DATA TERRA

ODATIS

SeBiMER
Bioinformatique marine

athENA
a tool for sequencing data management

Atelier technique #17

'Données bioinformatiques de diversité'

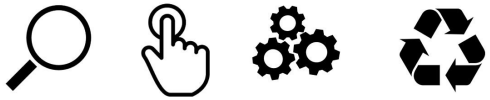
Pauline Auffret - SeBiMER

Tuesday, March 12th, 2024

contact@odatis-ocean.fr | www.odatis-ocean.fr

Contents

Introduction : be Findable Accessible Interoperable Reusable at Ifremer



Sequencing data life cycle at Ifremer

FAIR

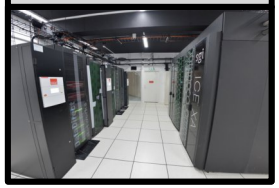
Experiment design

Data collection

Lab notebooks
BIGOOD
(BIology and GEology Ocean Database)

Data processing

Datarmor



Data analysis

DATAREF
Data storage and preservation

Disclosure & Publication

Sextant

CORE TRUST SEAL

doi

EMBL-EBI ENA

Data reuse

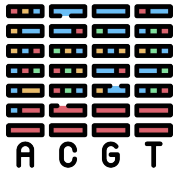
SeBiMER
Bioinformatique marine

Version control and code sharing
GitLab@Ifremer

DATA TERRA

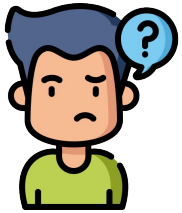
ODATIS

Sequencing data metadata

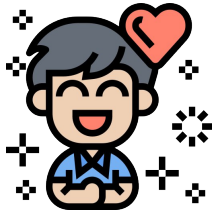


The importance of metadata

Metadata are **data about the data**



1



WHAT
WHO
WHEN
WHERE
HOW
WHY
…

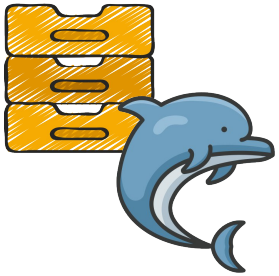
1. Example inspired by F. de Lamotte, IFB, *Introduction aux métadonnées*. https://ifb-elixirfr.github.io/IFB-FAIR-data-training/sequences/module3_sequence1_edition1_cours.html (Accessed on March 11, 2024)

What is athENA ?

athENA is a sequencing (meta)data management and brokering tool.

Three main goals :

- metadata collection in compliance with ENA metadata model ;
- sequencing data brokering to ENA database ;
- international indexing for Ifremer' sequencing data.



Local storage
(e.g. Dataref at Ifremer)

athENA

Metadata collection
Data brokering

ENA

Publicly available in international nucleotide databases

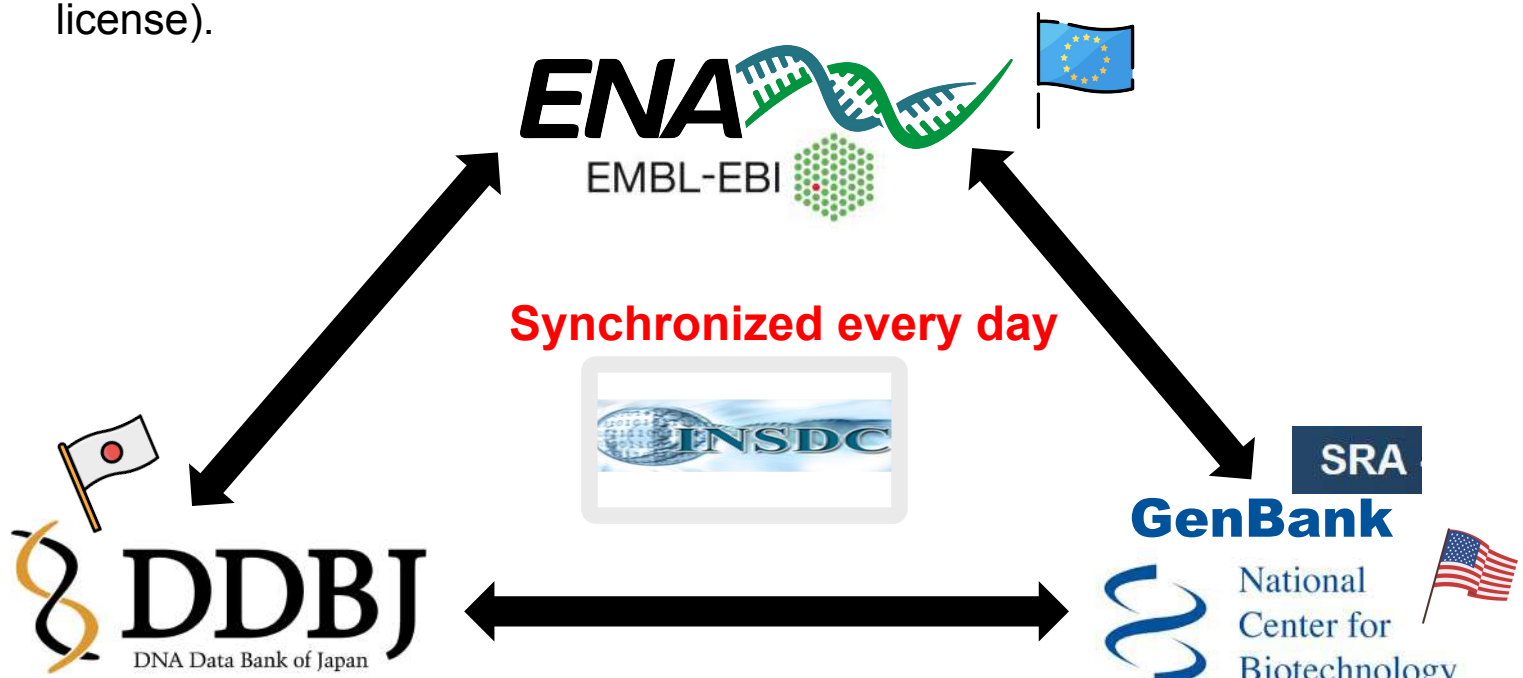
DATA TERRA

ODATIS

International referencing

The **International Nucleotide Sequence Database Collaboration (INSDC)** collects and disseminates international databases containing DNA and RNA sequences.

