

Biological Analysis Report

NW-DIST-2023-03-16-02

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 West**
Request ID: **RQ-2023-03-13-27**
Customer: **NW-DIST**
Project ID: **SMP**

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Date Certified: 21-APR-2023 14:14



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: 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: Oak Creek below Tommy Steele Road

Collection Date/Time: 03/15/2023 12:15

Field ID: G4NW0395

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2394516	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	52		MPN/100 mL		
2394522	SOP-PCR-1.0	HF183-qPCR**	3.3E+03	U	TSC/100 mL	P426669	
	SOP-PCR-1.2	GULL2-qPCR**	3.6E+03	U	TSC/100 mL	P426669	
	SOP-PCR-1.3	GFD-qPCR**	940	I	TSC/100 mL	P426669	
	SOP-PCR-1.4	BacR-qPCR**	3.0E+03	U	TSC/100 mL	P426669	
	SOP-PCR-1.5	DG3-qPCR**	2.6E+03	U	TSC/100 mL	P426669	

Ref. Method and Comment:
SOP-PCR-1.2: Precision data unavailable for Gull2-qPCR component.

Sample Location: Olive Branch below Mormon Temple Road

Collection Date/Time: 03/15/2023 11:50

Field ID: G4NW0161

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2394519	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	108		MPN/100 mL		

Sample Location: Davis Mill Creek below dam

Collection Date/Time: 03/15/2023 10:55

Field ID: G4NW0010

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2394520	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	85		MPN/100 mL		

Sample Location: Baggett Creek upstream of Hwy 90

Collection Date/Time: 03/15/2023 10:15

Field ID: G4NW0336

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2394518	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	41		MPN/100 mL		

Sample Location: Blackwater River upstream of Charles Booker Rd.

Collection Date/Time: 03/15/2023 12:35

Field ID: G4NW0012

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2394521	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	135		MPN/100 mL		

Sample Location: Wilkinson Creek upstream of Wilkinson Bluff Rd.

Collection Date/Time: 03/15/2023 09:55

Field ID: G4NW0357

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2394517	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	173		MPN/100 mL		
2394523	SOP-PCR-1.0	HF183-qPCR**	1.7E+03	U	TSC/100 mL	P426669	
	SOP-PCR-1.4	BacR-qPCR**	1.74E+04		TSC/100 mL	P426669	
	SOP-PCR-1.5	DG3-qPCR**	1.3E+03	U	TSC/100 mL	P426669	

**Quality Assurance Report
Method Blank Results**

Quality Assurance Report
Method Blank Results

Reference Method: SOP-PCR-1.0
Batch ID: P426669

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

Reference Method: SOP-PCR-1.2
Batch ID: P426669

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

Reference Method: SOP-PCR-1.3
Batch ID: P426669

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

Reference Method: SOP-PCR-1.4
Batch ID: P426669

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

Reference Method: SOP-PCR-1.5
Batch ID: P426669

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

Quality Assurance Report
Laboratory Control Sample Accuracy

Reference Method: SOP-PCR-1.0
Batch ID: P426669

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
HF183-qPCR	78.4	83.6	P/P	50 - 175

Reference Method: SOP-PCR-1.2
Batch ID: P426669

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GULL2-qPCR	100	84.1	P/P	50 - 300

Reference Method: SOP-PCR-1.3
Batch ID: P426669

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GFD-qPCR	87.4	113	P/P	50 - 175

Reference Method: SOP-PCR-1.4
Batch ID: P426669

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

Reference Method: SOP-PCR-1.4
Batch ID: P426669

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
BacR-qPCR	99.0	85.3	P/P	50 - 175

Reference Method: SOP-PCR-1.5
Batch ID: P426669

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
DG3-qPCR	99.8	80.7	P/P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0
Batch ID: P426669

