

Biological Analysis Report

SO-DIST-2021-04-06-01

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

Overflow Analyses Performed By:
Sanders Laboratories
10090 Bavaria Road
Ft. Myers, FL 33913
DOH Accreditation E85457

Event Description: **2021 SMP : Estero Cricks**
Request ID: **RQ-2021-04-05-63**
Customer: **SO-DIST**
Project ID: **SMP**

Send Reports to:
FL Dept. of Environmental Protection
FLDEP South District Office
2295 Victoria Ave. Suite 364
Fort Myers, FL 33901-3381
Attn: Chris Hormuth

For additional information please contact
Cheryl Swanson - Administrator
Jacqueline Savage - Bench Biology & Aquatic
Toxicity
Puja Jasrotia, Ph.D. - Molecular Biology & Taxonomy
Thekkekalathil Chandrasekhar, Ph.D, QA Officer
Phone (850) 245-8177

Certified by: Puja Jasrotia, Environmental Administrator

Date Certified: 07-MAY-2021 13:51



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: Mullock Creek near US41

Collection Date/Time: 04/05/2021 09:05

Field ID: G1SD0100

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2236216	SOP-PCR-1.0	HF183-qPCR**	1.7E+03	UJ	TSC/100 mL	P395993	SUR
	SOP-PCR-1.2	GULL2-qPCR**	3.6E+03	UJ	TSC/100 mL	P395993	MS, SUR
	SOP-PCR-1.3	GFD-qPCR**	3.1E+03	IJ	TSC/100 mL	P395993	SUR
	SOP-PCR-1.4	BacR-qPCR**	3.0E+03	UJ	TSC/100 mL	P395993	SUR
	SOP-PCR-1.5	DG3-qPCR**	2.6E+03	UJ	TSC/100 mL	P395993	SUR
2236218	Colilert-18	Escherichia Coli-Quanti-Tray	238	Q	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-1.2: Refer to the Lab Analysis Report for an explanation of QC Codes.

SOP-PCR-1.3: Duplicate was analyzed and result is 720 U. Refer to the Lab Analysis Report for an explanation of QC Codes.

SOP-PCR-1.4: 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: Mullock Creek upstream of US 41

Collection Date/Time: 04/05/2021 10:15

Field ID: G1SD0005

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2236217	SOP-PCR-1.0	HF183-qPCR**	1.7E+03	U	TSC/100 mL	P395993	
	SOP-PCR-1.2	GULL2-qPCR**	1.8E+03	U	TSC/100 mL	P395993	MS
	SOP-PCR-1.3	GFD-qPCR**	1.4E+03	I	TSC/100 mL	P395993	
	SOP-PCR-1.4	BacR-qPCR**	1.5E+03	U	TSC/100 mL	P395993	
	SOP-PCR-1.5	DG3-qPCR**	1.3E+03	U	TSC/100 mL	P395993	
2236219	Colilert-18	Escherichia Coli-Quanti-Tray	866		MPN/100 mL		

Quality Assurance Report Method Blank Results

Reference Method: SOP-PCR-1.0

Batch ID: P395993

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

Reference Method: SOP-PCR-1.2

Batch ID: P395993

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

Reference Method: SOP-PCR-1.3

Batch ID: P395993

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

Reference Method: SOP-PCR-1.4

Batch ID: P395993

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: P395993

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: P395993

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
HF183-qPCR	94.0	97.9	P/P	50 - 175

Reference Method: SOP-PCR-1.2
 Batch ID: P395993

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GULL2-qPCR	57.1	95.3	P/P	50 - 300

Reference Method: SOP-PCR-1.3
 Batch ID: P395993

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GFD-qPCR	69.0	92.4	P/P	50 - 175

Reference Method: SOP-PCR-1.4
 Batch ID: P395993

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
BacR-qPCR	94.1	95.5	P/P	50 - 175

Reference Method: SOP-PCR-1.5
 Batch ID: P395993

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
DG3-qPCR	92.3	107	P/P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0
 Batch ID: P395993