All data is **available** for free and unrestricted access, for any purpose, with no restrictions on analysis, redistribution, or re-publication of the data (CC-BY license).



The ENA metadata model

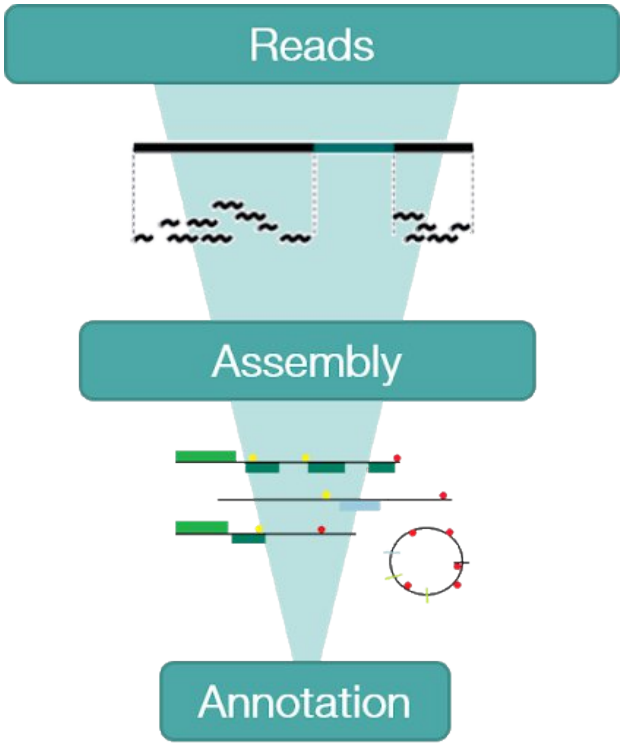
ENA : European Nucleotide Archive

ENA is based on the following principles :

- Three tiers data structure
- 6-object metadata model
- Sample metadata checklists
- Communities ontologies

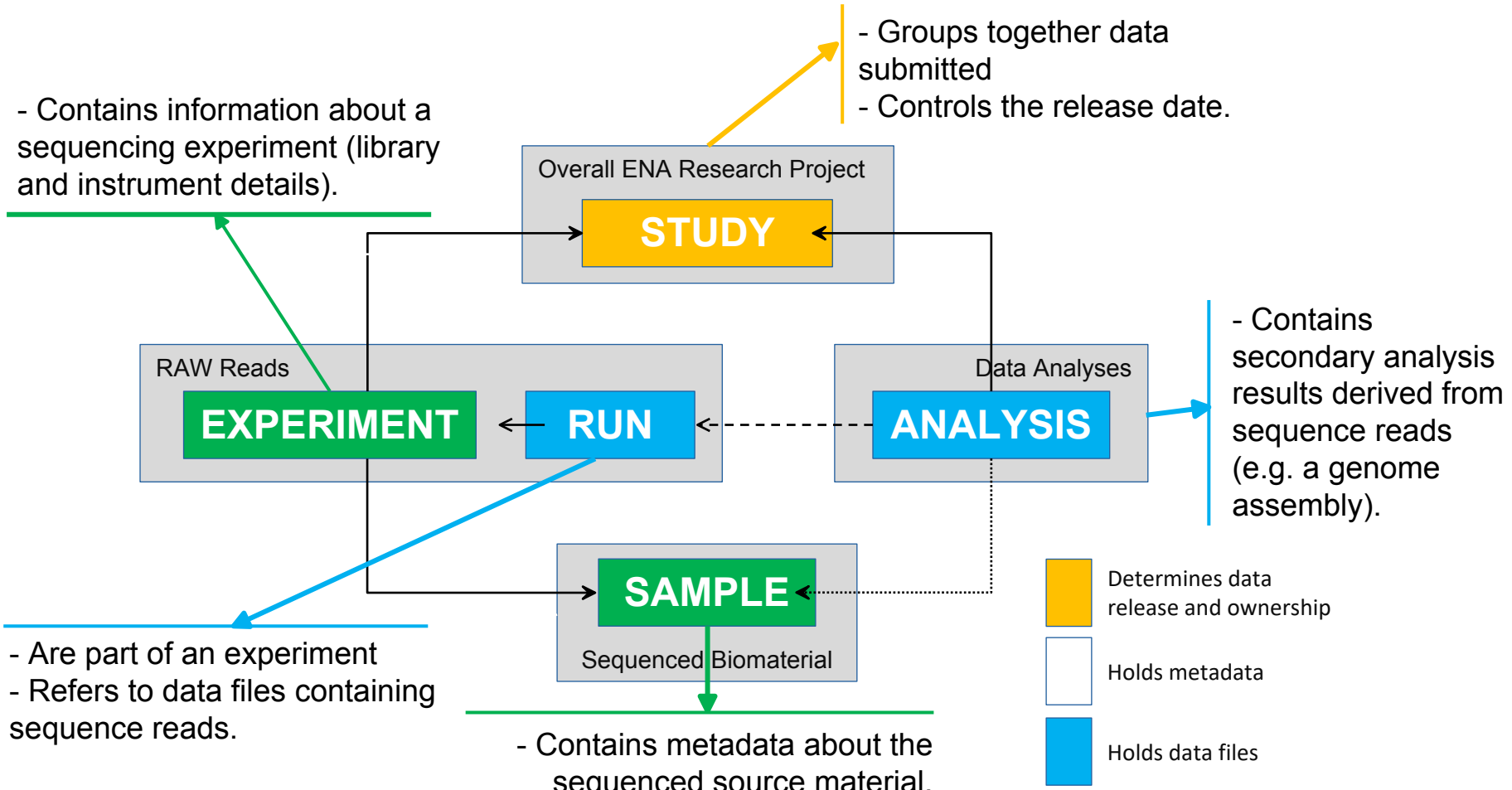
https://www.ebi.ac.uk/training/online/courses/ena-quick-tour/what-is-ena/

ENA 3 tiers data structure



https://www.ebi.ac.uk/training/online/courses/ena-quick-tour/what-is-ena/

ENA 6-object data structure

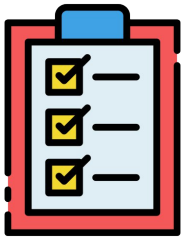


https://ena-docs.readthedocs.io/en/latest/submit/study.html

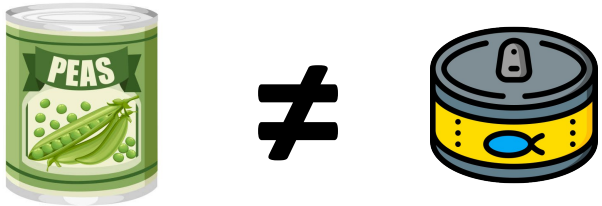
ENA sample checklists

ENA sample checklists ensure that a minimum amount of information is provided.

