SLOSEQ - Consolidation and Integration of Whole Genome Sequencing (WGS) into Routine Surveillance in Slovenia

Project progress report for Slovenia

European Commission

SYMPOSIUM

on Strengthening Genomic Sequencing Capacities
for Outbreak Preparedness and Response

4-5 June 2025

La Tricoterie
Rue Théodore Verhaegen 158,
1060 Bruxelles

#HealthUnion

EU in Emergencies @EC_HERA

Co-funded by
the European Union

NIJZ Nacionalni inštitut za javno zdravje

SLOSEQ project and project consortium

- Coordinatior: NATIONAL LABORATORY OF HEALTH, ENVIRONMENT AND FOOD, MARIBOR, SLOVENIA
  - Affiliated entities:
    - NATIONAL INSTITUTE OF PUBLIC HEALTH, Ljubljana, Slovenia
    - INSTITUTE OF MICROBIOLOGY AND IMMUNOLOGY, FACULTY OF MEDICINE, UNIVERSITY OF LJUBLJANA, Ljubljana, Slovenia
- Project budget 1.345.000,00 EUR, max grant 1.075.999,97 EUR
- Duration: 1.7.2023 – 30.6.2025

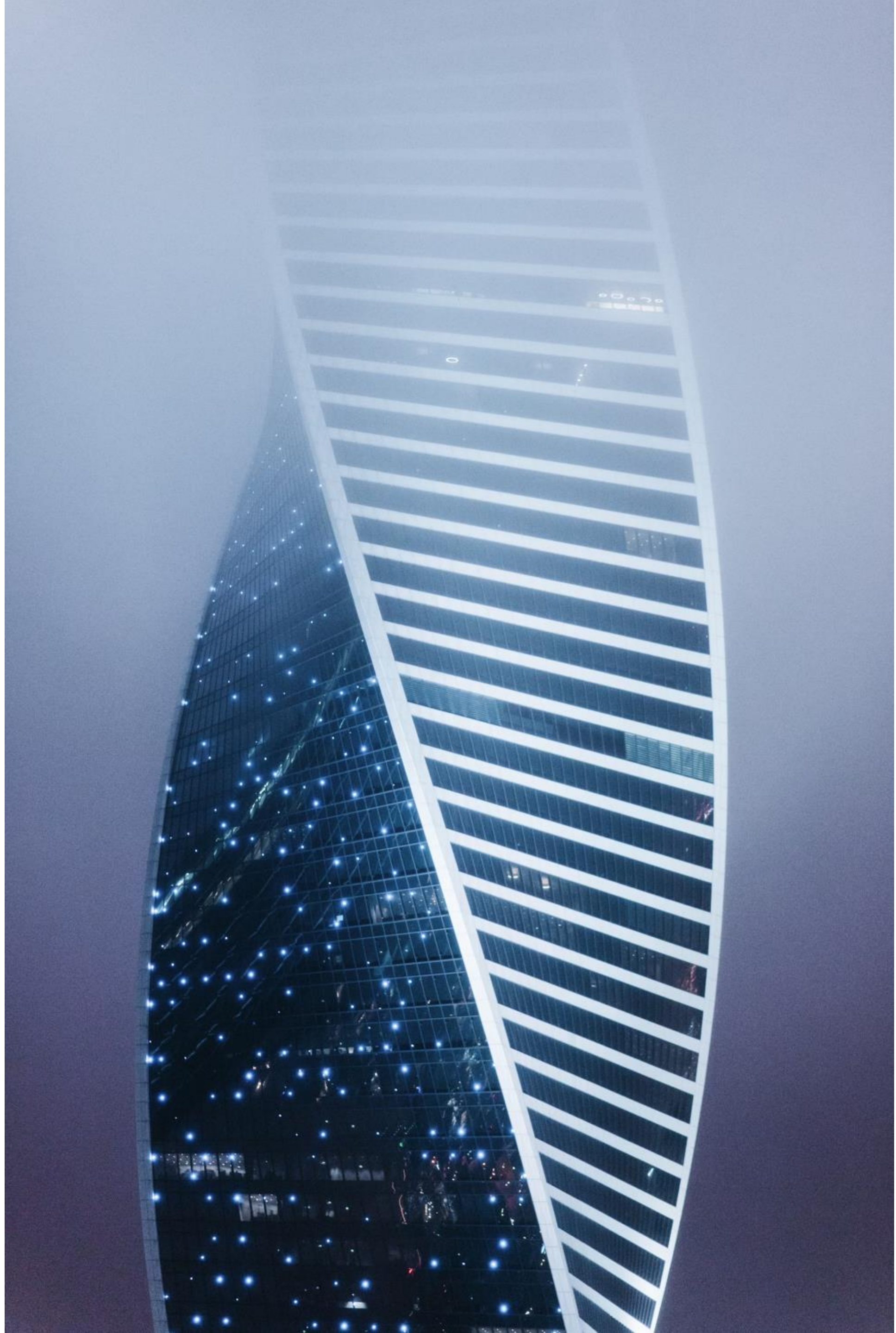
- Project WPs:
  - WP1: Coordination and project management
  - WP2: High-throughput detection of SARS-CoV-2, monitoring and detection of known and emerging new SARS-CoV-2 variants
  - WP3: Consolidation of WGS workflows and integration genomic surveillance for selected pathogens
  - WP4: Developing a data processing software
  - WP5: Identifying infrastructure for the national coordinating centre for AMR and preparation of protocols for its operation
  - WP6: Upgrade of the digital solutions for SARS-CoV-2 genomic surveillance and response
  - WP7: Communication and dissemination of project results
  - WP8: Project monitoring and evaluation
  - WP9: Sustainability of the project

