

Biological Analysis Report

CEN-DIST-2022-07-13-03

Florida Department of Environmental Protection
Central Laboratory
2600 Blair Stone Road
Tallahassee, FL 32399-2400
DOH Accreditation E31780

Event Description: **Run 10**
Request ID: **RQ-2022-07-11-30**
Customer: **CEN-DIST**
Project ID: **SMP**

Send Reports to:
FL Dept. of Environmental Protection
FL Dept. of Environmental Protection
3319 Maguire Blvd., Suite 232
Orlando, FL 32803-3767
Attn: Matthew Bearden

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: 24-AUG-2022 15:27

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: 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: Phelps Creek 85m N of Lake Mary Blvd.

Collection Date/Time: 07/12/2022 10:04

Field ID: G2CE0247

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2341089	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	5794.0	Q	MPN/100 mL	P416726	

Sample Location: Phelps Creek 25m S of Pine Ave.

Collection Date/Time: 07/12/2022 10:15

Field ID: G2CE0246

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2341090	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	65.0	Q	MPN/100 mL	P416726	

Sample Location: Sweetwater Creek @ US of DeLeon St.

Collection Date/Time: 07/12/2022 10:40

Field ID: G2CE0119

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2341087	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	488.4	Q	MPN/100 mL	P416726	
2341091	SOP-PCR-1.0	HF183-purified-qPCR**	370	UJ	TSC/100 mL	P417933	LCS, SUR
	SOP-PCR-1.2	GULL2-purified-qPCR**	400	UJ	TSC/100 mL	P417933	LCS, SUR
	SOP-PCR-1.3	GFD-purified-qPCR**	1.59E+03	J	TSC/100 mL	P417933	LCS, SUR
	SOP-PCR-1.4	BacR-purified-qPCR**	340	UJ	TSC/100 mL	P417933	LCS, SUR
	SOP-PCR-1.5	DG3-purified-qPCR**	290	UJ	TSC/100 mL	P417933	LCS, SUR

Ref. Method and Comment:

SOP-PCR-1.0: Precision data unavailable for HF183-purified-qPCR component. Refer to the Lab Analysis Report for an explanation of QC Codes.
 SOP-PCR-1.2: Precision data unavailable for GULL2-purified-qPCR component. Refer to the Lab Analysis Report for an explanation of QC Codes.
 SOP-PCR-1.3: Precision data unavailable for GFD-purified-qPCR component. Refer to the Lab Analysis Report for an explanation of QC Codes.
 SOP-PCR-1.4: Precision data unavailable for BacR-purified-qPCR component. Refer to the Lab Analysis Report for an explanation of QC Codes.
 SOP-PCR-1.5: Precision data unavailable for DG3-purified-qPCR component. Refer to the Lab Analysis Report for an explanation of QC Codes.

Sample Location: Sweetwater Creek @ 35m US of Cress Run

Collection Date/Time: 07/12/2022 10:58

Field ID: G2CE0139

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2341088	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	435.2	Q	MPN/100 mL	P416726	
2341092	SOP-PCR-1.0	HF183-purified-qPCR**	390	IJ	TSC/100 mL	P417933	LCS, SUR
	SOP-PCR-1.4	BacR-purified-qPCR**	340	UJ	TSC/100 mL	P417933	LCS, SUR
	SOP-PCR-1.5	DG3-purified-qPCR**	290	UJ	TSC/100 mL	P417933	LCS, SUR

Ref. Method and Comment:

SOP-PCR-1.0: Precision data unavailable for HF183-purified-qPCR component. Refer to the Lab Analysis Report for an explanation of QC Codes.
 SOP-PCR-1.4: Precision data unavailable for BacR-purified-qPCR component. Refer to the Lab Analysis Report for an explanation of QC Codes.
 SOP-PCR-1.5: Precision data unavailable for DG3-purified-qPCR component. Refer to the Lab Analysis Report for an explanation of QC Codes.

