

Biological Analysis Report

NW-DIST-2022-01-26-01

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: **Walton Bay Run**
Request ID: **RQ-2022-01-24-41**
Customer: **NW-DIST**
Project ID: **SMP**

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

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

Certified by: Cheryl A. Swanson, Program Administrator

Date Certified: 11-MAR-2022 14:38



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: Bay Branch below Coy Burgess Loop

Collection Date/Time: 01/25/2022 10:25

Field ID: G3NW0231

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2301679	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	332		MPN/100 mL		
2301686	SOP-PCR-1.0	HF183-qPCR**	1.7E+03	U	TSC/100 mL	P408914	
	SOP-PCR-1.2	GULL2-qPCR**	3.6E+03	U	TSC/100 mL	P408914	
	SOP-PCR-1.3	GFD-qPCR**	720	U	TSC/100 mL	P408914	
	SOP-PCR-1.4	BacR-qPCR**	3.0E+03	U	TSC/100 mL	P408914	
	SOP-PCR-1.5	DG3-qPCR**	1.3E+03	UJ	TSC/100 mL	P408914	LCS, SUR

Ref. Method and Comment:
 SOP-PCR-1.5: Refer to the Lab Analysis Report for an explanation of QC Codes.

Sample Location: Bay Branch upstream of HWY 331

Collection Date/Time: 01/25/2022 10:45

Field ID: G3NW0171

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2301680	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	98		MPN/100 mL		
2301687	SOP-PCR-1.0	HF183-qPCR**	1.7E+03	U	TSC/100 mL	P408914	
	SOP-PCR-1.2	GULL2-qPCR**	3.6E+03	U	TSC/100 mL	P408914	
	SOP-PCR-1.3	GFD-qPCR**	720	U	TSC/100 mL	P408914	
	SOP-PCR-1.4	BacR-qPCR**	3.0E+03	U	TSC/100 mL	P408914	
	SOP-PCR-1.5	DG3-qPCR**	1.3E+03	UJ	TSC/100 mL	P408914	LCS, SUR

Ref. Method and Comment:
 SOP-PCR-1.5: Refer to the Lab Analysis Report for an explanation of QC Codes.

Sample Location: Little Alaqua Creek Below 282 Bridge

Collection Date/Time: 01/25/2022 11:20

Field ID: G3NW0065

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2301681	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	148		MPN/100 mL		

Sample Location: Little Alaqua Creek above Eglin AFB Road 208

Collection Date/Time: 01/25/2022 11:35

Field ID: G3NW0453

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2301682	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	145		MPN/100 mL		

Sample Location: Lafayette Creek

Collection Date/Time: 01/25/2022 12:15

Field ID: G3NW0455

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2301684	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	327		MPN/100 mL		

Sample Location: Pine Log Creek Upstream of Highway 20

Collection Date/Time: 01/25/2022 13:00

Field ID: G3NW0454

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2301685	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	85		MPN/100 mL		

Sample Location: Seven Runs upstream of Hwy 81

Collection Date/Time: 01/25/2022 13:30

Field ID: G3NW0116

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2301683	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	75		MPN/100 mL		

Sample Location: Panther Creek upstream of SR280

Collection Date/Time: 01/25/2022 14:00

Field ID: G3NW0061

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2301677	SM 10200 H (mod.)	Chlorophyll-a, Corrected	0.82	U	ug/L	P409382	
		Phaeophytin-a	0.90	U	ug/L	P409382	
		Chlorophyll-a, Uncorrected	0.60	U	ug/L	P409382	
2301678	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	275		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: P409382

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: P408914

Component	Result	Code	Units
HF183-qPCR	3.3E+03	U	TSC/100 mL

Reference Method: SOP-PCR-1.2
Batch ID: P408914

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

Reference Method: SOP-PCR-1.3
Batch ID: P408914

Component	Result	Code	Units
GFD-qPCR	720	U	TSC/100 mL

Reference Method: SOP-PCR-1.4
Batch ID: P408914

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

Reference Method: SOP-PCR-1.5
Batch ID: P408914

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: P409382

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
Chlorophyll-a, Corrected	94.7		P	84 - 116

Reference Method: SOP-PCR-1.0
Batch ID: P408914

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
HF183-qPCR	87.0	89.0	P/P	50 - 175

Reference Method: SOP-PCR-1.2
Batch ID: P408914

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GULL2-qPCR	87.8	83.0	P/P	50 - 300

Reference Method: SOP-PCR-1.3
Batch ID: P408914

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GFD-qPCR	70.1	88.6	P/P	50 - 175

Reference Method: SOP-PCR-1.4
Batch ID: P408914

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
BacR-qPCR	86.9	61.4	P/P	50 - 175

