

Biological Analysis Report

SO-DIST-2020-09-22-02

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-2020-09-21-39**

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
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Certified by: Puja Jasrotia, Environmental Manager

Date Certified: 07-OCT-2020 13:42

A handwritten signature in black ink, appearing to be 'Puja Jasrotia', with a horizontal line underneath it.

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: Powell Creek

Collection Date/Time: 09/21/2020 08:15

Field ID: G3SD0165

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2197429	SOP-PCR-1.0	HF183-qPCR**	140	U	GEU/100 mL	P386924	
	SOP-PCR-1.2	GULL2-qPCR**	1.8E+03	U	TSC/100 mL	P386924	LCS
	SOP-PCR-1.3	GFD-purified-qPCR**	170	U	TSC/100 mL	P387144	LCS, MS
	SOP-PCR-1.4	BacR-qPCR**	1.5E+03	U	TSC/100 mL	P386924	
	SOP-PCR-1.5	DG3-qPCR**	1.3E+03	U	TSC/100 mL	P386924	
2197432	Colilert-18	Escherichia Coli-Quanti-Tray	2420	J	MPN/100 mL		

Ref. Method and Comment:

SOP-PCR-1.2: Precision data unavailable for Analysis Batch A101312

Colilert-18: All Quanti-Tray wells were positive. Result may be higher than reported

Sample Location: Popash Creek @ Pritchett Pkwy

Collection Date/Time: 09/21/2020 08:35

Field ID: G3SD0079

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2197430	SOP-PCR-1.0	HF183-qPCR**	140	U	GEU/100 mL	P386924	
	SOP-PCR-1.4	BacR-qPCR**	4.6E+03	I	TSC/100 mL	P386924	
	SOP-PCR-1.5	DG3-qPCR**	2.6E+03	U	TSC/100 mL	P386924	
2197433	Colilert-18	Escherichia Coli-Quanti-Tray	2420	J	MPN/100 mL		

Ref. Method and Comment:

Colilert-18: All Quanti-Tray wells were positive. Result may be higher than reported

Sample Location: Cypress Creek @ N River Rd

Collection Date/Time: 09/21/2020 09:15

Field ID: G3SD0084

Matrix: W-SURF-FRH

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2197431	SOP-PCR-1.0	HF183-purified-qPCR**	24	UJ	GEU/100 mL	P387144	LCS, SUR
	SOP-PCR-1.4	BacR-qPCR**	5.7E+03	I	TSC/100 mL	P386924	
	SOP-PCR-1.5	DG3-qPCR**	2.6E+03	U	TSC/100 mL	P386924	
2197434	Colilert-18	Escherichia Coli-Quanti-Tray	866		MPN/100 mL		

Ref. Method and Comment:

SOP-PCR-1.0: Precision data unavailable for Analysis Batch A101301. Refer to the Lab Analysis Report for an explanation of QC Codes.

Quality Assurance Report

Method Blank Results

Reference Method: SOP-PCR-1.0

Batch ID: P386924

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

Reference Method: SOP-PCR-1.0

Batch ID: P387144

Component	Result	Code	Units
HF183-purified-qPCR	47	U	GEU/100 mL

Quality Assurance Report Method Blank Results

Reference Method: SOP-PCR-1.2
Batch ID: P386924

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

Reference Method: SOP-PCR-1.3
Batch ID: P387144

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

Reference Method: SOP-PCR-1.4
Batch ID: P386924

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

Reference Method: SOP-PCR-1.5
Batch ID: P386924

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: P386924

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
HF183-qPCR	117	95.6	P/P	50 - 175

Reference Method: SOP-PCR-1.0
Batch ID: P387144

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
HF183-purified-qPCR	107	16.3	P/F	50 - 175

Reference Method: SOP-PCR-1.2
Batch ID: P386924

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GULL2-qPCR	96.7	25.3	P/F	50 - 300

Reference Method: SOP-PCR-1.3
Batch ID: P387144

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
GFD-purified-qPCR	90.1	6.04	P/F	50 - 175

Reference Method: SOP-PCR-1.4
Batch ID: P386924

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: P386924

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
BacR-qPCR	95.6	71.4	P/P	50 - 175

Reference Method: SOP-PCR-1.5
Batch ID: P386924

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
DG3-qPCR	82.9	80.7	P/P	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.0
Batch ID: P386924

