

Biological Analysis Report

SE-DIST-2023-07-28-01

Florida DEP Laboratory
2600 Blair Stone Road
Tallahassee, FL 32399-2400
DOH Accreditation E31780

Event Description: **Highlands Run**
Request ID: **RQ-2023-07-24-33**
Customer: **SE-DIST**
Project ID: **SMP**

Send Reports to:
FL Dept. of Environmental Protection
FL Dept. of Environmental Protection
2199 South Rock Road
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Certified by: Sarah Menz, Environmental Administrator

Date Certified: 11-SEP-2023 10:44

A handwritten signature in black ink, appearing to be 'Sarah Menz', with a long, sweeping horizontal stroke extending to the right.

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, Microbiology and Molecular.

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: INDIAN PRAIRIE CANAL UPSTREAM OF SR 70 **Collection Date/Time: 07/27/2023 09:45**

Field ID: G4SE0152

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2427492	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	124.6	Q	MPN/100 mL	P433224	

Sample Location: ARBUCKLE CREEK AT US HWY 98 **Collection Date/Time: 07/27/2023 11:00**

Field ID: G4SE0150

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2427488	SM 10150 B	Chlorophyll-a, Corrected	12		ug/L	P433336	
		Phaeophytin-a	5.2		ug/L	P433336	
		Chlorophyll-a, Uncorrected	15		ug/L	P433336	
2427490	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	1553.1	Q	MPN/100 mL	P433224	

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.

Sample Location: ARBUCKLE BRANCH AT ARBUCKLE CREEK RD **Collection Date/Time: 07/27/2023 11:30**

Field ID: G4SE0149

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2427489	SM 10150 B	Chlorophyll-a, Corrected	28		ug/L	P433336	
		Phaeophytin-a	9.1	I	ug/L	P433336	
		Chlorophyll-a, Uncorrected	34		ug/L	P433336	
2427491	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	648.8	Q	MPN/100 mL	P433224	
2427495	SOP-PCR-1.0	HF183-qPCR**	6.6E+03	U	TSC/100 mL	P432974	
	SOP-PCR-1.4	BacR-qPCR**	6.7E+04		TSC/100 mL	P432974	
	SOP-PCR-1.5	DG3-qPCR**	5.1E+03	U	TSC/100 mL	P432974	

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.

Sample Location: KISSIMMEE TRIBUTARY AT US 98 EAST OF BUCKHORN **Collection Date/Time: 07/27/2023 12:00**

Field ID: G4SE0155

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2427493	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	79.4	Q	MPN/100 mL	P433224	
2427496	SOP-PCR-1.0	HF183-qPCR**	2.2E+03	U	TSC/100 mL	P432974	
	SOP-PCR-1.2	GULL2-qPCR**	2.4E+03	U	TSC/100 mL	P432974	
	SOP-PCR-1.3	GFD-qPCR**	2.1E+03	I	TSC/100 mL	P432974	
	SOP-PCR-1.4	BacR-qPCR**	2.0E+03	U	TSC/100 mL	P432974	
	SOP-PCR-1.5	DG3-qPCR**	1.7E+03	U	TSC/100 mL	P432974	

Sample Location: KISSIMMEE TRIBUTARY AT US 98 WEST OF CORNWELL **Collection Date/Time: 07/27/2023 12:15**

Field ID: G4SE0154

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2427494	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	2419.6	JQ	MPN/100 mL	P433224	

Field ID: G4SE0154

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
Ref. Method and Comment: SM 9223 B Quanti-Tray: All Quanti-Tray wells were positive. Result may be higher than reported.							

Quality Assurance Report
Method Blank Results

Reference Method: SM 10150 B
Batch ID: P433336

Component	Result	Code	Units
Chlorophyll-a, Corrected	0.89	U	ug/L
Chlorophyll-a, Uncorrected	0.65	U	ug/L
Phaeophytin-a	0.98	U	ug/L

Reference Method: SM 9223 B Quanti-Tray
Batch ID: P433224

Component	Result	Code	Units
Escherichia Coli-Quanti-Tray	1.0	U	MPN/100 mL

Reference Method: SOP-PCR-1.0
Batch ID: P432974

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

Reference Method: SOP-PCR-1.2
Batch ID: P432974

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

Reference Method: SOP-PCR-1.3
Batch ID: P432974

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

Reference Method: SOP-PCR-1.4
Batch ID: P432974

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

Reference Method: SOP-PCR-1.5
Batch ID: P432974

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

Quality Assurance Report
Laboratory Control Sample Accuracy

Quality Assurance Report Laboratory Control Sample Accuracy

Reference Method: SM 10150 B
Batch ID: P433336

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
Chlorophyll-a, Corrected	101		P	84 - 116

