

Jun 26, 2024

## NCBI submission protocol for HPAI milk surveillance

 Forked from [NCBI submission protocol for microbial pathogen surveillance](#)

DOI

**[dx.doi.org/10.17504/protocols.io.q26g711b1gwz/v1](https://dx.doi.org/10.17504/protocols.io.q26g711b1gwz/v1)**

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DOI: **[dx.doi.org/10.17504/protocols.io.q26g711b1gwz/v1](https://dx.doi.org/10.17504/protocols.io.q26g711b1gwz/v1)**

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**Manuscript citation:**

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**Protocol status:** In development

**We are still developing and  
optimizing this protocol.**

**Created:** January 09, 2024

**Last Modified:** June 26, 2024

**Protocol Integer ID:** 100855

**Keywords:** NCBI submission, pathogen surveillance, H5N1, HPAI, Influenza A, milk



## Disclaimer

Please note that this protocol is public domain, which supersedes the CC-BY license default used by protocols.io.

## Abstract

**PURPOSE:** This DRAFT protocol provides detailed instructions on how to submit raw targeted amplicon sequence data and associated contextual data of H5N1 to NCBI. The protocol includes essential steps to create a new NCBI submission environment for your laboratory group, which is crucial to have in place before data are submitted. After these initial setups, the the remaining protocol focuses on step-by-step instructions for data submission

**GUIDANCE FOR NEW SUBMITTERS:** Before initiating your first data submission, there is significant preparatory work required. We advise designating a team member to spend several days setting up the necessary systems well before your anticipated first submission.

Watch NCBI's [10min video tutorial](#) describing general submission to SRA.

**ADVICE FOR FREQUENT/LARGE VOLUME SUBMISSIONS:** Start by following Step 1 to establish your NCBI submission environment. For ongoing or large-scale submissions, email [gb-admin@ncbi.nlm.nih.gov](mailto:gb-admin@ncbi.nlm.nih.gov) to arrange an account for API-based submissions.

### Version history:

## Before start

### This protocol has three sections:

- **Section 1:** Setting up NCBI accounts (for new users)
- **Section 2:** Data submission to BioSample for sample metadata and to SRA for raw reads and associated sequence metadata.
- **Section 3:** Detailed steps for creating a BioProject (usually done once during the account set-up)



## Establish submission environment at NCBI

### 1 Set up a new NCBI submission environment for your lab:

**1.1:** Create an NCBI user account

**1.2:** Set up an NCBI submission user group for your lab

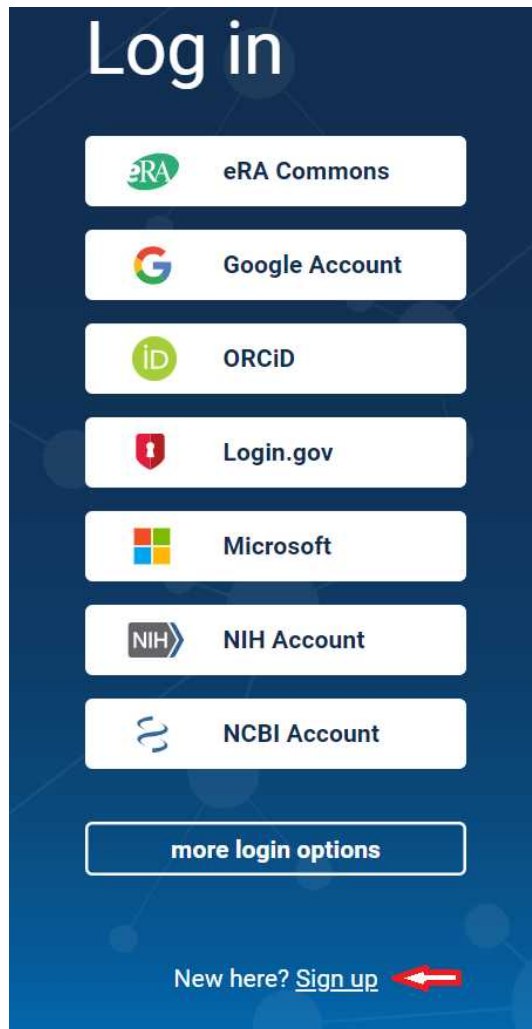
**1.4:** Bookmark the link to your submission portal

**1.5.** Identify or establish new BioProjects (detailed in **Step 3**)

#### **Ready for data submission:**

After these steps are complete you can proceed with data submission in **Step 2**.

- 1.1 **Create an NCBI user account** at NCBI: <https://www.ncbi.nlm.nih.gov/account>. This will be your own individual user account at NCBI.



The signup link is at the bottom of the page.

Choose a signup option that works for your institution.

## 1.2 Establish an NCBI submission user group for your laboratory.

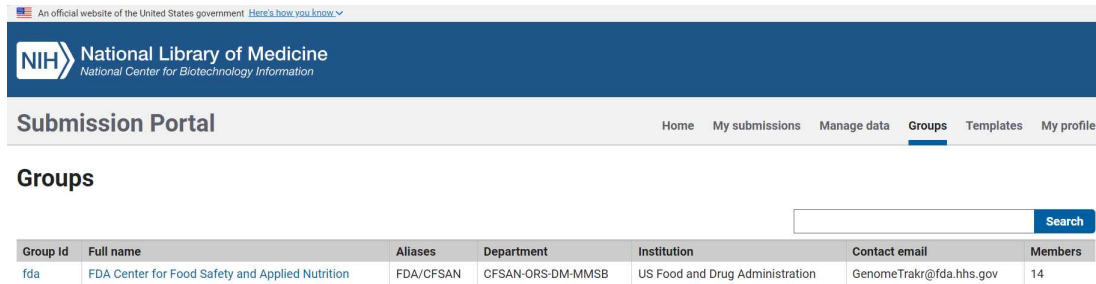
We recommend using this user group for *all* NCBI submissions related to your lab's pathogen genome surveillance submissions.

This approach will link data submitted by your lab to the ***user group*** and not to individuals doing the submissions, allowing anyone in the current submission group to perform updates or retractions and answer inquiries from the NCBI staff, even if there's been a complete turnover of staff since the original data were submitted.

User groups also ensure consistent data ownership across BioProjects, BioSamples, and sequence data. If your laboratory has non-overlapping research groups submitting and

managing data at NCBI, multiple user groups can be established, if needed, to manage these efforts separately.

Your laboratory might already have a submission group established! Sign into your personal NCBI account, then check the "Group" tab in the Submission Portal ([Submission Portal | NCBI | NLM | NIH](#)). Ask your colleagues to do the same to ensure your laboratory does not already have one in place.



