF183-qPCR	120		P	50 - 175
2133851	HF183-qPCR	118		P	50 - 175
2133852	HF183-qPCR	110		P	50 - 175
2133882	HF183-qPCR	111		P	50 - 175
2134510	HF183-qPCR	116		P	50 - 175
2134520	HF183-qPCR	114		P	50 - 175
2134521	HF183-qPCR	109		P	50 - 175

Reference Method: SOP-PCR-1.2
 Batch ID: P373742

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2133850	GULL2-qPCR	89.6		P	50 - 300
2133851	GULL2-qPCR	84.0		P	50 - 300
2133852	GULL2-qPCR	85.0		P	50 - 300
2133882	GULL2-qPCR	91.0		P	50 - 300

Reference Method: SOP-PCR-1.3
 Batch ID: P374479

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2131866	GFD-purified-qPCR	370		F	50 - 175
2131867	GFD-purified-qPCR	320	314	F/F	50 - 175
2132062	GFD-purified-qPCR	326		F	50 - 175
2132063	GFD-purified-qPCR	330		F	50 - 175
2132064	GFD-purified-qPCR	309		F	50 - 175
2133882	GFD-purified-qPCR	297		F	50 - 175
2138823	GFD-purified-qPCR	348		F	50 - 175
2138824	GFD-purified-qPCR	329		F	50 - 175
2138825	GFD-purified-qPCR	312		F	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.3

Batch ID: P374479

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2138829	GFD-purified-qPCR	316		F	50 - 175
2138830	GFD-purified-qPCR	279		F	50 - 175

Reference Method: SOP-PCR-1.4

Batch ID: P373742

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2133784	BacR-qPCR	102		P	50 - 175
2133799	BacR-qPCR	105		P	50 - 175
2133800	BacR-qPCR	104		P	50 - 175
2133801	BacR-qPCR	106		P	50 - 175
2133802	BacR-qPCR	122		P	50 - 175
2133803	BacR-qPCR	118	113	P/P	50 - 175
2133850	BacR-qPCR	130		P	50 - 175
2133851	BacR-qPCR	123		P	50 - 175
2133852	BacR-qPCR	119		P	50 - 175
2133882	BacR-qPCR	119		P	50 - 175

Reference Method: SOP-PCR-1.5

Batch ID: P373742

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2133784	DG3-qPCR	110		P	50 - 175
2133799	DG3-qPCR	113		P	50 - 175
2133800	DG3-qPCR	110		P	50 - 175
2133801	DG3-qPCR	104		P	50 - 175
2133802	DG3-qPCR	110		P	50 - 175
2133803	DG3-qPCR	107	96.4	P/P	50 - 175
2133850	DG3-qPCR	96.0		P	50 - 175
2133851	DG3-qPCR	99.8		P	50 - 175
2133852	DG3-qPCR	96.5		P	50 - 175
2133882	DG3-qPCR	102		P	50 - 175
2134655	DG3-qPCR	97.3		P	50 - 175

Quality Assurance Report Precision

Reference Method: SM 10200 H (mod.)

Batch ID: P374167

Replicated

Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2133627	Chlorophyll-a, Corrected	7.35	Sample	P	0 - 25
2133627	Chlorophyll-a, Uncorrected	7.75	Sample	P	0 - 25

Reference Method: SOP-PCR-1.2

Batch ID: P373742

Replicated

Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2132425	GULL2-qPCR	3.68	Spike	P	0 - 25

Quality Assurance Report Precision

Reference Method: SOP-PCR-1.3
Batch ID: P374479

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2131867	GFD-purified-qPCR	2.03	Spike	P	0 - 25

Reference Method: SOP-PCR-1.4
Batch ID: P373742

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2133803	BacR-qPCR	4.55	Spike	P	0 - 25

Reference Method: SOP-PCR-1.5
Batch ID: P373742

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2133803	DG3-qPCR	10.2	Spike	P	0 - 25

* Sample, spike and/or laboratory control sample precision (LCS) is reported.

Quality Assurance Report Surrogates

Lab Sample ID: 2133882
Field Sample ID: G5NW0051

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	101	P	50 - 300
SOP-PCR-1.2	SKETA-qPCR	95.1	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	101	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	179	P	50 - 300

Quality Assurance Report Summary

Ref. Method	Analyte	LCS % Recovery		MS % Recovery			LCS	Precision SMP	MS
SM 10200 H (mod.)	Chlorophyll-a, Corrected	96.9						7.35	
	Chlorophyll-a, Uncorrected							7.75	
SOP-PCR-1.0	HF183-qPCR	130	123	120	118	110			
SOP-PCR-1.2	GULL2-qPCR	82.7	96.6	111					
				89.6	84.0	85.0			3.68
SOP-PCR-1.3	GFD-purified-qPCR	49.5	139	91.0					
				370	320	314			2.03
SOP-PCR-1.4	BacR-qPCR	173	143	326					
				102	105	104			4.55
SOP-PCR-1.5	DG3-qPCR	139	112	106					
				110	113	110			10.2
				104					

Reference Method Descriptions

Method / DoH Cert #	Description	<u>Associated Samples</u>
SM 10200 H (mod.) / E31780	Phytoplankton chlorophyll-a corrected, uncorrected, and phaeophytin by spectrophotometry	2133877, 2133878
SM 9223 B Quanti-Tray / E71969	E. coli by Colilert-18/QT by UWF in Pensacola	2133879, 2133880, 2133881
SOP-PCR-1.0 / E31780	Detection and quantification of human-specific HF183 Bacteroides genetic marker with quantitative polymerase chain reaction	2133882
SOP-PCR-1.2 / E31780	Detection and quantification of coastal bird specific Catellicoccus marimammalium Gull2 genetic marker with quantitative polymerase chain reaction	2133882
SOP-PCR-1.3 / E31780	Detection and quantification of bird specific Helicobacter GFD genetic marker with quantitative polymerase chain reaction	2133882
SOP-PCR-1.4 / E31780	Detection and quantification of Ruminant specific Bacteroidetes BacR genetic marker with quantitative polymerase chain reaction	2133882
SOP-PCR-1.5 / E31780	Detection and quantification of canine-specific DG3 Bacteroides genetic marker with quantitative polymerase chain reaction	2133882

Preparation and Analysis Log

Ref. Method	Received Date	Prep Date/Time	Prepared By	Analysis Date/Time	Analyzed By	Associated Samples
SM 10200 H (mod.)	11/06/2019	11/06/2019 14:30	Peterson Janvier	11/13/2019 10:05	Yuliya Danyuk	2133877, 2133878
SM 9223 B Quanti-Tray	11/05/2019	11/05/2019 13:20		11/06/2019 10:42	UWF Lab	2133879, 2133880, 2133881
SOP-PCR-1.0	11/06/2019	11/06/2019 15:40	Angela Smith	11/25/2019 10:27	Lauren Campbell	2133882
SOP-PCR-1.2	11/06/2019	11/06/2019 15:40	Angela Smith	11/27/2019 11:10	Mailin Lopez	2133882
SOP-PCR-1.3	11/06/2019	11/06/2019 15:40	Lauren Campbell	12/03/2019 08:22	Mailin Lopez	2133882
SOP-PCR-1.4	11/06/2019	11/06/2019 15:40	Angela Smith	11/25/2019 15:44	Lauren Campbell	2133882
SOP-PCR-1.5	11/06/2019	11/06/2019 15:40	Angela Smith	11/27/2019 10:05	Lauren Campbell	2133882