

Instructions:

Each column that has a red triangle in the upper-right corner has a co

Some column headers have hyperlinks to NCBI webpages.

The YELLOW columns have drop-down menus that allow you to sele

You must save spreadsheet under second tab (SRA_data) as a T

If you created samples previously, provide accessions in the form of **SAMN#** in the column *sample_accession*. Otherwise provide the sample name used in the BioSample attributes spreadsheet.

Each row in the template represents a sequencing library with a unique combination of *sample + library + sequencing strategy + layout + instrument model*. Each row should have a **unique library_id** that is short and meaningful (like an ID you might use in lab).

When libraries are indeed identical (same combination of *sample + library + strategy + layout + instrument model*), all files should be placed in the same row. To do this simply enter the file names consecutively in the same row by adding more columns with headers filename2, filename3, etc.... PAIRED files must always be listed in the same row.

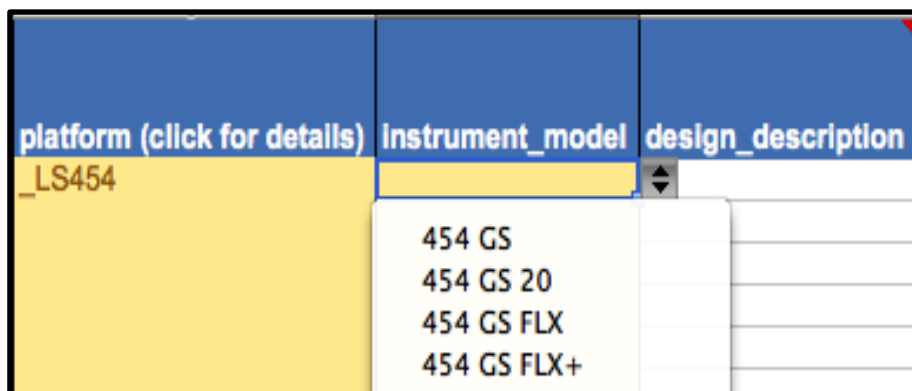
Provide **exact file names** (including extensions) in the *filename* columns.

File names must be **unique**.

red triangles indicate pop-up comments for that field

Many of the columns also have data checks - if you received a warnin

NOTE: There are data checks and autocomplete features in this sprea manually consult the platform and instrument information on the last



	454 GS FLX Titanium	
	454 GS Junior	
	unspecified	

Example Drop Down Menu

Helpful Hyperlinks:

SRA submission overview:

SRA submission in Submission Portal:

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comment that can be displayed if you hover over the header.

ect from a controlled vocabulary. Once specified for one row, these values can be copied-and-p

TSV (tab-delimited file) to upload the TSV file for the SRA metadata tab.

required for ALL data types	required for aligned data	paired-end data only
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g, please verify that you have entered a value from the drop-down menu. Example Below.

dsheet that are not compatible with Libre- and Open-Office. If you use one of these suites, please
page.

<http://www.ncbi.nlm.nih.gov/sra/docs/submit/>
<http://www.ncbi.nlm.nih.gov/sra/docs/submitportal/>

TSV (tab-delimited file) to upload the TSV file for the SRA metadata tab.

asted down.

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sample_name	library_ID	title	library_strategy
biosample_id	nwss_sample_idX	Targeted Amplification	on sequencing of was

<u>library_source</u>	<u>library_selection</u>	<u>library_layout</u>
stewater sample		

platform	instrument_model	design_description	filetype
ILLUMINA	Illumina iSeq 100	methods included in custom attributes	

filename

filename2 filename3 filename4

assembly fasta_file	enrichment_kit	amplicon_PCR_primer_scheme
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library_preparation_kit	amplicon_size	quality_control_method
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quality_control_method_version	quality_control_determination	quality_control_issues
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quality_control_details	dehosting_method	sequence_submitter_contact_email
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raw_sequence_data_processing_method

Column Name	Required?	Comments
sample_name	required for ALL data types	Targeted Amplification sequence
library_ID	required for ALL data types	
title	required for ALL data types	
<u>library_strategy</u>	required for ALL data types	
<u>library_source</u>	required for ALL data types	
<u>library_selection</u>	required for ALL data types	
library_layout	required for ALL data types	
<u>platform</u>	required for ALL data types	
instrument_model	required for ALL data types	
design_description	required for ALL data types	
filetype	required for ALL data types	
filename	required for ALL data types	
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filename3		
filename4		
assembly		
fasta_file		
enrichment_kit	required for ALL data types	
amplicon_PCR_primer_scheme	required for ALL data types	
library_preparation_kit	required for ALL data types	
amplicon_size		Would it work if both method a Is version required? Can be intentionally left blank Can be left blank if no issues po Any additional details one wish
quality_control_method		
quality_control_method_version		
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quality_control_details		
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sequence_submitter_contact_email		
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sed for amplification
or amplicon generation
eparation kit used for sequencing

nd version were in 1 cell?

