Clean Water Act Section 319(h) Nonpoint Source Grant Program

Fate and Transport of E. coli in Rural Texas Landscapes and Streams
TSSWCB Project Number 07-06
Revision #2

Quality Assurance Project Plan

Texas State Soil and Water Conservation Board

prepared by

Texas AgriLife Research
Texas Water Resources Institute

Effective Period: Upon EPA Approval through August 2011
(with annual updates required)

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A1 APPROVAL PAGE

Quality Assurance Project Plan for "*Fate and Transport of E. coli in Rural Texas Landscapes and Streams"*

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Name: R. Daren Harmel
Title: Agricultural Engineer; Investigator

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A3 DISTRIBUTION LIST

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List of Acronyms

ACS	American Chemical Society
ARS	USDA-Agricultural Research Service
AWRL	Ambient Water Reporting Limit
BST	Bacteria Source Tracking
CAR	Corrective Action Report
CD	Compact Disc
CFS	Cubic Feet Per Second
CFU	Colony-Forming Unit of Bacteria
C/N	Carbon to Nitrogen Ratio
COC	Chain of Custody
DI	De-ionized
DQO	Data Quality Objective
EMC	Event Mean Concentration
EPA	Environmental Protection Agency
Extension	Texas AgriLife Extension Service
g	Gram
GPS	Global Positioning System
ID	Identification
L	Liter
mL	Milliliter
NIST	National Institute of Standards and Technology
PM	Project Manager
QA	Quality Assurance
QC	Quality Control
QAO	Quality Assurance Officer
QAPP	Quality Assurance Project Plan
RPD	Relative Percent Difference
R/W	Readable/Writeable
SELECT	Spatially Explicit Load Enrichment Calculation Tool
SM	Standard Methods for Examination of Water and Wastewater, 20^{th} edition
SOP	Standard Operating Procedures
SSL	Spatial Sciences Laboratory
Research	Texas AgriLife Research
TAMU	Texas A&M University
TMDL	Total Maximum Daily Load
TSSWCB	Texas State Soil and Water Conservation Board
TWRI	Texas AgriLife Research, Texas Water Resources Institute
USDA	United States Department of Agriculture
UV	Ultraviolet
WPP	Watershed Protection Plan

A4 PROJECT/TASK ORGANIZATION

The following is a list of individuals and organizations participating in the project with their specific roles and responsibilities:

U.S. Environmental Protection Agency Region 6

Henry Brewer, USEPA Texas Nonpoint Source Project Officer

Responsible for overall performance and direction of the project at the federal level. Ensures that the project assists in achieving the goals of the clean water act (CWA). Reviews and approves the quality assurance project plan (QAPP), project progress, and deliverables.

Texas State Soil and Water Conservation Board (TSSWCB)

Pamela Casebolt, TSSWCB Project Manager

Responsible for ensuring that the project delivers data of known quality, quantity, and type on schedule to achieve project objectives. Provides the primary point of contact between the TWRI and the TSSWCB. Tracks and reviews deliverables to ensure that tasks in the work plan are completed as specified in the contract. Notifies the TSSWCB QAO of significant project nonconformances and corrective actions taken as documented in quarterly progress reports from TWRI Project Lead.

Donna Long, TSSWCB Quality Assurance Officer

Reviews and approves QAPP and any amendments or revisions and ensures distribution of approved/revised QAPPs to TSSWCB participants. Responsible for verifying that the QAPP is followed by the TWRI. Assists the TSSWCB Project Manager on QA-related issues. Coordinates reviews and approvals of QAPPs and amendments or revisions. Conveys QA problems to appropriate TSSWCB management. Monitors implementation of corrective actions. Coordinates and conducts audits

Texas AgriLife Research, Texas Water Resources Institute (TWRI)

B. L. Harris, TWRI Acting Director; TWRI Project Lead

The TWRI Project Lead is responsible for ensuring that tasks and other requirements in the contract are executed on time and with the quality assurance/quality control requirements in the system as defined by the contract and in the project QAPP; assessing the quality of subcontractor/participant work; and submitting accurate and timely deliverables to the TSSWCB Project Manager.

Lucas Gregory, TWRI Quality Assurance Officer (QAO)

Responsible for coordinating development and implementation of TWRI's QA program. Responsible for writing and maintaining QAPPs and monitoring its implementation. Responsible for QAPP distribution, including appendices and amendments. Ensures the data collected for the project is of known and acceptable quality and adheres to the specifications of the QAPP. Responsible for identifying, receiving, and maintaining project quality assurance records. Responsible for coordinating with the TSSWCB QAO to resolve QA-related issues. Notifies the TWRI Project Manager and TSSWCB Project Manager of particular circumstances which may adversely affect the quality of data. Coordinates the research and review of technical QA material and data related to water quality monitoring system design and analytical techniques. Conducts assessments of participating organizations during the life of the project as noted in Section C1. Implements or ensures implementation of corrective actions needed to resolve nonconformances noted during assessments.

Texas AgriLife Research (Research) – Biological & Ag Engineering

R. Karthikeyan, Research Project Lead

Responsible for ensuring tasks and other requirements in the contract are executed on time and with the QA/QC requirements in the system as defined by the contract work plan and in the QAPP. Responsible for verifying that the data produced are of known and acceptable quality. Responsible for ensuring adequate training and supervision of all activities involved in generating analytical data for this project. Responsible for the facilitation of audits and the implementation, documentation, verification, and reporting of corrective actions. Responsible for submitting accurate and timely data analyses and other materials for progress and final reports to TWRI. Responsible for supervision of laboratory personnel involved in generating analytical data for the project. Responsible for ensuring that laboratory personnel involved in generating analytical data have adequate training and a thorough knowledge of the QAPP and all SOPs specific to the analyses or task performed and/or supervised. Responsible for oversight of all laboratory operations ensuring that all QA/QC requirements are met, documentation related to the analysis is complete and adequately maintained, and that results are reported accurately. Responsible for ensuring corrective actions are implemented, documented, reported and verified.

Texas AgriLife Extension Service (Extension) – Biological & Ag Engineering

Saqib Mukhtar, Extension Project Lead

Responsible for ensuring that tasks and other requirements in the contract are executed on time and with the QA/QC requirements in the system as defined by the contract work plan and in the QAPP. Responsible for verifying that the data produced are of known and acceptable quality. Responsible for ensuring adequate training and supervision of all

activities involved in generating analytical data for this project. Responsible for the facilitation of audits and the implementation, documentation, verification, and reporting of corrective actions. Responsible for submitting accurate and timely data analyses and other materials for progress and final reports to TWRI.

Texas AgriLife Research (Research) – Wildlife & Fisheries Sciences

Roel Lopez, Research Project Co-Lead

Responsible for coordinating and supervising sanitary surveys and fecal / waste stream sampling activities in the Cedar Creek watershed. Responsible for ensuring that field personnel have adequate training and a thorough knowledge of standard operating procedures (SOPs) specific to the analysis or task performed and/or supervised.

Texas A&M University Spatial Sciences Laboratory (SSL)

