

Biological Analysis Report

NW-DIST-2020-07-22-01

Florida Department of Environmental Protection
Central Laboratory
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DOH Accreditation E31780

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

Event Description: **Holmes Run**
Request ID: **RQ-2020-07-20-55**
Customer: **NW-DIST**
Project ID: **SMP**

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Certified by: Jacqueline Savage, Environmental Administrator

Date Certified: 02-SEP-2020 10:54



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: Limestone Branch upstream of Mt. Ida Rd

Collection Date/Time: 07/21/2020 10:35

Field ID: G3NW0023

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2184570	SM 10200 H (mod.)	Chlorophyll-a, Corrected	0.89	U	ug/L	P385108	
		Phaeophytin-a	0.98	U	ug/L	P385108	
		Chlorophyll-a, Uncorrected	0.65	U	ug/L	P385108	
2184575	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	183		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: East Pitman Creek upstream of HWY 179

Collection Date/Time: 07/21/2020 11:05

Field ID: G3NW0157

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2184567	SM 10200 H (mod.)	Chlorophyll-a, Corrected	1.2	U	ug/L	P385108	
		Phaeophytin-a	1.3	U	ug/L	P385108	
		Chlorophyll-a, Uncorrected	1.2	I	ug/L	P385108	
2184572	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	262		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: Wrights Creek above Highway 177A

Collection Date/Time: 07/21/2020 11:35

Field ID: G3NW0444

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2184576	SOP-PCR-1.0	HF183-qPCR**	140	U	GEU/100 mL	P384933	
	SOP-PCR-1.2	GULL2-qPCR**	8.4E+03	U	TSC/100 mL	P384933	LCS
	SOP-PCR-1.3	GFD-purified-qPCR**	170	U	TSC/100 mL	P385132	LCS, MS
	SOP-PCR-1.4	BacR-qPCR**	1.5E+03	U	TSC/100 mL	P384933	
	SOP-PCR-1.5	DG3-qPCR**	1.3E+03	U	TSC/100 mL	P384933	
2186322	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	110		MPN/100 mL		

Sample Location: Sikes Creek upstream of Bonifay Gritney Rd

Collection Date/Time: 07/21/2020 12:00

Field ID: G3NW0159

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2184566	SM 10200 H (mod.)	Chlorophyll-a, Corrected	2.4	U	ug/L	P385108	
		Phaeophytin-a	2.6	U	ug/L	P385108	
		Chlorophyll-a, Uncorrected	1.8	U	ug/L	P385108	
2184571	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	189		MPN/100 mL		

Ref. Method and Comment:

SM 10200 H (mod.): Lab dup result is 2.4 U for Chl-a Corrected, 1.8 U for Chl-a Uncorrected, and 2.6 U for Phaeophytin-a. Precision data is unavailable due to the small amount of analyte in the QC sample.

Sample Location: Choctawhatchee River upstream HWY 90 boat ramp

Collection Date/Time: 07/21/2020 12:35

Field ID: G3NW0052

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
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Field ID: G3NW0052

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2184568	SM 10200 H (mod.)	Chlorophyll-a, Corrected	13		ug/L	P385108	
		Phaeophytin-a	0.90	U	ug/L	P385108	
		Chlorophyll-a, Uncorrected	13		ug/L	P385108	
2184573	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	20		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: Gum Creek upstream of River Road Collection Date/Time: 07/21/2020 13:00

Field ID: G3NW0439

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2184569	SM 10200 H (mod.)	Chlorophyll-a, Corrected	2.1	U	ug/L	P385108	
		Phaeophytin-a	2.2	U	ug/L	P385108	
		Chlorophyll-a, Uncorrected	1.5	U	ug/L	P385108	
2184574	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	309		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: P385108

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: P384933

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

Reference Method: SOP-PCR-1.2
 Batch ID: P384933

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

Reference Method: SOP-PCR-1.3
 Batch ID: P385132

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

Reference Method: SOP-PCR-1.4
 Batch ID: P384933

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

Quality Assurance Report Method Blank Results

Reference Method: SOP-PCR-1.5
Batch ID: P384933

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: P385108

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
Chlorophyll-a, Corrected	85.1		P	84 - 116

Reference Method: SOP-PCR-1.0
Batch ID: P384933

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
HF183-qPCR	129	117	P/P	50 - 175

Reference Method: SOP-PCR-1.2
Batch ID: P384933

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GULL2-qPCR	94.7	21.0	P/F	50 - 300