ip up
es to include about QC goes here
nan-read scrubber. Note if other methods are used instead

ssing"

Strategy

WGA	Random sequencing of the whole genome following non-pcr amplification
WGS	Random sequencing of the whole genome
WXS	genome
RNA-Seq	Random sequencing of whole transcriptome
miRNA-Seq	Random sequencing of small miRNAs
WCS	Random sequencing of a whole chromosome or other replicon isolated from a genome
CLONE	Genomic clone based (hierarchical) sequencing
POOLCLONE	Shotgun of pooled clones (usually BACs and Fosmids)
AMPLICON	Sequencing of overlapping or distinct PCR or RT-PCR products
CLONEEND	Clone end (5', 3', or both) sequencing
FINISHING	Sequencing intended to finish (close) gaps in existing coverage
ChIP-Seq	Direct sequencing of chromatin immunoprecipitates
MNase-Seq	Direct sequencing following MNase digestion
DNase-Hypersensitivity	Sequencing of hypersensitive sites, or segments of open chromatin that are more readily cleaved by DNaseI
Bisulfite-Seq	Sequencing following treatment of DNA with bisulfite to convert cytosine residues to uracil depending on methylation status
Tn-Seq	Sequencing from transposon insertion sites
EST	Single pass sequencing of cDNA templates
FL-cDNA	Full-length sequencing of cDNA templates
CTS	Concatenated Tag Sequencing
MRE-Seq	Methylation-Sensitive Restriction Enzyme Sequencing strategy
MeDIP-Seq	Methylated DNA Immunoprecipitation Sequencing strategy
MBD-Seq	Direct sequencing of methylated fractions sequencing strategy
Synthetic-Long-Read	
ATAC-seq	Assay for Transposase-Accessible Chromatin (ATAC) strategy is
ChIA-PET	Direct sequencing of proximity-ligated chromatin immunoprec
FAIRE-seq	Formaldehyde Assisted Isolation of Regulatory Elements. reveal
Hi-C	Chromosome Conformation Capture technique where a biotin-
ncRNA-Seq	Capture of other non-coding RNA types, including post-translat
RAD-Seq	
RIP-Seq	Direct sequencing of RNA immunoprecipitates (includes CLIP-S
SELEX	Systematic Evolution of Ligands by EXponential enrichment
ssRNA-seq	strand-specific RNA sequencing
Targeted-Capture	
Tethered Chromatin Conformation Capture	

OTHER	Library strategy not listed (please include additional info in the “design description”)
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Source

GENOMIC	Genomic DNA (includes PCR products from genomic DNA)
TRANSCRIPTOMIC	Transcription products or non genomic DNA (EST, cDNA, RT-PCR, screened libraries)
METAGENOMIC	Mixed material from metagenome
METATRANSCRIPTOMIC	Transcription products from community targets
SYNTHETIC	Synthetic DNA
VIRAL RNA	Viral RNA
GENOMIC SINGLE CELL	
TRANSCRIPTOMIC SINGLE CELL	
OTHER	Other, unspecified, or unknown library source material (please include additional info in the “design description”)

Selection

RANDOM	Random selection by shearing or other method
PCR	Source material was selected by designed primers
RANDOM PCR	Source material was selected by randomly generated primers
RT-PCR	Source material was selected by reverse transcription PCR
HMPR	Hypo-methylated partial restriction digest
MF	Methyl Filtrated
CF-S	Cot-filtered single/low-copy genomic DNA
CF-M	Cot-filtered moderately repetitive genomic DNA
CF-H	Cot-filtered highly repetitive genomic DNA
CF-T	Cot-filtered theoretical single-copy genomic DNA
MDA	Multiple displacement amplification
MSLL	Methylation Spanning Linking Library
cDNA	complementary DNA
ChIP	Chromatin immunoprecipitation
MNase	Micrococcal Nuclease (MNase) digestion
DNase	Deoxyribonuclease (MNase) digestion
Hybrid Selection	Selection by hybridization in array or solution Reproducible genomic subsets, often generated by restriction fragment size selection, containing a manageable number of loci to facilitate re-sampling
Reduced Representation	
Restriction Digest	DNA fractionation using restriction enzymes
5-methylcytidine antibody	Selection of methylated DNA fragments using an antibody raised against 5-methylcytosine or 5-methylcytidine (m5C)
MBD2 protein methyl-CpG binding domain	Enrichment by methyl-CpG binding domain
CAGE	Cap-analysis gene expression
RACE	Rapid Amplification of cDNA Ends
size fractionation	Physical selection of size appropriate targets
Padlock probes capture method	Circularized oligonucleotide probes