Project’s main objectives

ENHANCED GENOMIC SURVEILLANCE
- for extended list of known pathogens
- other public health threats (AMR)
- possible new epidemic/pandemic pathogens

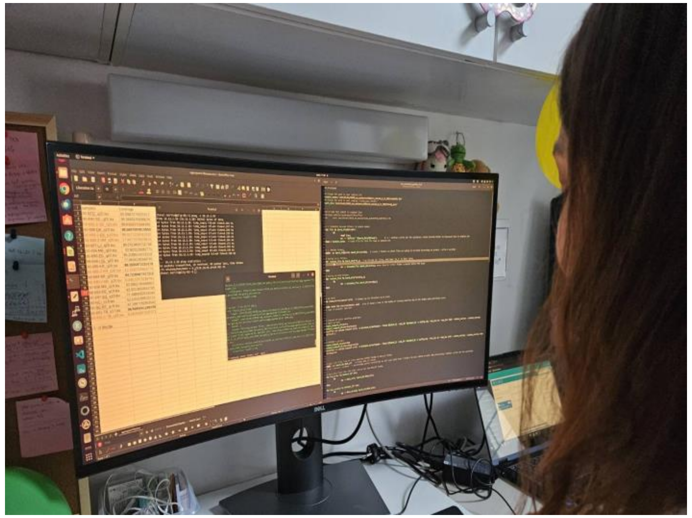
CONSOLIDATION OF METOHDS/PROTOCOLS
- At national level
- Synchronization with international guidelines
- Automatization of lab. procedures/bioinformatic tools
- Enhanced reporting (digitalization)

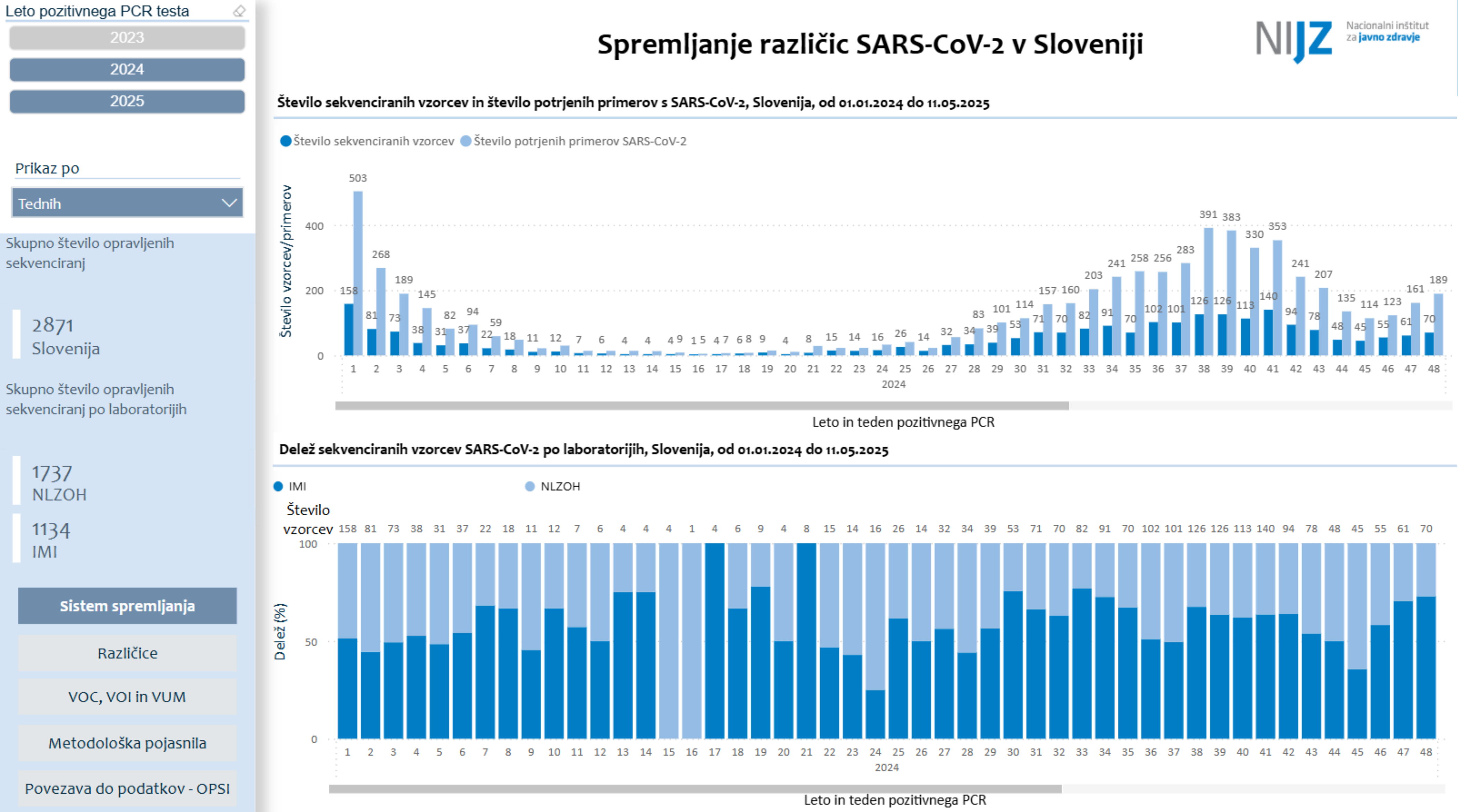
TO FOSTER AMR NATIONAL ACTION PLAN
- One-health approach – intersectoral collaboration
- Adjustment of national legislation
- Possibilities to include environmental surveillance