Reference Method: SOP-PCR-1.0
Batch ID: P432974

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
HF183-qPCR	80.8	99.7	P/P	50 - 175

Reference Method: SOP-PCR-1.2
Batch ID: P432974

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GULL2-qPCR	92.9	98.8	P/P	50 - 300

Reference Method: SOP-PCR-1.3
Batch ID: P432974

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GFD-qPCR	94.5	99.3	P/P	50 - 175

Reference Method: SOP-PCR-1.4
Batch ID: P432974

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
BacR-qPCR	108	69.2	P/P	50 - 175

Reference Method: SOP-PCR-1.5
Batch ID: P432974

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
DG3-qPCR	99.1	71.6	P/P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0
Batch ID: P432974

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2426361	HF183-qPCR	103	109	P/P	50 - 175
2426802	HF183-qPCR	96.7		P	50 - 175
2427046	HF183-qPCR	95.4		P	50 - 175
2427265	HF183-qPCR	94.0		P	50 - 175
2427495	HF183-qPCR	92.4		P	50 - 175
2427496	HF183-qPCR	88.6		P	50 - 175
2427519	HF183-qPCR	85.8		P	50 - 175
2427528	HF183-qPCR	107		P	50 - 175
2427529	HF183-qPCR	102		P	50 - 175
2427530	HF183-qPCR	113		P	50 - 175
2427531	HF183-qPCR	113		P	50 - 175
2427532	HF183-qPCR	111		P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0

Batch ID: P432974

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2427533	HF183-qPCR	115		P	50 - 175

Reference Method: SOP-PCR-1.2

Batch ID: P432974

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2426361	GULL2-qPCR	77.1	82.7	P/P	50 - 300
2426802	GULL2-qPCR	79.5		P	50 - 300
2427046	GULL2-qPCR	80.5		P	50 - 300
2427265	GULL2-qPCR	78.5		P	50 - 300
2427496	GULL2-qPCR	77.0		P	50 - 300
2427519	GULL2-qPCR	53.4		P	50 - 300
2427528	GULL2-qPCR	75.8		P	50 - 300
2427529	GULL2-qPCR	73.3		P	50 - 300
2427530	GULL2-qPCR	77.9		P	50 - 300
2427531	GULL2-qPCR	83.5		P	50 - 300
2427532	GULL2-qPCR	82.2		P	50 - 300
2427533	GULL2-qPCR	88.7		P	50 - 300

Reference Method: SOP-PCR-1.3

Batch ID: P432974

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2426361	GFD-qPCR	99.0	99.4	P/P	50 - 175
2426802	GFD-qPCR	95.4		P	50 - 175
2427046	GFD-qPCR	92.3		P	50 - 175
2427265	GFD-qPCR	88.5		P	50 - 175
2427496	GFD-qPCR	86.7		P	50 - 175
2427519	GFD-qPCR	90.4		P	50 - 175
2427528	GFD-qPCR	95.0		P	50 - 175
2427529	GFD-qPCR	85.0		P	50 - 175
2427530	GFD-qPCR	79.7		P	50 - 175
2427531	GFD-qPCR	77.3		P	50 - 175
2427532	GFD-qPCR	84.1		P	50 - 175
2427533	GFD-qPCR	87.8		P	50 - 175

Reference Method: SOP-PCR-1.4

Batch ID: P432974

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2426361	BacR-qPCR	102	95.4	P/P	50 - 175
2426802	BacR-qPCR	90.8		P	50 - 175
2427046	BacR-qPCR	86.3		P	50 - 175
2427265	BacR-qPCR	80.3		P	50 - 175
2427495	BacR-qPCR	69.6		P	50 - 175
2427496	BacR-qPCR	89.7		P	50 - 175
2427519	BacR-qPCR	89.8		P	50 - 175

Reference Method: SOP-PCR-1.5

Batch ID: P432974

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
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Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.5
Batch ID: P432974

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2426361	DG3-qPCR	105	109	P/P	50 - 175
2426802	DG3-qPCR	103		P	50 - 175
2427046	DG3-qPCR	105		P	50 - 175
2427265	DG3-qPCR	99.4		P	50 - 175
2427495	DG3-qPCR	97.7		P	50 - 175
2427496	DG3-qPCR	90.8		P	50 - 175
2427519	DG3-qPCR	92.6		P	50 - 175
2427528	DG3-qPCR	112		P	50 - 175
2427529	DG3-qPCR	110		P	50 - 175
2427530	DG3-qPCR	145		P	50 - 175
2427531	DG3-qPCR	115		P	50 - 175
2427532	DG3-qPCR	114		P	50 - 175
2427533	DG3-qPCR	120		P	50 - 175