- There is a **minimum amount of information** required during ENA sample registration and all samples must conform to a **defined checklist of expected metadata values**.



- These sample checklists have been developed to **meet the needs of different research communities**. Different communities have different requirements on the minimum metadata expected to describe biological samples.



ENA sample checklists

14 pre-selected checklists for Ifremer projects

Accession	Name
ERC000011	ENA default sample checklist
ERC000012	GSC MIxS air
ERC000013	GSC MIxS host associated
ERC000014	GSC MIxS human associated
ERC000015	GSC MIxS human gut
ERC000016	GSC MIxS human oral
ERC000017	GSC MIxS human skin
ERC000018	GSC MIxS human vaginal
ERC000019	GSC MIxS microbial mat biolfilm
ERC000020	GSC MIxS plant associated
ERC000021	GSC MIxS sediment
ERC000022	GSC MIxS soil
ERC000023	GSC MIxS wastewater sludge
ERC000024	GSC MIxS water
ERC000025	GSC MIxS miscellaneous natural or artificial environment
ERC000027	ENA Micro B3
ERC000028	ENA prokaryotic pathogen minimal sample checklist
ERC000029	ENA Global Microbial Identifier reporting standard checklist GMI_MDM:1.1
ERC000030	ENA Tara Oceans
ERC000031	GSC MIxS built environment
ERC000032	ENA Influenza virus reporting standard checklist
ERC000033	ENA virus pathogen reporting standard checklist

Accession	Name
ERC000034	ENA mutagenesis by carcinogen treatment checklist
ERC000035	ENA Crop Plant sample enhanced annotation checklist
ERC000036	ENA sewage checklist
ERC000037	ENA Plant Sample Checklist
ERC000038	ENA Shellfish Checklist
ERC000039	ENA parasite sample checklist
ERC000040	ENA UniEuk_EukBank Checklist
ERC000041	ENA Global Microbial Identifier Proficiency Test (GMI PT) checklist
ERC000043	ENA Marine Microalgae Checklist
ERC000044	COMPARE-ECDC-EFSA pilot human-associated reporting standard
ERC000045	COMPARE-ECDC-EFSA pilot food-associated reporting standard
ERC000047	GSC MIMAGS (Minimum Information about a Metagenome-Assembled Genome)
ERC000048	GSC MISAGS (Minimum Information about a Single Amplified Genome)
ERC000049	GSC MIUVIGS (Minimum Information about an Uncultivated Virus Genome)
ERC000050	ENA binned metagenome
ERC000051	PDX Checklist
ERC000052	HoloFood Checklist
ERC000053	Tree of Life Checklist

ENA sample checklists

14 pre-selected checklists for Ifremer projects

Accession	Name
ERC000011	**ENA default sample checklist**
ERC000012	GSC MIxS air
ERC000013	**GSC MIxS host associated**
ERC000014	GSC MIxS human associated
ERC000015	GSC MIxS human gut
ERC000016	GSC MIxS human oral
ERC000017	GSC MIxS human skin
ERC000018	GSC MIxS human vaginal
ERC000019	GSC MIxS microbial mat biolfilm
ERC000020	GSC MIxS plant associated
ERC000021	**GSC MIxS sediment**
ERC000022	**GSC MIxS soil**
ERC000023	**GSC MIxS wastewater sludge**
ERC000024	**GSC MIxS water**
ERC000025	**GSC MIxS miscellaneous natural or artificial environment**
ERC000027	ENA Micro B3
ERC000028	ENA prokaryotic pathogen minimal sample checklist
ERC000029	ENA Global Microbial Identifier reporting standard checklist GMI_MDM:1.1
ERC000030	**ENA Tara Oceans**
ERC000031	GSC MIxS built environment
ERC000032	ENA Influenza virus reporting standard checklist
ERC000033	ENA virus pathogen reporting standard checklist

Accession	Name
ERC000034	ENA mutagenesis by carcinogen treatment checklist
ERC000035	ENA Crop Plant sample enhanced annotation checklist
ERC000036	**ENA sewage checklist**
ERC000037	ENA Plant Sample Checklist
ERC000038	ENA Shellfish Checklist
ERC000039	**ENA parasite sample checklist**
ERC000040	ENA UniEuk_EukBank Checklist
ERC000041	ENA Global Microbial Identifier Proficiency Test (GMI PT) checklist
ERC000043	**ENA Marine Microalgae Checklist**
ERC000044	COMPARE-ECDC-EFSA pilot human-associated reporting standard
ERC000045	COMPARE-ECDC-EFSA pilot food-associated reporting standard
ERC000047	**GSC MIMAGS** (Minimum Information about a Metagenome-Assembled Genome)
ERC000048	**GSC MISAGS** (Minimum Information about a Single Amplified Genome)
ERC000049	GSC MIUVIGS (Minimum Information about an Uncultivated Virus Genome)
ERC000050	**ENA binned metagenome**
ERC000051	PDX Checklist
ERC000052	HoloFood Checklist
ERC000053	Tree of Life Checklist

DATA TERRA

ODATIS

ENA sample checklists

Accession	Name
ERC000011	ENA default sample checklist

In practice, we often use the default sample checklist because of lack of existing checklists adapted specifically to ifremer projects.