View of the "Groups" tab, when selected from the NCBI Submission Portal

Click on this link to verify your membership in NCBI user groups:

<https://submit.ncbi.nlm.nih.gov/groups/>

### Creating a new submission group:

1. On your NCBI profile page (<https://submit.ncbi.nlm.nih.gov/accounts/profile/>), scroll to the bottom of the page and click on the **"Create group for shared submissions"** button.

#### Note

The "Create group for shared submissions" button will not exist, if the user has not filled in all of the required profile information, marked with an asterisk (\*) on the profile page.

## Submission Portal

### Personal Information

★ First (given) name	Middle name	★ Last (family) name		
<input type="text" value="Candace"/>	<input type="text" value="Hope"/>	<input type="text" value="Bias"/>		
★ Email (primary) <a href="#">Change primary email</a>				
<input type="text" value="Candace.Bias@fda.hhs.gov"/>		<input checked="" type="checkbox"/> Send email notifications on submission status change		
Email (secondary)				
<input type="text"/>				
★ Institution	Institution URL	★ Department		
<input type="text" value="US Food &amp; Drug Administration"/>	<input type="text"/>	<input type="text" value="Center for Food Safety and Nutrition"/>		
Phone ?	Fax ?			
<input type="text"/>	<input type="text"/>			
★ Street	★ City	★ State/Province	★ Postal code	★ Country
<input type="text" value="Campus Drive"/>	<input type="text" value="College Park"/>	<input type="text" value="MD"/>	<input type="text" value="20740"/>	<input type="text" value="USA"/>

Save

Back

### Groups

FDA Center for Food Safety and Applied Nutrition ([admin](#), [read](#), [modify](#), [submit](#), [delete](#))

**14 members:** Anjanette Johnston, Yan Luo, Errol Strain, Ruth Timme, Justin Payne, Narjol Gonzalez-Escalona, Hugh Rand, Maria Balkey, Maria Hoffmann, Jayanthi Gangiredla, Julie Haendiges, Yu Wang, Elizabeth Hunter, you

Create group for shared submissions



2. On the resulting page, fill in the required information to create, at minimum, a short name, full name, and contact information for this submission group.



Profile Members Invites Submissions

Open Invites tab to invite collaborators to join your group.

★ Short name	★ Full name
<input type="text" value="candace.bias@fd."/>	<input type="text" value="Candace Bias's shared submissions"/>
Aliases	Description
<input type="text"/>	<input type="text"/>
URL	
<input type="text"/>	

★ Email	Alternative email			
<input type="text" value="Candace.Bias@fda.hhs.gov"/>	<input type="text"/>			
★ Institution	★ Department			
<input type="text" value="US Food &amp; Drug Administration"/>	<input type="text" value="Center for Food Safety and Nutrition"/>			
Phone ?	Fax ?			
<input type="text"/>	<input type="text"/>			
★ Street	★ City	★ State/Province	★ Postal code	★ Country
<input type="text" value="Campus Drive"/>	<input type="text" value="College Park"/>	<input type="text" value="MD"/>	<input type="text" value="20740"/>	<input type="text" value="USA"/>

Save

3. To invite members, use either the "**Invite members**" button at the top of the next, "Members" tab or from the "Invites" tab itself to navigate to the invite tab and add the invitees' emails to the text box. Click the "**Invite Members**" button when finished.



## Submission Portal

### Groups > Candace Bias's shared submissions

Profile Members Invites Submissions

All invited members will be granted with **read, write, submit** and **delete** permissions, and they will have **email notifications** enabled.

✱ Emails ⓘ

1000 characters allowed

Invite members


ⓘ No pending invites.

### 1.3 Managing your NCBI submission user group.

After a user group has been established it can be edited for membership and permissions by clicking in the "group" tab of the submission portal (<https://submit.ncbi.nlm.nih.gov/groups/>), then on the Group Id hyperlink, e.g "fda\_ny" in the above example.

Users with admin privileges can update contact information in the "Profile" tab and membership in the "Members" tab. New members can be invited by clicking on the "Invite members" link.





National Library of Medicine  
National Center for Biotechnology Information

Submission Portal

Home
My submissions
Manage data
**Groups**
Templates
My profile

Groups > fda

Profile
Members
Invites
Submissions

Invite members

Search

Save permissions changes

First name	Last name	Email	Receives email notifications	Read	Modify	Submit	Delete	Admin	Remove member
			<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<div>Remove</div>
			<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<div>Remove</div>

This user list should be kept current as members/staff enter and leave the laboratory.

Permissions levels:

- **READ:** primarily for collaborators who would like to view the submissions, but not edit them.
- **MODIFY, SUBMIT, DELETE:** Permissions to submit, modify, or retract data (members usually have all or none of these permissions)
- **ADMIN:** Can invite or remove members of the submission group. Ensure that at least one (or more) members of your group have ADMIN privileges.

The "Submissions" tab will show a breakdown of how many submissions have been made by this group:

## Submission Portal

### Groups > fda

Profile Members Invites Submissions

All submissions	109148
api	108144
bioproject	150
biosample	573
genbank	6
sra	96
wgs	105
wgs1	8
wgs_batch	66

- 1.4 **Bookmark “My submissions” at NCBI:** <https://submit.ncbi.nlm.nih.gov/subs/>. This is the page where you view and track current and past submissions.

An official website of the United States government [Here's how you know](#)

**NIH** National Library of Medicine  
National Center for Biotechnology Information

## Submission Portal

Home **My submissions** Manage data Groups Templates My profile

### Your submissions

Start a new submission

- GenBank
- Sequence Read Archive
- Genome
- TSA
- BioProject
- BioSample
- Supplementary Files
- API

Filter / Search

From date: YYYY-MM-DD To date: YYYY-MM-DD Status: Not deleted Sort by:  ☐ desc

Apps + Data archives +

Query

1 2 3 4 5 6 7 8 9 ... 4,359 4,360 4,361 4,362 Next >

Submission	Title	App	Group	Status	Updated
SUB13884823	GenomeTrakr Project: University of California, Davis   School of Veterinary Medicine	BioProject	fda	✓ BioProject: Processed PRJNA1024537 : GenomeTrakr Project: University of California, Davis   School of Veterinary Medicine	<a href="#">Manage data</a> 05:00
SUB480036	Salmonella enterica Genome sequencing	BioProject	fda	✓ BioProject: Processed PRJNA1024537 : GenomeTrakr Project: University of California, Davis   School of Veterinary Medicine	<a href="#">Manage data</a> Jan 15



## 1.5 Identify or establish new BioProjects (Umbrella and/or Data BioProjects)

**Umbrella BioProjects.** If you are already part of a surveillance network, (e.g. GenomeTrakr, NARMS, Vet-LIRN, NAHLN, PulseNet, or others), you should follow the guidance from each network coordinator for creating new BioProjects.

