

Biological Analysis Report

NW-DIST-2022-04-27-03

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-2022-04-25-28**
Customer: **NW-DIST**
Project ID: **SMP**

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Date Certified: 09-JUN-2022 14:23



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: Little Alaqua Creek Below 282 Bridge

Collection Date/Time: 04/26/2022 12:10

Field ID: G3NW0065

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2322246	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	266		MPN/100 mL		

Sample Location: Seven Runs upstream of HWY 81

Collection Date/Time: 04/26/2022 10:25

Field ID: G3NW0116

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2322243	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	181		MPN/100 mL		

Sample Location: Little Alaqua Creek above Eglin AFB Road 208

Collection Date/Time: 04/26/2022 12:25

Field ID: G3NW0453

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2322247	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	228		MPN/100 mL		

Sample Location: Pine Log Creek upstream of HWY 20

Collection Date/Time: 04/26/2022 10:50

Field ID: G3NW0454

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2322244	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	31		MPN/100 mL		

Sample Location: Lafayette Creek

Collection Date/Time: 04/26/2022 11:35

Field ID: G3NW0455

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2322245	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	331		MPN/100 mL		

Sample Location: Bay Branch upstream of HWY 331

Collection Date/Time: 04/26/2022 13:00

Field ID: G3NW0171

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2322242	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	121		MPN/100 mL		
2322248	SOP-PCR-1.0	HF183-qPCR**	3.3E+03	U	TSC/100 mL	P412650	
	SOP-PCR-1.2	GULL2-qPCR**	3.6E+03	U	TSC/100 mL	P412650	
	SOP-PCR-1.3	GFD-qPCR**	720	U	TSC/100 mL	P412650	
	SOP-PCR-1.4	BacR-qPCR**	3.0E+03	U	TSC/100 mL	P412650	
	SOP-PCR-1.5	DG3-qPCR**	2.6E+03	U	TSC/100 mL	P412650	

Ref. Method and Comment:

SOP-PCR-1.0: Precision data unavailable for HF183-qPCR component.

SOP-PCR-1.2: Precision data unavailable for Gull2-qPCR component.

SOP-PCR-1.3: Precision data unavailable for GFD-qPCR component.

SOP-PCR-1.4: Precision data unavailable for BacR-qPCR component.

SOP-PCR-1.5: Precision data unavailable for DG3-qPCR component.

Quality Assurance Report

Method Blank Results

Quality Assurance Report Method Blank Results

Reference Method: SOP-PCR-1.0
Batch ID: P412650

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

Reference Method: SOP-PCR-1.2
Batch ID: P412650

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

Reference Method: SOP-PCR-1.3
Batch ID: P412650

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

Reference Method: SOP-PCR-1.4
Batch ID: P412650

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

Reference Method: SOP-PCR-1.5
Batch ID: P412650

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: P412650

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
HF183-qPCR	91.9	85.4	P/P	50 - 175

Reference Method: SOP-PCR-1.2
Batch ID: P412650

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GULL2-qPCR	91.4	148	P/P	50 - 300

Reference Method: SOP-PCR-1.3
Batch ID: P412650

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GFD-qPCR	89.0	71.7	P/P	50 - 175

Reference Method: SOP-PCR-1.4
Batch ID: P412650

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: P412650

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
BacR-qPCR	52.3	71.4	P/P	50 - 175

Reference Method: SOP-PCR-1.5
Batch ID: P412650

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
DG3-qPCR	83.8	78.1	P/P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0
Batch ID: P412650

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2321141	HF183-qPCR	85.2		P	50 - 175
2321439	HF183-qPCR	80.9		P	50 - 175
2322177	HF183-qPCR	78.5		P	50 - 175
2322178	HF183-qPCR	89.8		P	50 - 175
2322179	HF183-qPCR	81.3		P	50 - 175
2322248	HF183-qPCR	74.7		P	50 - 175
2322346	HF183-qPCR	84.3		P	50 - 175
2322347	HF183-qPCR	85.4		P	50 - 175
2322575	HF183-qPCR	80.4		P	50 - 175
2322576	HF183-qPCR	79.4		P	50 - 175
2322666	HF183-qPCR	93.7		P	50 - 175
2322667	HF183-qPCR	101		P	50 - 175

Reference Method: SOP-PCR-1.2
Batch ID: P412650

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2321439	GULL2-qPCR	53.1		P	50 - 300
2322177	GULL2-qPCR	80.3		P	50 - 300
2322178	GULL2-qPCR	106		P	50 - 300
2322179	GULL2-qPCR	121		P	50 - 300
2322248	GULL2-qPCR	108		P	50 - 300
2322346	GULL2-qPCR	71.2		P	50 - 300
2322347	GULL2-qPCR	91.9		P	50 - 300
2322666	GULL2-qPCR	98.6		P	50 - 300
2322667	GULL2-qPCR	84.7		P	50 - 300

