

Biological Analysis Report

SW-DIST-2020-09-23-01

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

Event Description: **RUN 1.5: Cazz River GOM Pasco**

Request ID: **RQ-2020-09-21-52**

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: Bne Paswater

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: Jacqueline Savage, Environmental Administrator

Date Certified: 20-OCT-2020 07:33

A handwritten signature in black ink that reads "Jackie Savage". The signature is written in a cursive, flowing 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: Chlorophyll/Grain Size/BOD, 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: Gulf of Mexico Pasco

Collection Date/Time: 09/22/2020 11:25

Field ID: G5SW0159

Matrix: W-SURF-SLT

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2197573	SM 10200 H (mod.)	Chlorophyll-a, Corrected	3.7		ug/L	P388007	
		Phaeophytin-a	0.90	U	ug/L	P388007	
		Chlorophyll-a, Uncorrected	4.1		ug/L	P388007	
2197580	Enterolert/QT	Enterococci-Quanti-Tray	10.0	Q	MPN/100 mL	P387865	RPD
2197581	SOP-PCR-1.0	HF183-qPCR**	71	U	GEU/100 mL	P386924	
	SOP-PCR-1.2	GULL2-qPCR**	900	U	TSC/100 mL	P386924	LCS
	SOP-PCR-1.3	GFD-purified-qPCR**	84	U	TSC/100 mL	P387144	LCS, MS
	SOP-PCR-1.4	BacR-qPCR**	760	U	TSC/100 mL	P386924	
	SOP-PCR-1.5	DG3-qPCR**	640	U	TSC/100 mL	P386924	

Ref. Method and Comment:

SM 10200 H (mod.): Precision data is not available for at least one component due to the small amount of analyte in the QC sample. Refer to QA report for available precision data.

Enterolert/QT: Laboratory duplicate precision is not within criteria limits.

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

Sample Location: Chassahowitzka River @ Start of Wbid

Collection Date/Time: 09/22/2020 14:00

Field ID: G5SW0064

Matrix: W-SURF-SLT

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2197576	Enterolert/QT	Enterococci-Quanti-Tray	31.0	Q	MPN/100 mL	P387865	RPD
2197578	SM 9222 D	Fecal Coliforms-Membrane Filter	56	Q	cfu/100 mL	P387957	

Ref. Method and Comment:

Enterolert/QT: Laboratory duplicate precision is not within criteria limits.

Sample Location: Chassahowitzka River Downstream of Houses

Collection Date/Time: 09/22/2020 13:35

Field ID: G5SW0065

Matrix: W-SURF-SLT

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2197572	SM 10200 H (mod.)	Chlorophyll-a, Corrected	3.5	A	ug/L	P388007	
		Phaeophytin-a	2.6	I	ug/L	P388007	
		Chlorophyll-a, Uncorrected	5.2	A	ug/L	P388007	
2197574	Enterolert/QT	Enterococci-Quanti-Tray	20.5	AJQ	MPN/100 mL	P387865	RPD
2197575	SM 9222 D	Fecal Coliforms-Membrane Filter	54	AQ	cfu/100 mL	P387957	

Ref. Method and Comment:

SM 10200 H (mod.): Laboratory duplicate result is 2.5 I for Phaeophytin-a. Precision data is unavailable due to the small amount of analyte in the QC sample.

Enterolert/QT: Result is J-qualified due to a lab replicate precision failure.

Sample Location: Chassahowitzka River @ Island Compound

Collection Date/Time: 09/22/2020 13:50

Field ID: G5SW0066

Matrix: W-SURF-SLT

Sample ID	Ref. Method	Component	Result	Code	Units	Batch ID	QC Codes
2197577	Enterolert/QT	Enterococci-Quanti-Tray	20.0	Q	MPN/100 mL	P387865	RPD
2197579	SM 9222 D	Fecal Coliforms-Membrane Filter	27	BQ	cfu/100 mL	P387957	

Ref. Method and Comment:

Enterolert/QT: Laboratory duplicate precision is not within criteria limits.

Quality Assurance Report

Method Blank Results

Quality Assurance Report Method Blank Results

Reference Method: Enterolert/QT
Batch ID: P387865

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

Reference Method: SM 10200 H (mod.)
Batch ID: P388007

Component	Result	Code	Units
Chlorophyll-a, Corrected	0.82	U	ug/L
Chlorophyll-a, Uncorrected	0.60	U	ug/L
Phaeophytin-a	0.90	U	ug/L

Reference Method: SM 9222 D
Batch ID: P387957

Component	Result	Code	Units
Fecal Coliforms-Membrane Filter	2	U	cfu/100 mL

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

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

Reference Method: SOP-PCR-1.2
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

Quality Assurance Report Laboratory Control Sample Accuracy

Reference Method: SM 10200 H (mod.)
 Batch ID: P388007

Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
Chlorophyll-a, Corrected	98.2		P	84 - 116

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-1.2
 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
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

Quality Assurance Report Matrix Spike Accuracy

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
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

Quality Assurance Report Matrix Spike Accuracy

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

Spiked Sample	Component	% Rec.1	% Rec.2	Pass/Fail	Control Limits
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: Enterolert/QT
Batch ID: P387865