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2194195	HF183-qPCR	88.8	91.7	P/P	50 - 175
2194437	HF183-qPCR	90.2		P	50 - 175
2195672	HF183-qPCR	90.5		P	50 - 175
2195945	HF183-qPCR	92.8		P	50 - 175
2195946	HF183-qPCR	94.1		P	50 - 175
2197429	HF183-qPCR	80.6		P	50 - 175
2197430	HF183-qPCR	60.0		P	50 - 175
2197581	HF183-qPCR	88.7		P	50 - 175
2197653	HF183-qPCR	92.1		P	50 - 175

Reference Method: SOP-PCR-1.0
Batch ID: P387144

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2194514	HF183-purified-qPCR	128		P	50 - 175
2196599	HF183-purified-qPCR	98.9		P	50 - 175
2196600	HF183-purified-qPCR	107		P	50 - 175
2197431	HF183-purified-qPCR	104		P	50 - 175

Reference Method: SOP-PCR-1.2
Batch ID: P386924

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2194437	GULL2-qPCR	58.8		P	50 - 300
2195672	GULL2-qPCR	90.9		P	50 - 300
2197429	GULL2-qPCR	53.5		P	50 - 300
2197581	GULL2-qPCR	90.8		P	50 - 300
2197653	GULL2-qPCR	79.5		P	50 - 300

Reference Method: SOP-PCR-1.3
Batch ID: P387144

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2192796	GFD-purified-qPCR	127	120	P/P	50 - 175
2192797	GFD-purified-qPCR	99.3		P	50 - 175
2192864	GFD-purified-qPCR	33.7		F	50 - 175

Quality Assurance Report Matrix Spike Accuracy

Reference Method: SOP-PCR-1.3

Batch ID: P387144

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2192865	GFD-purified-qPCR	72.7		P	50 - 175
2193347	GFD-purified-qPCR	84.3		P	50 - 175
2193348	GFD-purified-qPCR	79.8		P	50 - 175
2194016	GFD-purified-qPCR	113		P	50 - 175
2194193	GFD-purified-qPCR	39.8		F	50 - 175
2194437	GFD-purified-qPCR	115		P	50 - 175
2195672	GFD-purified-qPCR	92.9		P	50 - 175
2197429	GFD-purified-qPCR	67.0		P	50 - 175
2197581	GFD-purified-qPCR	91.6		P	50 - 175
2197653	GFD-purified-qPCR	88.8		P	50 - 175

Reference Method: SOP-PCR-1.4

Batch ID: P386924

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2194195	BacR-qPCR	77.8	73.1	P/P	50 - 175
2194437	BacR-qPCR	66.1		P	50 - 175
2195672	BacR-qPCR	85.0		P	50 - 175
2195945	BacR-qPCR	80.0		P	50 - 175
2195946	BacR-qPCR	68.1		P	50 - 175
2196599	BacR-qPCR	67.8		P	50 - 175
2197429	BacR-qPCR	74.6		P	50 - 175
2197430	BacR-qPCR	77.0		P	50 - 175
2197431	BacR-qPCR	76.1		P	50 - 175
2197581	BacR-qPCR	77.8		P	50 - 175
2197653	BacR-qPCR	77.6		P	50 - 175

Reference Method: SOP-PCR-1.5

Batch ID: P386924

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
2194195	DG3-qPCR	93.7	94.6	P/P	50 - 175
2194437	DG3-qPCR	88.4		P	50 - 175
2195672	DG3-qPCR	101		P	50 - 175
2195945	DG3-qPCR	89.8		P	50 - 175
2195946	DG3-qPCR	94.3		P	50 - 175
2196599	DG3-qPCR	90.8		P	50 - 175
2197429	DG3-qPCR	81.4		P	50 - 175
2197430	DG3-qPCR	87.9		P	50 - 175
2197431	DG3-qPCR	89.1		P	50 - 175
2197581	DG3-qPCR	107		P	50 - 175
2197653	DG3-qPCR	89.7		P	50 - 175

Quality Assurance Report Precision

Reference Method: SOP-PCR-1.0

Batch ID: P386924

Replicated

Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
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Quality Assurance Report Precision

Reference Method: SOP-PCR-1.0
Batch ID: P386924

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2194195	HF183-qPCR	3.15	Spike	P	0 - 25