other	Other library enrichment, screening, or selection process (please include additional info in the "design description")
unspecified	Library enrichment, screening, or selection is not specified (please include additional info in the "design description")
cDNA_oligo_dT	
cDNA_randomPriming	
Inverse rRNA	depletion of ribosomal RNA by oligo hybridization.
Oligo-dT	enrichment of messenger RNA (mRNA) by hybridization to Olig
PolyA repeat fractionation	PolyA selection or enrichment for messenger RNA (mRNA); sho Selection for less repetitive (and more gene rich) sequence thrc

Platforms

_LS454	_LS454	ILLUMINA
ABI_SOLID	454 GS	HiSeq X Five
BGISEQ	454 GS 20	HiSeq X Ten
CAPILLARY	454 GS FLX	Illumina Genome Analyzer
COMPLETE_GENOMICS	454 GS FLX+	Illumina Genome Analyzer II
HELICOS	454 GS FLX Titanium	Illumina Genome Analyzer IIX
ILLUMINA	454 GS Junior	Illumina HiScanSQ
ION_TORRENT		Illumina HiSeq 1000
OXFORD_NANOPORE		Illumina HiSeq 1500
PACBIO_SMRT		Illumina HiSeq 2000
		Illumina HiSeq 2500
		Illumina HiSeq 3000
		Illumina HiSeq 4000
		Illumina iSeq 100
		Illumina NovaSeq 6000
		Illumina MiniSeq
		Illumina MiSeq
		NextSeq 500
		NextSeq 550
		NextSeq 1000
		NextSeq 2000
		 Illumina HiSeq X

used to study genome-wide chromatin accessibility. alternative method to DNase-seq that uses formaldehyde crosslinking and immunoprecipitation.

Identifies regions of open chromatin

A labeled nucleotide is incorporated at the ligation junction, enabling selective purification of chromatin regions associated with specific RNA types such as snRNA (small nuclear RNA) or snoRNA (small nucleolar RNA), or

other RNA types (e.g., HITS-CLIP and PAR-CLIP).

o-dT.

uld replace cDNA enumeration.

ough Cot filtration (CF) or other fractionation techniques based on DNA kinetics.

HELICOS

Helicos HeliScope

ABI_SOLID

AB 5500 Genetic Analyzer

AB 5500xl Genetic Analyzer

AB 5500x-WI Genetic Analyzer

AB SOLiD 3 Plus System

AB SOLiD 4 System

AB SOLiD 4hq System

AB SOLiD PI System

AB SOLiD System

AB SOLiD System 2.0

AB SOLiD System 3.0

COMPLETE_GENOMICS

Complete Genomics

PACBIO_SMRT

PacBio RS

PacBio RS II

PacBio Sequel

PacBio Sequel II

s an engineered Tn5 transposase to cleave DNA and to integrate primer DNA sequences into the cle

meric DNA ligation junctions followed by deep sequencing

expression regulation types such as siRNA (small interfering RNA) or piRNA/piwi/RNA (piwi-interacti

ION_TORRENT

Ion Torrent PGM
Ion Torrent Proton
Ion Torrent S5 XL
Ion Torrent S5

CAPILLARY

AB 310 Genetic Analyzer
AB 3130 Genetic Analyzer
AB 3130xL Genetic Analyzer
AB 3500 Genetic Analyzer
AB 3500xL Genetic Analyzer
AB 3730 Genetic Analyzer
AB 3730xL Genetic Analyzer

OXFORD_NANOPORE

GridION
MinION
PromethION

BGISEQ

BGISEQ-500
DNBSEQ-G400
DNBSEQ-T7
DNBSEQ-G50
MGISEQ-2000RS

aved genomic DNA

ng RNA).