WP2 High-throughput detection of SARS-CoV-2, monitoring and detection of known and emerging new SARS-CoV-2 variants

- Continuation of SARS CoV-2 variant screening (over 6000 strains sequenced in the period 07/2023-06/2025
- Integration of automatization in lab procedures enabled:
  - ➢ enhanced sequencing process
  - ➢ increased throughput,
  - ➢ enhanced accuracy,
  - ➢ improved consistency,
  - ➢ time efficiency
- Consolidation of protocols among institutions (ring trial) for Illumina and ONT platforms





Leto pozitivnega PCR testa

2023

2024

2025

Prikaz po

Tednih

Skupno število opravljenih sekvenciranj

2871
Slovenija

Skupno število opravljenih sekvenciranj po laboratorijih

1737
NLZOH

1134
IMI

Sistem spremljanja

Različice

VOC, VOI in VUM

Metodološka pojasnila

Povezava do podatkov - OPSI

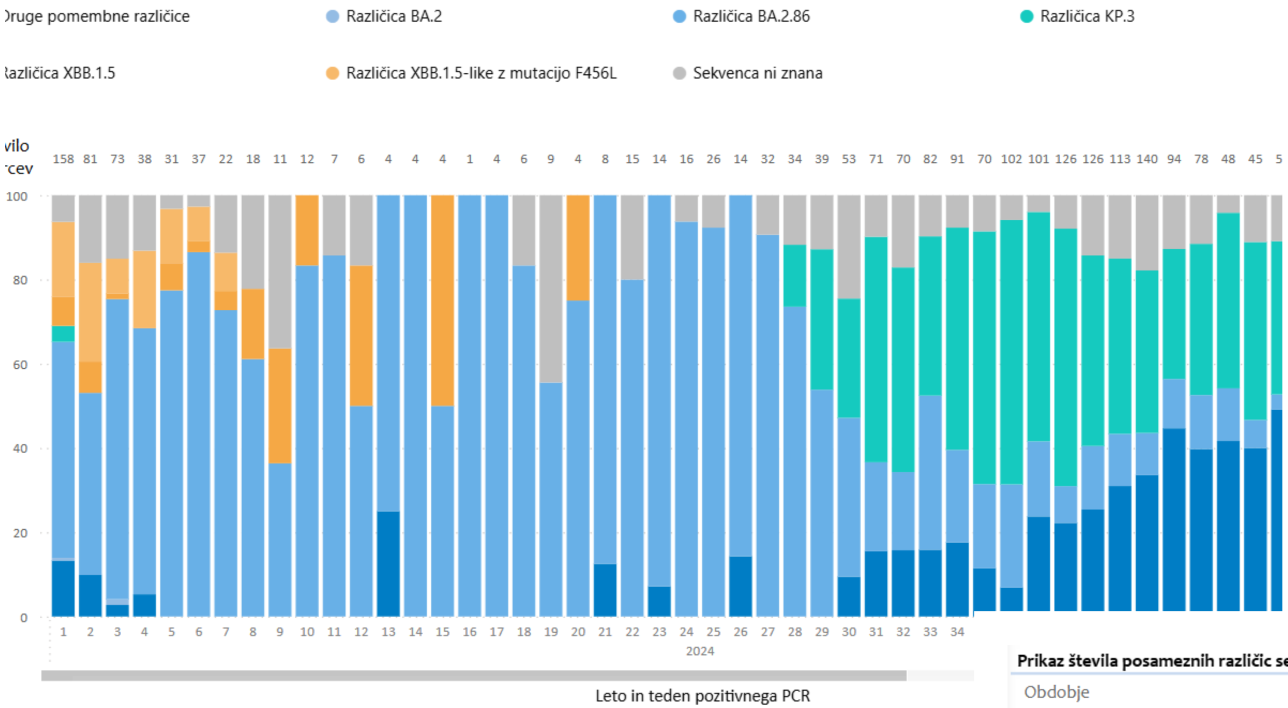
Spremljanje različic SARS-CoV-2 v Sloveniji