Quality Assurance Report Method Blank Results

Reference Method: SM 9223 B Quanti-Tray

Batch ID: P416726

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

Quality Assurance Report Method Blank Results

Reference Method: SOP-PCR-1.0
Batch ID: P417933

Component	Result	Code	Units
HF183-purified-qPCR	550	U	TSC/100 mL

Reference Method: SOP-PCR-1.2
Batch ID: P417933

Component	Result	Code	Units
GULL2-purified-qPCR	600	U	TSC/100 mL

Reference Method: SOP-PCR-1.3
Batch ID: P417933

Component	Result	Code	Units
GFD-purified-qPCR	120	U	TSC/100 mL

Reference Method: SOP-PCR-1.4
Batch ID: P417933

Component	Result	Code	Units
BacR-purified-qPCR	510	U	TSC/100 mL

Reference Method: SOP-PCR-1.5
Batch ID: P417933

Component	Result	Code	Units
DG3-purified-qPCR	430	U	TSC/100 mL

Quality Assurance Report Laboratory Control Sample Accuracy

Reference Method: SOP-PCR-1.0
Batch ID: P417933

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
HF183-purified-qPCR	90.6	4.29	P/F	50 - 175

Reference Method: SOP-PCR-1.2
Batch ID: P417933

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GULL2-purified-qPCR	91.2	2.07	P/F	50 - 300

Reference Method: SOP-PCR-1.3
Batch ID: P417933

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GFD-purified-qPCR	92.1	1.09	P/F	50 - 175

Reference Method: SOP-PCR-1.4
Batch ID: P417933

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
-----------	---------	---------	-----------	----------------

Quality Assurance Report Laboratory Control Sample Accuracy

Reference Method: SOP-PCR-1.4
Batch ID: P417933

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
BacR-purified-qPCR	83.5	5.13	P/F	50 - 175

Reference Method: SOP-PCR-1.5
Batch ID: P417933

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
DG3-purified-qPCR	85.4	6.69	P/F	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0
Batch ID: P417933

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2341091	HF183-purified-qPCR	95.2		P	50 - 175
2341092	HF183-purified-qPCR	94.0		P	50 - 175
2341750	HF183-purified-qPCR	92.8		P	50 - 175
2342111	HF183-purified-qPCR	95.9		P	50 - 175
2342265	HF183-purified-qPCR	100		P	50 - 175
2343442	HF183-purified-qPCR	102		P	50 - 175

Reference Method: SOP-PCR-1.2
Batch ID: P417933

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2341091	GULL2-purified-qPCR	105		P	50 - 300
2341750	GULL2-purified-qPCR	101		P	50 - 300
2342111	GULL2-purified-qPCR	99.1		P	50 - 300
2342265	GULL2-purified-qPCR	101		P	50 - 300

Reference Method: SOP-PCR-1.3
Batch ID: P417933

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2341091	GFD-purified-qPCR	99.4		P	50 - 175
2341750	GFD-purified-qPCR	98.0		P	50 - 175
2342111	GFD-purified-qPCR	103		P	50 - 175
2342265	GFD-purified-qPCR	105		P	50 - 175

Reference Method: SOP-PCR-1.4
Batch ID: P417933

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2341091	BacR-purified-qPCR	86.2		P	50 - 175
2341092	BacR-purified-qPCR	83.5		P	50 - 175
2341750	BacR-purified-qPCR	81.3		P	50 - 175
2342111	BacR-purified-qPCR	84.5		P	50 - 175
2342265	BacR-purified-qPCR	77.9		P	50 - 175
2343442	BacR-purified-qPCR	75.8		P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.5
Batch ID: P417933

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2341091	DG3-purified-qPCR	104		P	50 - 175
2341092	DG3-purified-qPCR	98.4		P	50 - 175
2341750	DG3-purified-qPCR	98.2		P	50 - 175
2342111	DG3-purified-qPCR	102		P	50 - 175
2342265	DG3-purified-qPCR	97.4		P	50 - 175
2343442	DG3-purified-qPCR	96.0		P	50 - 175

