

Biological Analysis Report

NW-DIST-2020-10-14-01

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: **Choctawhatchee Escambia Run**
Request ID: **RQ-2020-10-12-49**
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

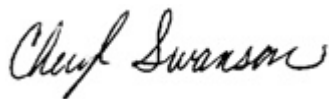
For additional information please contact
Cheryl Swanson - Administrator
Jacqueline Savage - Bench Biology & Aquatic
Toxicity
Puja Jasrotia, Ph.D. - Molecular Biology & Taxonomy
Thekkekalathil Chandrasekhar, Ph.D, QA Officer
Phone (850) 245-8177

This Report replaces the previous Report Serial Number: 0125474

Report Comment: Unauthorized to remove unnecessary "Q" qualifier.

Revision certified by: Cheryl A. Swanson, Program Administrator

Date Certified: 14-DEC-2020 15:27



NON-CONFORMANCE REPORT INCLUDED

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: Choctawhatchee River South of Carlisle Lake **Collection Date/Time: 10/12/2020 10:52**

Field ID: G3NW0130

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2201754	SM 10200 H (mod.)	Chlorophyll-a, Corrected	1.7	I	ug/L	P388912	
		Phaeophytin-a	0.90	U	ug/L	P388912	
		Chlorophyll-a, Uncorrected	1.9		ug/L	P388912	
2201757	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	96		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: Escambia River **Collection Date/Time: 10/12/2020 13:33**

Field ID: G4NW0405

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2201760	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	1050		MPN/100 mL		
2201769	SOP-PCR-1.0	HF183-qPCR**	280	U	GEU/100 mL	P387749	
	SOP-PCR-1.2	GULL2-qPCR**	3.6E+03	U	TSC/100 mL	P387749	
	SOP-PCR-1.3	GFD-purified-qPCR**	170	U	TSC/100 mL	P388232	LCS, RPD
	SOP-PCR-1.4	BacR-qPCR**	3.0E+03	U	TSC/100 mL	P387749	
	SOP-PCR-1.5	DG3-qPCR**	2.6E+03	U	TSC/100 mL	P387749	

Sample Location: Escambia River Downstream of McCostill Mill Creek **Collection Date/Time: 10/12/2020 13:47**

Field ID: G4NW0504

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2201755	SM 10200 H (mod.)	Chlorophyll-a, Corrected	1.8	I	ug/L	P388912	
		Phaeophytin-a	1.5	U	ug/L	P388912	
		Chlorophyll-a, Uncorrected	1.9	I	ug/L	P388912	
2201758	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	865		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: Escambia River Upstream of Mitchell Creek **Collection Date/Time: 10/12/2020 14:04**

Field ID: G4NW0404

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2201756	SM 10200 H (mod.)	Chlorophyll-a, Corrected	1.9	I	ug/L	P388912	
		Phaeophytin-a	1.5	U	ug/L	P388912	
		Chlorophyll-a, Uncorrected	2.2	I	ug/L	P388912	
2201759	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	780		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.

Non-Conformance Report

NCR ID: 8464

Event(s)

Job(s)

Sample(s)

Test(s)

Non-Conformance Report

NCR ID: 8464

Event(s)

NW-DIST-2020-10-14-01

Job(s)

Sample(s)

Test(s)

NCR Type: SHIPPING/RECEIVING

NCR Category: Samples arrived late

Observation: Samples arrived one or more days late by FedEx, samples received within the acceptable temperature range.

Resolution: Samples will be qualified as appropriate.

Authorized by/Date: Joshua Ayres 10/19/2020

The Non-Conformance Report details exceptions or problems encountered with the events/jobs/samples/test.
Please address questions to:

Chemistry Colin Wright (850) 245-8085
Biology Cheryl Swanson (850) 245-8177

Quality Assurance Report Method Blank Results

Reference Method: SM 10200 H (mod.)

Batch ID: P388912

Component	Result	Code	Units
Chlorophyll-a, Corrected	0.82	U	ug/L
Chlorophyll-a, Uncorrected	0.60	U	ug/L
Phaeophytin-a	0.90	U	ug/L

Reference Method: SOP-PCR-1.0

Batch ID: P387749

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

Reference Method: SOP-PCR-1.2

Batch ID: P387749

Component	Result	Code	Units
GULL2-qPCR	3.6E+03	U	TSC/100 mL

Reference Method: SOP-PCR-1.3

Batch ID: P388232

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

Reference Method: SOP-PCR-1.4

Batch ID: P387749

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

Reference Method: SOP-PCR-1.5

Batch ID: P387749

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: P388912

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
Chlorophyll-a, Corrected	96.4		P	84 - 116

Reference Method: SOP-PCR-1.0
 Batch ID: P387749

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
HF183-qPCR	104	85.9	P/P	50 - 175