Reference Method: SOP-PCR-1.3
Batch ID: P412650

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2321439	GFD-qPCR	87.3		P	50 - 175
2322177	GFD-qPCR	75.5		P	50 - 175
2322178	GFD-qPCR	81.1		P	50 - 175
2322179	GFD-qPCR	77.8		P	50 - 175
2322248	GFD-qPCR	85.4		P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.3
Batch ID: P412650

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2322346	GFD-qPCR	77.8		P	50 - 175
2322347	GFD-qPCR	77.6		P	50 - 175
2322575	GFD-qPCR	76.4		P	50 - 175
2322576	GFD-qPCR	75.8		P	50 - 175
2322666	GFD-qPCR	82.5		P	50 - 175
2322667	GFD-qPCR	69.5		P	50 - 175

Reference Method: SOP-PCR-1.4
Batch ID: P412650

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2321141	BacR-qPCR	58.0		P	50 - 175
2322178	BacR-qPCR	53.8		P	50 - 175
2322179	BacR-qPCR	66.6		P	50 - 175
2322248	BacR-qPCR	55.9		P	50 - 175
2322347	BacR-qPCR	63.1		P	50 - 175
2322575	BacR-qPCR	72.9		P	50 - 175
2322666	BacR-qPCR	66.2		P	50 - 175
2322667	BacR-qPCR	58.9		P	50 - 175

Reference Method: SOP-PCR-1.5
Batch ID: P412650

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2321141	DG3-qPCR	78.4		P	50 - 175
2321439	DG3-qPCR	61.3		P	50 - 175
2322177	DG3-qPCR	66.3		P	50 - 175
2322178	DG3-qPCR	72.9		P	50 - 175
2322179	DG3-qPCR	70.1		P	50 - 175
2322248	DG3-qPCR	70.7		P	50 - 175
2322346	DG3-qPCR	69.3		P	50 - 175
2322347	DG3-qPCR	64.4		P	50 - 175
2322666	DG3-qPCR	78.9		P	50 - 175
2322667	DG3-qPCR	72.7		P	50 - 175

Quality Assurance Report Surrogates

Lab Sample ID: 2322248
Field Sample ID: G3NW0171

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	70.6	P	50 - 300
SOP-PCR-1.2	SKETA-qPCR	128	P	50 - 300
SOP-PCR-1.3	SKETA-qPCR	92.1	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	86.1	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	74.2	P	50 - 300

Quality Assurance Report Summary

Ref. Method	Analyte	LCS % Recovery		MS % Recovery			LCS	Precision SMP	MS
SOP-PCR-1.0	HF183-qPCR	85.4	91.9	85.2	80.9	78.5			
SOP-PCR-1.2	GULL2-qPCR	148	91.4	53.1	80.3	106			
SOP-PCR-1.3	GFD-qPCR	71.7	89.0	87.3	75.5	81.1			
SOP-PCR-1.4	BacR-qPCR	71.4	52.3	58.0	53.8	66.6			
SOP-PCR-1.5	DG3-qPCR	78.1	83.8	78.4	61.3	66.3			

Reference Method Descriptions

Method	Description	Associated Samples
SM 9223 B Quanti-Tray	E. coli by Colilert-18/QT by UWF in Pensacola	2322242, 2322243, 2322244, 2322245, 2322246, 2322247
SOP-PCR-1.0	Detection and quantification of human-specific HF183 Bacteroides genetic marker with quantitative polymerase chain reaction	2322248
SOP-PCR-1.2	Detection and quantification of coastal bird specific Catellicoccus marimammalium Gull2 genetic marker with quantitative polymerase chain reaction	2322248
SOP-PCR-1.3	Detection and quantification of bird specific Helicobacter GFD genetic marker with quantitative polymerase chain reaction	2322248
SOP-PCR-1.4	Detection and quantification of Ruminant specific Bacteroidetes BacR genetic marker with quantitative polymerase chain reaction	2322248
SOP-PCR-1.5	Detection and quantification of canine-specific DG3 Bacteroides genetic marker with quantitative polymerase chain reaction	2322248

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	04/26/2022	04/26/2022 15:55		04/27/2022 12:20	UWF Lab	2322242, 2322243, 2322244, 2322245, 2322246, 2322247
SOP-PCR-1.0	04/27/2022	04/27/2022 13:00	Jennifer Mihalic	05/25/2022 14:08	DeAsia Armster	2322248
SOP-PCR-1.2	04/27/2022	04/27/2022 13:00	Jennifer Mihalic	05/27/2022 11:26	DeAsia Armster	2322248
SOP-PCR-1.3	04/27/2022	04/27/2022 13:00	Jennifer Mihalic	05/27/2022 13:50	Jennifer Mihalic	2322248
SOP-PCR-1.4	04/27/2022	04/27/2022 13:00	Jennifer Mihalic	05/26/2022 12:54	Jennifer Mihalic	2322248
SOP-PCR-1.5	04/27/2022	04/27/2022 13:00	Jennifer Mihalic	05/27/2022 10:16	DeAsia Armster	2322248