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2391600	HF183-qPCR	86.1	90.5	P/P	50 - 175
2392452	HF183-qPCR	90.8		P	50 - 175
2393010	HF183-qPCR	96.5		P	50 - 175
2393144	HF183-qPCR	90.8		P	50 - 175
2393145	HF183-qPCR	92.1		P	50 - 175
2393146	HF183-qPCR	84.6		P	50 - 175
2393460	HF183-qPCR	88.6		P	50 - 175
2393667	HF183-qPCR	87.0		P	50 - 175
2393910	HF183-qPCR	92.9		P	50 - 175
2393911	HF183-qPCR	86.8		P	50 - 175
2393985	HF183-qPCR	103		P	50 - 175
2394522	HF183-qPCR	95.8		P	50 - 175
2394523	HF183-qPCR	92.5		P	50 - 175

Reference Method: SOP-PCR-1.2
Batch ID: P426669

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2392452	GULL2-qPCR	59.1		P	50 - 300
2393010	GULL2-qPCR	81.3		P	50 - 300
2393144	GULL2-qPCR	25.4		F	50 - 300
2393145	GULL2-qPCR	66.1		P	50 - 300
2393460	GULL2-qPCR	78.2		P	50 - 300
2393667	GULL2-qPCR	54.5		P	50 - 300
2393910	GULL2-qPCR	82.5		P	50 - 300
2393911	GULL2-qPCR	82.3		P	50 - 300
2393985	GULL2-qPCR	76.7		P	50 - 300
2394522	GULL2-qPCR	82.1		P	50 - 300

Reference Method: SOP-PCR-1.3
Batch ID: P426669

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2392452	GFD-qPCR	91.6		P	50 - 175
2393010	GFD-qPCR	89.4		P	50 - 175
2393144	GFD-qPCR	92.4		P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.3

Batch ID: P426669

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2393145	GFD-qPCR	88.1		P	50 - 175
2393460	GFD-qPCR	93.6		P	50 - 175
2393667	GFD-qPCR	88.6		P	50 - 175
2393910	GFD-qPCR	94.0		P	50 - 175
2393911	GFD-qPCR	93.5		P	50 - 175
2393985	GFD-qPCR	87.9		P	50 - 175
2394522	GFD-qPCR	88.2		P	50 - 175

Reference Method: SOP-PCR-1.4

Batch ID: P426669

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2391600	BacR-qPCR	93.7	94.2	P/P	50 - 175
2392452	BacR-qPCR	79.9		P	50 - 175
2393010	BacR-qPCR	93.6		P	50 - 175
2393144	BacR-qPCR	69.3		P	50 - 175
2393145	BacR-qPCR	72.6		P	50 - 175
2393146	BacR-qPCR	64.4		P	50 - 175
2393460	BacR-qPCR	80.3		P	50 - 175
2393667	BacR-qPCR	73.6		P	50 - 175
2393910	BacR-qPCR	81.4		P	50 - 175
2393911	BacR-qPCR	78.5		P	50 - 175
2393985	BacR-qPCR	83.9		P	50 - 175
2394522	BacR-qPCR	77.3		P	50 - 175
2394523	BacR-qPCR	76.9		P	50 - 175

Reference Method: SOP-PCR-1.5

Batch ID: P426669

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2391600	DG3-qPCR	98.4	99.2	P/P	50 - 175
2392452	DG3-qPCR	91.5		P	50 - 175
2393010	DG3-qPCR	94.3		P	50 - 175
2393144	DG3-qPCR	78.8		P	50 - 175
2393145	DG3-qPCR	93.2		P	50 - 175
2393146	DG3-qPCR	86.5		P	50 - 175
2393460	DG3-qPCR	92.9		P	50 - 175
2393667	DG3-qPCR	85.0		P	50 - 175
2393910	DG3-qPCR	96.2		P	50 - 175
2393911	DG3-qPCR	94.9		P	50 - 175
2393985	DG3-qPCR	96.3		P	50 - 175
2394522	DG3-qPCR	99.8		P	50 - 175
2394523	DG3-qPCR	93.9		P	50 - 175

