

Biological Analysis Report

NW-DIST-2023-04-19-05

Florida DEP 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: **Laurel Hill East**
Request ID: **RQ-2023-04-17-26**
Customer: **NW-DIST**
Project ID: **SMP**

Send Reports to:
FL Dept. of Environmental Protection
160 W Government Street
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: Sarah Menz, Environmental Administrator

Date Certified: 25-MAY-2023 14:18

A handwritten signature in black ink, appearing to be 'Sarah Menz', written in a cursive style.

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: Pine Log Creek Upstream of Long Road

Collection Date/Time: 04/18/2023 10:12

Field ID: G4NW0315

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2401440	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	201		MPN/100 mL		

Sample Location: Turkey Creek Upstream of CR 2A

Collection Date/Time: 04/18/2023 11:20

Field ID: G4NW0527

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2401441	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	134		MPN/100 mL		

Sample Location: Gum Creek

Collection Date/Time: 04/18/2023 12:15

Field ID: G4NW0382

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2401439	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	175		MPN/100 mL		
2401445	SOP-PCR-1.0	HF183-qPCR**	3.3E+03	U	TSC/100 mL	P428319	
	SOP-PCR-1.2	GULL2-qPCR**	3.6E+03	U	TSC/100 mL	P428319	
	SOP-PCR-1.3	GFD-qPCR**	720	U	TSC/100 mL	P428319	
	SOP-PCR-1.4	BacR-qPCR**	3.0E+03	U	TSC/100 mL	P428319	
	SOP-PCR-1.5	DG3-qPCR**	2.6E+03	U	TSC/100 mL	P428319	

Sample Location: Gum Creek Above Bonita Drive

Collection Date/Time: 04/18/2023 12:45

Field ID: G4NW0381

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2401442	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	97		MPN/100 mL		

Sample Location: Narrows Creek West of Kings Lake Rd.

Collection Date/Time: 04/18/2023 11:42

Field ID: G4NW0013

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2401443	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	185		MPN/100 mL		

Sample Location: East Branch Pine Log Creek Above Bear Bay Flats Road

Collection Date/Time: 04/18/2023 10:40

Field ID: G4NW0208

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2401444	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	20		MPN/100 mL		
2401458	SM 10150 B	Chlorophyll-a, Corrected	6.1		ug/L	P429294	
		Phaeophytin-a	1.2	U	ug/L	P429294	
		Chlorophyll-a, Uncorrected	6.6		ug/L	P429294	

Ref. Method and Comment:

SM 10150 B: 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

Quality Assurance Report Method Blank Results

Reference Method: SM 10150 B
Batch ID: P429294

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: P428319

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

Reference Method: SOP-PCR-1.2
Batch ID: P428319

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

Reference Method: SOP-PCR-1.3
Batch ID: P428319

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

Reference Method: SOP-PCR-1.4
Batch ID: P428319

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

Reference Method: SOP-PCR-1.5
Batch ID: P428319

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

Quality Assurance Report Laboratory Control Sample Accuracy

Reference Method: SM 10150 B
Batch ID: P429294

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
Chlorophyll-a, Corrected	100		P	84 - 116

Reference Method: SOP-PCR-1.0
Batch ID: P428319

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
HF183-qPCR	89.4	92.1	P/P	50 - 175

Quality Assurance Report Laboratory Control Sample Accuracy

Reference Method: SOP-PCR-1.2
Batch ID: P428319

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GULL2-qPCR	131	86.2	P/P	50 - 300

Reference Method: SOP-PCR-1.3
Batch ID: P428319

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GFD-qPCR	103	84.7	P/P	50 - 175

Reference Method: SOP-PCR-1.4
Batch ID: P428319

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
BacR-qPCR	107	92.4	P/P	50 - 175

Reference Method: SOP-PCR-1.5
Batch ID: P428319

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
DG3-qPCR	104	87.7	P/P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0
Batch ID: P428319