Reference Method: SOP-PCR-1.5
Batch ID: P408914

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
DG3-qPCR	79.3	40.7	P/F	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0
Batch ID: P408914

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2301273	HF183-qPCR	84.7	78.1	P/P	50 - 175
2301313	HF183-qPCR	91.7		P	50 - 175
2301314	HF183-qPCR	81.8		P	50 - 175
2301315	HF183-qPCR	85.1		P	50 - 175
2301316	HF183-qPCR	82.3		P	50 - 175
2301317	HF183-qPCR	83.4		P	50 - 175
2301686	HF183-qPCR	97.9		P	50 - 175
2301687	HF183-qPCR	84.1		P	50 - 175
2302275	HF183-qPCR	82.6		P	50 - 175
2302714	HF183-qPCR	103		P	50 - 175
2302715	HF183-qPCR	101		P	50 - 175
2302732	HF183-qPCR	83.5		P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0
Batch ID: P408914

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2303015	HF183-qPCR	93.0		P	50 - 175

Reference Method: SOP-PCR-1.2
Batch ID: P408914

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2301273	GULL2-qPCR	92.2	90.0	P/P	50 - 300
2301316	GULL2-qPCR	85.0		P	50 - 300
2301686	GULL2-qPCR	93.1		P	50 - 300
2301687	GULL2-qPCR	90.2		P	50 - 300
2302275	GULL2-qPCR	77.1		P	50 - 300
2302714	GULL2-qPCR	61.6		P	50 - 300
2302715	GULL2-qPCR	82.9		P	50 - 300
2303015	GULL2-qPCR	68.6		P	50 - 300

Reference Method: SOP-PCR-1.3
Batch ID: P408914

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2301273	GFD-qPCR	83.1	82.9	P/P	50 - 175
2301316	GFD-qPCR	93.3		P	50 - 175
2301686	GFD-qPCR	108		P	50 - 175
2301687	GFD-qPCR	103		P	50 - 175
2302275	GFD-qPCR	95.3		P	50 - 175
2302714	GFD-qPCR	119		P	50 - 175
2302715	GFD-qPCR	118		P	50 - 175
2303015	GFD-qPCR	113		P	50 - 175

Reference Method: SOP-PCR-1.4
Batch ID: P408914

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2301273	BacR-qPCR	76.2	76.9	P/P	50 - 175
2301313	BacR-qPCR	63.5		P	50 - 175
2301314	BacR-qPCR	63.8		P	50 - 175
2301315	BacR-qPCR	65.7		P	50 - 175
2301316	BacR-qPCR	73.1		P	50 - 175
2301317	BacR-qPCR	76.4		P	50 - 175
2301686	BacR-qPCR	57.3		P	50 - 175
2301687	BacR-qPCR	62.3		P	50 - 175
2302275	BacR-qPCR	65.0		P	50 - 175
2302714	BacR-qPCR	64.6		P	50 - 175
2302715	BacR-qPCR	73.7		P	50 - 175
2302732	BacR-qPCR	76.2		P	50 - 175
2303015	BacR-qPCR	58.7		P	50 - 175

Reference Method: SOP-PCR-1.5
Batch ID: P408914

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2301273	DG3-qPCR	74.4	69.8	P/P	50 - 175
2301313	DG3-qPCR	57.5		P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.5

Batch ID: P408914

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2301314	DG3-qPCR	64.6		P	50 - 175
2301315	DG3-qPCR	66.9		P	50 - 175
2301316	DG3-qPCR	74.1		P	50 - 175
2301317	DG3-qPCR	75.2		P	50 - 175
2301686	DG3-qPCR	61.7		P	50 - 175
2301687	DG3-qPCR	68.1		P	50 - 175
2302275	DG3-qPCR	65.1		P	50 - 175
2302714	DG3-qPCR	51.0		P	50 - 175
2302715	DG3-qPCR	65.4		P	50 - 175
2302732	DG3-qPCR	70.2		P	50 - 175
2303015	DG3-qPCR	55.2		P	50 - 175

Quality Assurance Report Precision

Reference Method: SM 10200 H (mod.)

