

Biological Analysis Report

SO-DIST-2020-08-19-02

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: **Hatchee Cricks PT1**

Request ID: **RQ-2020-08-17-46**

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: Nicole Reistad

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 Manager

Date Certified: 11-SEP-2020 10:18



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: Popash Creek @ Pritchett Pkwy

Collection Date/Time: 08/18/2020 09:20

Field ID: G3SD0079

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2191167	SOP-PCR-1.0	HF183-qPCR**	140	U	GEU/100 mL	P385909	
	SOP-PCR-1.4	BacR-qPCR**	1.5E+03	U	TSC/100 mL	P385909	
	SOP-PCR-1.5	DG3-qPCR**	2.0E+03	I	TSC/100 mL	P385909	
2191171	Colilert-18	Escherichia Coli-Quanti-Tray	517		MPN/100 mL		

Sample Location: Yellow Fever Creek @ Littleton Rd

Collection Date/Time: 08/18/2020 10:10

Field ID: G3SD0149

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2191168	SOP-PCR-1.0	HF183-qPCR**	140	U	GEU/100 mL	P385909	
	SOP-PCR-1.2	GULL2-qPCR**	8.4E+03	U	TSC/100 mL	P385909	RPD
	SOP-PCR-1.3	GFD-purified-qPCR**	170	U	TSC/100 mL	P386857	LCS
	SOP-PCR-1.4	BacR-qPCR**	1.5E+03	U	TSC/100 mL	P385909	
	SOP-PCR-1.5	DG3-qPCR**	1.3E+03	U	TSC/100 mL	P385909	
2191172	Colilert-18	Escherichia Coli-Quanti-Tray	866		MPN/100 mL		

Sample Location: Powell Creek

Collection Date/Time: 08/18/2020 09:55

Field ID: G3SD0165

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2191169	SOP-PCR-1.0	HF183-qPCR**	140	U	GEU/100 mL	P385909	
	SOP-PCR-1.2	GULL2-qPCR**	8.4E+03	U	TSC/100 mL	P385909	RPD
	SOP-PCR-1.3	GFD-purified-qPCR**	170	U	TSC/100 mL	P386857	LCS
	SOP-PCR-1.4	BacR-qPCR**	1.5E+03	U	TSC/100 mL	P385909	
	SOP-PCR-1.5	DG3-qPCR**	1.3E+03	U	TSC/100 mL	P385909	
2191173	Colilert-18	Escherichia Coli-Quanti-Tray	816		MPN/100 mL		

Sample Location: Daughtrey Creek

Collection Date/Time: 08/18/2020 09:45

Field ID: G3SD0164

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2191170	SOP-PCR-1.0	HF183-qPCR**	140	U	GEU/100 mL	P385909	
	SOP-PCR-1.4	BacR-qPCR**	2.99E+05		TSC/100 mL	P385909	
	SOP-PCR-1.5	DG3-qPCR**	1.3E+03	U	TSC/100 mL	P385909	
2191174	Colilert-18	Escherichia Coli-Quanti-Tray	275		MPN/100 mL		

Quality Assurance Report

Method Blank Results

Reference Method: SOP-PCR-1.0
Batch ID: P385909

Component	Result	Code	Units
HF183-qPCR	280	U	GEU/100 mL

Quality Assurance Report Method Blank Results

Reference Method: SOP-PCR-1.2
Batch ID: P385909

Component	Result	Code	Units
GULL2-qPCR	1.7E+04	U	TSC/100 mL

Reference Method: SOP-PCR-1.3
Batch ID: P386857

Component	Result	Code	Units
GFD-purified-qPCR	340	U	TSC/100 mL

Reference Method: SOP-PCR-1.4
Batch ID: P385909

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

Reference Method: SOP-PCR-1.5
Batch ID: P385909

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: P385909

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
HF183-qPCR	110	115	P/P	50 - 175

Reference Method: SOP-PCR-1.2
Batch ID: P385909

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GULL2-qPCR	73.7	83.1	P/P	50 - 300

Reference Method: SOP-PCR-1.3
Batch ID: P386857

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GFD-purified-qPCR	89.8	2.56	P/F	50 - 175

Reference Method: SOP-PCR-1.4
Batch ID: P385909

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
BacR-qPCR	99.9	105	P/P	50 - 175

