

Biological Analysis Report

NW-DIST-2021-12-07-01

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: **Alaqua Whaler Run**
Request ID: **RQ-2021-12-06-46**
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 K 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: 19-JAN-2022 09:55



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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: Microbiology, 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: Joes Bayou

Collection Date/Time: 12/06/2021 11:35

Field ID: G3NW0182

Matrix: W-SURF-SLT

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2293466	SM 9222 D	Fecal Coliforms-Membrane Filter	25	BQ	cfu/100 mL	P407353	
2293467	Enterolert/QT	Enterococci-Quanti-Tray	10	U	MPN/100 mL		
	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	52		MPN/100 mL		

Ref. Method and Comment:

SM 9222 D: Precision data for QC sample is unavailable due to the colony count falling outside of method acceptable range.

Sample Location: Shaw Still Branch upstream E College Blvd

Collection Date/Time: 12/06/2021 12:40

Field ID: G3NW0162

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2293468	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	168		MPN/100 mL		
2293469	SOP-PCR-1.0	HF183-qPCR**	1.7E+03	U	TSC/100 mL	P406337	
	SOP-PCR-1.2	GULL2-qPCR**	1.8E+03	U	TSC/100 mL	P406337	
	SOP-PCR-1.3	GFD-qPCR**	360	U	TSC/100 mL	P406337	
	SOP-PCR-1.4	BacR-qPCR**	1.5E+03	U	TSC/100 mL	P406337	
	SOP-PCR-1.5	DG3-qPCR**	1.3E+03	U	TSC/100 mL	P406337	

Quality Assurance Report Method Blank Results

Reference Method: SM 9222 D

Batch ID: P407353

Component	Result	Code	Units
Fecal Coliforms-Membrane Filter	2	U	cfu/100 mL

Reference Method: SOP-PCR-1.0

Batch ID: P406337

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

Reference Method: SOP-PCR-1.2

Batch ID: P406337

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

Reference Method: SOP-PCR-1.3

Batch ID: P406337

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

Reference Method: SOP-PCR-1.4

Batch ID: P406337

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

Quality Assurance Report Method Blank Results

Reference Method: SOP-PCR-1.5
 Batch ID: P406337

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: P406337

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
HF183-qPCR	76.3	103	P/P	50 - 175

Reference Method: SOP-PCR-1.2
 Batch ID: P406337

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GULL2-qPCR	101	79.0	P/P	50 - 300
GULL2-qPCR	75.0	98.1	P/P	50 - 300

Reference Method: SOP-PCR-1.3
 Batch ID: P406337

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GFD-qPCR	95.3	76.3	P/P	50 - 175

Reference Method: SOP-PCR-1.4
 Batch ID: P406337

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
BacR-qPCR	89.4	90.8	P/P	50 - 175

Reference Method: SOP-PCR-1.5
 Batch ID: P406337

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
DG3-qPCR	75.8	81.6	P/P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0
 Batch ID: P406337

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2289534	HF183-qPCR	80.5		P	50 - 175
2289535	HF183-qPCR	76.9		P	50 - 175
2289673	HF183-qPCR	76.7		P	50 - 175
2291846	HF183-qPCR	68.9		P	50 - 175
2292416	HF183-qPCR	76.6		P	50 - 175
2293469	HF183-qPCR	89.2	72.3	P/P	50 - 175
2293814	HF183-qPCR	79.0		P	50 - 175
2294014	HF183-qPCR	83.4		P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.2
 Batch ID: P406337

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2292416	GULL2-qPCR	72.1		P	50 - 300
2293469	GULL2-qPCR	102	104	P/P	50 - 300
2294014	GULL2-qPCR	51.6		P	50 - 300

Reference Method: SOP-PCR-1.3
 Batch ID: P406337

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2291846	GFD-qPCR	99.1		P	50 - 175
2292416	GFD-qPCR	86.1		P	50 - 175
2293469	GFD-qPCR	90.5	89.6	P/P	50 - 175
2294014	GFD-qPCR	69.7		P	50 - 175

Reference Method: SOP-PCR-1.4
 Batch ID: P406337

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2289534	BacR-qPCR	82.8		P	50 - 175
2289535	BacR-qPCR	75.4		P	50 - 175
2289673	BacR-qPCR	75.2		P	50 - 175
2291846	BacR-qPCR	70.3		P	50 - 175
2292416	BacR-qPCR	60.3		P	50 - 175
2293469	BacR-qPCR	72.6	76.1	P/P	50 - 175
2293814	BacR-qPCR	73.5		P	50 - 175
2294014	BacR-qPCR	62.7		P	50 - 175

Reference Method: SOP-PCR-1.5
 Batch ID: P406337

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2289534	DG3-qPCR	80.4		P	50 - 175
2289535	DG3-qPCR	77.3		P	50 - 175
2289673	DG3-qPCR	78.1		P	50 - 175
2292416	DG3-qPCR	71.5		P	50 - 175
2293469	DG3-qPCR	81.7	81.5	P/P	50 - 175
2293814	DG3-qPCR	74.4		P	50 - 175
2294014	DG3-qPCR	82.1		P	50 - 175