R. Srinivasan, SSL Project Co-Lead

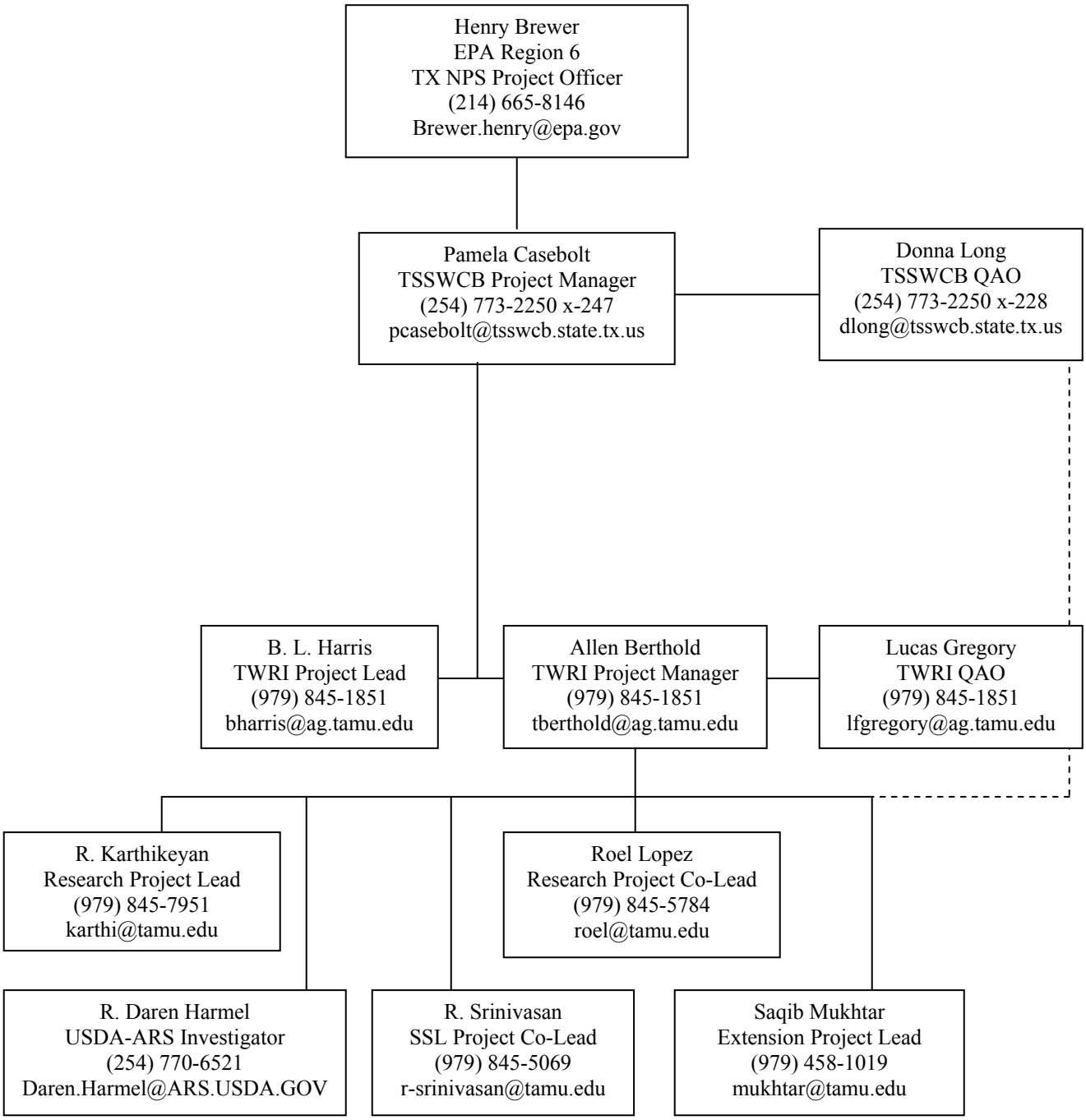
Responsible for general technical oversight of activities involved in collecting and incorporating project results into the SELECT model and other models.

USDA-Agricultural Research Service (ARS)

R. Daren Harmel, Investigator

Responsible for coordinating and supervising field sampling activities in Resley Creek. Responsible for ensuring that field personnel have adequate training and a thorough knowledge of standard operating procedures (SOPs) specific to the analysis or task performed and/or supervised.

Figure A4.1 Organization Chart



A5 PROBLEM DEFINITION/BACKGROUND

As of 2006, 310 water bodies in Texas were impaired because they did not meet bacteria criteria established by the state to protect contact recreation use (freshwater and saltwater) and/or oyster water use. The freshwater contact recreation use criterion used to determine impairment includes both a geometric mean for indicator bacteria, *Escherichia coli* (*E. coli*), of 126 colonies per 100 mL and a single sample maximum of 394 colonies per 100 mL.

A bacteria TMDL task-force was formed in the State of Texas to evaluate the current TMDL development process, address current weaknesses in the process, and develop a roadmap for further scientific research needed to reduce uncertainty in watershed modeling. There are several recommendations provided in the task-force document (Draft Four, 06/04/2007) to "scientifically" address the current "uncertainties" in bacteria TMDL development and implementation. (http://twri.tamu.edu/bacteriatmdl/). The objectives of this proposal are formulated based on several key recommendations compiled by task force experts. The key recommendations include identifying and characterizing *E. coli* sources and monitoring fate and transport of *E. coli* in impaired watersheds and streams. This project proposes a "holistic plan" for TMDL development that includes identification, characterization, and quantification of *E. coli* sources in impaired watersheds and monitoring the fate and transport of *E. coli* in these impaired watersheds. Project outputs will help to decrease uncertainties in *E. coli* load estimation from various sources and simulation of fate and transport processes of *E. coli* in watersheds and streams. The overall outcome of this project will help in developing scientifically sound TMDLs.

The first bacteria TMDL task-force recommendation that this project will focus on is conducting a sanitary survey of the watershed. Inventory sanitary surveys identify various potential *E. coli* sources in an impaired watershed are indispensable in TMDL development. In the majority of rural and agricultural stream-impairments due to bacteria, specifically *E. coli*, the specific sources and accurate quantities from each source have not been accurately determined. This lack of information makes managing the impaired waterway difficult and expensive. Currently, the most widely used approach to determine the source of *E. coli* is Bacteria Source Tracking (BST). This approach is good in determining the source of the impairment, but not in determining the load produced by specific sources. Moreover, BST methods are very labor intensive and expensive. On the other hand, inventory sanitary surveys identify various potential *E. coli* sources in impaired watersheds, are simpler and less expensive. *E. coli* load estimation tools such as Spatially Explicit Load Enrichment Calculation Tool (SELECT) estimates *E. coli* loads from various sources using literature values and expert knowledge. *E. coli* content of feces has been reported in literature for certain domestic and wildlife species and has been summarized in several reports used in TMDL development. However, this information has not been the focus of the reported research and therefore has not undergone extensive peer review. Consequently, there is a high level of uncertainty in identifying *E. coli* loads and sources for use in watershed modeling and *E. coli* load estimation tools. This project will conduct inventory sanitary surveys to identify potential *E. coli* sources in the Cedar Creek (1209G) watershed. Then, *E. coli* in various identified sources in the inventory survey will be characterized and quantified.

Accurate identification, characterization, and quantification of *E. coli* sources in the impaired Cedar Creek watershed will improve the predictions of SELECT, which is currently developed and successfully applied in TMDL development for an impaired watershed in the State of Texas.

The Bacteria TMDL Task Force also emphasized the need for additional studies that focus on developing a better understanding of *E. coli* fate and transport processes. Currently, knowledge of these processes is limited and contributes to significant uncertainties in the modeling of these processes. Fate and transport of *E. coli* in rural and agricultural landscapes is largely dependent on various environmental factors and management practices. Dominant environmental factors that affect *E. coli* transport in landscapes (e.g., waste source, soil type, temperature, rainfall, moisture content, nutrient status, etc) and persistence, growth, re-growth, and survival in landscapes need to be identified. Re-growth of *E. coli* in landscapes due to favorable environmental conditions (e.g., rainfall after dry weather conditions) is one of the major fate processes that influence *E. coli* concentrations in streams. The influence of different environmental variables on growth kinetics of *E. coli* and re-growth also need to be thoroughly studied and demonstrated to strengthen watershed models that simulate *E. coli* fate and transport in landscapes. The kinetic parameters obtained from the monitoring task of this project will be used to validate and improve fate and transport models used in TMDL development and implementation plans. Growth kinetics, survival rates, and re-growth are critical factors to accurately model the fate and transport of bacteria in watersheds.

Re-suspension of *E. coli* in streams (e.g. scouring of streambed sediments due to high flows) is one of the major fate processes that influence stream impairment. Unfortunately this process is not well studied or understood. The effect of rainfall and runoff on survival and growth of *E. coli* in streams and streambed sediments and subsequent re-suspension of *E. coli* in streams need to be quantified to properly assess the impairment of a stream. Parameters obtained from the monitoring study on Resley Creek (1221A) will be used to improve in-stream hydrodynamic processes modeled by fate and transport models. Information gained from this demonstration task will have an impact on TMDL development and the types of BMPs recommended to decrease bacterial contamination issues in agricultural watersheds.

Identifying, characterizing, and quantifying *E. coli* loads resulting from various sources are critical tasks in TMDL development for any impaired watershed. Monitoring and assessing the fate and transport processes of *E. coli* in landscapes and streams and monitoring the effects of environmental factors on fate and transport processes are required to develop and validate watershed models that utilize process-based fate and transport subroutines. Effective communication of findings is a crucial task that will not be overlooked in this project. Concise, easy to read publications and brochures will be developed to inform public, stakeholders, regulators, and authorities about the findings of the study. This project will combine all these vital aspects of TMDL development process to ultimately enhance the understanding of fate and transport of *E. coli* in rural landscapes and streams.