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amplicon_PCR_primer_scheme

ARTIC v5.4.2

ARTIC v5.3.2

ARTIC v5.3

ARTIC v5.2.0

ARTIC v5.1.0

ARTIC v5.0.0

ARTIC v4.1

ARTIC v4

ARTIC v3

ARTIC v2

ARTIC v1

NEB VarSkip Short / VarSkip Short v1a

NEB VarSkip Short v2 / VarSkip v2a

NEB VarSkip Short v2b

NEB VarSkip / VarSkip Long / VarSkip Long 1a

Midnight

Midnight v2

Midnight v3

AmpliSeq SARS-CoV-2 Research Panel (Illumina)

ARTIC SARS-CoV-2 Amplicon Panel (IDT)

CleanPlex SARS-CoV-2 FLEX Kit (Paragon Genomics)

CleanPlex SARS-CoV-2 Kit (Paragon Genomics)

CleanPlex SARS-CoV-2 Emerging Variants Panel Add-on v2 (Paragon Genom

CleanPlex Respiratory Virus Research Panel v2 (Paragon Genomics)

Clear Dx FlexPro: Wastewater (SARS-CoV-2) (Clear Labs)

Clear Dx SARS-CoV-2 WGS v2.0 (Clear Labs)

Clear Dx SARS-CoV-2 WGS v3.0 (Clear Labs)

COVID Mini v14: 576 samples (ONT)

COVIDSeq Assay (96 samples) (Illumina)

COVIDSeq Test (RUO Version) (Illumina)

COVIDSeq v3 Primer Pool (Illumina)
COVIDSeq v4 Primer Pool (Illumina)
EasySeq SARS-CoV-2 WGS Library Prep Kit v4 (NimaGen)
EasySeq SARS-CoV-2 WGS Library Prep Kit v5 (NimaGen)
HIFIVIRAL SARS-COV-2 (PacBio)
Ion AmpliSeq SARS-CoV-2 sequencing (Thermo Fisher)
Midnight RT PCR Expansion (ONT)
NEBNext ARTIC SARS-CoV-2 Companion Kit - ARTIC v3 (NEB)
NEBNext ARTIC SARS-CoV-2 Companion Kit - VarSkip Short v2 (NEB)
NEBNext ARTIC SARS-CoV-2 FS Library Prep Kit - ARTIC v3 (NEB)
NEBNext ARTIC SARS-CoV-2 FS Library Prep Kit - VarSkip Short v2 (NEB)
NEBNext ARTIC SARS-CoV-2 Library Prep Kit - ARTIC v3 (NEB)
NEBNext ARTIC SARS-CoV-2 RT-PCR Module - ARTIC v3 (NEB)
NEBNext ARTIC SARS-CoV-2 RT-PCR Module - VarSkip Short v2 (NEB)
QIAseq DIRECT SARS-CoV-2 (Qiagen)
QIAseq DIRECT SARS-CoV-2 Boosted (Qiagen)
SNAP - Swift Normalase Amplicon SARS-COV-2 Additional Genome Coverage
SNAP - Swift Normalase Amplicon SARS-COV-2 Panel (Swift Biosciences)
SNAP - Swift Normalase Amplicon SARS-COV-2 S Gene Panel (Swift Biosciences)
xGen SARS-CoV-2 Amplicon Panel (IDT)
xGen SARS-CoV-2 Midnight Amplicon Panel (IDT)
xGen SARS-CoV-2 S-gene Amplicon Panel (IDT)

amplicon_PCR_primer_scheme_no_longer_included

Cotten
Eden
Gonzalez-Reiche
Itokawa
Resende
SARS-CoV-2 SNAP primer pool

UNZA Sanger
Midnight 1200/Freed primers

library_preparation_kit

Covidmini kit
Illumina DNA prep
Illumina Nextera XT
Illumina TrueSeq
Illumina RNA Prep with Enrichment, (L) Tagmentation
Illumina COVIDSeq Assay
Ligation sequencing kit
Midnight expansion kit
NEBNext ARTIC SARS-CoV-2 Library Prep Kit (Illumina)
NEBNext Ultra II FS DNA Library Prep for Illumina
QIAseq DIRECT Unique Dual Index prep
Rapid barcoding kit

dehosting_method

ncov-dehoster
Nanostripper 0.1alpha
SRA human read removal tool

ics)

Panel (Swift Biosciences)

es)

quality_control_method_name

GalaxyTrakr SSQuAWK
CFSAN Wastewater Analysis Pipeline (C-WAP)

quality_control_method_version

quality_control_determination

no quality control issues identified
minor quality control issues identified
sequence flagged for potential quality control issues
not performed

quality_control_issues

low quality sequence
sequenced contaminated
low average genome coverage
low % genome captured
read lengths shorter than expected
sequence amplification artifacts
low signal to noise ratio
low coverage of characteristic mutatio

quality_control_details

ons

Update from v5.7 to v5.8

Date Updated: 2025-04-21

1. The **"enrichment_kit"** field is now a free-text field, allowing users to provide the name of the
2. The **"amplicon_PCR_primer_scheme"** field has been updated: outdated primer scheme optio

PCR kit used for amplification instead of selecting from a predefined drop-down list.

ns were removed, and the list was expanded to include primers and kits used for amplicon gene

eration.