NIJZ Nacionalni inštitut za javno zdravje

Število sekvenciranih vzorcev in število potrjenih primerov s SARS-CoV-2, Slovenija, od 01.01.2024 do 11.05.2025

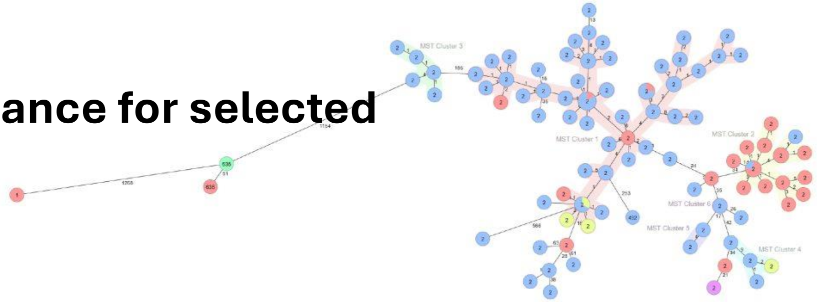
Delež sekvenciranih vzorcev SARS-CoV-2 po laboratorijih, Slovenija, od 01.01.2024 do 11.05.2025

ež posameznih različic sekvenciranih vzorcev SARS-CoV-2, Slovenija, od 01.01.2024 do 11.05.2025



Prikaz števila posameznih različic sekvenciranih vzorcev SARS-CoV-2, Slovenija, od 01.01.2024 do 11.05.2025

Obdobje																							2024		
Različica	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25
XFC																									
XFG																									
Različica BA.2	**1**		**1**																						
BA.2.1	1		1																						
Različica BA.2.86	**81**	**35**	**52**	**24**	**24**	**32**	**16**	**11**	**4**	**10**	**6**	**3**	**3**	**4**	**2**	**1**	**4**	**5**	**5**	**3**	**7**	**12**	**13**	**15**	**24**
BA.2			1																						
BA.2.86					1																				
BA.2.86.1	18	3	4	2	3	4	1																		
JN.1	45	19	28	11	10	17	5	6	3	10	4	1	2	2		1		2							1
JN.1.1	3	2		1	3			2														1			
JN.1.10						1																			
JN.1.11																									
JN.1.11.1											1														
JN.1.16														1			3	2		1	1	1	1	4	3
JN.1.16.1															1			1	1			2			1
JN.1.18													1									1			1
JN.1.18.2															1										
JN.1.18.3																					1				
JN.1.18.5																									
JN.1.18.6																									
JN.1.22																									
JN.1.4	11	7	17	9	7	4	6	3	1			1									2				
JN.1.4.7														1											
JN.1.48.1																									1
Skupaj	**158**	**81**	**73**	**38**	**31**	**37**	**22**	**18**	**11**	**12**	**7**	**6**	**4**	**4**	**4**	**1**	**4**	**6**	**9**	**4**	**8**	**15**	**14**	**16**	**26**



WP3: Consolidation of WGS workflows and integration genomic surveillance for selected pathogens

- Centralized WGS workflows at NLZOH sequencing sites
- Implementation of WGS surveillance for selected bacterial pathogens of public health concern:
  - CRE isolates (centralized strain collection and sequencing database)
  - Foodborne and zoonotic bacterial pathogens (*E. coli*, *L. monocytogenes*, *Salmonella sp*., *Campylobacter spp*.)
  - Bacteria in VPI (*B. pertusis*, *S. pneumonae*, *N. meningitidis)*
  - Respiratory viruses (Influenza, RSV – in process for integrated surveillance with SARS CoV-2)
- Consolidation of protocols among institutions (ring trial)