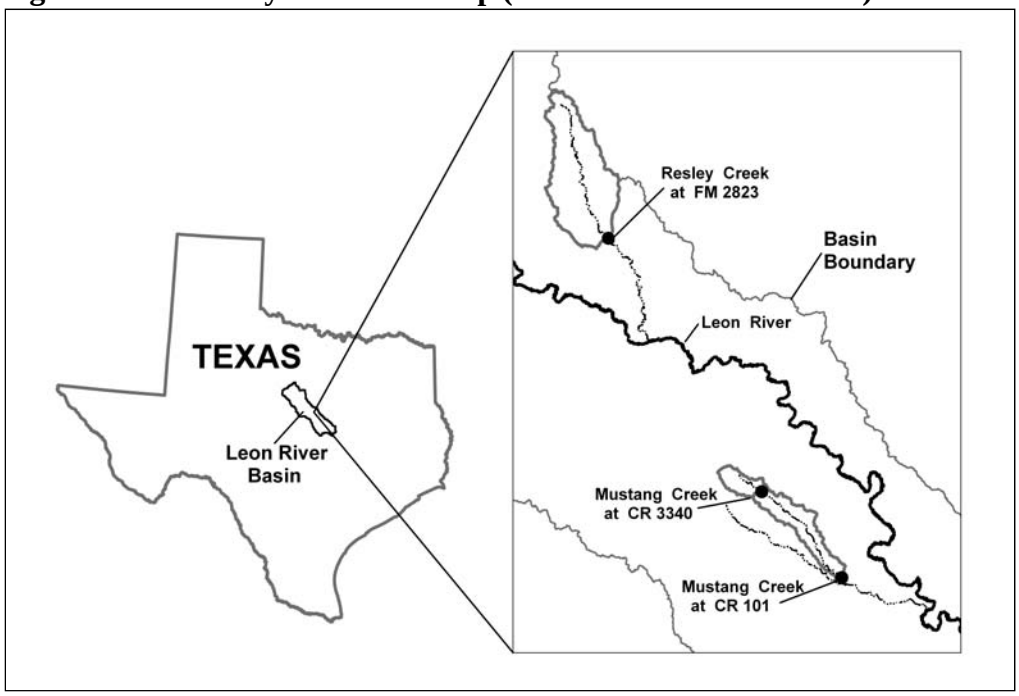
A6 PROJECT/TASK DESCRIPTION

The first portion of this monitoring and assessment project (Tasks 3, 4, & 5) will involve conducting sanitary surveys in the Cedar Creek watershed twice a year (winter and summer) to identify various dominant and relevant *E. coli* sources (cattle, poultry, deer, feral hogs, etc). The developed sanitary surveys will be updated with inputs from local experts (e.g. wildlife experts and enthusiasts, stakeholders, county agents, farmers, citizens). *E. coli* loads resulting from waste streams will be quantified for each identified source in the Cedar Creek watershed by utilizing the *E. coli* concentration determined (Task 4) and calculated population densities for the corresponding source from sanitary surveys (Task 3) (for example if "dairy cows" are a source, excreted manure, flushed manure, separated solids, lagoon wastewater, composted dairy manure, etc will be waste streams). *E. coli* concentration resulting from characterized waste streams for all dominant identified sources (five maximum) will be quantified. Random and representative samples of identified waste streams and fecal material of identified species will be collected. Four sub-samples will be collected for each waste stream/fecal material and a composite sample will be made on-site. Each composite sample will be transferred to a sterile 1 L bottle. All sample bottles will be stored in a cooler at 4°C and transported to the laboratory for *E. coli* analysis. *E. coli* from fecal samples will be elutriated using the method developed by Muirhead et al. (2005) and analyzed for *E. coli* by modified mTEC agar method (Method 1603; USEPA, 2002). Briefly, each composite sample collected will be extracted with sterile phosphate-buffered dilution water and filtered through a filter paper for *E. coli* incubation and enumeration using EPA method 1603.

This demonstration project also addresses another major concern stated by the Bacteria TMDL Task Force. This involves monitoring the survival, growth, re-growth, and die-off of *E. coli* in water under different environmental conditions. Currently, there is a significant knowledge gap about the fate of *E. coli* from various sources under different environmental conditions. Composite samples from each waste stream and species will be subjected to different temperatures (0°C, 10° C, 25° C, and 50°C), moisture conditions (0%, 5%, 25%, 50%, and 75% dry-basis), and pH (acidic, neutral, and alkaline). Growth and die-off at above mentioned environmental conditions will be monitored. Once optimum conditions are identified, re-growth will be monitored by bringing back the environmental conditions for the above mentioned scenarios to optimum conditions.

E. coli survival, growth, re-growth, and die-off in stream sediments (Task 6) and re-suspension of *E. coli* in streams are also poorly understood. In many cases, the re-suspension of *E. coli* has been suspected as a significant source of *E. coli* measured in the stream. Physical disturbances from humans, animals, waterfowl, fish, large rainfall events, and other events can significantly increase the amount of suspended sediments in the stream and thus may have a significant impact on the amount of *E. coli* suspended in the water column. Data will be collected from Resley Creek at FM2823, located in the Leon River watershed (Figure A6.1) under Baseflow and storm event conditions. Analysis will result in information about survival, growth, re-growth, and die off under all conditions.

Figure A6.1. Resley Creek Site Map (Task 6 Demonstration Site)



Sediment samples will be collected from 24 random locations along a 10-m segment of Resley Creek at FM 2823 and composited into one sample. Three sub-samples will be taken from the composite and brought to the laboratory in sterile containers kept at 4°C. These samples will be used to monitor survival, growth, re-growth, and die-off of *E. coli* under different temperatures (0°C, 10°C, 25°C, and 35°C), nutrient conditions (three different concentrations of organic carbon), pH (acidic, neutral, and alkaline), light intensity (three different light intensities), and chlorination (three residual chlorine concentration levels).

Presence of *E. coli* in stream water column and re-suspension of *E. coli* in Resley Creek will be monitored during four storm (runoff) events and four baseflow sampling events (two each during winter and summer for both baseflow and storm sampling). An automatic water sample collection system will be used to collect storm event samples. Streambed samples will be collected during or immediately following storm event water quality sampling. Sediment samples will be collected in sterile containers, stored at 4°C, transported to laboratory for *E. coli* analysis. Ambient baseflow samples will be collected along with samples immediately downstream of an area in which the streambed is mechanically agitated. Triplicate grab samples of water will be taken immediately prior to and immediately downstream following mechanical agitation. All samples will be stored in coolers at 4°C and transported to the laboratory for *E. coli* analysis.

Findings from this monitoring and demonstration study will be presented in a series of technical and non-technical publications. The fact sheets generated from this study will concisely present information related to fate and transport of *E. col*i in impaired watersheds and TMDL development process.

Additionally, all materials related to the project will be posted on a website developed specifically for this project. The website will also contain a brief description of the project, the need for the project, provide contact information for all parties involved, goals and objectives and any project updates that may occur. Findings from all Tasks, and specifically Task 6, will support the development and implementation of a WPP (TSSWCB 319 project 06-12) and TMDLs in the Leon River watershed. To produce timely results, the project will follow the timeline in Table A6.1.

Table A6.1. Project Plan Milestones

Task	Project Milestones	Agency	Start	End
1.1	Prepare quarterly reports	TWRI	10/07	8/11
1.2	Conduct quarterly meetings with project participants	TWRI	10/07	8/11
1.3	Conduct meetings with TSSWCB project manager as needed	TWRI	10/07	8/11
1.4	Submit Reimbursement Forms	TWRI	10/07	8/11
1.5	Develop and maintain website	TWRI	10/07	8/11
1.6	Prepare final report	TWRI, Research	04/10	8/11
1.7	Prepare publications for educational purposes	TWRI, Research	10/07	8/11
2.1	Develop QAPP	TWRI	10/07	04/08
2.2	QAPP Annual Revision #1	TWRI	02/09	05/09
2.2	QAPP Annual Revision #2	TWRI	02/10	08/10
3.1	Select watershed	Research	10/07	12/07
3.2	Conduct reconnaissance survey of watershed	Research	03/08	05/08
3.3	Conduct sanitary survey to identify winter wildlife sources	Research	12/08	02/09
3.4	Conduct sanitary survey to identify winter livestock & poultry sources	Research	12/08	02/09
3.5	Verify and update winter surveys	Research	12/08	02/09
3.6	Conduct sanitary survey to identify summer wildlife sources	Research	05/08	09/08
3.7	Conduct sanitary survey to identify summer livestock & poultry sources	Research	05/08	09/08
3.8	Verify and update summer surveys	Research	09/08	11/08
4.1	Collect winter fecal samples	Research	12/08	02/09
4.2	Extract winter fecal samples for *E. coli*	Research	12/08	02/09
4.3	Enumerate winter *E. coli*	Research	12/08	02/09
4.4	Calculate winter *E. coli* load	Research	02/09	04/09
4.5	Collect summer fecal samples	Research	07/08	09/08
4.6	Extract summer fecal samples for *E. coli*	Research	07/08	09/08
4.7	Enumerate summer *E. coli*	Research	07/08	09/08
4.8	Calculate summer *E. coli* load	Research	09/08	11/08
5.1	Prepare samples collected under task 4.5 for study	Research	09/08	8/11
5.2	Measure growth kinetics of *E. coli* under various conditions	Research	09/08	8/11
5.3	Measure survival of *E. coli* under various conditions	Research	09/08	8/11
5.4	Measure re-growth of *E. coli* under various conditions	Research	05/09	8/11
6.1	Collect water samples during summer for 2 runoff events	Research	05/08	05/10
6.2	Collect water samples during winter for 2 runoff events	Research	05/08	05/10
6.3	Collect stream sediments after each water sample collection	Research	05/08	05/10
6.4	Analyze water and stream sediment for *E. coli* concentrations	Research	05/08	8/11
6.5	Measure growth kinetics, survival, and re-growth of *E. coli* in stream sediments under different environment conditions	Research	06/08	8/11
6.6	Mechanically disturb stream sediments 4 times (2/summer & 2/winter, collect water and sediment samples and analyze for *E. coli*	Research	06/08	8/11

A7 QUALITY OBJECTIVES AND CRITERIA

Table A7.1 outlines measurement performance specifications needed to support project goals of:

1. Identifying, characterizing, and quantifying *E. coli* loads from various sources in an impaired watershed,
2. Monitoring survival, growth, re-growth, and die-off of *E. coli* under different environmental conditions,
3. Monitoring re-suspension of *E. coli* in streams,
4. Developing and disseminating clear and concise educational materials that can be used to educate the public on bacterial issues in the state, and
5. Strengthening spatially explicit load allocation tools and validate and improve process-based pathogen transport models used in TMDL development by providing scientific data.

