

Biological Analysis Report

SW-DIST-2022-09-07-01

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

Overflow Analyses Performed By:
Benchmark EnvironAnalytical, Inc.
1711 12th Street East
Palmetto, FL 34221
DOH Accreditation E84167

Event Description: **Big Slough**
Request ID: **RQ-2022-09-05-71**
Customer: **SW-DIST**
Project ID: **SMP**

Send Reports to:
FL Dept. of Environmental Protection
FL Dept. of Environmental Protection
13051 N. Telecom Parkway
Temple Terrace, FL 33637-0926
Attn: Ben Paswater

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-OCT-2022 15:30

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: 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: Big Slough Canal (Potable Portion)@ Route 72 Collection Date/Time: 09/06/2022 09:40

Field ID: G3SW0115

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2354276	SOP-PCR-1.0	HF183-qPCR**	3.3E+03	UJ	TSC/100 mL	P419056	SUR
	SOP-PCR-1.4	BacR-qPCR**	1.6E+04		TSC/100 mL	P419056	
	SOP-PCR-1.5	DG3-qPCR**	2.6E+03	UJ	TSC/100 mL	P419056	SUR
2354278	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	110		MPN/100 mL		

Ref. Method and Comment:
 SOP-PCR-1.0: Refer to the Lab Analysis Report for an explanation of QC Codes.
 SOP-PCR-1.5: Refer to the Lab Analysis Report for an explanation of QC Codes.

Sample Location: Big Slough Canal in Preserve

Collection Date/Time: 09/06/2022 11:20

Field ID: G3SW0117

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2354277	SOP-PCR-1.0	HF183-qPCR**	3.3E+03	U	TSC/100 mL	P419056	
	SOP-PCR-1.4	BacR-qPCR**	2.1E+04		TSC/100 mL	P419056	
	SOP-PCR-1.5	DG3-qPCR**	2.6E+03	U	TSC/100 mL	P419056	
2354279	SM 9223 B Quanti-Tray	Escherichia Coli-Quanti-Tray	134		MPN/100 mL		

Quality Assurance Report Method Blank Results

Reference Method: SOP-PCR-1.0
Batch ID: P419056

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

Reference Method: SOP-PCR-1.4
Batch ID: P419056

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

Reference Method: SOP-PCR-1.5
Batch ID: P419056

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: P419056

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
HF183-qPCR	89.9	91.9	P/P	50 - 175

Reference Method: SOP-PCR-1.4
Batch ID: P419056

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: P419056

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
BacR-qPCR	119	83.8	P/P	50 - 175

Reference Method: SOP-PCR-1.5
Batch ID: P419056

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
DG3-qPCR	106	91.1	P/P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0
Batch ID: P419056

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2354008	HF183-qPCR	81.3		P	50 - 175
2354060	HF183-qPCR	77.0	79.3	P/P	50 - 175
2354154	HF183-qPCR	85.4		P	50 - 175
2354155	HF183-qPCR	83.4		P	50 - 175
2354276	HF183-qPCR	94.9		P	50 - 175
2354277	HF183-qPCR	92.3		P	50 - 175
2355043	HF183-qPCR	90.3		P	50 - 175
2355661	HF183-qPCR	82.6		P	50 - 175
2355662	HF183-qPCR	81.3		P	50 - 175
2355663	HF183-qPCR	82.8		P	50 - 175
2355948	HF183-qPCR	77.6		P	50 - 175
2355949	HF183-qPCR	76.1		P	50 - 175

Reference Method: SOP-PCR-1.4
Batch ID: P419056

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2354060	BacR-qPCR	80.7	76.9	P/P	50 - 175
2354154	BacR-qPCR	93.5		P	50 - 175
2354155	BacR-qPCR	76.8		P	50 - 175
2354276	BacR-qPCR	67.3		P	50 - 175
2354277	BacR-qPCR	72.3		P	50 - 175
2355043	BacR-qPCR	77.8		P	50 - 175
2355661	BacR-qPCR	63.5		P	50 - 175
2355662	BacR-qPCR	75.6		P	50 - 175
2355663	BacR-qPCR	83.4		P	50 - 175
2355948	BacR-qPCR	71.1		P	50 - 175
2355949	BacR-qPCR	67.1		P	50 - 175
2356005	BacR-qPCR	85.5		P	50 - 175