2388 bacterial isolates were sequenced

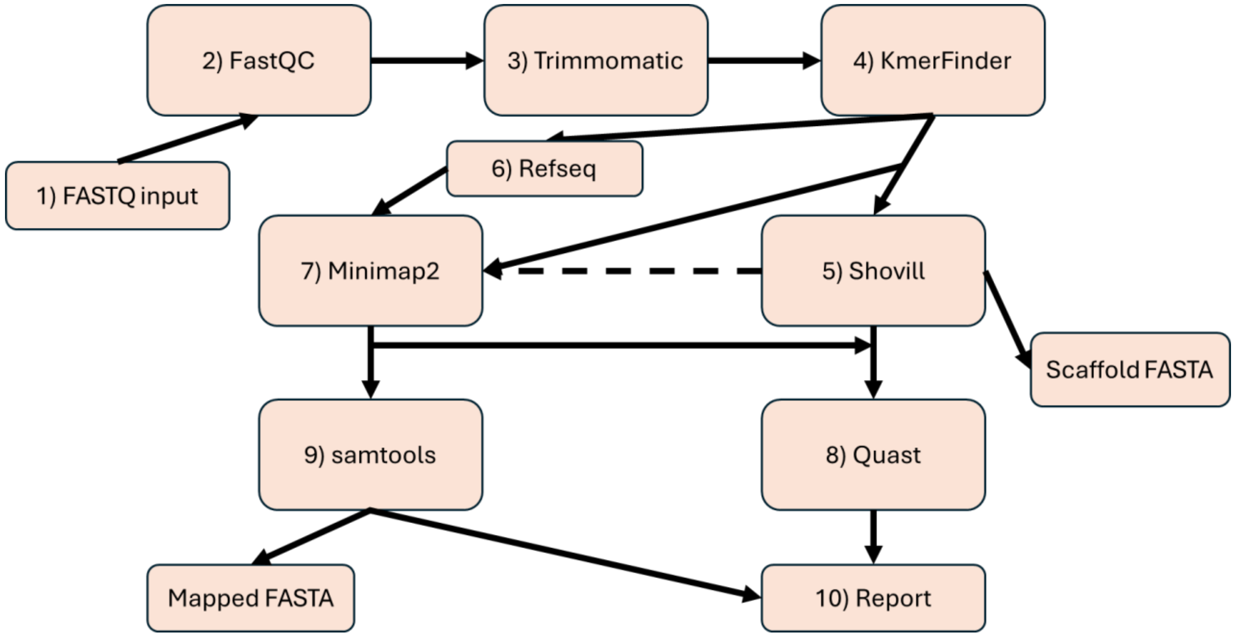
Shared in NCBI SRA, ENA; EpiPulse, Bordetella database, PubMLST

WP4: **Developing a data processing software**

Genome assembly harmonization pipeline

- The main idea of the pipeline is to allow users to *de novo* assemble bacterial genomes with a standardised pipeline that allows users to produce genome assemblies with minimal user input and knowledge of advanced bioinformatics.
- Harmonization of assemblies in mind, to allow for the same starting point for further analysis
- Illumina and Nanopore data
- The produced pipeline offers the possibility to achieve genome assemblies that are further used by other tools for public health applications, such as cgMLST analysis
- Follow us on https://www.nlzoh.si/ and social media for the final version and URL to access the tool. Additionally, a Singularity package will be deposited on Github for local usage.