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2197574	Enterococci-Quanti-Tray	102	Sample	F	0 - 80

Reference Method: SM 10200 H (mod.)
Batch ID: P388007

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2197572	Chlorophyll-a, Corrected	4.23	Sample	P	0 - 25
2197572	Chlorophyll-a, Uncorrected	4.25	Sample	P	0 - 25

Reference Method: SM 9222 D
Batch ID: P387957

Replicated Lab Sample	Component	% RSD/RPD	Sample/Spike/LCS*	Pass/Fail	Control Limits
2197575	Fecal Coliforms-Membrane Filter	7.41	Sample	P	0 - 50

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

Quality Assurance Report Precision

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: 2197581
Field Sample ID: G5SW0159

Reference Method	Surrogate	% Rec.	Pass/Fail	Control Limits
SOP-PCR-1.0	SKETA-qPCR	91.1	P	50 - 300
SOP-PCR-1.2	SKETA-qPCR	91.5	P	50 - 300
SOP-PCR-1.4	SKETA-qPCR	95.0	P	50 - 300
SOP-PCR-1.5	SKETA-qPCR	167	P	50 - 300

Quality Assurance Report Summary

Ref. Method	Analyte	LCS % Recovery		MS % Recovery			LCS	Precision SMP	MS
Enterolert/QT	Enterococci-Quanti-Tray							102	
SM 10200 H (mod.)	Chlorophyll-a, Corrected	98.2						4.23	
	Chlorophyll-a, Uncorrected							4.25	
SM 9222 D	Fecal Coliforms-Membrane Filter							7.41	
SOP-PCR-1.0	HF183-qPCR	95.6	117	88.8	91.7	90.2			3.15
				90.5					
SOP-PCR-1.2	GULL2-qPCR	25.3	96.7	79.5	58.8	90.9			
				53.5					
SOP-PCR-1.3	GFD-purified-qPCR	6.04	90.1	127	120	99.3			5.86
				33.7					
SOP-PCR-1.4	BacR-qPCR	71.4	95.6	77.8	73.1	66.1			6.20
				85.0					
SOP-PCR-1.5	DG3-qPCR	80.7	82.9	93.7	94.6	88.4			0.962
				101					

Reference Method Descriptions

Method	Description	Associated Samples
Enterolert/QT	Enterococci by Enterolert Quanti-Tray method - non-chlorinated water systems	2197574, 2197576, 2197577, 2197580
SM 10200 H (mod.)	Phytoplankton chlorophyll-a corrected, uncorrected, and phaeophytin by spectrophotometry	2197572, 2197573
SM 9222 D	Fecal coliforms by membrane filter method - non-chlorinated water systems	2197575, 2197578, 2197579
SOP-PCR-1.0	Detection and quantification of human-specific HF183 Bacteroides genetic marker with quantitative polymerase chain reaction	2197581
SOP-PCR-1.2	Detection and quantification of coastal bird specific Catellicoccus marimammalium Gull2 genetic marker with quantitative polymerase chain reaction	2197581
SOP-PCR-1.3	Detection and quantification of bird specific Helicobacter GFD genetic marker with quantitative polymerase chain reaction	2197581

Reference Method Descriptions

Method	Description	<u>Associated Samples</u>
SOP-PCR-1.4	Detection and quantification of Ruminant specific Bacteroidetes BacR genetic marker with quantitative polymerase chain reaction	2197581
SOP-PCR-1.5	Detection and quantification of canine-specific DG3 Bacteroides genetic marker with quantitative polymerase chain reaction	2197581

Preparation and Analysis Log

Ref. Method	Received Date	Prep Date/Time	Prepared By	Analysis Date/Time	Analyzed By	Associated Samples
Enterolert/QT	09/23/2020	09/23/2020 13:41	Jennifer Mihalic	09/24/2020 16:35	Jessica Fetgatter	2197574, 2197576, 2197577, 2197580
SM 10200 H (mod.)	09/23/2020	09/23/2020 13:45	Jessica Sherman	09/29/2020 14:11	Yuliya Danyuk	2197572, 2197573
SM 9222 D	09/23/2020	09/23/2020 13:50	Jessica Fetgatter	09/24/2020 14:11	Jessica Fetgatter	2197575, 2197578, 2197579
SOP-PCR-1.0	09/23/2020	09/23/2020 13:00	Mailin Lopez	09/28/2020 12:36	Lauren Campbell	2197581
SOP-PCR-1.2	09/23/2020	09/23/2020 13:00	Mailin Lopez	10/01/2020 10:37	Mailin Lopez	2197581
SOP-PCR-1.3	09/23/2020	09/23/2020 13:00	Lauren Campbell	10/01/2020 15:58	Mailin Lopez	2197581
SOP-PCR-1.4	09/23/2020	09/23/2020 13:00	Mailin Lopez	09/29/2020 12:34	Lauren Campbell	2197581
SOP-PCR-1.5	09/23/2020	09/23/2020 13:00	Mailin Lopez	09/29/2020 14:58	Lauren Campbell	2197581