Table A7.1. Data Quality Objectives for Measurement Data

Parameter	Units	Method Type	Enumeration Method	Method Description	AWRL	Precision of Laboratory Duplicates	Percent Complete
E. coli in water	CFU/ 100 mL	Membrane filter culture on modified mTEC agar	EPA 1603 (EPA, 2002)	Membrane Filter	1	3.27* ΣRlog/n	90
E. coli in feces and/or waste stream	CFU/ g dry weight	Samples will be elutriated & then membrane filter cultured on modified mTEC agar	EPA 1603 (EPA, 2002); Muirhead et al (2005)	Membrane Filter	1	3.27* ΣRlog/n	90
E. coli in sediment	CFU/ g dry weight	Membrane filter culture on modified mTEC agar	EPA 1603 (EPA 2002); Byappanahalli et al (2003)	Membrane Filter	1	3.27* ΣRlog/n	90

Ambient Water Reporting Limits (AWRLs)

The AWRL establishes the reporting specification at or below which data for a parameter must be reported based on given freshwater screening criteria. The AWRLs specified in Table A7.1 are the program-defined reporting specifications for each analyte and yield data of acceptable quality for assessment.

Precision

Precision of laboratory data is a measure of the reproducibility of a result from repeated analyses. It is defined as a measure of the closeness with which multiple analyses of a given sample agree with each other. Precision is assessed by repeated analyses of a sample. For quantitative microbiological analyses, the method used for calculating precision is outlined in Standard Methods for the Examination of Water and Wastewater, 20th Edition, section 9020 B.8.b.

$$RPD_{bacteria} = (\log X_1 - \log X_2)$$

The $RPD_{bacteria}$ should be lower than 3.27 ΣRlog/n, where Rlog is the difference in the natural log of duplicates for the first 15 positive samples.

Accuracy

Accuracy is a statistical measurement of correctness and includes components of systemic error. A measurement is considered accurate when the result reported does not differ from the true situation. Performance limits are specified in Table A7.1. An additional element of accuracy is the absence of contamination. This is determined through the analysis of blank samples of sterile water processed in a manner identical to the sample. Requirements for blank samples are discussed in Section B5.

Representativeness

Data collected under this project will be considered representative of ambient water quality conditions. Representativeness is a measure of how accurately a monitoring program reflects the actual water quality conditions typical of a receiving waterbody. The representativeness of the data is dependent on 1) the sampling locations, 2) the number of samples collected, 3) the number of years and seasons when sampling is performed, 4) the number of depths sampled, and 5) the sampling procedures. Site selection procedures will assure that the measurement data represent the conditions at the site. The goal for meeting total representation of the water body and watershed is tempered by the availability of time, site accessibility, and funding. Representativeness will be measured with the completion of sample collection in accordance with the approved QAPP.

Comparability

The comparability of the data produced is predetermined by the commitment of the staff to use only approved procedures as described in this QAPP. Comparability is also guaranteed by reporting all ambient, high flow, and QC data for evaluation by others.

Completeness

The completeness of the data is a measure of how much of the data is available for use compared with the total potential data. Ideally, 100% of the data would be available. However, the possibility of unavailable data due to accidents, weather, insufficient sample volume, broken or lost samples, etc. is to be expected. Therefore, it will be a general goal of the project(s) that 90% data completion is achieved.

A8 SPECIAL TRAINING/CERTIFICATION

No special certifications are required. However, field personnel will receive training in proper sampling. Before actual sampling occurs, field personnel will demonstrate their ability to properly perform field sampling procedures. Laboratory analysts have a combination of experience, education, and training to demonstrate knowledge of their function. To perform analyses for the TSSWCB, each laboratory analyst must demonstrate their capability to conduct each test that the analyst performs before analyzing samples and annually thereafter.

A9 DOCUMENTS AND RECORDS

Hard copies of general maintenance records, all field data sheets, chain of custody forms (COCs), laboratory data entry sheets, calibration logs, electronic forms of all project data, and corrective action reports (CARs) will be archived by each laboratory for at least five years. All electronic data will be backed up on an external hard drive monthly, compact disks weekly, and are simultaneously saved in an external network folder and the computer's hard drive. A blank CAR form is presented in Appendix A, a blank COC form is presented in Appendix B, and a blank bacteriological data log sheet is presented in Appendix C.

Quarterly progress reports will note activities conducted in connection with the water quality monitoring program, items or areas identified as potential problems, and any variations or supplements to the QAPP. CARs will be utilized when necessary. CARs that result in any changes or variations from the QAPP will be made known to pertinent project personnel and documented in an update or amendment to the QAPP. All quarterly progress reports and QAPP revisions will be distributed to personnel listed in Section A3.

The TSSWCB may elect to take possession of records at the conclusion of the specified retention period.

QAPP Revision and Amendments

Until the work described is completed, this QAPP shall be revised as necessary and reissued annually on the anniversary date, or revised and reissued within 120 days of significant changes, whichever is sooner. The last approved versions of QAPPs shall remain in effect until revised versions have been fully approved; the revision must be submitted to the TSSWCB for approval before the last approved version has expired. If the entire QAPP is current, valid, and accurately reflects the project goals and the organization's policy, the annual re-issuance may be done by a certification that the plan is current. This will be accomplished by submitting a cover letter stating the status of the QAPP and a copy of new, signed approval pages for the QAPP.

QAPP amendments may be necessary to reflect changes in project organization, tasks, schedules, objectives and methods; address deficiencies and nonconformance; improve operational efficiency; and/or accommodate unique or unanticipated circumstances. Written requests for amendments are directed from the TWRI Project Leader to the TSSWCB PM and are effective immediately upon approval by the TSSWCB PM and QAO, and EPA Project Officer. Amendments to the QAPP and the reasons for the changes will be documented and distributed to all individuals on the QAPP distribution list by the TWRI Project Leader or designee. Amendments shall be reviewed, approved, and incorporated into a revised QAPP during the annual revision process.

B1 SAMPLING PROCESS DESIGN

The main goals of the sampling are to:

1. Identify, characterize, and quantify *E. coli* loads resulting from various sources in an impaired watershed,
2. Monitor survival, growth, re-growth, and die-off of *E. coli* under different environmental conditions, and
3. Monitor re-suspension of *E. coli* in streams.

To achieve goals 1 and 2, data collection efforts will involve conducting summer and winter sanitary surveys in the Cedar Creek watershed (Table B1.1). Based on the results of the sanitary survey, fecal samples from the top 5 sources will be collected. *E. coli* levels in collected fecal samples will then be extracted and enumerated upon arrival at the lab and then daily thereafter following exposure to varying temperatures, moisture conditions, and pH levels to assess the survival growth, re-growth, and die-off of *E. coli* in fecal matter.

Table B1.1. Sample Sites and Monitoring Frequencies

Creek	Location Description	Monitoring Frequencies (per fiscal year) for each Parameter Group			
		Summer '08	Winter '08	Summer '09	Winter '09
Cedar	Brazos and Robertson county	Sanitary Survey Waste (*E. coli* source) collection			Sanitary Survey Waste (*E. coli* source) collection
Resley	FM 2823	Streambed disturbance Storm flow water Storm flow sediment Baseflow water Baseflow sediment	Streambed disturbance Storm flow water Storm flow sediment Baseflow water Baseflow sediment	Streambed disturbance Storm flow water Storm flow sediment Baseflow water Baseflow sediment	Streambed disturbance Storm flow water Storm flow sediment Baseflow water Baseflow sediment

Information gained will be used to develop and disseminate clear and concise educational materials that can be used to educate the public on bacterial issues in the state, strengthen spatially explicit load allocation tools, and validate and improve process-based pathogen transport models used in TMDL development by providing scientific data collected in this project. The constituents that will be measured are shown in Table B1.2.

Table B1.2. Constituents

Parameter	Status	Reporting Units
Escherichia coli in water	Critical	CFU per 100 milliliters (CFU/100 mL)
Escherichia coli in fecal matter	Critical	CFU per g of dry fecal matter (CFU/g)
Escherichia coli in sediment	Critical	CFU per g of dry sediment (CFU/g)

B2 SAMPLING METHODS

Source Identification and Fecal Sample Collection – Cedar Creek Watershed

The first portion of this project will involve conducting sanitary surveys in the Cedar Creek twice a year (winter and summer) to identify various dominant and relevant *E. coli* sources (e.g. cattle, poultry, deer, feral hogs). Sanitary surveys will be developed with input from local experts (e.g. wildlife experts and enthusiasts, stakeholders, county agents, farmers, citizens) and utilize cameras, fecal plots, and trapping (described below).