ILLUMINA



2) FastQC

3) Trimmomatic

4) KmerFinder

1) FASTQ input

6) Refseq

7) Minimap2

5) Shovill

Scaffold FASTA

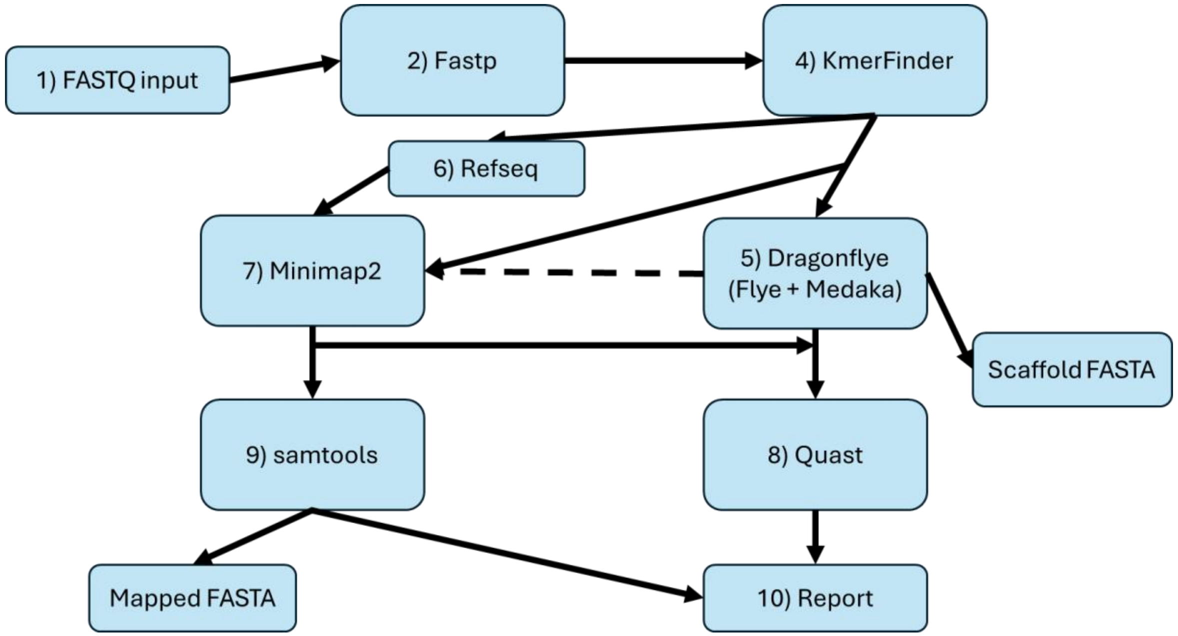
9) samtools

8) Quast

Mapped FASTA

10) Report

NANOPORE



1) FASTQ input

2) Fastp

4) KmerFinder

6) Refseq

7) Minimap2

5) Dragonflye
(Flye + Medaka)