An update is scheduled for March 15, 2024 :

4 new MIxS checklists have been added to ENA:

- GSC MIxS Agriculture
- GSC MIxS Food and Production
- GSC MIxS Symbiont
- GSC MIxS Hydrocarbon

Ontologies

Ontologies are interoperable, logically well-defined, machine readable controlled vocabularies

Ontology is a **structured set of terms and concepts** of a particular domain specifying the **relationships** between these terms and their properties.

Each term of an ontology must have a definition to be sure of the associated meaning.

An ontology presents a **hierarchical structure** and the set of terms is anchored by a high-level term, the root.

Ontologies

Examples :

- The GENEONTOLOGY (GO) aims to maintain and develop controlled vocabulary of **gene and gene product attributes**

- EDAM is an ontology of well established, familiar concepts that are prevalent within bioinformatics, including **types of data and data identifiers, data formats, operations and topics.**

- ENVO is an ontology which represents knowledge about **environments, environmental processes, ecosystems, habitats, and related entities**

- and so on : ontologies OLS ONTOLOGY SEARCH

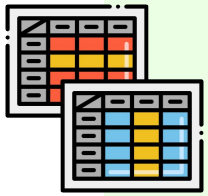
athENA in practice

athENA in a nutshell

nextflow

athENA

Metadata collection



- User friendly **Excel** sheets
- Drop-down lists with controlled vocabulary
- **ENA standards**

Metadata validation

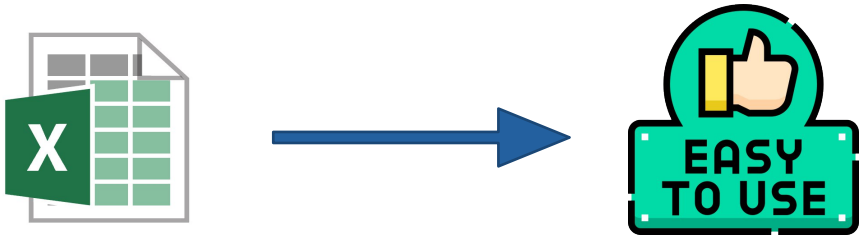
- Python pipeline to convert Excel sheets to XML files
- Metadata validation using controlled vocabulary, checklilsts requirements and format

ENA submission

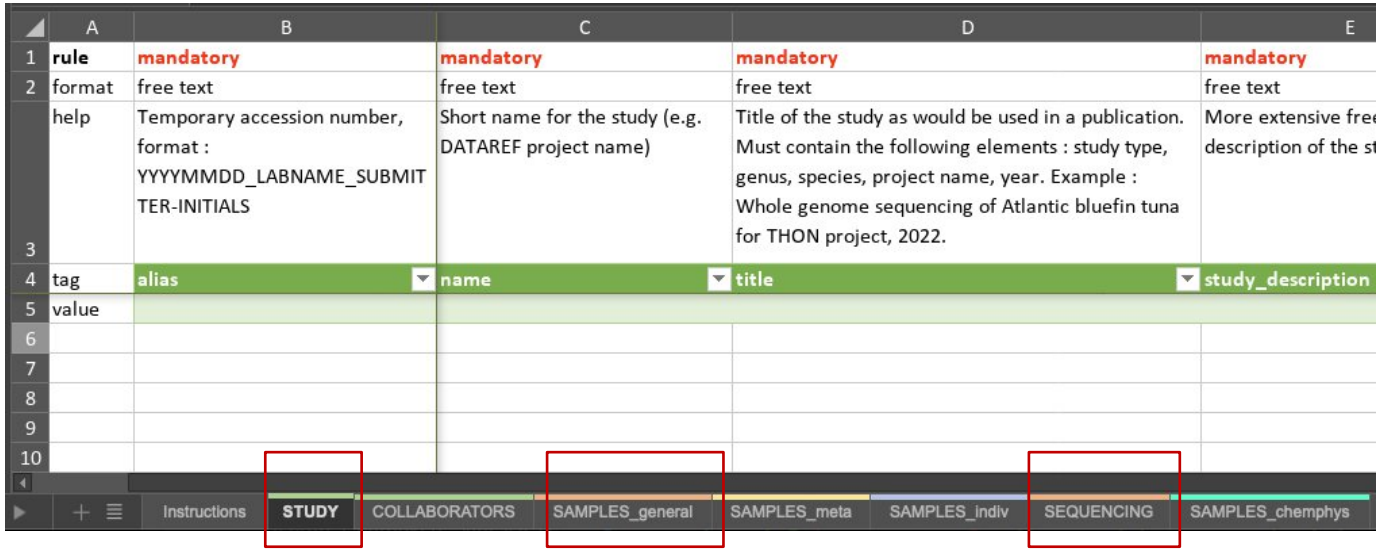
- Data upload to ENA server using cURL
- Embargo max 2 years
- Accession IDS delivered as soon as data are uploaded

Metadata collection via an Excel file

- **content** : 7 editable sheets corresponding to ENA metadata model
- **rules** : mandatory / optional fields according to selected checklists
- **format** : free text / controlled vocabulary / drop-down lists
- **joker values for mandatory fields** : “not collected” ; “not provided”, “not applicable” ; “restricted access”



Metadata collection via an Excel file



	A	B	C	D	E
1	rule	mandatory	mandatory	mandatory	mandatory
2	format	free text	free text	free text	free text
3	help	Temporary accession number, format : YYYYMMDD_LABNAME_SUBMITTER-INITIALS	Short name for the study (e.g. DATAREF project name)	Title of the study as would be used in a publication. Must contain the following elements : study type, genus, species, project name, year. Example : Whole genome sequencing of Atlantic bluefin tuna for THON project, 2022.	More extensive free description of the st
4	tag	alias	name	title	study_description
5	value				
6					
7					
8					
9					
10					

Instructions | STUDY | COLLABORATORS | SAMPLES_general | SAMPLES_meta | SAMPLES_indiv | SEQUENCING | SAMPLES_chemphys