Cameras

AgriLife Research will set up approximately 30 remote digital cameras in the study area for 14-21 consecutive days during study seasons. Random locations for cameras will be decided by overlaid grid. Previous research suggests that certain species (i.e., white-tailed deer [*Odocoileus virginianus*], feral hogs [*Sus scrofa*], raccoons [*Procyon lotor*], opossums [*Didelphys virginiana*]]) generally have high population densities and are carriers. Research has been conducted to ascertain the fecal deposition rates of these species (Rowland et al. 1984, Acevedo et al. 2006). Based on an extensive literature review, we will use infrared triggered cameras to aid in determining relative abundances of mid-size to large mammals present (Trolle 2003). We will select approximately 30 random sample points and place remotely-operated infrared digital cameras for 14 consecutive days during study seasons for the 2 years of the study. Cameras will be placed at observed wildlife trails or openings suitable for camera placement (Claridge et al. 2004, Trolle and Kéry 2005).

Fecal Plots

We will use a random design to place 70-80 individual 600 m^2 transects within each study property (160 transects total per season). These plots will be recorded with handheld Global Positioning Systems (GPS) and entered into a GIS. Floodplain-scale estimates of the amount of species-specific fecal material will be extrapolated (Morrison et al. 2001) This will be supplemented by literature that approximates the fecal deposition rates of relevant species. These transects will: 1) serve as a rough check for the relative abundance estimates derived from the cameras, 2) aid in determination of species depositing fecal material, and 3) reveal the fecal deposition rate of each species. This will be supplemented by literature that approximates the fecal deposition rates of species.

Trapping

AgriLife Research will trap meso-mammals using a grid-design with 250 meter spacing between traps that has been shown to adequately sample animals that are highly attracted to aromatic baits (e.g., raccoons and opossums). They will use randomly located trap arrays in order to capture armadillos, rabbits, and skunks (e.g., species less attracted to baits). Focal species will be largely determined by species represented in fecal quadrat surveys. Relative abundances of trapped species will be calculated based on trap success (Geier and Best 1980, Fryxell et al. 1998). We will also conduct live-trapping of meso-carnivores in order to test fecal shedding rates (McCleery et al. 2005).

Animals will be released safely and once clear of the area, technicians will collect approximately 30 grams of feces (Table B2.1). After releasing animals from the trap and collecting a fecal sample, the cage will be cleaned and moved to prevent possible cross contamination of subsequent fecal samples. Traps will be closed every morning and reopened every evening during each trap session to prevent animals from being confined in cages in daylight hours. Traps will be set in shaded areas to reduce heat stress on the animals and for their safety. During periods of high temperature, trapping may be rescheduled.

Fecal Collection and Handling

Waste streams will be quantified for identified sources in the Cedar Creek watershed using *E. coli* concentration in collected waste samples and determined population densities from sanitary surveys. *E. coli* numbers from characterized waste streams for all dominant identified sources (5 maximum) will be quantified as follows: Random and representative samples of the 5 dominant sources will be collected by compositing four sub-samples each source on-site in a sterile 1 L bottle. All sample bottles will be stored in a cooler at 4°C and transported to the laboratory for *E. coli* analysis.

All collection and handling of fecal specimens will be performed using all safety precautions (wearing protective gears such as nitrile gloves will be strictly enforced). Specimens will be handled aseptically to ensure sample quality and minimize exposure of personnel to pathogens. All fecal material and waste collected will be placed in screw capped sterile containers (Table B2.1). Containers will be labeled with: Name of collector, date, species, GPS location, and photo of specimen before collection. Fecal specimens will be placed in an insulated cooler and transported to the College Station lab. All fecal material will be cultured within 24 hours of collection.

Table B2.1. Container Types, Preservation Requirements, Temperature, Sample Size, and Holding Time Requirements.

Parameter	Matrix	Container	Preservation	Temperature	Sample Size	Holding Time
E. coli	water	sterile plastic bag	none	4°C	125 mL	6 hours[1]
E. coli	sediment	sterile plastic bottle	none	4°C	30 g	24 hours
Fecal specimen	feces	sterile plastic bottle	none	4°C	30 g	48 hours

[1] 6 hours to deliver to laboratory. In the case that the 6-hour holding time is not met, the *E. coli* quantitative count will be flagged.

In-Stream Sampling Procedures – Resley Creek

Storm Event Water Sampling

In-stream storm event (runoff) flow water sampling will be conducted twice each in summer and winter at Resley Creek to determine water column concentrations and evaluate the effect of re-suspension of *E. coli* in stream sediments on stream water quality. Flow-weighted composite samples from Resley Creek will be collected using an ISCO 6712 full-size portable sampler with an eight bottle configuration into sterile two liter polyethylene bottles (Table B2.1). This will allow calculation of event mean concentrations (EMC) of bacteria for each rainfall event. The Program for the ISCO sampler on Resley Creek may be found in Appendix D.

Baseflow Water Sampling

In-stream Baseflow water sampling will be conducted twice each in summer and winter at Resley Creek to determine water column concentrations and evaluate the effect of re-suspension of *E. coli* in stream sediments on stream water quality. Triplicate water samples will be collected prior to and following mechanical agitation of stream sediment. Water samples will be collected directly from the stream (midway in the water column) into sterile Whirlpack® bags (Table B2.1). The sample container will be held upstream of the sampler and care will be exercised to avoid direct contact with bottom sediments and the surface micro layer of water, which may be enriched with bacteria and not representative of the water column. The top one inch of water will be squeezed from the bag before whirling and sealing. This airspace will help mix the sample when it is shaken just before making dilutions and membrane filtration.

Sediment Sampling

Concurrent with each water sampling event described above (both storm event and baseflow), sediment samples will be collected to assess the *E. coli* levels in the stream sediment. Sediment samples will be collected from the upper 2 cm using a sediment corer at 24 locations along transects in the wetted perimeter of Resley Creek at FM 2823 and composited into one sample. Three sub-samples will then be taken from the composite and each will placed into sterile 1 L bottles and transported to the lab for analysis.

Processes to Prevent Cross Contamination

To prevent cross-contamination, samples will be collected directly into sample containers. QC samples (Section B5) are also evaluated to verify that cross-contamination has not occurred.

Documentation of Field Sampling Activities

Field sampling activities are documented in field notebooks. For all visits, station ID, location, sampling time, sampling date, and sample collector's name/signature are recorded. Detailed observational data are also recorded including, but not limited to water appearance, weather, biological activity, stream uses, unusual odors, and specific sample information.

Recording Data

All field and laboratory personnel follow the basic rules for recording information including: (1) writing legibly in indelible, waterproof ink with no modifications, write-overs or cross-outs; (2) correcting errors with a single line followed by an initial and date; and (3) closing-out incomplete pages with an initialed and dated diagonal line.

Deviations from Sampling Method Requirements or Sample Design, and Corrective Action

Examples of deviations from sampling method requirements or sample design include but are not limited to such things as inadequate sample volume due to spillage or container leaks, failure to preserve samples appropriately, contamination of a sample bottle during collection, storage temperature and holding time exceedance, sampling at the wrong site, etc. Any deviations will invalidate resulting data and may require corrective action. Corrective action may include for samples to be discarded and re-collected. It is the responsibility of the TWRI Project Lead and QAO to ensure that the actions and resolutions to the problems are documented and that records are maintained in accordance with this QAPP. In addition, these actions and resolutions will be conveyed to the TSSWCB Project Manager both verbally and in writing in the project progress reports and by completion of a corrective action report (CAR).

Corrective Action Reports (CARs) document: root cause(s); programmatic impact(s); specific corrective action(s) to address any deviations; action(s) to prevent recurrence; individual(s) responsible for each action; the timetable for completion of each action; and the means by which completion of each corrective action will be documented. CARs will be included with project progress reports. In addition, significant conditions (i.e., situations which, if uncorrected, could have a serious effect on safety or on the validity or integrity of data) will be reported to the TSSWCB immediately both verbally and in writing.

B3 SAMPLE HANDLING AND CUSTODY

Chain-of-Custody

Proper sample handling and custody procedures ensure the custody and integrity of samples beginning at the time of sampling and continuing through transport, sample receipt, preparation, and analysis. The COC form is used to document sample handling during transfer from the field to the laboratory. The sample number, location, date, changes in possession and other pertinent data will be recorded in indelible ink on the COC. The sample collector will sign the COC and transport it with the sample to the laboratory. At the laboratory, samples are inventoried against the accompanying COC. Any discrepancies will be noted at that time and the COC will be signed for acceptance of custody. In the instance that the field sample collector and laboratory sample processor are one in the same, a field-to-lab COC will be unnecessary. A copy of a blank COC form used on this project is included as Appendix B.