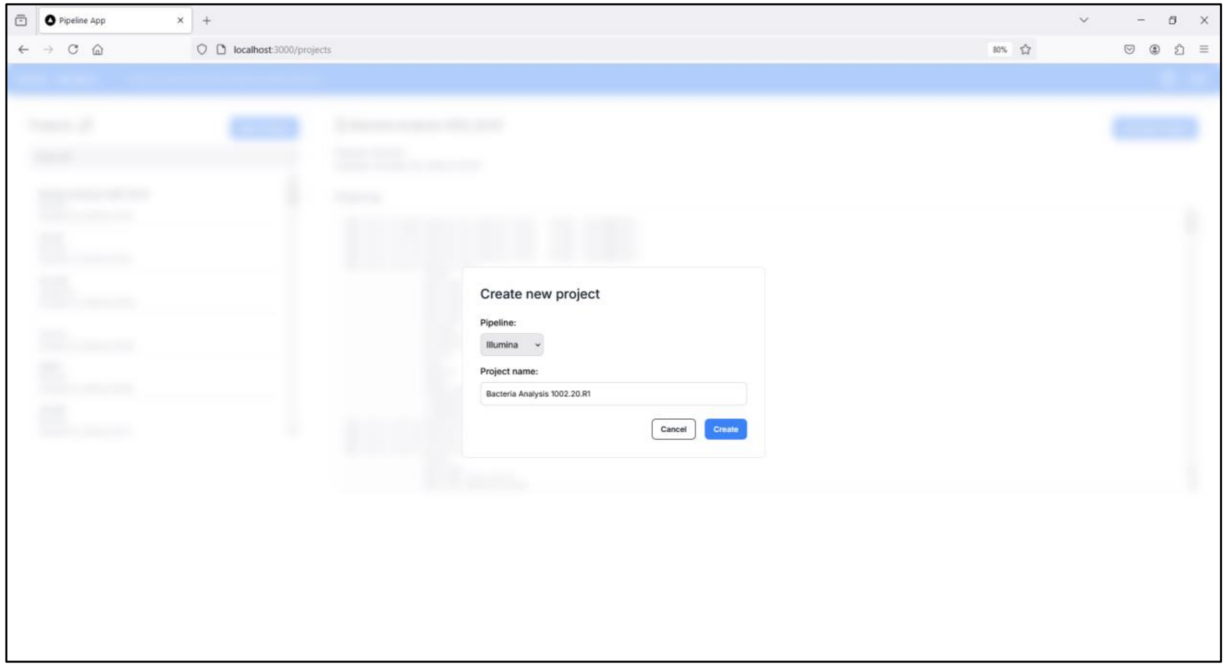
Scaffold FASTA

9) samtools

8) Quast

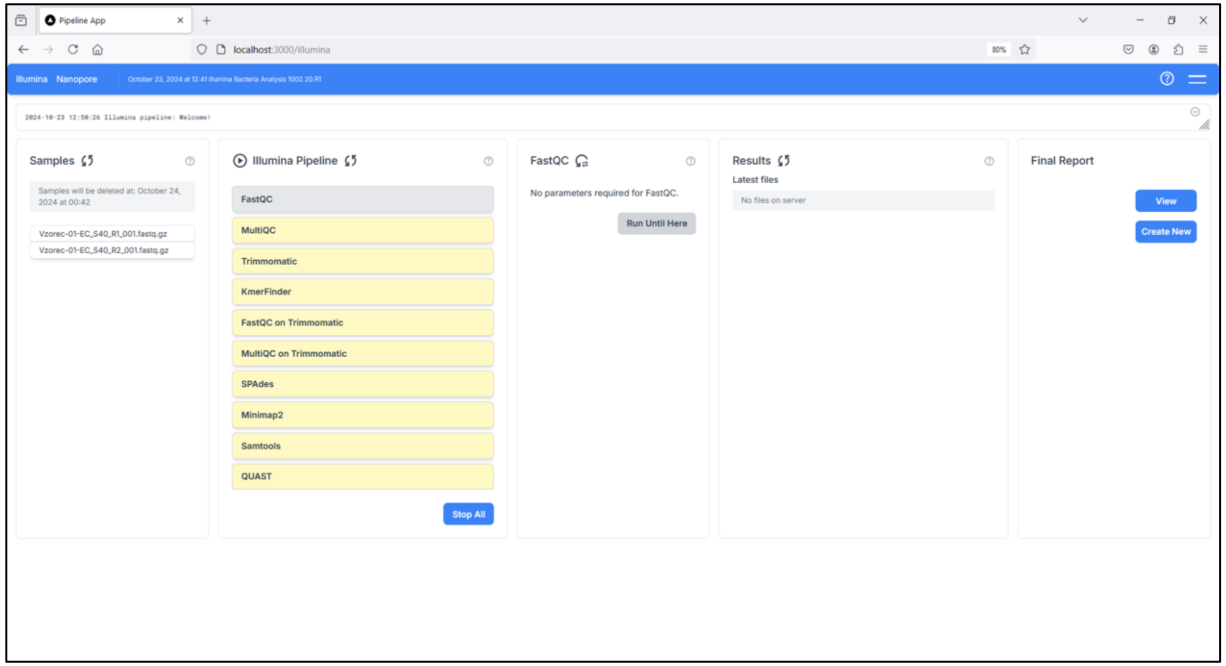
Mapped FASTA

10) Report



Pipeline: Illumina

Project name: Bacteria Analysis 1002.20.R1



October 23, 2024 at 12:41 Illumina Bacteria Analysis 1002.20.R1

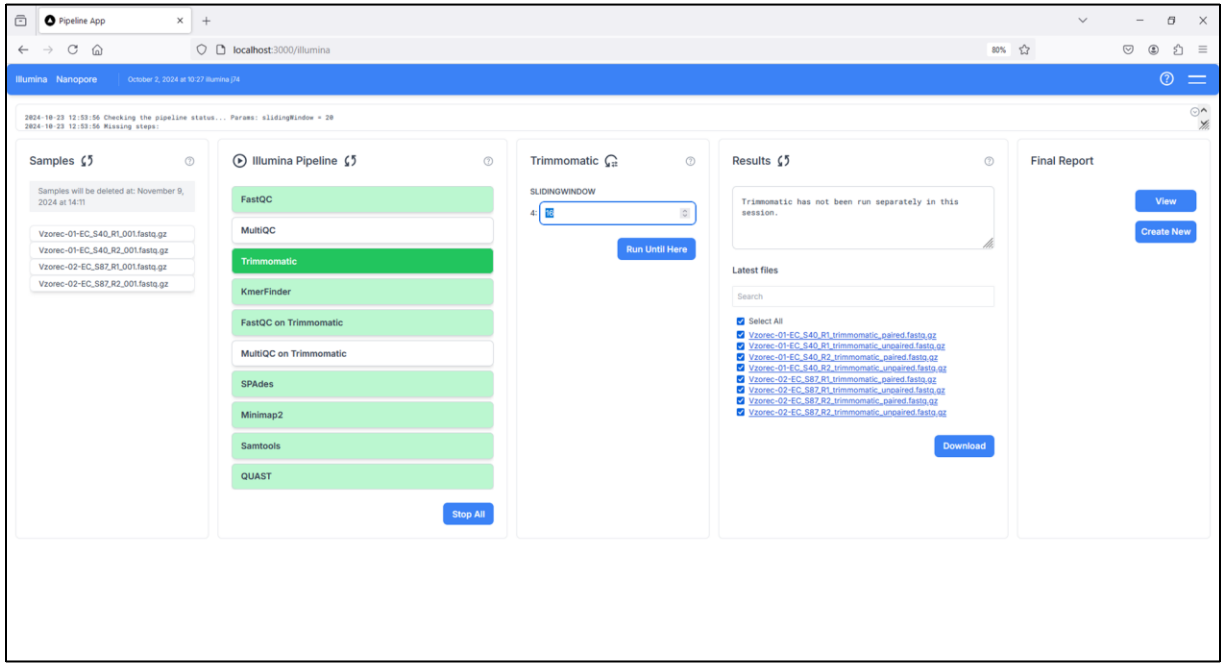
```
2024-10-23 12:50:26 Illumina pipeline: Welcome!
```

Samples will be deleted at: October 24, 2024 at 00:42

Vzorec-01-EC_S40_R1_001.fastq.gz
Vzorec-01-EC_S40_R2_001.fastq.gz

No parameters required for FastQC.