Reference Method: SOP-PCR-1.5
Batch ID: P385909

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

Quality Assurance Report Laboratory Control Sample Accuracy

Reference Method: SOP-PCR-1.5
Batch ID: P385909

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
DG3-qPCR	97.2	99.5	P/P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0
Batch ID: P385909

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2189481	HF183-qPCR	90.4		P	50 - 175
2189776	HF183-qPCR	104	106	P/P	50 - 175
2189777	HF183-qPCR	97.3		P	50 - 175
2190856	HF183-qPCR	97.6		P	50 - 175
2191060	HF183-qPCR	101		P	50 - 175
2191167	HF183-qPCR	101		P	50 - 175
2191168	HF183-qPCR	100		P	50 - 175
2191169	HF183-qPCR	102		P	50 - 175
2191170	HF183-qPCR	93.1		P	50 - 175
2191545	HF183-qPCR	95.6		P	50 - 175
2192167	HF183-qPCR	101		P	50 - 175
2192168	HF183-qPCR	109		P	50 - 175
2192169	HF183-qPCR	94.9		P	50 - 175

Reference Method: SOP-PCR-1.2
Batch ID: P385909

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2189776	GULL2-qPCR	64.9	87.3	P/P	50 - 300
2189777	GULL2-qPCR	80.0		P	50 - 300
2190856	GULL2-qPCR	74.8		P	50 - 300
2191060	GULL2-qPCR	87.2		P	50 - 300
2191168	GULL2-qPCR	82.3		P	50 - 300
2191169	GULL2-qPCR	79.6		P	50 - 300
2191545	GULL2-qPCR	59.6		P	50 - 300
2192168	GULL2-qPCR	65.8		P	50 - 300
2192169	GULL2-qPCR	67.2		P	50 - 300

Reference Method: SOP-PCR-1.3
Batch ID: P386857

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2189776	GFD-purified-qPCR	127	137	P/P	50 - 175
2189777	GFD-purified-qPCR	113		P	50 - 175
2190856	GFD-purified-qPCR	146		P	50 - 175
2191060	GFD-purified-qPCR	133		P	50 - 175
2191168	GFD-purified-qPCR	135		P	50 - 175
2191169	GFD-purified-qPCR	121		P	50 - 175
2191545	GFD-purified-qPCR	79.8		P	50 - 175
2192168	GFD-purified-qPCR	100		P	50 - 175
2192169	GFD-purified-qPCR	84.8		P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.4
Batch ID: P385909

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2189481	BacR-qPCR	78.0		P	50 - 175
2189776	BacR-qPCR	94.6	92.5	P/P	50 - 175
2189777	BacR-qPCR	83.8		P	50 - 175
2190856	BacR-qPCR	84.9		P	50 - 175
2191060	BacR-qPCR	90.4		P	50 - 175
2191167	BacR-qPCR	86.1		P	50 - 175
2191168	BacR-qPCR	90.6		P	50 - 175
2191169	BacR-qPCR	87.9		P	50 - 175
2191170	BacR-qPCR	86.1		P	50 - 175
2191545	BacR-qPCR	73.8		P	50 - 175
2192167	BacR-qPCR	83.9		P	50 - 175
2192168	BacR-qPCR	90.1		P	50 - 175
2192169	BacR-qPCR	84.7		P	50 - 175

Reference Method: SOP-PCR-1.5
Batch ID: P385909

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2189481	DG3-qPCR	89.7		P	50 - 175
2189776	DG3-qPCR	102	102	P/P	50 - 175
2189777	DG3-qPCR	90.3		P	50 - 175
2190856	DG3-qPCR	88.8		P	50 - 175
2191060	DG3-qPCR	101		P	50 - 175
2191167	DG3-qPCR	90.9		P	50 - 175
2191168	DG3-qPCR	92.8		P	50 - 175
2191169	DG3-qPCR	94.4		P	50 - 175
2191170	DG3-qPCR	93.6		P	50 - 175
2191545	DG3-qPCR	92.8		P	50 - 175
2192167	DG3-qPCR	80.0		P	50 - 175
2192168	DG3-qPCR	103		P	50 - 175
2192169	DG3-qPCR	94.0		P	50 - 175

Quality Assurance Report Precision

Reference Method: SOP-PCR-1.0
Batch ID: P385909

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2189776	HF183-qPCR	1.13	Spike	P	0 - 25

Reference Method: SOP-PCR-1.2
Batch ID: P385909

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2189776	GULL2-qPCR	29.4	Spike	F	0 - 25