If you need to establish a new umbrella BioProject, follow instructions in **Step 3** with modifications for creating a new Umbrella BioProject (**Step 3.12**).

**Data BioProjects.** Does your laboratory have an established data BioProject for this effort? If not, please follow the instructions in **Step 3** for creating a new one.

### Note

#### More information:

Learn more about **data** vs **umbrella** BioProjects in **Step 3**.

## 1.6 Download and populate metadata templates

Link to **VERY DRAFT** BioSample template (suggested picklist terms are in separate tab "vocabulary"):



OneHealth-Milk\_v0.1.xlsx

Link to **VERY DRAFT** SRA metadata template (picklist terms need to be added for H5N1 methods):



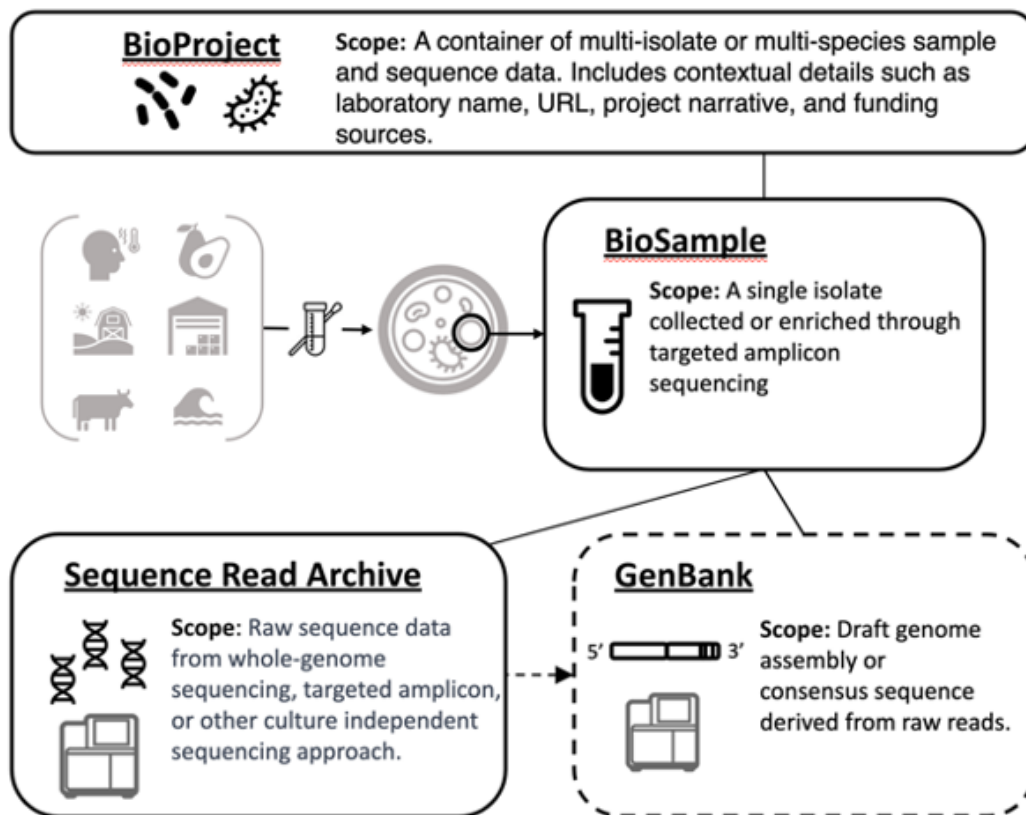
SRA-metadata-custom\_Jun26.xlsx

These templates are at the starting draft stage. Feel free to comment here for adding new picklist terms and/or attributes.

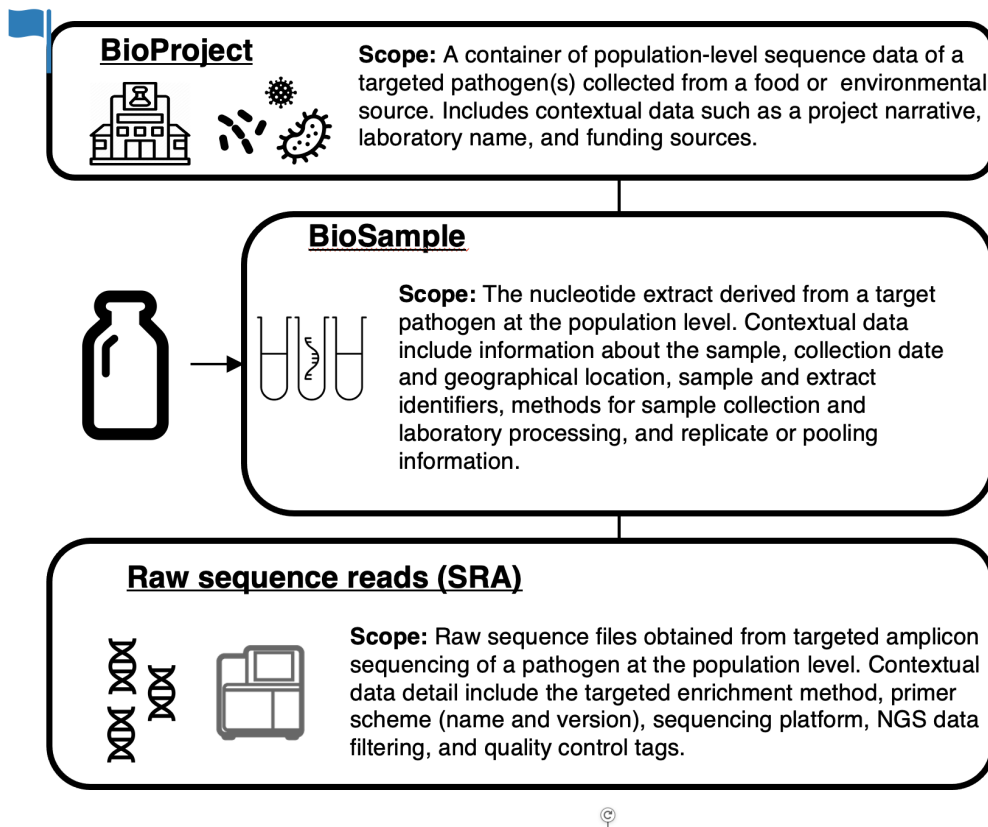
## Data submission (BioSample and SRA)

## 2 Data submission (Sample metadata, SRA metadata, and raw sequence data), compliant with the Pathogen DOM data structure.