Overall ENA Research Project

STUDY

SAMPLE

Sequenced Biomaterial

RAW Reads

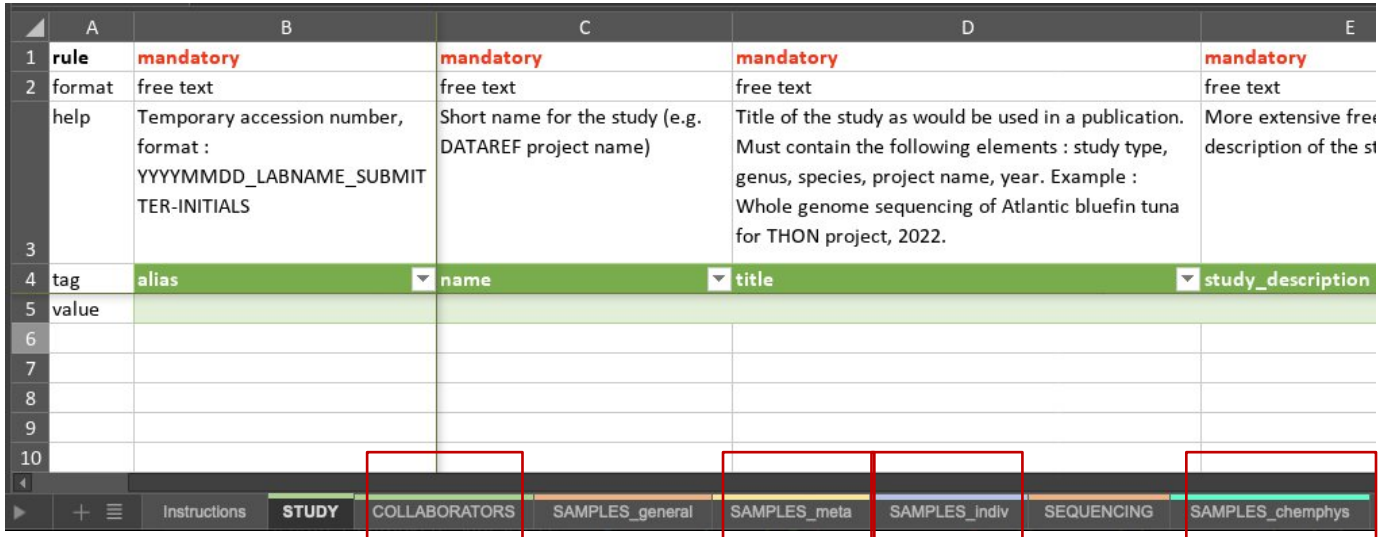
EXPERIMENT **RUN**

Data Analyses

ANALYSIS

mandatory **mandatory** **mandatory** **optional**

Metadata collection via an Excel file



	A	B	C	D	E
1	rule	mandatory	mandatory	mandatory	mandatory
2	format	free text	free text	free text	free text
3	help	Temporary accession number, format : YYYYMMDD_LABNAME_SUBMITTER-INITIALS	Short name for the study (e.g. DATAREF project name)	Title of the study as would be used in a publication. Must contain the following elements : study type, genus, species, project name, year. Example : Whole genome sequencing of Atlantic bluefin tuna for THON project, 2022.	More extensive free description of the st
4	tag	alias	name	title	study_description
5	value				
6					
7					
8					
9					
10					

Instructions | STUDY | COLLABORATORS | SAMPLES_general | SAMPLES_meta | SAMPLES_indiv | SEQUENCING | SAMPLES_chemphys

List here owner of data

STUDY

mandatory

ENA mandatory fields only for metaB projects

SAMPLE

Optional metadata when applied:
- description of individuals
- physico-chemical information

SAMPLE

optional

Metadata collection via an Excel file

scientific_name	common_name	taxon_id	collection_date	isolation_source
seawater metagenome	seawater metagenome	1561972	2020-11-23	control sample
seawater metagenome	seawater metagenome	1561972	2020-11-23	control sample
seawater metagenome	seawater metagenome	1561972	2020-09-03	coastal sea water

sample salinity	sample temperature
39.2	24.7
38.8	16.0
39.9	13.4

country	locality	latitude	longitude
France	Baie des Veys	49.3258	-1.1127
France	Etang de Thau	43.4561	3.6723
France	Etang de Thau	43.4561	3.6723

environment (biom	environment (feature	environment (material	target gene	target subfragmer
ocean	not applicable	coastal sea water	16S rRNA	V4-V5
ocean	not applicable	coastal sea water	16S rRNA	V4-V5
sea	mediterranean sea	sea water	16S rRNA	V4-V5

PLATFORM	FILETYPE	pcr primers	multiplex id	adapters
ILLUMINA	fastq	F:GTGYCAGCMGCCGCGGTAA-R:CCGYCAATTYMTTTRAGTTT	AGTTTG	F:CTTTCCCTACACGACGCTCTTCCGATCT-R:GGAGTTCAG
ILLUMINA	fastq	F:GTGYCAGCMGCCGCGGTAA-R:CCGYCAATTYMTTTRAGTTT	GGACGG	F:CTTTCCCTACACGACGCTCTTCCGATCT-R:GGAGTTCAG
ILLUMINA	fastq	F:GTGYCAGCMGCCGCGGTAA-R:CCGYCAATTYMTTTRAGTTT	TCAGCG	F:CTTTCCCTACACGACGCTCTTCCGATCT-R:GGAGTTCAG

ENA programmatic submission

3 routes for ENA submissions, all appropriate, maybe complementary :

- **Interactive Submissions** : filling out by web forms or off-line downloaded template spreadsheets and uploaded to ENA. Most accessible submission route.

- **Command Line Submissions** : Webin-CLI program. Validates submissions. Allow you maximum control of the process.

- **Programmatic Submissions** : data must be in XML format. Send to ENA by Webin Portal or cURL.

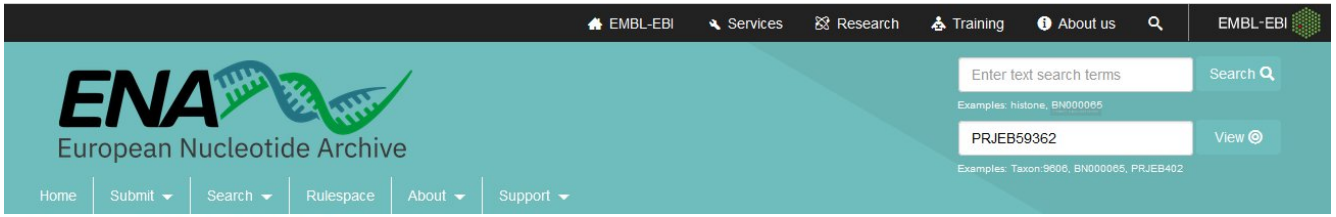
ENA programmatic submission

	Interactive	Webin-CLI	Programmatic
Study	Y	N	Y
Sample	Y	N	Y
Read data	Y	Y	Y
Genome Assembly	N	Y	N
Transcriptome Assembly	N	Y	N
Template Sequence	N	Y	N
Other Analyses	N	N	Y

athENA uses the programmatic route to submit raw reads, and Webin-CLI for genomes.