Quality Assurance Report Precision

Reference Method: SOP-PCR-1.3
Batch ID: P386857

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2189776	GFD-purified-qPCR	7.81	Spike	P	0 - 25

Reference Method: SOP-PCR-1.4
Batch ID: P385909

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2189776	BacR-qPCR	2.20	Spike	P	0 - 25

Reference Method: SOP-PCR-1.5
Batch ID: P385909

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2189776	DG3-qPCR	0.278	Spike	P	0 - 25

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

Quality Assurance Report Surrogates

Lab Sample ID: 2191167
Field Sample ID: G3SD0079

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	89.4	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	96.7	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	90.9	P	50 - 300

Lab Sample ID: 2191168
Field Sample ID: G3SD0149

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	97.5	P	50 - 300
SOP-PCR-1.2	SKETA-qPCR	138	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	110	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	82.4	P	50 - 300

Lab Sample ID: 2191169
Field Sample ID: G3SD0165

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	95.8	P	50 - 300
SOP-PCR-1.2	SKETA-qPCR	133	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	108	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	88.3	P	50 - 300

Quality Assurance Report Surrogates

Lab Sample ID: 2191170
 Field Sample ID: G3SD0164

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	82.3	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	107	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	75.0	P	50 - 300

Quality Assurance Report Summary

Ref. Method	Analyte	LCS % Recovery		MS % Recovery			LCS	Precision SMP	MS
SOP-PCR-1.0	HF183-qPCR	115	110	90.4	104	106			1.13
SOP-PCR-1.2	GULL2-qPCR	83.1	73.7	97.3	64.9	87.3	80.0		29.4
SOP-PCR-1.3	GFD-purified-qPCR	2.56	89.8	74.8	127	137	113		7.81
SOP-PCR-1.4	BacR-qPCR	105	99.9	146	78.0	94.6	92.5		2.20
SOP-PCR-1.5	DG3-qPCR	99.5	97.2	83.8	89.7	102	102		0.278
				90.3					

Reference Method Descriptions

Method	Description	Associated Samples
Colilert-18	E. coli by Colilert-18/QT by Sanders in Fort Myers	2191171, 2191172, 2191173, 2191174
SOP-PCR-1.0	Detection and quantification of human-specific HF183 Bacteroides genetic marker with quantitative polymerase chain reaction	2191167, 2191168, 2191169, 2191170
SOP-PCR-1.2	Detection and quantification of coastal bird specific Catellicoccus marimammalium Gull2 genetic marker with quantitative polymerase chain reaction	2191168, 2191169
SOP-PCR-1.3	Detection and quantification of bird specific Helicobacter GFD genetic marker with quantitative polymerase chain reaction	2191168, 2191169
SOP-PCR-1.4	Detection and quantification of Ruminant specific Bacteroidetes BacR genetic marker with quantitative polymerase chain reaction	2191167, 2191168, 2191169, 2191170
SOP-PCR-1.5	Detection and quantification of canine-specific DG3 Bacteroides genetic marker with quantitative polymerase chain reaction	2191167, 2191168, 2191169, 2191170

Preparation and Analysis Log

Ref. Method	Received Date	Prep Date/Time	Prepared By	Analysis Date/Time	Analyzed By	Associated Samples
Colilert-18	08/18/2020	08/18/2020 13:58		08/19/2020 10:00	Sanders-Fort Myers	2191171, 2191172, 2191173, 2191174
SOP-PCR-1.0	08/19/2020	08/19/2020 14:30	Angela Smith	08/31/2020 16:40	Lauren Campbell	2191167, 2191168, 2191169, 2191170
SOP-PCR-1.2	08/19/2020	08/19/2020 14:30	Angela Smith	09/03/2020 09:47	Mailin Lopez	2191168, 2191169
SOP-PCR-1.3	08/19/2020	08/19/2020 14:30	Lauren Campbell	09/04/2020 13:22	Mailin Lopez	2191168, 2191169
SOP-PCR-1.4	08/19/2020	08/19/2020 14:30	Angela Smith	09/01/2020 09:50	Lauren Campbell	2191167, 2191168, 2191169, 2191170
SOP-PCR-1.5	08/19/2020	08/19/2020 14:30	Angela Smith	09/02/2020 11:17	Lauren Campbell	2191167, 2191168, 2191169, 2191170