Quality Assurance Report Precision

Reference Method: SM 9223 B Quanti-Tray
Batch ID: P416726

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2340776	Escherichia Coli-Quanti-Tray	13.9	Sample	P	0 - 80
2340833	Escherichia Coli-Quanti-Tray	1.36	Sample	P	0 - 80

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

Quality Assurance Report Surrogates

Lab Sample ID: 2341091
Field Sample ID: G2CE0119

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	8.46	F	50 - 300
SOP-PCR-1.2	SKETA-qPCR	12.4	F	30 - 300
SOP-PCR-1.3	SKETA-qPCR	10.2	F	30 - 300
SOP-PCR-1.4	SKETA-qPCR	10.5	F	30 - 300
SOP-PCR-1.5	SKETA-qPCR	9.42	F	50 - 300

Lab Sample ID: 2341092
Field Sample ID: G2CE0139

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	8.53	F	50 - 300
SOP-PCR-1.4	SKETA-qPCR	9.71	F	30 - 300
SOP-PCR-1.5	SKETA-qPCR	9.17	F	50 - 300

Quality Assurance Report Summary

Ref. Method	Analyte	LCS % Recovery		MS % Recovery			Precision		
							LCS	SMP	MS
SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray							13.9	1.36
SOP-PCR-1.0	HF183-purified-qPCR	4.29	90.6	95.2	94.0	92.8			
				95.9					

Quality Assurance Report Summary

Ref. Method	Analyte	LCS % Recovery		MS % Recovery			LCS	Precision SMP	MS
SOP-PCR-1.2	GULL2-purified-qPCR	2.07	91.2	105	101	99.1			
SOP-PCR-1.3	GFD-purified-qPCR	1.09	92.1	99.4	98.0	103			
SOP-PCR-1.4	BacR-purified-qPCR	5.13	83.5	86.2	83.5	81.3			
SOP-PCR-1.5	DG3-purified-qPCR	6.69	85.4	104	98.4	98.2			

Reference Method Descriptions

Method	Description	Associated Samples
SM 9223 B Quanti-Tray	Escherichia coli by Colilert 18 Quanti-Tray method - non-chlorinated water systems	2341087, 2341088, 2341089, 2341090
SOP-PCR-1.0	Detection and quantification of human-specific HF183 Bacteroides genetic marker with quantitative polymerase chain reaction	2341091, 2341092
SOP-PCR-1.2	Detection and quantification of coastal bird specific Catellicoccus marimammalium Gull2 genetic marker with quantitative polymerase chain reaction	2341091
SOP-PCR-1.3	Detection and quantification of bird specific Helicobacter GFD genetic marker with quantitative polymerase chain reaction	2341091
SOP-PCR-1.4	Detection and quantification of Ruminant specific Bacteroidetes BacR genetic marker with quantitative polymerase chain reaction	2341091, 2341092
SOP-PCR-1.5	Detection and quantification of canine-specific DG3 Bacteroides genetic marker with quantitative polymerase chain reaction	2341091, 2341092

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	07/13/2022	07/13/2022 16:31	Ethan Wickline	07/14/2022 10:34	Austin Nolan	2341087, 2341088, 2341089, 2341090
SOP-PCR-1.0	07/13/2022	07/13/2022 13:10	DeAsia Armster	08/16/2022 13:24	DeAsia Armster	2341091, 2341092
SOP-PCR-1.2	07/13/2022	07/13/2022 13:10	DeAsia Armster	08/19/2022 09:16	DeAsia Armster	2341091
SOP-PCR-1.3	07/13/2022	07/13/2022 13:10	DeAsia Armster	08/22/2022 09:33	Roberta Tatti	2341091
SOP-PCR-1.4	07/13/2022	07/13/2022 13:10	DeAsia Armster	08/17/2022 14:10	DeAsia Armster	2341091, 2341092
SOP-PCR-1.5	07/13/2022	07/13/2022 13:10	DeAsia Armster	08/18/2022 09:06	DeAsia Armster	2341091, 2341092