ENA published data with athENA

STUDY



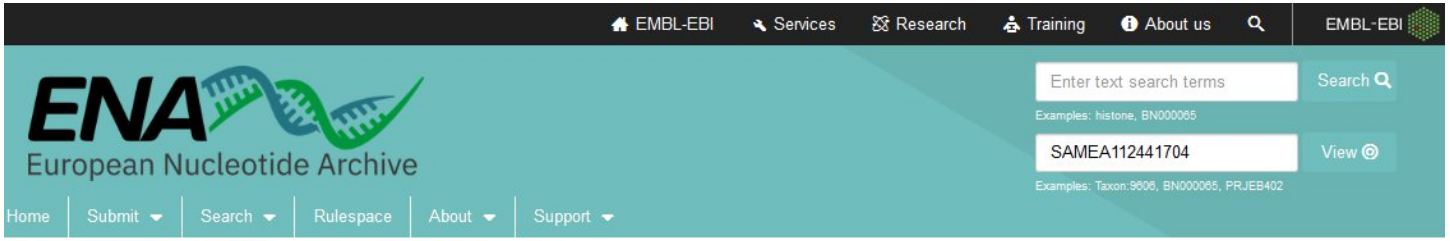
Project: PRJEB59362

Transgenerational exposure to ocean acidification impacts the hepatic transcriptome of European sea bass

Secondary Study Accession:	ERP144411
Study Title:	RNAseq analysis reveals that transgenerational exposure to ocean acidification impacts the hepatic transcriptome of European sea bass (Dicentrarchus labrax) Show Less
Center Name:	Ifremer
Study Name:	LIVACID
Broker Name:	SeBiMER
COLLABORATORS:	Mazurais David,Servili Arianna,Mouchel Olivier,Zambonino Jose
INSTITUTE_NAME:	IFREMER_RBE_PHYTNESS
CENTER_PROJECT_NAME:	LIVACID
STUDY_TYPE:	RNASeq
STUDY_ABSTRACT:	Physiological effects of ocean acidification associated to elevated CO2 concentrations in seawater is the subject of numerous studies in teleost fish. While short time impact of ocean acidification on acid-base exchange and energy metabolism are relatively well described, the longer-term effects are much less known. In this study, we investigated the effect of transgenerational exposure to ocean acidification on the hepatic transcriptome of European sea bass (Dicentrarchus labrax). Show Less
ENA-FIRST-PUBLIC:	2023-02-09
ENA-LAST-UPDATE:	2023-02-09