Reference Method: SOP-PCR-1.2
 Batch ID: P387749

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GULL2-qPCR	90.4	90.8	P/P	50 - 300

Reference Method: SOP-PCR-1.3
 Batch ID: P388232

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GFD-purified-qPCR	104	3.94	P/F	50 - 175

Reference Method: SOP-PCR-1.4
 Batch ID: P387749

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
BacR-qPCR	93.6	88.7	P/P	50 - 175

Reference Method: SOP-PCR-1.5
 Batch ID: P387749

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
DG3-qPCR	89.6	158	P/P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0
 Batch ID: P387749

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2197889	HF183-qPCR	91.7	95.2	P/P	50 - 175
2198126	HF183-qPCR	82.4		P	50 - 175
2198129	HF183-qPCR	84.6		P	50 - 175
2198811	HF183-qPCR	93.8		P	50 - 175
2198812	HF183-qPCR	96.5		P	50 - 175
2199831	HF183-qPCR	87.1		P	50 - 175
2199832	HF183-qPCR	89.8		P	50 - 175
2199833	HF183-qPCR	97.5		P	50 - 175
2200720	HF183-qPCR	79.3		P	50 - 175
2201065	HF183-qPCR	77.4		P	50 - 175
2201210	HF183-qPCR	131		P	50 - 175
2201211	HF183-qPCR	103		P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0

Batch ID: P387749

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2201769	HF183-qPCR	88.2		P	50 - 175

Reference Method: SOP-PCR-1.2

Batch ID: P387749

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2197889	GULL2-qPCR	85.4	88.5	P/P	50 - 300
2198126	GULL2-qPCR	78.4		P	50 - 300
2198129	GULL2-qPCR	64.0		P	50 - 300
2198811	GULL2-qPCR	67.8		P	50 - 300
2198812	GULL2-qPCR	88.8		P	50 - 300
2199831	GULL2-qPCR	72.2		P	50 - 300
2199832	GULL2-qPCR	77.2		P	50 - 300
2199833	GULL2-qPCR	80.7		P	50 - 300
2200720	GULL2-qPCR	79.2		P	50 - 300
2201065	GULL2-qPCR	50.7		P	50 - 300
2201769	GULL2-qPCR	89.0		P	50 - 300

Reference Method: SOP-PCR-1.3

Batch ID: P388232

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2197889	GFD-purified-qPCR	118		P	50 - 175
2198126	GFD-purified-qPCR	61.2		P	50 - 175
2198129	GFD-purified-qPCR	99.0		P	50 - 175
2198811	GFD-purified-qPCR	54.7		P	50 - 175
2198812	GFD-purified-qPCR	90.0		P	50 - 175
2199831	GFD-purified-qPCR	126		P	50 - 175
2199832	GFD-purified-qPCR	135		P	50 - 175
2199833	GFD-purified-qPCR	107	171	P/P	50 - 175
2200720	GFD-purified-qPCR	120		P	50 - 175
2201065	GFD-purified-qPCR	113		P	50 - 175
2201769	GFD-purified-qPCR	107		P	50 - 175
2201853	GFD-purified-qPCR	138		P	50 - 175
2203492	GFD-purified-qPCR	70.8		P	50 - 175
2203493	GFD-purified-qPCR	101		P	50 - 175
2203494	GFD-purified-qPCR	113		P	50 - 175
2203770	GFD-purified-qPCR	99.8		P	50 - 175
2203940	GFD-purified-qPCR	91.4		P	50 - 175
2203980	GFD-purified-qPCR	133		P	50 - 175
2204785	GFD-purified-qPCR	143		P	50 - 175

Reference Method: SOP-PCR-1.4

Batch ID: P387749

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2197889	BacR-qPCR	72.1	74.4	P/P	50 - 175
2198126	BacR-qPCR	64.2		P	50 - 175
2198129	BacR-qPCR	65.0		P	50 - 175
2198811	BacR-qPCR	75.5		P	50 - 175
2198812	BacR-qPCR	75.9		P	50 - 175
2199831	BacR-qPCR	72.4		P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.4

Batch ID: P387749

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2199832	BacR-qPCR	71.1		P	50 - 175
2199833	BacR-qPCR	80.2		P	50 - 175
2200720	BacR-qPCR	55.6		P	50 - 175
2201065	BacR-qPCR	64.6		P	50 - 175
2201210	BacR-qPCR	88.5		P	50 - 175
2201211	BacR-qPCR	83.5		P	50 - 175
2201769	BacR-qPCR	70.0		P	50 - 175