For single isolate or expectation of pathogen clonality within the sample. For example, milk samples on-farm, collected from an animal or from a bulk tank.



Environmental Pathogen DOM for samples with reasonable expectation of population-level sequence data. Consensus sequences not expected from these data. For example, milk sampled from silos or from Retail Milk cartons.

#### Note

Arrange your submissions according to their corresponding BioProjects, ensuring that each submission workflow is dedicated to a single BioProject. In cases where your data encompass multiple BioProjects, initiate a distinct submission for each BioProject separately.

## 2.1 **\*\*Pre-Submission Data Quality Control\*\***

Verify that your sequence data meet the established quality control (QC) thresholds specific to your surveillance network.

<placeholder for QC check protocol>



2.2 Navigate to the **My Submissions** page in the NCBI Submission Portal:  
<https://submit.ncbi.nlm.nih.gov/subs/>

Click "Sequence Read Archive" to start a submission.

**Submission Portal** Home **My submissions** Manage data Groups Templates My profile

**Your submissions**

Start a new submission

- GenBank
- Sequence Read Archive
- Genome
- TSA
- BioProject
- BioSample
- Supplementary Files
- API

**Filter / Search**

From date: YYYY-MM-DD To date: YYYY-MM-DD Status: Not deleted Sort by:  ☐ desc

Apps:  Data archives:

Query:

1 2 3 4 5 6 7 8 9 ... 4,422 4,423 4,424 4,425 Next >

Submission	Title	App	Group	Status	Updated
SUB14137051	CDC HAI-Seq Gram-negative bacteria, Jan 05 '24	Sequence Read Archive (SRA)	scdhec_pngt	BioSample: <b>Processed</b> Successfully loaded (7 objects) Download attributes file with BioSample accessions SRA: <b>Processing</b>	15:00
SUB14140138	CDC NWSS - SARS-CoV-2 Wastewater Sequencing, Jan 08 '24	Sequence Read Archive (SRA)	wa_gt	BioSample: <b>Processed</b> Successfully loaded (33 objects) Download attributes file with BioSample accessions SRA: <b>Processed</b> (33 objects) Download metadata file with SRA accessions View and manage my SRA submission data	07:02
SUB480036	Salmonella enterica Genome sequencing	BioProject	fda	BioProject: <b>Processed</b> PRJNA242847 : GenomeTrakr Project: USDA - Food Safety and Inspection Service (TaxID: 28901) Locus Tag Prefixes: • A0J62 (SAMN04532063) • A0J63 (SAMN04532066) locustagprefix.txt	Jan 08

2.3 Click the "New submission" button.

**Submission Portal** Home **My submissions** Manage data Groups Templates My profile

**Sequence Read Archive (SRA)**

Short description and brief instructions

Options to upload data:

- Upload via Cloud: Amazon S3 or Google Cloud
- Upload via Aspera command line or FTP
- Upload via Web browser or Aspera browser plugin

**Filter / Search**

From date: YYYY-MM-DD To date: YYYY-MM-DD Status: Not deleted Sort by:  ☐ desc

Data archives:

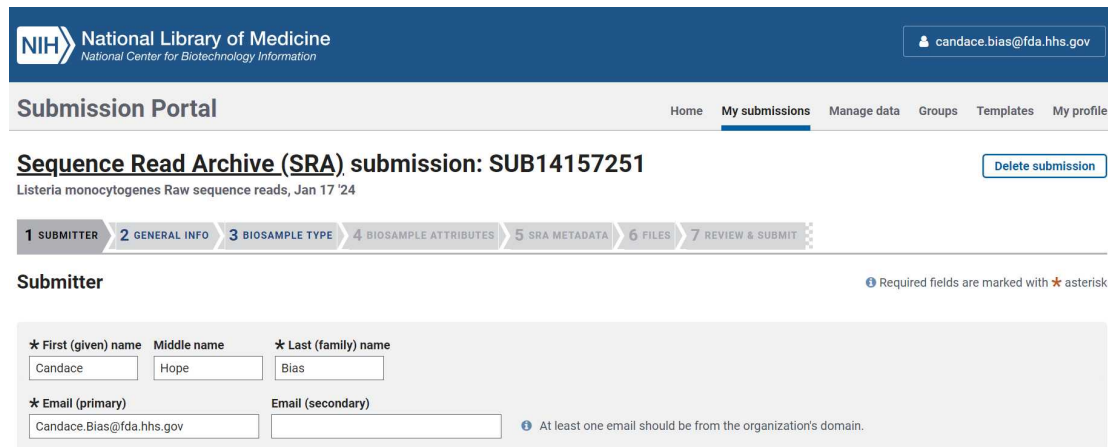
Query:

1 2 3 4 5 6 7 8 9 ... 38 39 40 41 Next >

## 2.4 SUBMITTER tab:

Populate with submitter info. The “submitter” is the name of the person AND user group, who is physically doing the submissions, not a supervisor or PI.

Select the appropriate submission group name (see **Step 1.2** for creating a new submission group), and describe the submitting organization or laboratory name. This will be auto-populated from the contact info you included in your NCBI user account. Click "Continue" to proceed.



The screenshot shows the NIH Submission Portal interface. At the top, the NIH logo and "National Library of Medicine" are displayed. A user profile for "candace.bias@fda.hhs.gov" is in the top right. The main header is "Submission Portal" with navigation links: Home, My submissions (active), Manage data, Groups, Templates, and My profile. Below this, the submission title is "Sequence Read Archive (SRA) submission: SUB14157251" with a "Delete submission" button. A progress bar shows steps 1 through 7, with "1 SUBMITTER" highlighted. The "Submitter" section contains form fields for:
 

- First (given) name: Candace
- Middle name: Hope
- Last (family) name: Bias
- Email (primary): Candace.Bias@fda.hhs.gov
- Email (secondary): [empty]

 A note at the bottom states: "At least one email should be from the organization's domain."

## 2.5 GENERAL INFO tab:

**1. BioProject:** Did you already have a data BioProject for this effort? If not please follow instructions in **Step 3** for creating a new data or umbrella BioProject. Return back to this sub-step with the **data BioProject** accession in hand.

Click "**Yes**" and paste in your data BioProject accession, e.g. PRJNA614995. Note: Be sure not to use an umbrella BioProject. Select the appropriate BioProject under the umbrella. Otherwise, you will receive an error and not be able to proceed.

**2. BioSample:** Click "NO" here. You will be registering BioSamples within this current submission.



**3. Release date:** Choose "Release immediately following processing".

4. Click **Continue**.

## Sequence Read Archive (SRA) submission: SUB14157251

Listeria monocytogenes Raw sequence reads, Jan 17 '24



### General Information

#### BioProject

BioProject describes the goal of your research effort.