Quality Assurance Report Precision

Quality Assurance Report Precision

Reference Method: SOP-PCR-1.0
Batch ID: P426669

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2391600	HF183-qPCR	5.00	Spike	P	0 - 25

Reference Method: SOP-PCR-1.3
Batch ID: P426669

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2391600	GFD-qPCR	3.70	Spike	P	0 - 25

Reference Method: SOP-PCR-1.4
Batch ID: P426669

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2391600	BacR-qPCR	0.459	Spike	P	0 - 25

Reference Method: SOP-PCR-1.5
Batch ID: P426669

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2391600	DG3-qPCR	0.790	Spike	P	0 - 25

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

Quality Assurance Report Surrogates

Lab Sample ID: 2394522
Field Sample ID: G4NW0395

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	87.1	P	50 - 300
SOP-PCR-1.2	SKETA-qPCR	91.1	P	50 - 300
SOP-PCR-1.3	SKETA-qPCR	92.0	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	92.3	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	80.0	P	50 - 300

Lab Sample ID: 2394523
Field Sample ID: G4NW0357

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	70.9	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	84.7	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	65.7	P	50 - 300

Quality Assurance Report Summary

Ref. Method	Analyte	LCS % Recovery		MS % Recovery			LCS	Precision SMP	MS
SOP-PCR-1.0	HF183-qPCR	83.6	78.4	86.1	90.5	90.8			5.00
SOP-PCR-1.2	GULL2-qPCR	84.1	100	59.1	81.3	66.1			
SOP-PCR-1.3	GFD-qPCR	113	87.4	91.6	89.4	92.4			3.70
SOP-PCR-1.4	BacR-qPCR	85.3	99.0	93.7	94.2	79.9			0.459
SOP-PCR-1.5	DG3-qPCR	80.7	99.8	98.4	99.2	91.5			0.790

Reference Method Descriptions

Method	Description	Associated Samples
SM 9223 B Quanti-Tray	E. coli by Colilert-18/QT by UWF in Pensacola	2394516, 2394517, 2394518, 2394519, 2394520, 2394521
SOP-PCR-1.0	Detection and quantification of human-specific HF183 Bacteroides genetic marker with quantitative polymerase chain reaction	2394522, 2394523
SOP-PCR-1.2	Detection and quantification of coastal bird specific Catellicoccus marimammalium Gull2 genetic marker with quantitative polymerase chain reaction	2394522
SOP-PCR-1.3	Detection and quantification of bird specific Helicobacter GFD genetic marker with quantitative polymerase chain reaction	2394522
SOP-PCR-1.4	Detection and quantification of Ruminant specific Bacteroidetes BacR genetic marker with quantitative polymerase chain reaction	2394522, 2394523
SOP-PCR-1.5	Detection and quantification of canine-specific DG3 Bacteroides genetic marker with quantitative polymerase chain reaction	2394522, 2394523

Preparation and Analysis Log

Ref. Method	Received Date	Prep Date/Time	Prepared By	Analysis Date/Time	Analyzed By	Associated Samples
SM 9223 B Quanti-Tray	03/15/2023	03/15/2023 16:35		03/16/2023 12:40	UWF Lab	2394516, 2394517, 2394518, 2394519, 2394520, 2394521
SOP-PCR-1.0	03/16/2023	03/16/2023 14:35	Claudia Castro	03/29/2023 09:41	Roberta Tatti	2394522, 2394523
SOP-PCR-1.2	03/16/2023	03/16/2023 14:35	Claudia Castro	03/31/2023 13:54	Jennifer Mihalic	2394522
SOP-PCR-1.3	03/16/2023	03/16/2023 14:35	Claudia Castro	04/04/2023 13:30	Roberta Tatti	2394522
SOP-PCR-1.4	03/16/2023	03/16/2023 14:35	Claudia Castro	03/30/2023 09:10	Roberta Tatti	2394522, 2394523
SOP-PCR-1.5	03/16/2023	03/16/2023 14:35	Claudia Castro	03/30/2023 14:20	Roberta Tatti	2394522, 2394523