Reference Method: SOP-PCR-1.3
Batch ID: P385132

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GFD-purified-qPCR	106	12.7	P/F	50 - 175

Reference Method: SOP-PCR-1.4
Batch ID: P384933

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
BacR-qPCR	98.5	89.2	P/P	50 - 175

Reference Method: SOP-PCR-1.5
Batch ID: P384933

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
DG3-qPCR	96.1	168	P/P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0
Batch ID: P384933

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2184371	HF183-qPCR	103	108	P/P	50 - 175
2184372	HF183-qPCR	96.5		P	50 - 175
2184373	HF183-qPCR	85.3		P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0

Batch ID: P384933

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2184576	HF183-qPCR	148		P	50 - 175
2184832	HF183-qPCR	98.3		P	50 - 175
2184833	HF183-qPCR	96.6		P	50 - 175
2184834	HF183-qPCR	101		P	50 - 175
2185504	HF183-qPCR	86.7		P	50 - 175
2186533	HF183-qPCR	108		P	50 - 175
2186840	HF183-qPCR	94.8		P	50 - 175
2186923	HF183-qPCR	104		P	50 - 175
2186924	HF183-qPCR	103		P	50 - 175
2187410	HF183-qPCR	131		P	50 - 175

Reference Method: SOP-PCR-1.2

Batch ID: P384933

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2184371	GULL2-qPCR	93.4	93.3	P/P	50 - 300
2184373	GULL2-qPCR	68.8		P	50 - 300
2184576	GULL2-qPCR	89.7		P	50 - 300
2185504	GULL2-qPCR	50.7		P	50 - 300
2186533	GULL2-qPCR	73.8		P	50 - 300
2186923	GULL2-qPCR	71.7		P	50 - 300
2186924	GULL2-qPCR	52.1		P	50 - 300

Reference Method: SOP-PCR-1.3

Batch ID: P385132

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2181555	GFD-purified-qPCR	94.1		P	50 - 175
2181556	GFD-purified-qPCR	55.0		P	50 - 175
2183113	GFD-purified-qPCR	106		P	50 - 175
2184015	GFD-purified-qPCR	80.4		P	50 - 175
2184016	GFD-purified-qPCR	89.2		P	50 - 175
2184371	GFD-purified-qPCR	94.4	115	P/P	50 - 175
2184373	GFD-purified-qPCR	39.2		F	50 - 175
2184576	GFD-purified-qPCR	104		P	50 - 175
2185504	GFD-purified-qPCR	80.3		P	50 - 175
2186533	GFD-purified-qPCR	57.6		P	50 - 175
2186923	GFD-purified-qPCR	56.2		P	50 - 175
2186924	GFD-purified-qPCR	58.4		P	50 - 175

Reference Method: SOP-PCR-1.4

Batch ID: P384933

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2184371	BacR-qPCR	82.7	86.0	P/P	50 - 175
2184372	BacR-qPCR	70.6		P	50 - 175
2184373	BacR-qPCR	72.7		P	50 - 175
2184576	BacR-qPCR	86.9		P	50 - 175
2184832	BacR-qPCR	70.1		P	50 - 175
2184833	BacR-qPCR	77.8		P	50 - 175
2184834	BacR-qPCR	80.4		P	50 - 175
2185504	BacR-qPCR	59.2		P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.4
Batch ID: P384933

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2186533	BacR-qPCR	69.3		P	50 - 175
2186840	BacR-qPCR	54.2		P	50 - 175
2186923	BacR-qPCR	79.9		P	50 - 175
2186924	BacR-qPCR	69.3		P	50 - 175
2187410	BacR-qPCR	75.1		P	50 - 175

Reference Method: SOP-PCR-1.5
Batch ID: P384933

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2184371	DG3-qPCR	102	106	P/P	50 - 175
2184372	DG3-qPCR	95.1		P	50 - 175
2184373	DG3-qPCR	97.4		P	50 - 175
2184576	DG3-qPCR	82.0		P	50 - 175
2184832	DG3-qPCR	106		P	50 - 175
2184833	DG3-qPCR	82.1		P	50 - 175
2184834	DG3-qPCR	95.5		P	50 - 175
2185504	DG3-qPCR	71.0		P	50 - 175
2186533	DG3-qPCR	96.0		P	50 - 175
2186840	DG3-qPCR	72.7		P	50 - 175
2186923	DG3-qPCR	71.3		P	50 - 175
2186924	DG3-qPCR	90.0		P	50 - 175
2187410	DG3-qPCR	85.2		P	50 - 175