ENA published data with athENA

SAMPLE

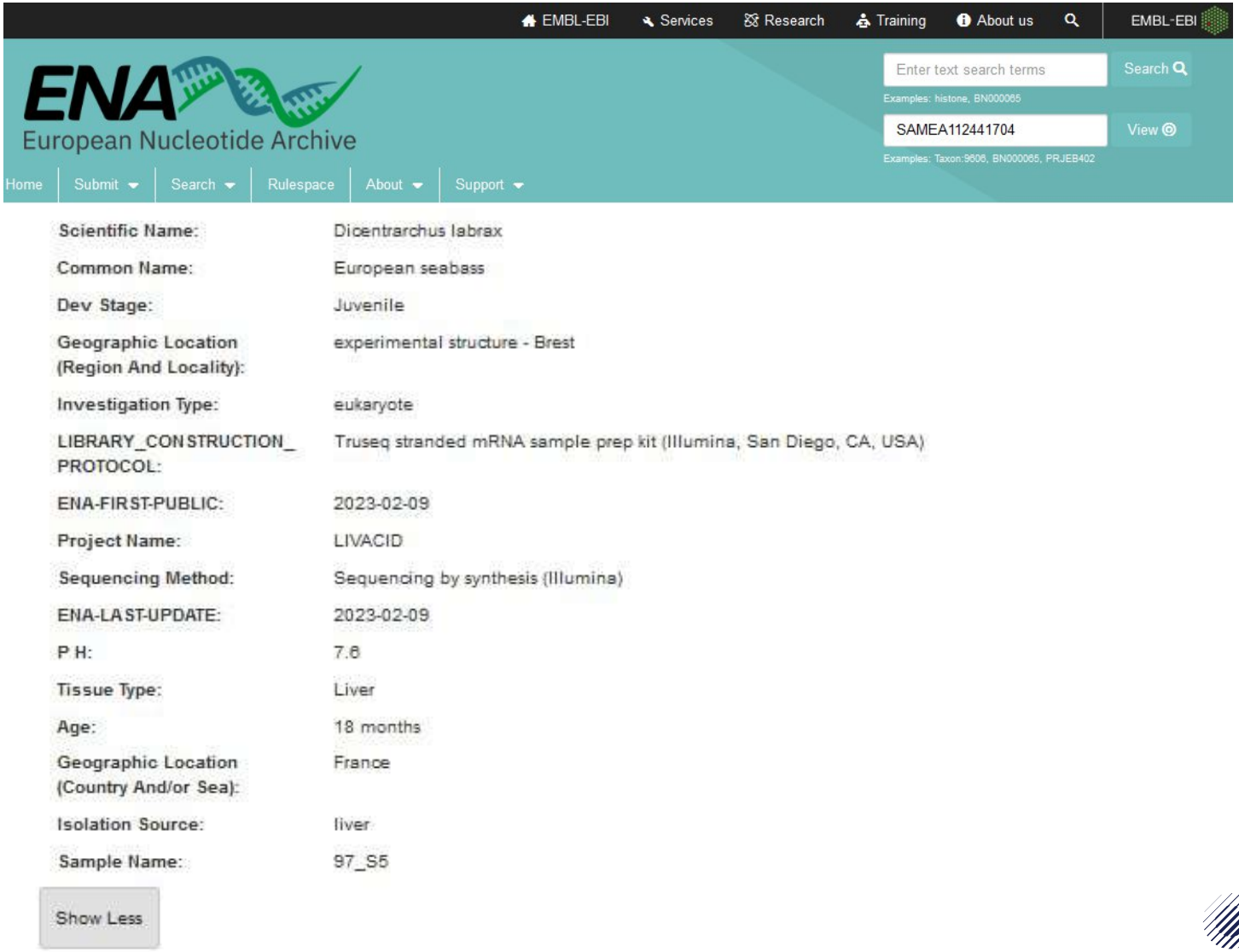


Biosample: SAMEA112441704

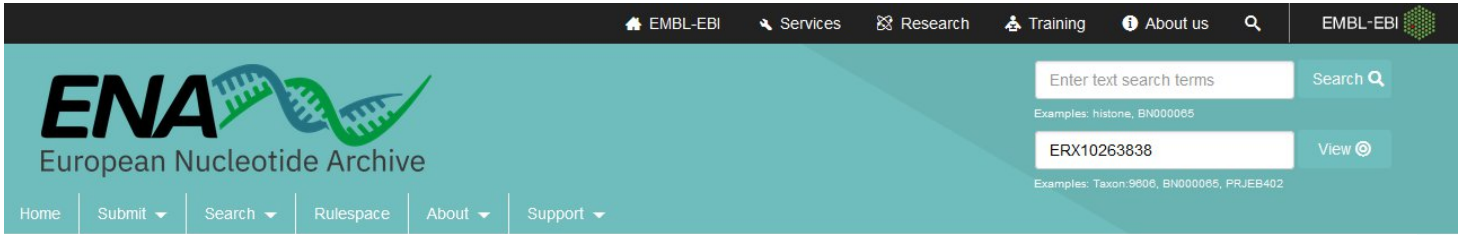
97_Liv

Organism:	Dicentrarchus labrax (European seabass);Dicentrarchus labrax
Sample Accession:	SAMEA112441704
Sample Title:	97_Liv
Center Name:	Ifremer
Sample Alias:	sam_97_S5
Checklist:	ERC000011
Broker Name:	Bioinformatics Core Facility of Ifremer (French Research Institute for Exploitation of the Sea)
ENA-CHECKLIST:	ERC000011
Collection Date:	2019-10-15
Sample Description:	Acid condition sampling day 1
Environmental Sample:	No
Aquaculture Origin:	AOAR (Aquaculture Origin Aquaculture Raised)
Geographic Location (Longitude):	not provided
Geographic Location (Latitude):	not provided