Quality Assurance Report Precision

Reference Method: SOP-PCR-1.0
 Batch ID: P406337

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2293469	HF183-qPCR	20.9	Spike	P	0 - 25

Quality Assurance Report Precision

Reference Method: SOP-PCR-1.2
Batch ID: P406337

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2293469	GULL2-qPCR	2.54	Spike	P	0 - 25

Reference Method: SOP-PCR-1.3
Batch ID: P406337

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2293469	GFD-qPCR	0.942	Spike	P	0 - 25

Reference Method: SOP-PCR-1.4
Batch ID: P406337

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2293469	BacR-qPCR	4.76	Spike	P	0 - 25

Reference Method: SOP-PCR-1.5
Batch ID: P406337

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2293469	DG3-qPCR	0.301	Spike	P	0 - 25

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

Quality Assurance Report Surrogates

Lab Sample ID: 2293469
Field Sample ID: G3NW0162

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	101	P	50 - 300
SOP-PCR-1.0	SKETA-qPCR	92.9	P	50 - 300
SOP-PCR-1.2	SKETA-qPCR	113	P	50 - 300
SOP-PCR-1.2	SKETA-qPCR	101	P	50 - 300
SOP-PCR-1.3	SKETA-qPCR	96.6	P	50 - 300
SOP-PCR-1.3	SKETA-qPCR	96.6	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	100	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	91.1	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	98.2	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	97.1	P	50 - 300

Quality Assurance Report Summary

Ref. Method	Analyte	LCS % Recovery	MS % Recovery	Precision
			LCS	SMP MS

Quality Assurance Report Summary

Ref. Method	Analyte	LCS % Recovery			MS % Recovery			LCS	Precision SMP	MS
SOP-PCR-1.0	HF183-qPCR	103	76.3		80.5	76.9	76.7			20.9
SOP-PCR-1.2	GULL2-qPCR	101	98.1	75.0	72.1	102	104			2.54
SOP-PCR-1.3	GFD-qPCR	79.0			51.6					
SOP-PCR-1.4	BacR-qPCR	76.3	95.3		99.1	86.1	90.5			0.942
SOP-PCR-1.5	DG3-qPCR	89.4	90.8		82.8	75.4	75.2			4.76
					70.3					
		81.6	75.8		80.4	77.3	78.1			0.301
					71.5					

Reference Method Descriptions

Method	Description	Associated Samples
Enterolert/QT	Enterococci by Enterolert/QT by UWF in Pensacola	2293467
SM 9222 D	Fecal coliforms by membrane filter method - non-chlorinated water systems	2293466
SM 9223 B Quanti-Tray	E. coli by Colilert-18/QT by UWF in Pensacola	2293467, 2293468
SOP-PCR-1.0	Detection and quantification of human-specific HF183 Bacteroides genetic marker with quantitative polymerase chain reaction	2293469
SOP-PCR-1.2	Detection and quantification of coastal bird specific Catellicoccus marimammalium Gull2 genetic marker with quantitative polymerase chain reaction	2293469
SOP-PCR-1.3	Detection and quantification of bird specific Helicobacter GFD genetic marker with quantitative polymerase chain reaction	2293469
SOP-PCR-1.4	Detection and quantification of Ruminant specific Bacteroidetes BacR genetic marker with quantitative polymerase chain reaction	2293469
SOP-PCR-1.5	Detection and quantification of canine-specific DG3 Bacteroides genetic marker with quantitative polymerase chain reaction	2293469

Preparation and Analysis Log

Ref. Method	Received Date	Prep Date/Time	Prepared By	Analysis Date/Time	Analyzed By	Associated Samples
Enterolert/QT	12/06/2021	12/06/2021 15:10		12/07/2021 15:40	UWF Lab	2293467
SM 9222 D	12/07/2021	12/07/2021 15:45	Carina A. Tull	12/08/2021 16:05	Carina A. Tull	2293466
SM 9223 B Quanti-Tray	12/06/2021	12/06/2021 15:20		12/07/2021 11:21	UWF Lab	2293467, 2293468
SOP-PCR-1.0	12/07/2021	12/07/2021 14:00	Luke Hudson	12/14/2021 11:13	DeAsia Armster	2293469
SOP-PCR-1.2	12/07/2021	12/07/2021 14:00	Luke Hudson	12/15/2021 14:51	DeAsia Armster	2293469
SOP-PCR-1.3	12/07/2021	12/07/2021 14:00	Luke Hudson	12/16/2021 11:45	DeAsia Armster	2293469
SOP-PCR-1.4	12/07/2021	12/07/2021 14:00	Luke Hudson	12/14/2021 15:31	DeAsia Armster	2293469
SOP-PCR-1.5	12/07/2021	12/07/2021 14:00	Luke Hudson	12/15/2021 09:38	DeAsia Armster	2293469