★ Do you already have a BioProject accession number for this research?

☒ Yes ☐ No (a BioProject will be created within this submission)

★ Existing BioProject

PRJNA295367

PRJNA295367: Listeria monocytogenes Raw sequence reads

#### BioSample

The BioSample records the detailed biological and physical properties of the sample that was sequenced. A BioSample can be used in more than one BioProject since it should be used for all the data that were obtained from that sample. Usually SRA data sets are generated from more than one sample.

★ Do you already have BioSample accession numbers for these samples?

☐ Yes ☒ No (BioSamples will be created within this submission)

#### Release date

Note: Release of BioProject or BioSample is also triggered by the release of linked data.

★ When should this submission be released to the public?

☒ Release immediately following processing  
☐ Release on specified date or upon publication, whichever is first

Please allow 24-48 hours for propagation of the data to the NCBI SRA public site.

**Continue**

Example of filled in "General Info" tab. Please use the BioProject accession necessary for your organism and project.

## 2.6 BIOSAMPLE TYPE tab:

Choose the appropriate metadata package here for your sample (which sample template did you populate?)





We recommend using the **One Heath Enteric** package for milk samples.

⇒ [go to step #1.6](#)

National Library of Medicine  
National Center for Biotechnology Information

Submission Portal

Sequence Read Archive (SRA) submission: SUB14157251  
Listeria monocytogenes Raw sequence reads, Jan 17 '24

1 SUBMITTER 2 GENERAL INFO 3 BIOSAMPLE TYPE 4 BIOSAMPLE ATTRIBUTES 5 SRA METADATA 6 FILES 7 REVIEW & SUBMIT

Sample Type

★ Select the package that best describes your samples.

All packages Packages for MAG submitters Packages for metagenome submitters

(Optional) Filter packages by organism name

To filter for relevant BioSample packages, enter the full scientific name of the organism of your samples.

- If your BioSamples are derived from a species not represented in NCBI's Taxonomy database, enter the genus-level name, e.g., *Escherichia*.
- If your BioSamples are derived from more than one organism, enter the common species, genus, or family, e.g., *Enterobacteriaceae*.
- If your BioSamples are metagenomic/environmental, or metagenome-assembled genomes (MAG), select the appropriate tab above.
- For more information about organism names, see [Organism information](#).

NCBI packages [More...](#)

☐ SARS-CoV-2: clinical or host-associated  
Use for SARS-CoV-2 samples that are relevant to public health. Required attributes include those considered useful for the rapid analysis and trace back of SARS-CoV-2 cases.

☐ SARS-CoV-2: wastewater surveillance  
Use for SARS-CoV-2 wastewater surveillance samples that are relevant to public health. Required attributes include those considered useful for the rapid analysis and trace back of SARS-CoV-2 cases.

☐ Pathogen  
Use for pathogen samples that are relevant to public health. Required attributes include those considered useful for the rapid analysis and trace back of pathogens.

☒ One Health Enteric  
Use for microbial isolates that are collected for genomic surveillance of enteric pathogens. Sample spaces include the following: 1. human/animal hosts; 2. food samples; 3. food facilities; 4. environmental samples (farm, water, and the environment).  
US public health agencies have created customized versions of this package that include more specific guidance, controlled vocabulary picklists, and sub-packages for each of the 4 sample types.

- [GitHub repository](#)
- Validation for the OHE package

GSC MixS packages for genomes, metagenomes, and marker sequences [More...](#)

☐ MIGS Cultured Bacterial/Archaeal  
Use for cultured bacterial or archaeal genomic sequences. Organism must have lineage [Bacteria](#) or [Archaea](#).

☐ MIGS Eukaryotic  
Use for eukaryotic genomic sequences. Organism must have lineage [Eukaryota](#).

☐ MIGS Viral  
Use for virus genomic sequences. Organism must have lineage [Viruses](#).

☐ MIMAG Metagenome-assembled Genome  
Use for metagenome-assembled genome sequences produced using computational binning tools that group sequences into individual organism genome assemblies starting from metagenomic data sets. Organism cannot contain the term 'metagenome'. Use the MIUVIG package for virus genomes. Before creating BioSamples for prokaryotic and eukaryotic MAGs, please read and follow the [MAG submission instructions](#).

☐ MIMARKS Specimen  
Use for any type of marker gene sequences, eg, 16S, 18S, 23S, 28S rRNA or COI obtained from cultured or voucher-identifiable specimens. Organism cannot contain the term 'metagenome'.

☐ MIMARKS Survey related  
Use for any type of marker gene sequences, eg, 16S, 18S, 23S, 28S rRNA or COI obtained directly from the environment, without culture or identifier of the organism. Organism must be

Example "BioSample Type" tab. Note "One Health Enteric," recommended for GenomeTrakr submissions, has been selected.


## 2.7 BIOSAMPLE ATTRIBUTES tab:



Choose "Upload a file using Excel or text format (tab-delimited) that includes the attributes for each of your BioSamples".

Then click "Choose File" and browse to your populated metadata template.

#### Note

If you have not yet populated and validated your GenomeTrakr BioSample metadata template, 

**Antibiogram data:** please provide if you have it!

Click "**Continue**".

## Sequence Read Archive (SRA) submission: SUB14157251

Listeria monocytogenes Raw sequence reads, Jan 17 '24

1 SUBMITTER 2 GENERAL INFO 3 BIOSAMPLE TYPE 4 BIOSAMPLE ATTRIBUTES 5 SRA METADATA 6 FILES 7 REVIEW & SUBMIT

### Attributes

Package One Health Enteric; version 1.0

★ How do you want to provide your BioSample attributes?

- ☐ Use built-in table editor
- ☒ Upload a file using Excel or text format (tab-delimited) that includes the attributes for each of your BioSamples

★ Attributes file

OneHealthEntericMetadata\_listeria\_240118.xlsx 144.4 kB 2024-01-18 10:44

Delete

📎 Template for BioSample package One Health Enteric; version 1.0

[Download Excel](#) or [download TSV](#)

For column explanations and examples, please see the [sample attributes page](#).

For more information, please see [creating sample attribute file](#).

### Antibiogram — pathogen MIC (optional)

[Choose file](#) or drag and drop it here

📎 [Download Antibiogram Excel template](#)

Once the spreadsheet is completed, save as a 'Text (Tab-delimited)' file, and upload it.

For more information, please see [providing pathogen MIC antibiogram data](#).

### Antibiogram — mycobacterial non-MIC (optional)

[Choose file](#) or drag and drop it here

📎 [Download Antibiogram Excel template](#)

Once the spreadsheet is completed, save as a 'Text (Tab-delimited)' file, and upload it.