Quality Assurance Report Precision

Reference Method: SOP-PCR-1.0
Batch ID: P432974

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2426361	HF183-qPCR	6.09	Spike	P	0 - 25

Reference Method: SOP-PCR-1.2
Batch ID: P432974

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2426361	GULL2-qPCR	6.95	Spike	P	0 - 25

Reference Method: SOP-PCR-1.3
Batch ID: P432974

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2426361	GFD-qPCR	0.487	Spike	P	0 - 25

Reference Method: SOP-PCR-1.4
Batch ID: P432974

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2426361	BacR-qPCR	6.67	Spike	P	0 - 25

Reference Method: SOP-PCR-1.5
Batch ID: P432974

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2426361	DG3-qPCR	3.90	Spike	P	0 - 25

Quality Assurance Report
Precision

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

Quality Assurance Report
Surrogates

Lab Sample ID: 2427495
Field Sample ID: G4SE0149

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	90.8	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	91.8	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	87.3	P	50 - 300

Lab Sample ID: 2427496
Field Sample ID: G4SE0155

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	126	P	50 - 300
SOP-PCR-1.2	SKETA-qPCR	99.5	P	50 - 300
SOP-PCR-1.3	SKETA-qPCR	97.5	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	101	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	112	P	50 - 300

Quality Assurance Report
Summary

Ref. Method	Analyte	LCS % Recovery		MS % Recovery			LCS	Precision SMP	MS
SM 10150 B	Chlorophyll-a, Corrected	101							
SOP-PCR-1.0	HF183-qPCR	99.7	80.8	103	109	96.7			6.09
				95.4					
SOP-PCR-1.2	GULL2-qPCR	98.8	92.9	77.1	82.7	79.5			6.95
				80.5					
SOP-PCR-1.3	GFD-qPCR	99.3	94.5	99.0	99.4	95.4			0.487
				92.3					
SOP-PCR-1.4	BacR-qPCR	69.2	108	102	95.4	90.8			6.67
				86.3					
SOP-PCR-1.5	DG3-qPCR	71.6	99.1	105	109	103			3.90
				105					

Reference Method Descriptions

Method	Description	Associated Samples
SM 10150 B	Phytoplankton chlorophyll-a corrected, uncorrected, and phaeophytin by spectrophotometry	2427488, 2427489
SM 9223 B Quanti-Tray	Escherichia coli by Colilert 18 Quanti-Tray method - non-chlorinated water systems	2427490, 2427491, 2427492, 2427493, 2427494
SOP-PCR-1.0	Detection and quantification of human-specific HF183 Bacteroides genetic marker with quantitative polymerase chain reaction	2427495, 2427496
SOP-PCR-1.2	Detection and quantification of coastal bird specific Catellicoccus marimammalium Gull2 genetic marker with quantitative polymerase chain reaction	2427496
SOP-PCR-1.3	Detection and quantification of bird specific Helicobacter GFD genetic marker with quantitative polymerase chain reaction	2427496
SOP-PCR-1.4	Detection and quantification of Ruminant specific Bacteroidetes BacR genetic marker with quantitative polymerase chain reaction	2427495, 2427496

Reference Method Descriptions

Method	Description	<u>Associated Samples</u>
SOP-PCR-1.5	Detection and quantification of canine-specific DG3 Bacteroides genetic marker with quantitative polymerase chain reaction	2427495, 2427496

Preparation and Analysis Log

Ref. Method	Received Date	Prep Date/Time	Prepared By	Analysis Date/Time	Analyzed By	Associated Samples
SM 10150 B	07/28/2023	07/28/2023 13:39	Keanu Thijm	08/02/2023 08:51	Aidan Cronin	2427488, 2427489
SM 9223 B Quanti-Tray	07/28/2023	07/28/2023 15:02	Julia Herten	07/29/2023 12:38	Callie Williams	2427490, 2427491, 2427492, 2427493, 2427494
SOP-PCR-1.0	07/28/2023	07/28/2023 13:40	Claudia Castro	08/07/2023 12:37	Madeline Gruver	2427495, 2427496
SOP-PCR-1.2	07/28/2023	07/28/2023 13:40	Claudia Castro	08/09/2023 14:25	Roberta Tatti	2427496
SOP-PCR-1.3	07/28/2023	07/28/2023 13:40	Claudia Castro	08/10/2023 10:08	Roberta Tatti	2427496
SOP-PCR-1.4	07/28/2023	07/28/2023 13:40	Claudia Castro	08/08/2023 15:11	Madeline Gruver	2427495, 2427496
SOP-PCR-1.5	07/28/2023	07/28/2023 13:40	Claudia Castro	08/08/2023 17:19	Madeline Gruver	2427495, 2427496