Quality Assurance Report Precision

Reference Method: SOP-PCR-1.0
Batch ID: P384933

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2184371	HF183-qPCR	4.78	Spike	P	0 - 25

Reference Method: SOP-PCR-1.2
Batch ID: P384933

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2184371	GULL2-qPCR	0.102	Spike	P	0 - 25

Reference Method: SOP-PCR-1.3
Batch ID: P385132

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2184371	GFD-purified-qPCR	19.5	Spike	P	0 - 25

Quality Assurance Report Precision

Reference Method: SOP-PCR-1.4
 Batch ID: P384933

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2184371	BacR-qPCR	3.93	Spike	P	0 - 25

Reference Method: SOP-PCR-1.5
 Batch ID: P384933

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2184371	DG3-qPCR	3.07	Spike	P	0 - 25

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

Quality Assurance Report Surrogates

Lab Sample ID: 2184576
 Field Sample ID: G3NW0444

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	100	P	50 - 300
SOP-PCR-1.2	SKETA-qPCR	103	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	97.0	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	105	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	85.1							
SOP-PCR-1.0	HF183-qPCR	117	129	103	108	96.5			4.78
				85.3					
SOP-PCR-1.2	GULL2-qPCR	21.0	94.7	93.4	93.3	68.8			0.102
				89.7					
SOP-PCR-1.3	GFD-purified-qPCR	12.7	106	94.1	55.0	106			19.5
				80.4					
SOP-PCR-1.4	BacR-qPCR	89.2	98.5	82.7	86.0	70.6			3.93
				72.7					
SOP-PCR-1.5	DG3-qPCR	168	96.1	102	106	95.1			3.07
				97.4					

Reference Method Descriptions

Method	Description	Associated Samples
SM 10200 H (mod.)	Phytoplankton chlorophyll-a corrected, uncorrected, and phaeophytin by spectrophotometry	2184566, 2184567, 2184568, 2184569, 2184570
SM 9223 B Quanti-Tray	E. coli by Colilert-18/QT by UWF in Pensacola	2184571, 2184572, 2184573, 2184574, 2184575, 2186322
SOP-PCR-1.0	Detection and quantification of human-specific HF183 Bacteroides genetic marker with quantitative polymerase chain reaction	2184576

Reference Method Descriptions

Method	Description	<u>Associated Samples</u>
SOP-PCR-1.2	Detection and quantification of coastal bird specific <i>Catelliboccus marimammalium</i> Gull2 genetic marker with quantitative polymerase chain reaction	2184576
SOP-PCR-1.3	Detection and quantification of bird specific <i>Helicobacter</i> GFD genetic marker with quantitative polymerase chain reaction	2184576
SOP-PCR-1.4	Detection and quantification of Ruminant specific <i>Bacteroidetes</i> BacR genetic marker with quantitative polymerase chain reaction	2184576
SOP-PCR-1.5	Detection and quantification of canine-specific DG3 <i>Bacteroides</i> genetic marker with quantitative polymerase chain reaction	2184576

Preparation and Analysis Log

Ref. Method	Received Date	Prep Date/Time	Prepared By	Analysis Date/Time	Analyzed By	Associated Samples
SM 10200 H (mod.)	07/22/2020	07/22/2020 15:07	Jessica Fetgatter	07/23/2020 09:20	Yuliya Danyuk	2184566, 2184567, 2184568, 2184569, 2184570
SM 9223 B Quanti-Tray	07/21/2020	07/21/2020 16:00		07/22/2020 13:02	UWF Lab	2184571, 2184572, 2184573, 2184574, 2184575, 2186322
SOP-PCR-1.0	07/22/2020	07/22/2020 14:15	Mailin Lopez	08/04/2020 14:04	Lauren Campbell	2184576
SOP-PCR-1.2	07/22/2020	07/22/2020 14:15	Mailin Lopez	08/07/2020 11:44	Mailin Lopez	2184576
SOP-PCR-1.3	07/22/2020	07/22/2020 14:15	Lauren Campbell	08/07/2020 15:28	Mailin Lopez	2184576
SOP-PCR-1.4	07/22/2020	07/22/2020 14:15	Mailin Lopez	08/05/2020 10:52	Lauren Campbell	2184576
SOP-PCR-1.5	07/22/2020	07/22/2020 14:15	Mailin Lopez	08/05/2020 13:58	Lauren Campbell	2184576