For more information, please see [providing mycobacterial non-MIC antibiogram data](#).

Continue

NCBI will do a validation check on your metadata. Resolve any red "errors" reported back by editing the spreadsheet and replacing the uploaded file. Review any yellow "Warnings" and proceed if everything looks ok.



## Note

If you are using the One Health Enteric Package BioSample metadata template downloaded from the CFSAN Biostatistics GitHub and receiving an error like the one below, an the empty, original version of the template may have been uploaded. Try again with your completed template.

### Sequence Read Archive (SRA) submission: SUB14159955

PulseNet Salmonella enterica Genome sequencing, Jan 18 '24

1 SUBMITTER

2 GENERAL INFO

3 BIOSAMPLE TYPE

4 BIOSAMPLE ATTRIBUTES

5 SRA METADATA

6 FILES

7

## Attributes



Error: Non-empty value in column without header: "Version Number: 1.3"



## Note

If you have followed One Health submission guidance and included the sub-species and serovar to the "Organism name" field, you may see the warning pictured below. You do not need to do anything in response to this warning. It exists merely to tell you that the "sub species" and "serovar" fields have been created in addition to the "Organism name" field. The metadata will be preserved in the Organism name as well as used to populate the new serovar and sub species attributes.



**Warning:** Provided taxonomy information was revised according to NCBI Taxonomy database rules. Please contact [biosamplehelp@ncbi.nlm.nih.gov](mailto:biosamplehelp@ncbi.nlm.nih.gov) if you have any questions.

Sample name	Organism name	Changed	Attribute name	Attribute value
SAL-23-VL-LA-PA-RYAN-0002	Salmonella enterica subsp. enterica serovar 4,[5],12:i:-	Yes	serovar	4,[5],12:i:-
SAL-23-VL-LA-PA-RYAN-0002	Salmonella enterica subsp. enterica serovar 4,[5],12:i:-	Yes	sub species	enterica
SAL-23-VL-LA-PA-RYAN-0003	Salmonella enterica subsp. enterica serovar Dublin	Yes	serovar	Dublin
SAL-23-VL-LA-PA-RYAN-0003	Salmonella enterica subsp. enterica serovar Dublin	Yes	sub species	enterica

Click "**Continue**".

## 2.8 SRA METADATA tab:

**Choose:** "Upload a file using Excel or text format (tab-delimited)".



## Sequence Read Archive (SRA) submission: SUB9477908

GenomeTrakr Project: US Food and Drug Administration, Apr 14 '21

1 SUBMITTER 2 GENERAL INFO 3 BIOSAMPLE TYPE 4 BIOSAMPLE ATTRIBUTES 5 SRA METADATA 6 FILES 7 REVIEW & SUBMIT

### SRA metadata

For more detailed help with SRA submission please read the [SRA Submission Wizard Help](#).

★ How do you want to provide your metadata?

- ☐ Use built-in table editor
- ☒ Upload a file using Excel or text format (tab-delimited)

★ Metadata file

SRA\_metadata-GTexample.xlsx 53.7 kB 2021-04-14 17:11

Delete

Use one of these ways to fill out SRA metadata:

[Download Excel spreadsheet](#) (designed to make it easier to select the correct metadata values), edit, save and then upload the modified Excel file.

Continue

Upload your populated **SRA metadata template** ( [go to step #1.6](#) for where to get this file)

Click "**Continue**".

NCBI will do a validation check on your sequence metadata. Resolve any red "errors" reported back by editing the spreadsheet and replacing the uploaded file. Review any yellow "Warnings" and proceed if everything looks ok.

Click "**Continue**".

## 2.9 Files tab:

Each laboratory will establish its own path for transferring files. Select the radio button corresponding to the means you will use.

In general, selecting the web browser option should work for uploading ~48 sequences at a time. For a more stable internet connection, your laboratory can use FTP or Aspera. Directions for doing so pop up after clicking the FTP radio button. Firewalls may prevent use of Aspera or AWS routes of submission.





### Note

It is generally not recommended to check the "Autofinish submission" box as this would not allow you to make corrections, if needed.

## Sequence Read Archive (SRA) submission: SUB14157251

Listeria monocytogenes Raw sequence reads, Jan 17 '24

1 SUBMITTER 2 GENERAL INFO 3 BIOSAMPLE TYPE 4 BIOSAMPLE ATTRIBUTES 5 SRA METADATA 6 FILES 7 REVIEW & SUBMIT

### Files

- Each file must be listed in the [SRA metadata table you uploaded](#). If you are uploading a tar archive, list each file name, not the archive name.
- Unique file names that do not contain any sensitive information should be used for all files. File names as submitted appear publicly when data is retrieved from the cloud.
- Files can be compressed using **gzip** or **bzip2**, and may be submitted in a **tar** archive, but archiving or compressing your files is not required. **Do not use zip!**

#### ★ How do you want to provide files for this submission?

- ☐ Web browser upload via HTTP or Aspera Connect plugin  
Do not use web browser HTTP upload if you are uploading files over 10 GB or more than 300 files.
- ☐ FTP or Aspera Command Line file preload  
All files for a submission must be uploaded into a single folder.
- ☐ AWS or GCP bucket

#### ☐ Autofinish submission

- i** Try to complete the submission automatically. Basic checks will be done on the files in the background. If there are no errors, this submission will be submitted for you. If the checks fail, you will get email notification to come back to fix the problems.

Continue

## 2.10 REVIEW & SUBMIT tab:

Check over your entire submission, then click "**Submit**."

If corrections are needed, you can go back and select individual tabs to edit your submission.



## Note

If you are having trouble finalizing your submission, contact the relevant NCBI database for assistance, and include your submission ID in the email subject (SUB#####):

**BioSample** (for source metadata issues): biosamplehelp@ncbi.nlm.nih.gov

**SRA** (for raw sequence or sequence metadata issues): sra@ncbi.nlm.nih.gov

## 2.11 BioSample accessions:

BioSample accessions will be automatically created upon submission and will be available on the “my submissions” page of the Submission portal by clicking on “## objects” within the submission record. You can also download by clicking the “Download attributes file with BioSample accessions”. Accessions will start with SAMNxxxxxxx. You will also receive an email within 12 hours, but typically much faster, containing these same accessions.

## 2.12 SRA Accessions:

SRA run accessions will be available on the “My Submissions” page of the Submission portal by clicking on “## objects” within the submission record. You can also download by clicking the “Download metadata file with SRA accession”. Accessions will start with SRRxxxxxxx.” You

will also receive an email with these same accessions within 24 hours, but typically much faster, containing these same accessions.