Sample Labeling

Samples will be labeled on the container with an indelible, waterproof marker. Label information will include site identification, date, sampler's initials, and time of sampling. The COC form will accompany all sets of sample containers.

Sample Handling

Following collection, samples will be placed on ice in an insulated cooler for transport to the laboratory. At the laboratory, samples will be placed in a refrigerated cooler dedicated to sample storage. The Laboratory Director has the responsibility to ensure that holding times are met. The holding time is documented on the COC. Any problem will be documented with a CAR.

Failures in Chain-of-Custody and Corrective Action

All failures associated with chain-of-custody procedures as described in this QAPP are immediately reported to the TWRI Project Lead and QAO. These include such items as delays in transfer, resulting in holding time violations; violations of sample preservation requirements; incomplete documentation, including signatures; possible tampering of samples; broken or spilled samples, etc. The TWRI Project Lead and QAO will determine if the procedural violation may have compromised the validity of the resulting data. Any failures that have reasonable potential to compromise data validity will invalidate data and the sampling event should be repeated. The resolution of the situation will be reported to the TSSWCB Project Manager in the project progress report. Corrective action reports will be prepared by the TWRI Project Lead and QAO and submitted to the TSSWCB Project Manager along with project progress report.

B4 ANALYTICAL METHODS

Fecal/Waste Stream Sample Preparation

E. coli will be extracted from fecal/waste stream samples upon arrival at the lab to assess initial *E. coli* levels. *E. coli* will be extracted from the fecal/waste stream samples by placing 1 g of sample in 99 mL of sterile phosphate-buffered dilution water. Serial dilutions of the fecal/waste stream sample and water solution will then be prepared and analyzed using the membrane filtration technique described in "Water Sample Analysis" (below).

The fecal/waste stream samples will then be subjected to temperatures of 0^{o}C, 10^{o}C, 25^{o}C, and 50^{o}C; moisture conditions of 0%, 5%, 25%, 50%, and 75% dry basis; and acidic, neutral, and alkaline pH conditions. Samples will be incubated in an isocratic temperature-controlled incubator. Temperatures will be varied by adjusting the temperature-control knob of the incubator. Initial moisture content of the samples will be determined. Sterile DI water will be added to increase the moisture content or samples will be dried to decrease the moisture content. Initial pH of the samples will be determined. Acid or base solution will be added to adjust the pH to acidic or alkaline conditions. Growth and die-off of *E. coli* at these environmental conditions will be monitored through daily *E. coli* extractions. Once optimum conditions are identified, re-growth will be monitored by bringing back the environmental conditions for each of the scenarios described above to optimum conditions until a complete growth curve has been completed.

All handling of fecal specimens and cultures will be performed using a Class 2 biological safety cabinet to minimize the exposure of laboratory personnel to pathogens.

Sediment Sample Preparation

E. coli will be extracted from sediment samples upon arrival at the lab by placing 1 g of sample in 99 mL of sterile phosphate-buffered dilution water. Serial dilutions of the sediment and water solution will then be prepared and analyzed using the membrane filtration technique described in "Water Sample Analysis" (below).

Sediment samples will then be subjected to temperatures of 0°C, 10°C, 25°C, and 35°C; three different concentrations of organic carbon; acidic, neutral, and alkaline pH conditions; three different light intensities; and three residual chlorine concentration levels. Samples will be incubated in an isocratic temperature-controlled incubator. Temperatures will be varied by adjusting the temperature-control knob of the incubator. Organic carbon in the form of compost (C/N ratio will be characterized before) will be added at three different concentrations. Acid or base solution will be added to adjust the pH to acidic or alkaline conditions. Three different light intensities will be provided using fluorescent lights. Three different chlorine concentrations will be simulated using bleach solution. Growth and die-off of *E. coli* at these environmental conditions will be monitored through daily *E. coli* extractions. Once optimum conditions are identified, re-growth will be monitored by bringing back the environmental conditions for each of the scenarios described above to optimum conditions until a complete growth curve has been completed.

Water Sample Analysis

E. coli in water, sediment, and fecal samples will be isolated and enumerated by laboratory personnel using modified mTEC agar, EPA Method 1603 [EPA/821/R-02/023. September 2002. *Escherichia coli* in Water by Membrane Filtration Using Modified Membrane-Thermotolerant *Escherichia coli* (modified m-TEC) Agar]. The modified mTEC method is a single-step method that uses one medium and does not require testing using any other substrate. The modified medium contains a chromogen, 5-bromo-6-chloro-3-indolyl-ß-D-glucuronide, which is catabolized to glucuronic acid and a red- or magenta-colored compound by *E. coli* that produce the enzyme ß-D-glucuronidase.

All laboratory sampling areas and equipment (incubator and filtering apparatus) will be sterilized with at least one or in any combination of the following methods--ethyl alcohol, bleach, UV light, or autoclave. All disposables will be placed in a heat-resistant biohazard bag and autoclaved prior to disposal.

Failures in Measurement Systems and Corrective Actions

Failures in field and laboratory measurement systems involve, but are not limited to such things as instrument malfunctions, failures in calibration, blank contamination, quality control samples outside QAPP defined limits, etc. In many cases, the field technician or lab analyst will be able to correct the problem. If the problem is resolvable by the field technician or lab analyst, then they will document the problem on the field data sheet or laboratory record and complete the analysis. If the problem is not resolvable, then it is conveyed to the Research Project Lead, who will make the determination in coordination with the TWRI QAO. If the analytical system failure may compromise the sample results, the resulting data will not be reported to the TSSWCB as part of this project. The nature and disposition of the problem is reported on the data report. The TWRI Project Lead and QAO will include this information in the CAR and submit with the Progress Report which is sent to the TSSWCB Project Manager.

B5 QUALITY CONTROL

Table A7.1 in Section A7 lists the required accuracy, precision, and completeness limits for the parameters of interest. It is the responsibility of the Project Leader to verify that the data are representative. The Project Leader also has the responsibility of determining that the 90% completeness criteria is met, or will justify acceptance of a lesser percentage. All incidents requiring corrective action will be documented through use of CARs (Appendix A). Laboratory audits, sampling site audits, and quality assurance of field sampling methods will be conducted by the TSSWCB QAO or their designee.

Laboratory Blanks

Laboratory blanks consist of 100-mL aliquots of sterile distilled water that are processed in the same manner as a field sample, at the beginning and the end of a sample set. They are used to assess the sterilization techniques employed throughout the sample process. Laboratory blanks will be included at the beginning and the end of the sample set for each sampling event. The analysis of laboratory blanks should yield a value of no colonies detected.

Positive Control

The Lab will maintain live *E. coli* in tryptic soy broth and kept refrigerated until needed. Each time a set of samples is run a positive control will be performed in the lab using the same media and 1 mL of live *E. coli* which will be added to 99 mL of sterile distilled water that will be run through the filter funnel system and the filter placed on the media. This control should always be positive for *E. coli* after recommended incubation time.

Laboratory Duplicate

Laboratory duplicates are used to assess precision. A laboratory duplicate is prepared by splitting aliquots of a single sample in the laboratory. Both samples are carried through the entire preparation and analytical process. Bacteriological duplicate analyses are performed on samples from the sample bottle on a 10% basis, or at a minimum rate of one per batch. Acceptability criteria are outlined in Table A7.1 of Section A7.

A bacteriological duplicate is considered to be a special type of laboratory duplicate. Results of bacteriological duplicates are evaluated by calculating the logarithm of each result and determining the range of each pair. The method, outlined in Standard Methods for the Examination of Water and Wastewater, 20th Edition, section 9020 B.8.b. is as follows:

$$RPD_{bacteria} = (\log X_1 - \log X_2)$$

The $RPD_{bacteria}$ should be lower than 3.27 ΣRlog/n as defined in Table A7.1, where Rlog is the difference in the natural log of duplicates for the first 15 positive samples. Precision limits for bacteriological analyses apply to samples with concentrations >10 org/100 ml.

Failures in Quality Control and Corrective Action

Notations of blank contamination will be noted in quarterly reports and the final report. Corrective action will involve identification of the possible cause (where possible) of the contamination failure. Any failure that has potential to compromise data validity will invalidate data, and the sampling event should be repeated. The resolution of the situation will be reported to the TSSWCB in the quarterly progress report. The CAR's will be maintained by the Project Leader and the TSSWCB PM.

B6 INSTRUMENT/EQUIPMENT TESTING, INSPECTION AND MAINTENANCE

To minimize downtime of all measurement systems, spare parts for field and laboratory equipment will be kept in the laboratory, and all field measurement and sampling equipment, in addition to all laboratory equipment, must be maintained in a working condition. All field and laboratory equipment will be tested, maintained, and inspected in accordance with manufacturer's instructions and recommendation in Standard Methods for the Examination of Water and Wastewater, 20th Edition. Maintenance and inspection logs will be kept on each piece of laboratory equipment and general maintenance checklists will be filled out for field sampling equipment, by the field technician, prior to each sampling event.