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2399390	HF183-qPCR	81.0		P	50 - 175
2399391	HF183-qPCR	82.6		P	50 - 175
2399480	HF183-qPCR	85.3		P	50 - 175
2399527	HF183-qPCR	82.6	82.6	P/P	50 - 175
2399793	HF183-qPCR	93.0		P	50 - 175
2399861	HF183-qPCR	90.3		P	50 - 175
2400270	HF183-qPCR	85.0		P	50 - 175
2400271	HF183-qPCR	90.6		P	50 - 175
2400272	HF183-qPCR	84.0		P	50 - 175
2400437	HF183-qPCR	90.5		P	50 - 175
2400438	HF183-qPCR	94.3		P	50 - 175
2400450	HF183-qPCR	107		P	50 - 175
2401445	HF183-qPCR	79.6		P	50 - 175

Reference Method: SOP-PCR-1.2
Batch ID: P428319

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2399390	GULL2-qPCR	30.3		F	50 - 300
2399391	GULL2-qPCR	74.5		P	50 - 300
2399480	GULL2-qPCR	91.4		P	50 - 300
2399861	GULL2-qPCR	87.8		P	50 - 300
2400271	GULL2-qPCR	84.0		P	50 - 300
2400272	GULL2-qPCR	80.1		P	50 - 300

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.2

Batch ID: P428319

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2400437	GULL2-qPCR	77.6		P	50 - 300
2400438	GULL2-qPCR	95.9		P	50 - 300
2400450	GULL2-qPCR	89.4		P	50 - 300
2401445	GULL2-qPCR	67.4		P	50 - 300

Reference Method: SOP-PCR-1.3

Batch ID: P428319

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2399390	GFD-qPCR	101		P	50 - 175
2399391	GFD-qPCR	99.7		P	50 - 175
2399480	GFD-qPCR	98.4		P	50 - 175
2399861	GFD-qPCR	94.9		P	50 - 175
2400271	GFD-qPCR	111		P	50 - 175
2400272	GFD-qPCR	110		P	50 - 175
2400437	GFD-qPCR	99.4		P	50 - 175
2400438	GFD-qPCR	108		P	50 - 175
2400450	GFD-qPCR	108		P	50 - 175
2401445	GFD-qPCR	102		P	50 - 175

Reference Method: SOP-PCR-1.4

Batch ID: P428319

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2399390	BacR-qPCR	81.9		P	50 - 175
2399391	BacR-qPCR	89.0		P	50 - 175
2399480	BacR-qPCR	103		P	50 - 175
2399527	BacR-qPCR	66.0	62.7	P/P	50 - 175
2399793	BacR-qPCR	94.8		P	50 - 175
2399861	BacR-qPCR	99.1		P	50 - 175
2400270	BacR-qPCR	94.5		P	50 - 175
2400271	BacR-qPCR	91.6		P	50 - 175
2400272	BacR-qPCR	90.3		P	50 - 175
2400437	BacR-qPCR	94.0		P	50 - 175
2400438	BacR-qPCR	103		P	50 - 175
2400450	BacR-qPCR	105		P	50 - 175
2401445	BacR-qPCR	79.7		P	50 - 175

Reference Method: SOP-PCR-1.5

Batch ID: P428319

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2399390	DG3-qPCR	75.8		P	50 - 175
2399391	DG3-qPCR	91.5		P	50 - 175
2399480	DG3-qPCR	93.6		P	50 - 175
2399527	DG3-qPCR	90.2	88.0	P/P	50 - 175
2399793	DG3-qPCR	94.7		P	50 - 175
2399861	DG3-qPCR	92.2		P	50 - 175
2400270	DG3-qPCR	91.8		P	50 - 175
2400271	DG3-qPCR	100		P	50 - 175
2400272	DG3-qPCR	94.4		P	50 - 175
2400437	DG3-qPCR	93.4		P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.5
Batch ID: P428319

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2400438	DG3-qPCR	101		P	50 - 175
2400450	DG3-qPCR	102		P	50 - 175
2401445	DG3-qPCR	81.7		P	50 - 175

Quality Assurance Report Precision

Reference Method: SOP-PCR-1.0
Batch ID: P428319

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2399527	HF183-qPCR	0.101	Spike	P	0 - 25

Reference Method: SOP-PCR-1.2
Batch ID: P428319

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2399527	GULL2-qPCR	8.09	Spike	P	0 - 25