**U.S. National Library of Medicine**  
National Center for Biotechnology Information

**Submission Portal**

Home **My submissions** Manage data Groups Templates My profile

**Sequence Read Archive (SRA)** [New submission](#)

Short description and brief instructions +

Options to preload data:

- Aspera browser plugin upload +
- Aspera command line and FTP upload options +
- Amazon S3 instructions +

Filter / Search

From date To date Status Not deleted Sort by desc

Data archives +

Query Search Clear

30 submissions

Submission	Title	Group	Status	Updated
SUB5616822	GenomeTrakr Project: US Food and Drug Administration, May 14 '19	fda	✓ SRA: Processed (2 objects) • SRR9052981 • SRR9052982 Download metadata file with SRA accessions View and manage my SRA submission data	May 14
SUB5110190	GenomeTrakr Project: Bacillus cereus, New York State Department of Health, Wadsworth Center, Jan 31 '19	fda_ny	✓ SRA: Processed (6 objects) Download metadata file with SRA accessions View and manage my SRA submission data	Jan 31
SUB5106005	GenomeTrakr Project: NY State Dept. of Health, Wadsworth Center, Jan 30 '19	fda_ny	✓ SRA: Processed (20 objects) Download metadata file with SRA accessions View and manage my SRA submission data	Jan 30

## 2.13 Important data stewardship and curation notes:

- Develop an internal method for storing and tracking your BioSample and SRR accessions! They are required for making future updates to your records.
- For updates, corrections, or retractions to your BioSample and SRA records, some edits can be made within the submission portal and others need to be done via email.

### Safety information

**Caution:** It is possible for a single BioSample to have more than one SRR ID. Two scenarios include:

- Two runs were submitted for the same isolate/BioSample, which is not generally recommended for surveillance. Follow **Step 3** in the NCBI curation protocol to retract one of them).
- if the initial submission was retracted and a new run was submitted. It's important to keep track of both IDs, even if one was retracted.

## BioProject Creation

### 3 Create a new BioProject

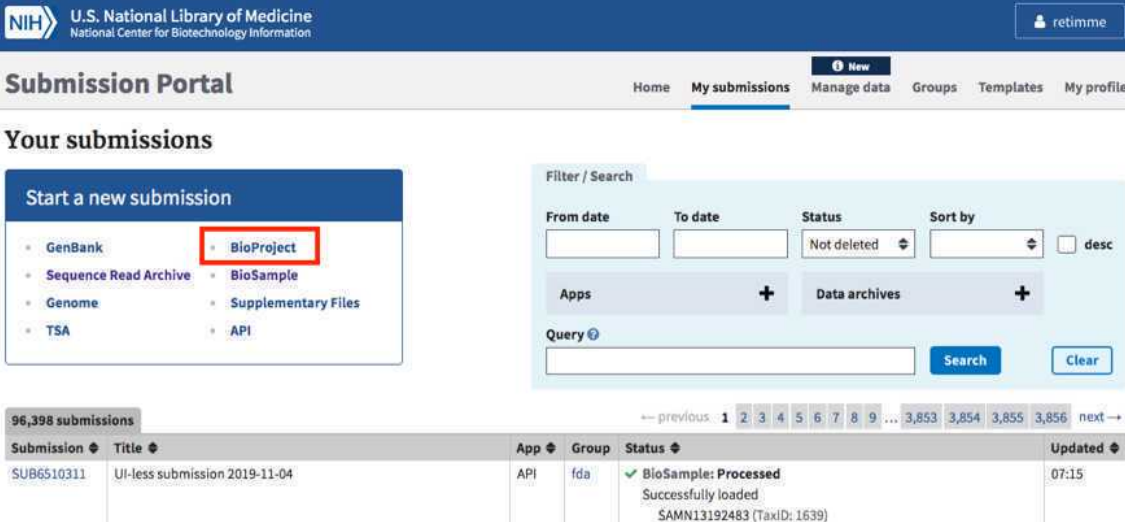
BioProjects are an organizing tool at NCBI that pulls together different kinds of data submitted across multiple NCBI databases. Each BioProject has a unique URL, providing a home page with a title, description, links to lab websites, publications, and funding resources associated with a particular project, along with links to the deposited data. A basic **data BioProject** holds actual sequence data, assemblies, and their associated metadata. An **umbrella BioProject** is a way to group two or more data BioProjects together, which is useful for coordinating disease surveillance and for looking across the grouped BioProjects in a single view.

This portion of the protocol describes the steps for creating a new data BioProject linked to an existing umbrella BioProject (usually established by a coordinating group, e.g. GenomeTrakr, NARMS, Vet-LIRN).

\*If you need to create a new Umbrella BioProject, modifications are summarized in Step **3.12**.

#### 3.1

**Navigate to the “My Submissions” page, <https://submit.ncbi.nlm.nih.gov/subs/>, and click “BioProject” in the “Start a new submission” box.**



The screenshot shows the NCBI Submission Portal interface. At the top, there's a header with the NIH logo and navigation links. Below the header, the 'Submission Portal' section is visible. On the left, under 'Your submissions', there's a 'Start a new submission' box. In this box, the 'BioProject' option is highlighted with a red rectangle. To the right of this box, there's a 'Filter / Search' section with various filters and a search bar. Below these, a table of submissions is displayed, showing columns for Submission ID, Title, App, Group, Status, and Updated. The first row shows a submission with ID SUB6510311, titled 'UI-less submission 2019-11-04', with App 'API' and Group 'fda'. The status is 'BioSample: Processed' and 'Successfully loaded', with a TaxID of 1639.

#### 3.2 Click the “New submission” box:



**NIH** U.S. National Library of Medicine  
National Center for Biotechnology Information

**Submission Portal** Home My submissions Manage data Groups Templates My profile

**BioProject** New submission

**ATTN:** to update an existing record or recent submission, please [email your request](#) with your BioProject ID or Submission ID included. **Do not** create new submission to update an existing submission!

Short description and brief instructions +

Filter / Search

From date To date Status Sort by

Not deleted desc

Data archives +

Query ? Search Clear

187 submissions

Submission	Title	Group	Status	Updated
SUB320199	Listeria monocytogenes Database: isolates from FDA-CFSAN	fda	BioProject: Processed PRJNA215355 : Listeria monocytogenes Database: isolates from FDA-CFSAN (TaxID: 1639) locustagprefix.txt	Nov 03

### 3.3 **Submitter tab:**

Populate with submitter info. An NCBI "submitter" is the name of the person or submission group who is managing the submissions, not a supervisor or PI.

Select the appropriate submission group name (see **Step 1.2** for creating a new submission group), and describe the submitting organization or laboratory name. This will be auto-populated from the contact info you included in your NCBI user account.