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2236216	HF183-qPCR	81.1	89.9	P/P	50 - 175
2236217	HF183-qPCR	81.7		P	50 - 175
2237642	HF183-qPCR	90.6		P	50 - 175
2238882	HF183-qPCR	81.9		P	50 - 175
2238883	HF183-qPCR	80.9		P	50 - 175
2238922	HF183-qPCR	77.1		P	50 - 175
2238923	HF183-qPCR	78.1		P	50 - 175
2238924	HF183-qPCR	82.5		P	50 - 175
2239037	HF183-qPCR	94.9		P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0

Batch ID: P395993

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2239038	HF183-qPCR	84.0		P	50 - 175
2239658	HF183-qPCR	90.9		P	50 - 175
2240112	HF183-qPCR	76.8		P	50 - 175
2240113	HF183-qPCR	87.0		P	50 - 175

Reference Method: SOP-PCR-1.2

Batch ID: P395993

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2236216	GULL2-qPCR	63.8	69.8	P/P	50 - 300
2236217	GULL2-qPCR	64.5		P	50 - 300
2237642	GULL2-qPCR	61.4		P	50 - 300
2238922	GULL2-qPCR	19.1		F	50 - 300
2238923	GULL2-qPCR	66.0		P	50 - 300
2238924	GULL2-qPCR	35.8		F	50 - 300
2239037	GULL2-qPCR	59.2		P	50 - 300
2239038	GULL2-qPCR	61.3		P	50 - 300

Reference Method: SOP-PCR-1.3

Batch ID: P395993

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2236216	GFD-qPCR	94.8	86.5	P/P	50 - 175
2236217	GFD-qPCR	87.6		P	50 - 175
2237642	GFD-qPCR	92.6		P	50 - 175
2238922	GFD-qPCR	101		P	50 - 175
2238923	GFD-qPCR	85.7		P	50 - 175
2238924	GFD-qPCR	97.3		P	50 - 175
2239037	GFD-qPCR	96.6		P	50 - 175
2239038	GFD-qPCR	97.6		P	50 - 175

Reference Method: SOP-PCR-1.4

Batch ID: P395993

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2236216	BacR-qPCR	81.7	85.8	P/P	50 - 175
2236217	BacR-qPCR	83.7		P	50 - 175
2237642	BacR-qPCR	82.8		P	50 - 175
2238882	BacR-qPCR	77.4		P	50 - 175
2238883	BacR-qPCR	81.0		P	50 - 175
2238922	BacR-qPCR	59.6		P	50 - 175
2238923	BacR-qPCR	71.3		P	50 - 175
2238924	BacR-qPCR	73.4		P	50 - 175
2239037	BacR-qPCR	84.1		P	50 - 175
2239038	BacR-qPCR	77.1		P	50 - 175
2239658	BacR-qPCR	70.6		P	50 - 175

Reference Method: SOP-PCR-1.5

Batch ID: P395993

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2236216	DG3-qPCR	82.6	78.1	P/P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.5
 Batch ID: P395993

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2236217	DG3-qPCR	84.0		P	50 - 175
2237642	DG3-qPCR	91.0		P	50 - 175
2238882	DG3-qPCR	87.6		P	50 - 175
2238883	DG3-qPCR	87.2		P	50 - 175
2238922	DG3-qPCR	80.8		P	50 - 175
2238923	DG3-qPCR	89.2		P	50 - 175
2238924	DG3-qPCR	79.8		P	50 - 175
2239037	DG3-qPCR	91.1		P	50 - 175
2239038	DG3-qPCR	84.8		P	50 - 175
2239658	DG3-qPCR	86.8		P	50 - 175
2240112	DG3-qPCR	79.7		P	50 - 175
2240113	DG3-qPCR	85.2		P	50 - 175

Quality Assurance Report Precision

Reference Method: SOP-PCR-1.0
 Batch ID: P395993

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2236216	HF183-qPCR	10.4	Spike	P	0 - 25

Reference Method: SOP-PCR-1.2
 Batch ID: P395993

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2236216	GULL2-qPCR	8.91	Spike	P	0 - 25

Reference Method: SOP-PCR-1.3
 Batch ID: P395993

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2236216	GFD-qPCR	8.96	Spike	P	0 - 25

Reference Method: SOP-PCR-1.4
 Batch ID: P395993

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2236216	BacR-qPCR	4.88	Spike	P	0 - 25