Reference Method: SOP-PCR-1.3
Batch ID: P428319

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2399527	GFD-qPCR	2.83	Spike	P	0 - 25

Reference Method: SOP-PCR-1.4
Batch ID: P428319

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2399527	BacR-qPCR	5.18	Spike	P	0 - 25

Reference Method: SOP-PCR-1.5
Batch ID: P428319

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2399527	DG3-qPCR	2.44	Spike	P	0 - 25

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

Quality Assurance Report Surrogates

Quality Assurance Report Surrogates

Lab Sample ID: 2401445
 Field Sample ID: G4NW0382

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	83.8	P	50 - 300
SOP-PCR-1.2	SKETA-qPCR	110	P	50 - 300
SOP-PCR-1.3	SKETA-qPCR	100	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	107	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	86.3	P	50 - 300

Quality Assurance Report Summary

Ref. Method	Analyte	LCS % Recovery		MS % Recovery			LCS	Precision SMP	MS
SM 10150 B	Chlorophyll-a, Corrected	100							
SOP-PCR-1.0	HF183-qPCR	92.1	89.4	93.0	94.3	107			0.101
				79.6					
SOP-PCR-1.2	GULL2-qPCR	86.2	131	30.3	74.5	91.4			8.09
				87.8					
SOP-PCR-1.3	GFD-qPCR	84.7	103	101	99.7	98.4			2.83
				94.9					
SOP-PCR-1.4	BacR-qPCR	107	92.4	81.9	89.0	103			5.18
				66.0					
SOP-PCR-1.5	DG3-qPCR	87.7	104	94.7	75.8	91.5			2.44
				93.6					

Reference Method Descriptions

Method	Description	Associated Samples
SM 10150 B	Phytoplankton chlorophyll-a corrected, uncorrected, and phaeophytin by spectrophotometry	2401458
SM 9223 B Quanti-Tray	E. coli by Colilert-18/QT by UWF in Pensacola	2401439, 2401440, 2401441, 2401442, 2401443, 2401444
SOP-PCR-1.0	Detection and quantification of human-specific HF183 Bacteroides genetic marker with quantitative polymerase chain reaction	2401445
SOP-PCR-1.2	Detection and quantification of coastal bird specific Catellicoccus marimammalium Gull2 genetic marker with quantitative polymerase chain reaction	2401445
SOP-PCR-1.3	Detection and quantification of bird specific Helicobacter GFD genetic marker with quantitative polymerase chain reaction	2401445
SOP-PCR-1.4	Detection and quantification of Ruminant specific Bacteroidetes BacR genetic marker with quantitative polymerase chain reaction	2401445
SOP-PCR-1.5	Detection and quantification of canine-specific DG3 Bacteroides genetic marker with quantitative polymerase chain reaction	2401445

Preparation and Analysis Log

Ref. Method	Received Date	Prep Date/Time	Prepared By	Analysis Date/Time	Analyzed By	Associated Samples
SM 10150 B	04/19/2023	04/19/2023 14:11	Tynan A Branch	05/01/2023 10:21	Callie Williams	2401458
SM 9223 B Quanti-Tray	04/18/2023	04/18/2023 15:35		04/19/2023 12:32	UWF Lab	2401439, 2401440, 2401441, 2401442, 2401443, 2401444
SOP-PCR-1.0	04/19/2023	04/19/2023 15:05	Claudia Castro	04/21/2023 10:24	Roberta Tatti	2401445
SOP-PCR-1.2	04/19/2023	04/19/2023 15:05	Claudia Castro	05/09/2023 13:11	Jennifer Mihalic	2401445
SOP-PCR-1.3	04/19/2023	04/19/2023 15:05	Claudia Castro	04/28/2023 15:38	Jennifer Mihalic	2401445
SOP-PCR-1.4	04/19/2023	04/19/2023 15:05	Claudia Castro	04/25/2023 09:39	Roberta Tatti	2401445
SOP-PCR-1.5	04/19/2023	04/19/2023 15:05	Claudia Castro	04/26/2023 08:58	Roberta Tatti	2401445