Reference Method: SOP-PCR-1.5
Batch ID: P419056

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2354060	DG3-qPCR	97.5	91.6	P/P	50 - 175
2354154	DG3-qPCR	98.2		P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.5
Batch ID: P419056

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2354155	DG3-qPCR	97.7		P	50 - 175
2354276	DG3-qPCR	76.0		P	50 - 175
2354277	DG3-qPCR	90.2		P	50 - 175
2355043	DG3-qPCR	99.9		P	50 - 175
2355662	DG3-qPCR	91.4		P	50 - 175
2355663	DG3-qPCR	98.1		P	50 - 175
2355948	DG3-qPCR	87.1		P	50 - 175
2355949	DG3-qPCR	83.1		P	50 - 175
2356005	DG3-qPCR	91.7		P	50 - 175

Quality Assurance Report Precision

Reference Method: SOP-PCR-1.0
Batch ID: P419056

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2354060	HF183-qPCR	2.98	Spike	P	0 - 25

Reference Method: SOP-PCR-1.4
Batch ID: P419056

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2354060	BacR-qPCR	4.77	Spike	P	0 - 25

Reference Method: SOP-PCR-1.5
Batch ID: P419056

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2354060	DG3-qPCR	6.18	Spike	P	0 - 25

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

Quality Assurance Report Surrogates

Lab Sample ID: 2354276
Field Sample ID: G3SW0115

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	35.1	F	50 - 300
SOP-PCR-1.4	SKETA-qPCR	66.4	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	43.8	F	50 - 300

Quality Assurance Report Surrogates

Lab Sample ID: 2354277
 Field Sample ID: G3SW0117

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	58.0	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	86.2	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	61.0	P	50 - 300

Quality Assurance Report Summary

Ref. Method	Analyte	LCS % Recovery		MS % Recovery			Precision SMP	MS
SOP-PCR-1.0	HF183-qPCR	91.9	89.9	81.3	77.0	79.3		2.98
SOP-PCR-1.4	BacR-qPCR	83.8	119	80.7	76.9	93.5		4.77
SOP-PCR-1.5	DG3-qPCR	91.1	106	97.7	76.0	90.2		6.18
				85.4				
				76.8				
				99.9				

Reference Method Descriptions

Method	Description	Associated Samples
SM 9223 B Quanti-Tray	E. coli by Colilert/QT by BEA in Palmetto	2354278, 2354279
SOP-PCR-1.0	Detection and quantification of human-specific HF183 Bacteroides genetic marker with quantitative polymerase chain reaction	2354276, 2354277
SOP-PCR-1.4	Detection and quantification of Ruminant specific Bacteroidetes BacR genetic marker with quantitative polymerase chain reaction	2354276, 2354277
SOP-PCR-1.5	Detection and quantification of canine-specific DG3 Bacteroides genetic marker with quantitative polymerase chain reaction	2354276, 2354277

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	09/06/2022	09/06/2022 15:47		09/07/2022 16:50	Benchmark EnviroAnalytical - Palmetto	2354278, 2354279
SOP-PCR-1.0	09/07/2022	09/07/2022 13:15	Claudia Castro	10/03/2022 16:07	Roberta Tatti	2354276, 2354277
SOP-PCR-1.4	09/07/2022	09/07/2022 13:15	Claudia Castro	10/04/2022 13:22	Roberta Tatti	2354276, 2354277
SOP-PCR-1.5	09/07/2022	09/07/2022 13:15	Claudia Castro	10/04/2022 14:48	Roberta Tatti	2354276, 2354277