Reference Method: SOP-PCR-1.3
Batch ID: P387144

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2192796	GFD-purified-qPCR	5.86	Spike	P	0 - 25

Reference Method: SOP-PCR-1.4
Batch ID: P386924

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2194195	BacR-qPCR	6.20	Spike	P	0 - 25

Reference Method: SOP-PCR-1.5
Batch ID: P386924

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2194195	DG3-qPCR	0.962	Spike	P	0 - 25

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

Quality Assurance Report Surrogates

Lab Sample ID: 2197429
Field Sample ID: G3SD0165

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	75.7	P	50 - 300
SOP-PCR-1.2	SKETA-qPCR	90.4	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	92.7	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	132	P	50 - 300

Lab Sample ID: 2197430
Field Sample ID: G3SD0079

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	57.7	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	94.0	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	139	P	50 - 300

Lab Sample ID: 2197431
Field Sample ID: G3SD0084

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
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Quality Assurance Report Surrogates

Lab Sample ID: 2197431
Field Sample ID: G3SD0084

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	10.0	F	50 - 300
SOP-PCR-1.4	SKETA-qPCR	89.4	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	121	P	50 - 300

Quality Assurance Report Summary

Ref. Method	Analyte	LCS % Recovery		MS % Recovery			LCS	Precision SMP	MS
SOP-PCR-1.0	HF183-purified-qPCR	16.3	107	98.9	107	104			
	HF183-qPCR	95.6	117	88.8	91.7	90.2			3.15
SOP-PCR-1.2	GULL2-qPCR	25.3	96.7	79.5	58.8	90.9			
SOP-PCR-1.3	GFD-purified-qPCR	6.04	90.1	127	120	99.3			5.86
SOP-PCR-1.4	BacR-qPCR	71.4	95.6	77.8	73.1	66.1			6.20
SOP-PCR-1.5	DG3-qPCR	80.7	82.9	93.7	94.6	88.4			0.962

Reference Method Descriptions

Method	Description	Associated Samples
Colilert-18	E. coli by Colilert-18/QT by Sanders in Fort Myers	2197432, 2197433, 2197434
SOP-PCR-1.0	Detection and quantification of human-specific HF183 Bacteroides genetic marker with quantitative polymerase chain reaction	2197429, 2197430
SOP-PCR-1.0	Detection and quantification of human-specific HF183 Bacteroides genetic marker with quantitative polymerase chain reaction	2197431
SOP-PCR-1.2	Detection and quantification of coastal bird specific Catellicoccus marimammalium Gull2 genetic marker with quantitative polymerase chain reaction	2197429
SOP-PCR-1.3	Detection and quantification of bird specific Helicobacter GFD genetic marker with quantitative polymerase chain reaction	2197429
SOP-PCR-1.4	Detection and quantification of Ruminant specific Bacteroidetes BacR genetic marker with quantitative polymerase chain reaction	2197429, 2197430, 2197431
SOP-PCR-1.5	Detection and quantification of canine-specific DG3 Bacteroides genetic marker with quantitative polymerase chain reaction	2197429, 2197430, 2197431

Preparation and Analysis Log

Ref. Method	Received Date	Prep Date/Time	Prepared By	Analysis Date/Time	Analyzed By	Associated Samples
Colilert-18	09/21/2020	09/21/2020 13:55		09/22/2020 09:00	Sanders-Fort Myers	2197432, 2197433, 2197434
SOP-PCR-1.0	09/22/2020	09/22/2020 13:15	Lauren Campbell	09/30/2020 13:36	Lauren Campbell	2197431
	09/22/2020	09/22/2020 13:15	Mailin Lopez	09/28/2020 12:36	Lauren Campbell	2197429, 2197430
SOP-PCR-1.2	09/22/2020	09/22/2020 13:15	Mailin Lopez	10/01/2020 10:37	Mailin Lopez	2197429
SOP-PCR-1.3	09/22/2020	09/22/2020 13:15	Lauren Campbell	10/01/2020 15:58	Mailin Lopez	2197429
SOP-PCR-1.4	09/22/2020	09/22/2020 13:15	Mailin Lopez	09/29/2020 12:34	Lauren Campbell	2197429, 2197430, 2197431
SOP-PCR-1.5	09/22/2020	09/22/2020 13:15	Mailin Lopez	09/29/2020 14:58	Lauren Campbell	2197429, 2197430, 2197431