Records of all tests, inspections, and maintenance will be maintained and log sheets kept showing time, date, and analyst signature. These records will be available for inspection by the TSSWCB.

Failures in any testing, inspections, or calibration of equipment will result in a CAR and resolution of the situation will be reported to the TSSWCB in the quarterly report. The CARs will be maintained by the Project Leader and the TSSWCB PM.

Table B6.1. Equipment Inspection and Maintenance Requirements

Equipment	Relevant Testing, Inspection and Maintenance Requirement
Thermometers	SM 9020 B 3.a
Water deionization units	SM 9020 B 3.d
Media dispensing apparatus	SM 9020 B 3.f
Autoclaves	SM 9020 B 3.h
Refrigerator	SM 9020 B 3.i
Ultra Low Freezer	SM 9020 B 3.j
Membrane filter equipment	SM 9020 B 3.k
Ultraviolet sterilization lamps	SM 9020 B 3.l
Biological safety cabinet	SM 9020 B 3.m
Incubators	SM 9020 B 3.o
Glassware and plastic ware	SM 9020 B 4.a
Utensils and containers	SM 9020 B 4.b
Dilution water bottles	SM 9020 B 4.c

B7 INSTRUMENT/EQUIPMENT CALIBRATION AND FREQUENCY

All instruments or devices used in obtaining environmental data will be calibrated prior to use. Each instrument has a specialized procedure for calibration and a specific type of standard used to verify calibration.

All calibration procedures will meet the requirements specified in the EPA approved methods of analysis. The frequency of calibration as well as specific instructions applicable to the analytical methods recommended by the equipment manufacturer will be followed. All information concerning calibration will be recorded in a calibration logbook by the person performing the calibration along with the date and will be accessible for verification during either a laboratory or field audit.

All instruments or devices used in obtaining environmental data will be used according to appropriate laboratory or field practices. Written copies of SOPs are available for review upon request.

Standards used for instrument or method calibrations shall be of known purity and be National Institute of Standards and Technology (NIST) traceable whenever possible. When NIST traceability is not available, standards shall be of American Chemical Society (ACS) or reagent grade quality, or of the best attainable grade. All certified standards will be maintained traceable with certificates on file in the laboratory. Dilutions from all standards will be recorded in the standards log book and given unique identification numbers. The date, analyst initials, stock sources with lot number and manufacturer, and how dilutions were prepared will also be recorded in the standards log book.

Failures in any testing, inspections, or calibration of equipment will result in a CAR and resolution of the situation will be reported to the TSSWCB in the quarterly report. The CARs will be maintained by the Project Leader and the TSSWCB PM.

B8 INSPECTION/ACCEPTANCE OF SUPPLIES AND CONSUMABLES

All standards, reagents, media, plates, filters, and other consumable supplies are purchased from manufacturers with performance guarantees, and are inspected upon receipt for damage, missing parts, expiration date, and storage and handling requirements. Labels on reagents, chemicals, and standards will be examined to ensure they are of appropriate quality, initialed by staff member and marked with receipt date. Volumetric glassware will be inspected to ensure class "A" classification, where required. Media will be checked as described in quality control procedures. All supplies will be stored as per manufacturer labeling and discarded past expiration date. In general, supplies for microbiological analysis are received pre-sterilized, used as received, and not re-used.

B9 NON-DIRECT MEASUREMENTS

Water quality determinations at sampling sites will be based upon data collected during the time frame of this project. However, quality assured data collected within the Cedar Creek and Resley Creek watersheds through other projects will be used as supplemental information to meet data quality objectives (see Section A7). This data from other projects will be referred to as historical data. Included in this are nitrate, ammonium, and phosphorus runoff data collected by the USDA-ARS from Resley Creek at FM2823. USDA-ARS bi-weekly monitoring of Resley Creek at FM2823, CR309, and CR394 for *E. coli*, nitrate, ammonium, and phosphorus levels will also be utilized as appropriate to assist in the evaluation of findings from the fate and transport project.

B10 DATA MANAGEMENT

Field Collection and Management of Routine Samples

Field staff will visit Resley Creek 4 times a year for portions of 2 years to collect summer and winter storm event flow and baseflow water and sediment samples. In addition, this site will be visited at least monthly to maintain high flow sampling equipment. Field staff will visit the Cedar Creek watershed semi-annually in year 1 to perform summer and winter sanitary surveys and fecal collections. Site identification, date, time, personnel, and any comment concerning weather or conditions at the site are noted in the field notebook. A field notebook is filled out in the field for each site visit.

Samples collected at each site will be labeled and placed in an iced, insulated chest for transportation to the laboratory. A COC form will be used if the collecting technician is in fact not the same person receiving samples into the lab. Site name, time of collection, comments, and other pertinent data are copied from the field notebook to the COC. The COC and accompanying sample bags/bottles are submitted to laboratory analyst, with relinquishing and receiving personnel both signing and dating the COC. All samples transported or mailed to the Lab will be accompanied by COC sheets filled out by the field technician.

All COC, field observations, and bacteriological data will be manually entered into an electronic spreadsheet. The electronic spreadsheet will be created in Microsoft Excel software on an IBM-compatible microcomputer with a Windows XP Operating System. The project spreadsheet will be maintained on the computer's hard drive, which is also simultaneously saved in a network folder. All pertinent data files will be backed up monthly on an external hard drive. Current data files will be backed up on R/W CD's weekly and stored in separate area away from the computer.

Original data recorded on paper files will be stored for at least five years. Electronic data files will be archived to CD after approximately one year, and then stored with the paper files for the remaining 4 years.

Laboratory Data

All field samples will be logged upon receipt, COC's (if applicable) will be checked for number of samples, proper and exact identification number, signatures, dates, and type of analysis specified. The field technician will be notified if any discrepancy is found and proper corrections made. All samples will be stored at 4ºC until analysis. Bacteriological samples will be given a unique identification number and logged into an electronic spreadsheet. Enumerated bacteriological data will be manually entered into the spreadsheet for electronic storage. The electronic spreadsheet will be created in Microsoft Excel software on an IBM-compatible microcomputer with the Windows XP Operating System The project spreadsheet will be maintained on the computer's hard drive, which is also simultaneously saved in an external network folder. All pertinent data files will be backed up monthly on an external hard drive.

Current data files will be backed up on R/W CDs weekly and stored in separate area away from the computer. At least 10% of all data manually entered in the database will be reviewed for accuracy by the Project Leader to ensure that there are no transcription errors. Hard copies of data will be printed and housed in the laboratory for a period of five years. Any COC's and bacteriological records related to QA/QC of bacteriological procedures will be housed at the Lab.

Data Validation

Following review of laboratory data, any data entry that is not representative of environmental conditions, because it was generated through poor field or laboratory practices, will not be submitted to the TSSWCB. This determination will be made by the Project Leader, Project Co-Leaders, TSSWCB QAO, and other personnel having direct experience with the data collection effort. This coordination is essential for the identification of valid data and the proper evaluation of that data. The validation will include the checks specified in Table D2.1.

Data Dissemination

At the conclusion of the project, the Research Project Leader will provide a copy of the complete project electronic spreadsheet via recordable CD media to the TSSWCB PM, along with the final report. The TSSWCB may elect to take possession of all project records. However, summaries of the data will be presented in the final project report.

C1 ASSESSMENTS AND RESPONSE ACTIONS

The following table presents types of assessments and response actions for data collection activities applicable to the QAPP.

Table C1.1 Assessments and Response Actions

Assessment Activity	Approximate Schedule	Responsible Party	Scope	Response Requirements
Status Monitoring Oversight, etc.	Continuous	TWRI Project Manager	Monitoring of project status and records to ensure requirements are being fulfilled. Monitoring and review of laboratory performance and data quality	Report to TSSWCB in Quarterly Report. Ensure project requirements are being fulfilled.
Laboratory Inspections	Dates to be determined by TSSWCB QAO	TSSWCB QAO	Analytical and quality control procedures employed at laboratory	30 days to respond in writing to TSSWCB to address corrective actions
Monitoring Systems Audit	Dates to be determined by TSSWCB	TSSWCB QAO	Field sampling, handling and measurement; facility review; and data management as they relate to project	30 days to respond in writing to TSSWCB to address corrective actions

Corrective Action

The TWRI Project Leader is responsible for implementing and tracking corrective action procedures as a result of audit findings. Records of audit findings and corrective actions are maintained by the TSSWCB Project Manager and TWRI QAO. Corrective action documentation will be submitted to the TSSWCB Project Manager with the progress report.

If audit findings and corrective actions cannot be resolved, then the authority and responsibility for terminating work is specified in agreements or contracts between participating organizations.