Reference Method: SOP-PCR-1.5
 Batch ID: P395993

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2236216	DG3-qPCR	5.55	Spike	P	0 - 25

Quality Assurance Report Precision

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

Quality Assurance Report Surrogates

Lab Sample ID: 2236216
Field Sample ID: G1SD0100

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	22.2	F	50 - 300
SOP-PCR-1.0	SKETA-qPCR	37.8	F	50 - 300
SOP-PCR-1.2	SKETA-qPCR	45.9	F	50 - 300
SOP-PCR-1.2	SKETA-qPCR	29.6	F	50 - 300
SOP-PCR-1.3	SKETA-qPCR	49.7	F	50 - 300
SOP-PCR-1.3	SKETA-qPCR	28.2	F	50 - 300
SOP-PCR-1.4	SKETA-qPCR	52.2	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	32.6	F	50 - 300
SOP-PCR-1.5	SKETA-qPCR	50.7	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	31.1	F	50 - 300

Lab Sample ID: 2236217
Field Sample ID: G1SD0005

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	76.3	P	50 - 300
SOP-PCR-1.2	SKETA-qPCR	84.5	P	50 - 300
SOP-PCR-1.3	SKETA-qPCR	79.7	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	94.1	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	86.1	P	50 - 300

Quality Assurance Report Summary

Ref. Method	Analyte	LCS % Recovery		MS % Recovery			Precision SMP	MS
SOP-PCR-1.0	HF183-qPCR	97.9	94.0	81.1	81.7	90.6		10.4
SOP-PCR-1.2	GULL2-qPCR	95.3	57.1	63.8	69.8	64.5		8.91
SOP-PCR-1.3	GFD-qPCR	92.4	69.0	94.8	86.5	87.6		8.96
SOP-PCR-1.4	BacR-qPCR	95.5	94.1	81.7	85.8	83.7		4.88
SOP-PCR-1.5	DG3-qPCR	107	92.3	82.6	78.1	84.0		5.55

Reference Method Descriptions

Method	Description	Associated Samples
Colilert-18	E. coli by Colilert-18/QT by Sanders in Fort Myers	2236218, 2236219
SOP-PCR-1.0	Detection and quantification of human-specific HF183 Bacteroides genetic marker with quantitative polymerase chain reaction	2236216, 2236217
SOP-PCR-1.2	Detection and quantification of coastal bird specific Catellicoccus marimammalum Gull2 genetic marker with quantitative polymerase chain reaction	2236216, 2236217

Reference Method Descriptions

Method	Description	<u>Associated Samples</u>
SOP-PCR-1.3	Detection and quantification of bird specific Helicobacter GFD genetic marker with quantitative polymerase chain reaction	2236216, 2236217
SOP-PCR-1.4	Detection and quantification of Ruminant specific Bacteroidetes BacR genetic marker with quantitative polymerase chain reaction	2236216, 2236217
SOP-PCR-1.5	Detection and quantification of canine-specific DG3 Bacteroides genetic marker with quantitative polymerase chain reaction	2236216, 2236217

Preparation and Analysis Log

Ref. Method	Received Date	Prep Date/Time	Prepared By	Analysis Date/Time	Analyzed By	Associated Samples
Colilert-18	04/05/2021	04/05/2021 17:23		04/06/2021 11:51	Sanders-Fort Myers	2236218, 2236219
SOP-PCR-1.0	04/06/2021	04/06/2021 13:35	DeAsia Armster	04/19/2021 11:19	Mailin Lopez	2236216, 2236217
SOP-PCR-1.2	04/06/2021	04/06/2021 13:35	DeAsia Armster	04/22/2021 08:17	Mailin Lopez	2236216, 2236217
SOP-PCR-1.3	04/06/2021	04/06/2021 13:35	DeAsia Armster	04/26/2021 13:26	Mailin Lopez	2236216, 2236217
SOP-PCR-1.4	04/06/2021	04/06/2021 13:35	DeAsia Armster	04/20/2021 11:50	Mailin Lopez	2236216, 2236217
SOP-PCR-1.5	04/06/2021	04/06/2021 13:35	DeAsia Armster	04/20/2021 10:37	Mailin Lopez	2236216, 2236217