ENA published data with athENA



ENA published data with athENA



EXPERIMENT

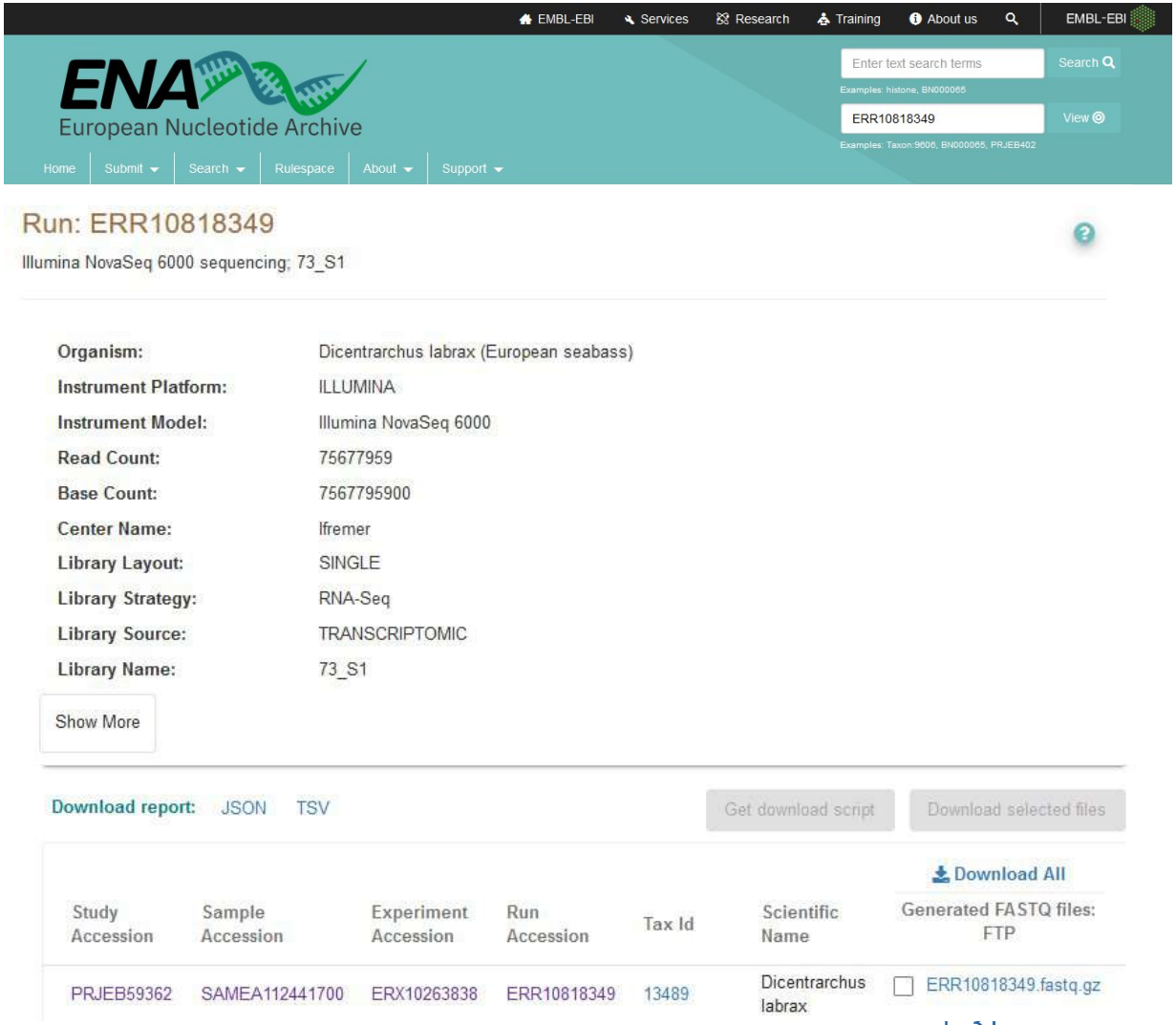
Experiment: ERX10263838

Illumina NovaSeq 6000 sequencing; 73_S1

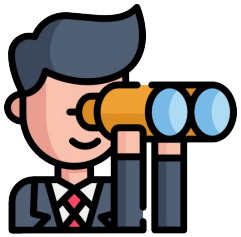
Organism:	Dicentrarchus labrax (European seabass)
Experiment Accession:	ERX10263838
Instrument Platform:	ILLUMINA
Instrument Model:	Illumina NovaSeq 6000
Center Name:	Ifremer
Library Layout:	SINGLE
Library Strategy:	RNA-Seq
Library Source:	TRANSCRIPTOMIC
Library Name:	73_S1
Library Selection:	cDNA_oligo_dT
Broker Name:	SeBiMER
LIBRARY_CONSTRUCTION_PROTOCOL:	Truseq stranded mRNA sample prep kit (Illumina, San Diego, CA, USA)

ENA published data with athENA

RUN



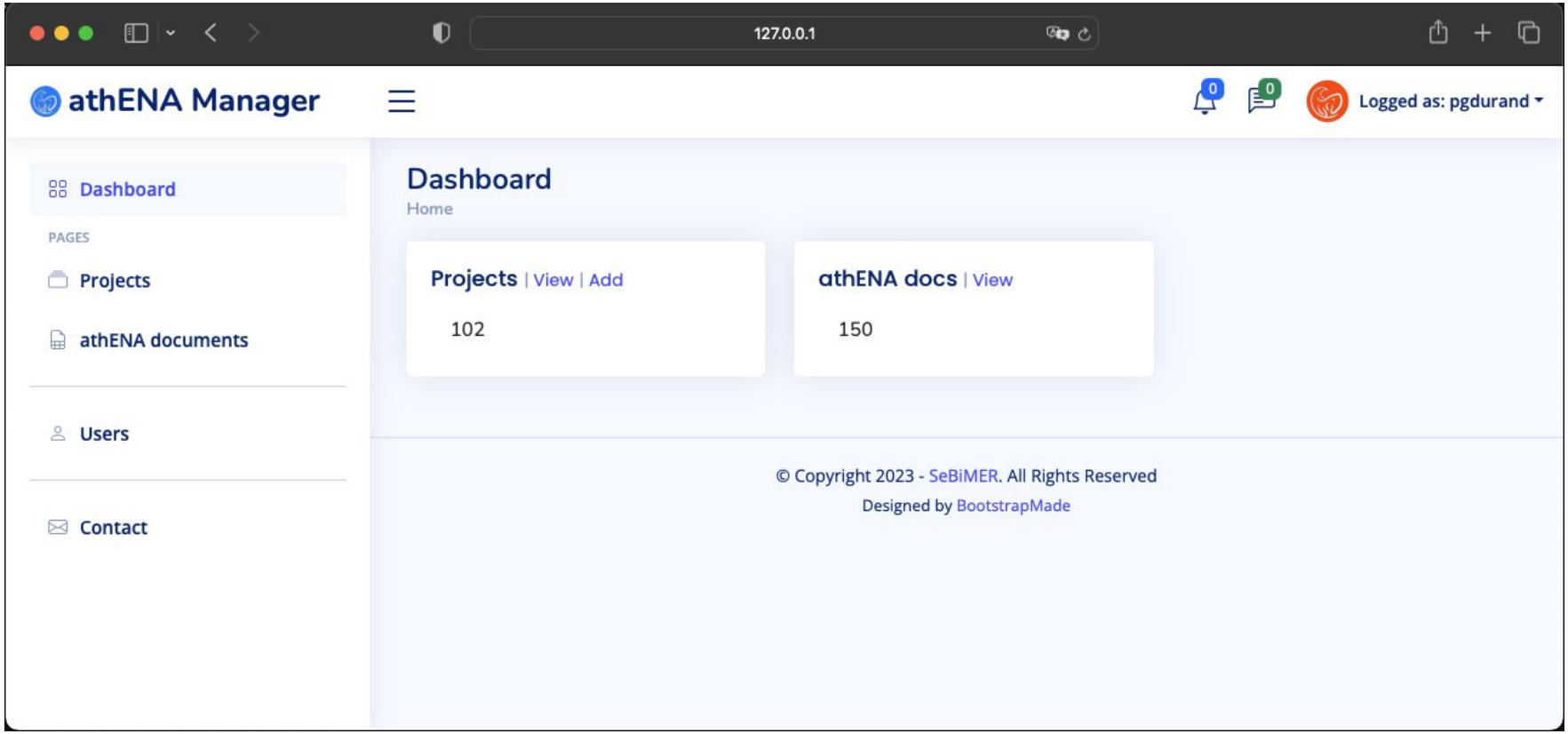
athENA : what's next ?



New features (work in progress)

- Programmatic generation of Excel templates
- Template versionning
- Integration of genome submission to main pipeline
- Specific templates (Single-cell)
- Genome annotations

athENA manager (Patrick Durand)



athENA manager (Patrick Durand)

Credits

SeBiMER
Bioinformatique marine

SISMER team

Elisabeth Hellec
M2 student + CDD
SeBiMER

Alizée bardon
M2 student + CDD
SeBiMER

Julie Clément
IHPE Perpignan

Bernard de Massy
Institute of Human Genetics,
Montpellier

Frédéric Bigey
INRAe
Montpellier

All icons in the previous slides made by Freepik from www.flaticon.com.

Thank you !

Pauline Auffret pauline.auffret@ifremer.fr

https://orcid.org/0009-0001-3834-7670

https://gitlab.ifremer.fr/bioinfo/workflows/athena

To sum-up