### 3.4 **Project type tab:**

Project data type: Genome sequencing and assembly.

Sample scope:

For a **Data BioProject**. Select multi-species. This will allow you to submit multiple different species to the BioProject.

### 3.5 **Target tab:**

For a **Data BioProject**. Populate ONLY the Organism name here:

**For targeted-pathogen BioProjects:**

Organism name = Include a Genus name, e.g., *Salmonella* sp.

**For non-targeted pathogens**

Organism name = "**Viruses**"

Create a description of the scope of the project (e.g. "enteric bacteria").

**3.6 General info tab:**

Click "Release immediately following processing".

Include a brief title describing the effort.

- **Data BioProject Title:** e.g., "GenomeTrakr Project: NY State Dept. of Health, Wadsworth Center".

Public Description: e.g., "Targeted amplicon sequencing of Influenza A (H5N1) as part of XXXX surveillance or research effort."

Relevance: environmental.

Is your project part of a larger initiative that is already registered at NCBI?

- **Data BioProjects.** Click "Yes" and include a brief description and umbrella BioProject accession number (see **Step 1.5**). This will properly link your data project to the umbrella.

**Note**

Note: We advise against linking data BioProjects to multiple umbrella BioProjects.



## BioProject submission: SUB12915263

FDA BioProject: bacterial pathogens



This submission was copied from SUB12914688.

1 SUBMITTER 2 PROJECT TYPE 3 TARGET 4 GENERAL INFO 5 BIOSAMPLE 6 PUBLICATIONS 7 REVIEW & SUBMIT

### General Info

#### Release date

**Note:** Release of BioProject or BioSample is also triggered by the release of linked data.

#### ★ When should this submission be released to the public?

- ☒ Release immediately following processing  
☐ Release on specified date or upon publication, whichever is first

#### ★ Project title

FDA BioProject: bacterial pathogens

#### ★ Public description

Whole genome sequencing of pure-cultured bacterial pathogens for FDA surveillance and outbreak investigations.

#### Relevance

Environmental

#### ★ Is your project part of a larger initiative which is already registered with NCBI?

- ☐ No ☒ Yes (not very common)

#### ★ Initiative description

GenomeTrakr umbrella for diverse pathogen species

#### ★ BioProject accession

PRJNA706684

If you are registering a project that is part of an initiative which is already registered in the BioProject database, then please tell us the existing BioProject accession and provide a general description of the larger initiative. For example, the ENCODE project and Human Microbiome project have several subprojects. This information is needed for project linking.

### External links

#### Description

#### URL

Delete

[Add another link](#)

### Select your grants

Use this tool to look up grants from many subscribed governmental funding agencies (eg NIH, CDC, FDA and VA) and some non-governmental funding sources (eg HHMI). You can search by grant number, title or grantee name. If your grant is not included, you can select the 'Add grants manually' option within this tool to add your grant.

[Add grants](#)

#### Consortium name

#### Consortium URL

#### Data provider

#### Data provider URL

Delete



 Add another data provider

### 3.7 ***BioSample tab:***

Leave blank!! You will create biosamples separately.

### 3.8 ***Publications tab:***

If relevant, include publications from your laboratory.

### 3.9 ***Review and Submit tab:***

Check if everything looks correct and edit if necessary, then click “**submit.**”



## Submission Portal

### BioProject submission: SUB12915263

FDA BioProject: bacterial pathogens



This submission was copied from SUB12914688.

1 SUBMITTER 2 PROJECT TYPE 3 TARGET 4 GENERAL INFO 5 BIOSAMPLE 6 PUBLICATIONS 7 REVIEW & SUBMIT

#### Review & Submit

This BioProject submission will be released **immediately following processing**.

##### Submitter

Submitter Ruth Timme  
ruth.timme@fda.hhs.gov

Submitting organization US Food and Drug Administration

##### Project type

Sample scope Multispecies

Target description Bacterial pathogens not included in the major taxa scoped for GenomeTrakr.

##### Target

Organism name (taxid) bacterial pathogens

##### General information

###### Project details

Parent project ID PRJNA706684

Parent project description GenomeTrakr umbrella for diverse pathogen species

Project type genome sequencing

Title FDA BioProject: bacterial pathogens

Description Whole genome sequencing of pure-cultured bacterial pathogens for FDA surveillance and outbreak investigations.

Relevance Environmental

Example for a new non-targeted BioProject

- 3.10 **The BioProject accession** will be available within a few minutes on the “My Submissions” page of the Submission portal in the format “PRJNAxxxxxx.” You will also receive an email containing the new accession.





3.11 If you are part of a coordinated surveillance effort, please alert the coordinating body that a new BioProject was created under an existing umbrella.

### 3.12 Creating a new Umbrella BioProject:

**Proceed as outlined in the above steps with the following modifications:**

#### PROJECT TYPE tab:

For an ***Umbrella BioProject***. Select multi-species. This will allow you to link multiple data BioProjects representing different species under a single umbrella.

-----

#### TARGET tab:

For an ***Umbrella BioProject***. Leave the Organism name field blank. Include a list or description of species you intend to include in this effort. E.g. "bacterial foodborne pathogens", or "SARS-Cov-2"

-----

#### GENERAL INFO tab:

***Umbrella BioProject*** Title: e.g. "Microbial pathogen surveillance at NY State Dept. of Health, Wadsworth Center."

Is your project part of a larger initiative that is already registered at NCBI?

- For an Umbrella BioProject: click "NO"

-----

The last step is to email [bioprojecthelp@ncbi.nlm.nih](mailto:bioprojecthelp@ncbi.nlm.nih):

Example email:

**Note**

"Dear BioProject and PD help teams,

Please convert the PRJNA##### to an Umbrella BioProject. Our laboratory will be submitting data under the XXX effort (SARS-CoV-2, GenomeTrakr, Vet-LIRN, NARMS, HAI, or more general pathogen surveillance).

I'd be happy to provide any additional details you might need.

Thank you, "

After the conversion is complete you can use the new Umbrella accession to properly link any new data BioProjects being created.

### 3.13 **Important data stewardship and curation notes:**

- Develop an internal method for storing and tracking your BioProject accessions! They are required for every BioSample and sequence data submission to ensure proper linkage.
- Bookmark URLs for each of your data BioProjects to monitor the public-facing view of your submissions.  
e.g. Virginia DCLS's GenomeTrakr *Salmonella* BP:  
<https://www.ncbi.nlm.nih.gov/bioproject/219491>
- For updates to your BioProjects, follow the guidance provided in the NCBI Curation Protocol. Some edits can be made within the submission portal and others need to be done via email.