C2 REPORTS TO MANAGEMENT

Quarterly progress reports will be generated by TWRI personnel and will note activities conducted in connection with the water quality monitoring program, items or areas identified as potential problems, and any variation or supplement to the QAPP. The CARs forms will be utilized when necessary (Appendix A) and will be maintained in an accessible location for reference at TWRI. The CARs that result in changes or variations from the QAPP will be made known to pertinent project personnel, documented in an update or amendment to the QAPP and distributed to personnel listed in Section A3. Following any audit performed by the TWRI, a report of findings, recommendations and responses are sent to the TSSWCB Project Manager in the quarterly/monthly progress report.

Field measurements and all sampling for the project will be done according to the QAPP. However, if the procedures and guidelines established in this QAPP are not successful, corrective action is required to ensure that conditions adverse to quality data will be identified promptly and corrected as soon as possible. Corrective actions include identification of root causes of problems and successful correction of identified problems. The CARs will be filled out to document the problems and the remedial action taken.

Laboratory data reports contain the results of all analyses, as well as specified QC measures listed in section B5. This information is reviewed by the Research Project Lead and TWRI QAO and compared to the pre-specified acceptance criteria to determine acceptability of data. This information is available for inspection by the TSSWCB.

D1 DATA REVIEW, VERIFICATION, AND VALIDATION

All data obtained from field and laboratory measurements will be reviewed and verified for conformance to project requirements, and then validated against the data quality objectives which are listed in Section A7. Only those data which are supported by appropriate quality control data and meet the data quality objectives defined for this project will be considered acceptable. This data will be submitted to the TSSWCB.

The procedures for verification and validation of data are described in Section D2, below. The Research Project Lead is responsible for ensuring that field data are properly reviewed and verified for integrity. The Research Project Lead is responsible for ensuring that laboratory data are scientifically valid, defensible, of acceptable precision and accuracy, and reviewed for integrity. The Research Project Lead and TWRI QAO will be responsible for ensuring that all data are properly reviewed and verified, validated, and submitted in the required format as described by the TSSWCB Project Manager. Finally, the Research Project Lead and TWRI QAO are responsible for validating that all data to be reported meet the objectives of the project and are suitable for reporting to TSSWCB.

D2 VERIFICATION AND VALIDATION METHODS

All field and laboratory data will be reviewed, verified and validated to ensure they conform to project specifications and meet the conditions of end use as described in Section A7. The staff and management of the respective field, laboratory, and data management tasks are responsible for the integrity, validation and verification of the data each task generates or handles throughout each process. The field and laboratory tasks ensure the verification of raw data, electronically generated data, and data on chain-of-custody forms and hard copy output from instruments.

Verification, validation and integrity review of data will be performed using self-assessments and peer review, as appropriate to the project task, followed by technical review by the manager of the task. The data to be verified (listed by task in Table D2.1) are evaluated against project specifications (Section A7) and are checked for errors, especially errors in transcription, calculations, and data input. Potential outliers are identified by examination for unreasonable data. If a question arises or an error or potential outlier is identified, the manager of the task responsible for generating the data is contacted to resolve the issue. Issues which can be corrected are corrected and documented electronically or by initialing and dating the associated paperwork. If an issue cannot be corrected, the Research Project Lead consults with the TWRI Project Lead to establish the appropriate course of action, or the data associated with the issue are rejected.

The Research Project Lead is responsible for validating that the verified data are scientifically valid, legally defensible, of known precision, accuracy, integrity, meet the data quality objectives of the project, and are reportable to TSSWCB. One element of the validation process involves evaluating the data for anomalies. The Research Project Lead and/or TWRI QAO may designate other experienced water quality experts familiar with the water bodies under investigation to perform this evaluation. Any suspected errors or anomalous data must be addressed by the manager of the task associated with the data, before data validation can be completed.

A second element of the validation process is consideration of any findings identified during the monitoring systems audit conducted by the TWRI QAO or TSSWCB QAO assigned to the project. Any issues requiring corrective action must be addressed, and the potential impact of these issues on previously collected data will be assessed. Finally, the Research Project Lead and/or TWRI QAO validates that the data meet the data quality objectives of the project and are suitable for reporting to the TSSWCB.

Table D2.1. Data Verification Procedures

Data to be Verified	Research Project Lead	TSSWCB PM/QAO
Collection and analysis techniques consistent with SOPs and QAPP	X	X
QC samples collected for all parameters as prescribed in the QAPP	X	X
Field documentation complete	X	X
Instrument calibration data complete	X	X
Bacteriological records complete	X	X
Sample documentation complete	X	X
Sample identifications	X	X
Chain of custody complete/acceptable	X	X
Sample preservation and handling	X	X
Holding times	X	X
QC samples analyzed at required frequencies	X	X
QC samples within acceptance limits	X	X
Instrument readings/printouts	X	X
Calculations	X	X
Laboratory data verification for integrity, precision, accuracy, and validation	X	X
Laboratory data reports	X	X
Data entered in required format	X	X
Site ID number assigned	X	X
Absence of transcription error	X	X
Reasonableness of data	X	X
Electronic submittal errors	X	X
Sampling and analytical data gaps	X	X

D3 RECONCILIATION WITH USER REQUIREMENTS

Data that have been reviewed, verified, and validated will be summarized for accuracy and their ability to meet the DQO's of the project and the informational needs of water quality agency decision-makers. These summaries will be included in the final report.

These data, and data collected by other organizations, will subsequently be analyzed and used for model development/refinement. Thus, data which do not meet requirements will not be submitted to the TSSWCB nor will be considered appropriate for any of the uses noted above.

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APPENDIX A. CORRECTIVE ACTION REPORT

Corrective Action Report

CAR #:_____

Date:_____ Area/Location:_____

Reported by:_____ Activity:_____

State the nature of the problem, nonconformance, or out-of-control situation:

_____
_____
_____
_____
_____

Possible causes:

_____
_____
_____

Recommended corrective action:

_____
_____
_____

CAR routed to:_____

Received by:_____

Corrective Actions taken:

_____
_____
_____
_____

Has problem been corrected? YES NO

Immediate Supervisor:_____

Project Leader:_____

Quality Assurance Officer:_____

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APPENDIX B. CHAIN-OF-CUSTODY FORM

TEXAS AGRILIFE RESEARCH
CHAIN OF CUSTODY RECORD

Project Name:					# of containers	Analyses Required											
Station ID	Date	Time (24hr)	Matrix	Description													Sample ID

Relinquished by: (Signature)	Date:	Time:	Received by: (Signature)	Date:	Time:	Laboratory remarks:
Relinquished by: (Signature)	Date:	Time:	Received by: (Signature)	Date:	Time:	Lab log #
Relinquished by: (Signature)	**Date:**	**Time:**	**Received for lab by: (Signature)**	**Date:**	**Time:**	**Laboratory Name:**

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APPENDIX C.
BACTERIOLOGICAL DATA LOG SHEET

Bacteriological Data Log Sheet-Membrane Filter

Sample Location	Sample Date	Sampler Initials	Time			Volume Filtered	Colony Count			Temperature		Analyst Initials	Comments
			Collected	Filtered	Counted		Colony Count X	100 mL / vol. filtered	#/100 mL	Initial ©	Final ©		

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APPENDIX D.
ISCO PROGRAM
FOR RESLEY CREEK
HIGH FLOW MONITORING

PROGRAM NAME:
"RESCK 2823"
SITE DESCRIPTION:
"RC 2823 "

UNITS SELECTED:
LENGTH: ft

UNITS SELECTED:
FLOW RATE: cfs
FLOW VOLUME: Mgal

BUBBLER MODULE:
DATA POINTS
"DATA SET 1"
8 POINTS ENTERED

---

15 MINUTE
DATA INTERVAL

8, 2.00 lit BTLS
45 ft SUCTION LINE
AUTO SUCTION HEAD
1 RINSES, 0 RETRIES

ONE-PART PROGRAM

PACING:
FLOW, EVERY
48.46 Mgal
NO SAMPLE AT START

DISTRIBUTION:
4 SAMPLES/BOTTLE
RUN CONTINUOUSLY

VOLUME:
450 ml SAMPLES

ENABLE:
LEVEL >1.650 ft

ENABLE:
ONCE ENABLED,
STAY ENABLED
NO SAMPLE AT ENABLE

ENABLE:
0 MINUTE DELAY TO
START OF SAMPLING

ENABLE:
0 PAUSE & RESUMES

NO DELAY TO START

LIQUID DETECT ON

QUICK VIEW/CHANGE

TAKE MEASUREMENTS
EVERY 1 MINUTES

DUAL SAMPLER OFF
BTL FULL DETECT OFF
TIMED BACKLIGHT

EVENT MARK SENT
DURING PUMP CYCLE

PUMP COUNTS FOR
EACH PURGE CYCLE:
200 PRE-SAMPLE
AUTO POST-SAMPLE

NO PERIODIC
SERIAL OUTPUT

INTERROGATOR
CONNECTOR
POWER ALWAYS ON

NO RAIN GAUGE

NO SDI-12 SONDE

AUTO SDI-12 SCAN OFF

I/O1= NONE
I/O2= NONE
I/O3= NONE

---

0 ANALOG OUTPUTS

NO DIALOUT
CONDITIONS SET