Batch ID: P409382

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2301615	Chlorophyll-a, Corrected	11.6	Sample	P	0 - 25
2301615	Chlorophyll-a, Uncorrected	11.2	Sample	P	0 - 25

Reference Method: SOP-PCR-1.0

Batch ID: P408914

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2301273	HF183-qPCR	8.14	Spike	P	0 - 25

Reference Method: SOP-PCR-1.2

Batch ID: P408914

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2301273	GULL2-qPCR	2.42	Spike	P	0 - 25

Reference Method: SOP-PCR-1.3

Batch ID: P408914

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2301273	GFD-qPCR	0.252	Spike	P	0 - 25

Reference Method: SOP-PCR-1.4

Batch ID: P408914

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2301273	BacR-qPCR	0.948	Spike	P	0 - 25

Quality Assurance Report Precision

Reference Method: SOP-PCR-1.5
 Batch ID: P408914

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2301273	DG3-qPCR	6.50	Spike	P	0 - 25

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

Quality Assurance Report Surrogates

Lab Sample ID: 2301686
 Field Sample ID: G3NW0231

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	97.3	P	50 - 300
SOP-PCR-1.2	SKETA-qPCR	101	P	50 - 300
SOP-PCR-1.3	SKETA-qPCR	95.1	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	88.3	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	37.6	F	50 - 300

Lab Sample ID: 2301687
 Field Sample ID: G3NW0171

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	92.6	P	50 - 300
SOP-PCR-1.2	SKETA-qPCR	87.3	P	50 - 300
SOP-PCR-1.3	SKETA-qPCR	90.4	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	88.8	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	37.6	F	50 - 300

Quality Assurance Report Summary

Ref. Method	Analyte	LCS % Recovery		MS % Recovery			LCS	Precision SMP	MS
SM 10200 H (mod.)	Chlorophyll-a, Corrected	94.7						11.6	
	Chlorophyll-a, Uncorrected							11.2	
SOP-PCR-1.0	HF183-qPCR	89.0	87.0	101	83.5	93.0			8.14
				84.7					
SOP-PCR-1.2	GULL2-qPCR	83.0	87.8	77.1	61.6	82.9			2.42
				68.6					
SOP-PCR-1.3	GFD-qPCR	88.6	70.1	83.1	82.9	93.3			0.252
				108					
SOP-PCR-1.4	BacR-qPCR	61.4	86.9	76.2	76.9	63.5			0.948
				63.8					
SOP-PCR-1.5	DG3-qPCR	40.7	79.3	74.4	69.8	57.5			6.50
				64.6					

Reference Method Descriptions

Method	Description	Associated Samples
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Reference Method Descriptions

Method	Description	<u>Associated Samples</u>
SM 10200 H (mod.)	Phytoplankton chlorophyll-a corrected, uncorrected, and phaeophytin by spectrophotometry	2301677
SM 9223 B Quanti-Tray	E. coli by Colilert-18/QT by UWF in Pensacola	2301678, 2301679, 2301680, 2301681, 2301682, 2301683, 2301684, 2301685
SOP-PCR-1.0	Detection and quantification of human-specific HF183 Bacteroides genetic marker with quantitative polymerase chain reaction	2301686, 2301687
SOP-PCR-1.2	Detection and quantification of coastal bird specific Catellicoccus marimammalium Gull2 genetic marker with quantitative polymerase chain reaction	2301686, 2301687
SOP-PCR-1.3	Detection and quantification of bird specific Helicobacter GFD genetic marker with quantitative polymerase chain reaction	2301686, 2301687
SOP-PCR-1.4	Detection and quantification of Ruminant specific Bacteroidetes BacR genetic marker with quantitative polymerase chain reaction	2301686, 2301687
SOP-PCR-1.5	Detection and quantification of canine-specific DG3 Bacteroides genetic marker with quantitative polymerase chain reaction	2301686, 2301687

Preparation and Analysis Log

Ref. Method	Received Date	Prep Date/Time	Prepared By	Analysis Date/Time	Analyzed By	Associated Samples
SM 10200 H (mod.)	01/26/2022	01/26/2022 13:52	Jillian G Hanley	02/02/2022 08:51	Joel Wharton	2301677
SM 9223 B Quanti-Tray	01/25/2022	01/25/2022 16:50		01/26/2022 13:15	UWF Lab	2301678, 2301679, 2301680, 2301681, 2301682, 2301683, 2301684, 2301685
SOP-PCR-1.0	01/26/2022	01/26/2022 14:10	Luke Hudson	02/23/2022 14:18	DeAsia Armster	2301686, 2301687
SOP-PCR-1.2	01/26/2022	01/26/2022 14:10	Luke Hudson	03/04/2022 12:05	DeAsia Armster	2301686, 2301687
SOP-PCR-1.3	01/26/2022	01/26/2022 14:10	Luke Hudson	03/07/2022 10:16	DeAsia Armster	2301686, 2301687
SOP-PCR-1.4	01/26/2022	01/26/2022 14:10	Luke Hudson	03/01/2022 15:10	DeAsia Armster	2301686, 2301687
SOP-PCR-1.5	01/26/2022	01/26/2022 14:10	Luke Hudson	03/03/2022 09:56	DeAsia Armster	2301686, 2301687