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Version 1

Standardized Sequencing Guidance for Microbiological Control Strains V.1

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Protocol status: Working

We use this protocol and it's working

Created: March 13, 2025

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Protocol Integer ID: 124351

Keywords: GenomeTrakr, Quality control, control strains, reference database, standardized sequencing guidance for microbiological control strain, microbiological control strain, sequencing laboratory control, standardized sequencing guidance, processing foodborne pathogen isolate, foodborne pathogen isolate, sequencing, genomic diversity of control, genomic diversity, laboratory control, public health laboratory, significant false positives in lab, bioproject, laboratory, significant false positive, new quality control step, strain

Abstract

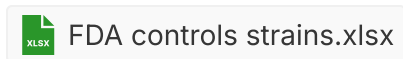
This protocol provides guidance for sequencing laboratory control strains and submitting those data to NCBI. The BioProject established for this project aims to capture the genetic/genomic diversity of control strains being used in public health laboratories. This resource will support a new quality control step for identifying rare but significant false positives in labs processing foodborne pathogen isolates, helping to reduce food waste and unnecessary product disposal.

- 1 The **Standardized Sequencing Guidance for Control Strains** protocol provides an SOP for generating high-quality whole-genome sequence data from bacterial control strains used in public health, academic, and industry laboratories sequencing bacterial isolates of foodborne pathogens.

By systematically sequencing and depositing these control strain genomes into a publicly accessible reference database at the National Center for Biotechnology Information (NCBI), this initiative aims to document the genomic diversity of strains used across laboratories globally. The resulting dataset will support new quality control measures to detect and mitigate rare but significant false-positive sample test results that could lead to unwarranted public health actions, such as food recalls or product disposal.

This protocol establishes a standardized approach to control strain sequencing, ensuring consistency across laboratories. The resulting database will serve both public health agencies as well as industry laboratories conducting food testing.

- 2 **Scope for control strains:** All actively used microbiological laboratory control strains are eligible for inclusion in this project. See Attached Table for a list of control strains used in FDA laboratories:



3 **Laboratory Methods:**

Control strain isolates should be processed according to standard GenomeTrakr or PulseNet laboratory procedures.

GenomeTrakr WGS workflow protocol collection provided here:

Protocol

NAME
GenomeTrakr WGS Protocol Collection and Workflow for MiSeq

CREATED BY
Tina Pfefer

Preview

4 Quality Assessment

Check that WGS sequence data meet the GenomeTrakr thresholds listed in the following protocol. Laboratories are welcome to use MicroRunQC or any other equivalent QC pipeline:

Protocol


NAME
**Quality control assessment for microbial genomes:
GalaxyTrakr MicroRunQC workflow**

CREATED BY
Ruth Timme

Preview

5 Metadata guidance:

Use the latest version of the **One Health Enteric BioSample package** and adhere to the guidelines in the following table:

Populate the CORE package attributes only.

A	B
Attribute Name	Guidance and examples
sample_name	Isolate ID provided by the laboratory
bioproject_accession	PRJNA981165
isolate_name_alias	other strain IDs for the isolate, separated by "; "
strain	ATCC #####
culture_collection	ATCC:#####
reference_material	laboratory control strain

A	B
organism	example "Salmonella enterica"
collected_by	Laboratory name where control strain is being used
collection_date	Collection date for sampling the control strain
cult_isol_date	BLANK
geo_loc_name	USA:State of laboratory location
isolation_source	laboratory control strain
source_type	Other
samp_collect_device	BLANK
purpose_of_sampling	Research
project_name	example "GenomeTrakr"
IFSAC_category	BLANK
lat_lon	BLANK
serotype	BLANK
serovar	BLANK
sequenced_by	Lab name performing the sequencing
description	Acquisition date of control strain = YYYY-MM-DD

Table: Metadata guidelines for control strain sequencing

6 NCBI submission:



Protocol



NAME

NCBI submission protocol for microbial pathogen surveillance

CREATED BY

Ruth Timme

Preview

BioProject: For labs collaborating with GenomeTrakr on this project, please use **PRJNA981165** (*FYI: these sequences will not appear in Pathogen Detection*).