Reference Method: SOP-PCR-1.5

Batch ID: P387749

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2197889	DG3-qPCR	98.4	97.8	P/P	50 - 175
2198126	DG3-qPCR	94.6		P	50 - 175
2198129	DG3-qPCR	101		P	50 - 175
2198811	DG3-qPCR	101		P	50 - 175
2198812	DG3-qPCR	115		P	50 - 175
2199831	DG3-qPCR	90.6		P	50 - 175
2199832	DG3-qPCR	102		P	50 - 175
2199833	DG3-qPCR	98.9		P	50 - 175
2200720	DG3-qPCR	96.9		P	50 - 175
2201065	DG3-qPCR	79.8		P	50 - 175
2201210	DG3-qPCR	103		P	50 - 175
2201211	DG3-qPCR	107		P	50 - 175
2201769	DG3-qPCR	91.7		P	50 - 175

Quality Assurance Report Precision

Reference Method: SOP-PCR-1.0

Batch ID: P387749

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2197889	HF183-qPCR	3.85	Spike	P	0 - 25

Reference Method: SOP-PCR-1.2

Batch ID: P387749

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2197889	GULL2-qPCR	3.48	Spike	P	0 - 25

Reference Method: SOP-PCR-1.3

Batch ID: P388232

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2199833	GFD-purified-qPCR	45.9	Spike	F	0 - 25

Quality Assurance Report Precision

Reference Method: SOP-PCR-1.4
 Batch ID: P387749

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2197889	BacR-qPCR	3.15	Spike	P	0 - 25

Reference Method: SOP-PCR-1.5
 Batch ID: P387749

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2197889	DG3-qPCR	0.564	Spike	P	0 - 25

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

Quality Assurance Report Surrogates

Lab Sample ID: 2201769
 Field Sample ID: G4NW0405

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	60.5	P	50 - 300
SOP-PCR-1.2	SKETA-qPCR	79.6	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	79.4	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	56.2	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	96.4							
SOP-PCR-1.0	HF183-qPCR	85.9	104	91.7	95.2	82.4			3.85
				84.6					
SOP-PCR-1.2	GULL2-qPCR	90.8	90.4	85.4	88.5	78.4			3.48
				64.0					
SOP-PCR-1.3	GFD-purified-qPCR	3.94	104	126	135	107			45.9
				171					
SOP-PCR-1.4	BacR-qPCR	88.7	93.6	72.1	74.4	64.2			3.15
				65.0					
SOP-PCR-1.5	DG3-qPCR	158	89.6	98.4	97.8	94.6			0.564
				101					

Reference Method Descriptions

Method	Description	Associated Samples
SM 10200 H (mod.)	Phytoplankton chlorophyll-a corrected, uncorrected, and phaeophytin by spectrophotometry	2201754, 2201755, 2201756
SM 9223 B Quanti-Tray	E. coli by Colilert-18/QT by UWF in Pensacola	2201757, 2201758, 2201759, 2201760
SOP-PCR-1.0	Detection and quantification of human-specific HF183 Bacteroides genetic marker with quantitative polymerase chain reaction	2201769
SOP-PCR-1.2	Detection and quantification of coastal bird specific Catellicoccus marimammalium Gull2 genetic marker with quantitative polymerase chain reaction	2201769

Reference Method Descriptions

Method	Description	<u>Associated Samples</u>
SOP-PCR-1.3	Detection and quantification of bird specific Helicobacter GFD genetic marker with quantitative polymerase chain reaction	2201769
SOP-PCR-1.4	Detection and quantification of Ruminant specific Bacteroidetes BacR genetic marker with quantitative polymerase chain reaction	2201769
SOP-PCR-1.5	Detection and quantification of canine-specific DG3 Bacteroides genetic marker with quantitative polymerase chain reaction	2201769

Preparation and Analysis Log

Ref. Method	Received Date	Prep Date/Time	Prepared By	Analysis Date/Time	Analyzed By	Associated Samples
SM 10200 H (mod.)	10/14/2020	10/14/2020 10:52	Jessica Sherman	10/19/2020 09:20	Jessica Fetgatter	2201754, 2201755, 2201756
SM 9223 B Quanti-Tray	10/12/2020	10/12/2020 16:15		10/13/2020 13:29	UWF Lab	2201757, 2201758, 2201759, 2201760
SOP-PCR-1.0	10/14/2020	10/14/2020 13:00	Lauren Campbell	10/19/2020 12:11	Lauren Campbell	2201769
SOP-PCR-1.2	10/14/2020	10/14/2020 13:00	Lauren Campbell	10/23/2020 08:44	Mailin Lopez	2201769
SOP-PCR-1.3	10/14/2020	10/14/2020 13:00	Lauren Campbell	10/29/2020 11:06	Mailin Lopez	2201769
SOP-PCR-1.4	10/14/2020	10/14/2020 13:00	Lauren Campbell	10/20/2020 10:27	Lauren Campbell	2201769
SOP-PCR-1.5	10/14/2020	10/14/2020 13:00	Lauren Campbell	10/21/2020 10:32	Lauren Campbell	2201769