No files on server



October 2, 2024 at 10:27 Illumina j74

```
2024-10-23 12:53:56 Checking the pipeline status... Params: slidingWindow = 20
2024-10-23 12:53:56 Missing steps:
```

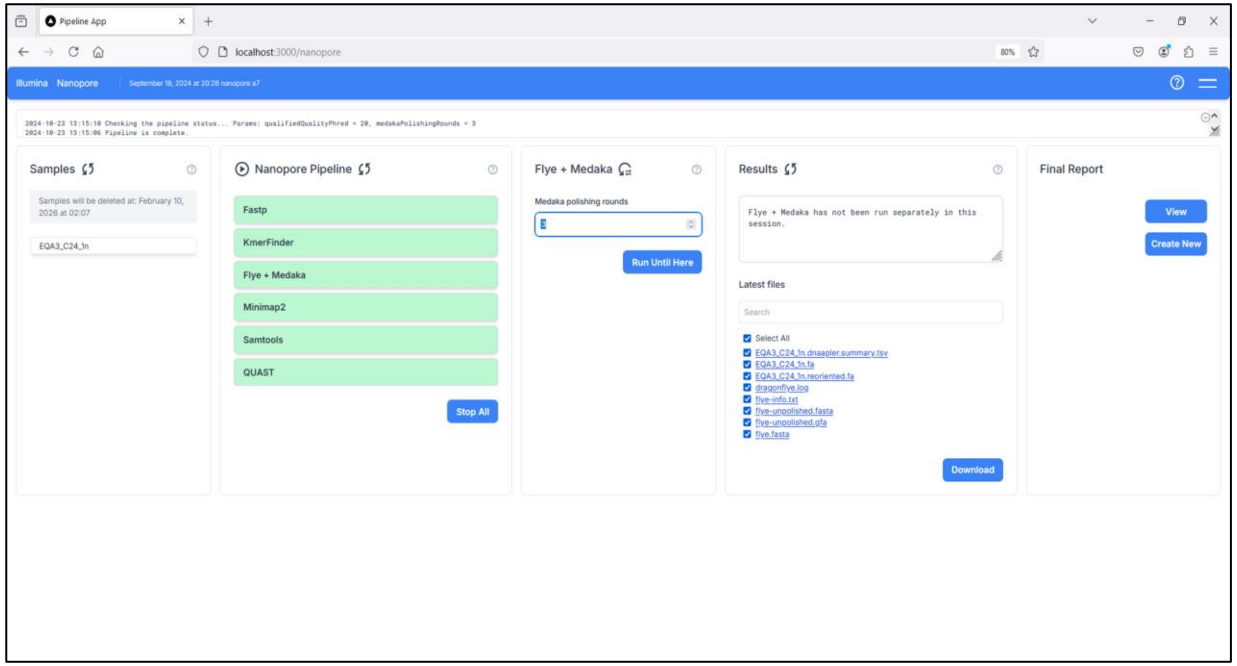
Samples will be deleted at: November 9, 2024 at 14:11

Vzorec-01-EC_S40_R1_001.fastq.gz
Vzorec-01-EC_S40_R2_001.fastq.gz
Vzorec-02-EC_S87_R1_001.fastq.gz
Vzorec-02-EC_S87_R2_001.fastq.gz

SLIDINGWINDOW
4: 16

```
Trimmomatic has not been run separately in this
session.
```

☑ Select All
☑ Vzorec-01-EC_S40_R1_trimmomatic_paired.fastq.gz
☑ Vzorec-01-EC_S40_R1_trimmomatic_unpaired.fastq.gz
☑ Vzorec-01-EC_S40_R2_trimmomatic_paired.fastq.gz
☑ Vzorec-01-EC_S40_R2_trimmomatic_unpaired.fastq.gz
☑ Vzorec-02-EC_S87_R1_trimmomatic_paired.fastq.gz
☑ Vzorec-02-EC_S87_R1_trimmomatic_unpaired.fastq.gz
☑ Vzorec-02-EC_S87_R2_trimmomatic_paired.fastq.gz
☑ Vzorec-02-EC_S87_R2_trimmomatic_unpaired.fastq.gz



September 18, 2024 at 20:26 nanopore a7

```
2024-10-23 13:15:10 Checking the pipeline status... Params: qualifiedQualityPhred = 20, medakaPolishingRounds = 3
2024-10-23 13:15:06 Pipeline is complete.
```

Samples will be deleted at: February 10, 2026 at 02:07

EQA3_C24_1n

Medaka polishing rounds: 3

```
Flye + Medaka has not been run separately in this
session.
```

☑ Select All
☑ EQA3_C24_1n.dnaapler.summary.tsv
☑ EQA3_C24_1n.fa
☑ EQA3_C24_1n.reoriented.fa
☑ dragonflye.log
☑ flye-info.txt
☑ flye-unpolished.fasta
☑ flye-unpolished.gfa
☑ flye.fasta

WP5 Identifying infrastructure for the national coordinating centre for AMR and preparation of protocols for its operation

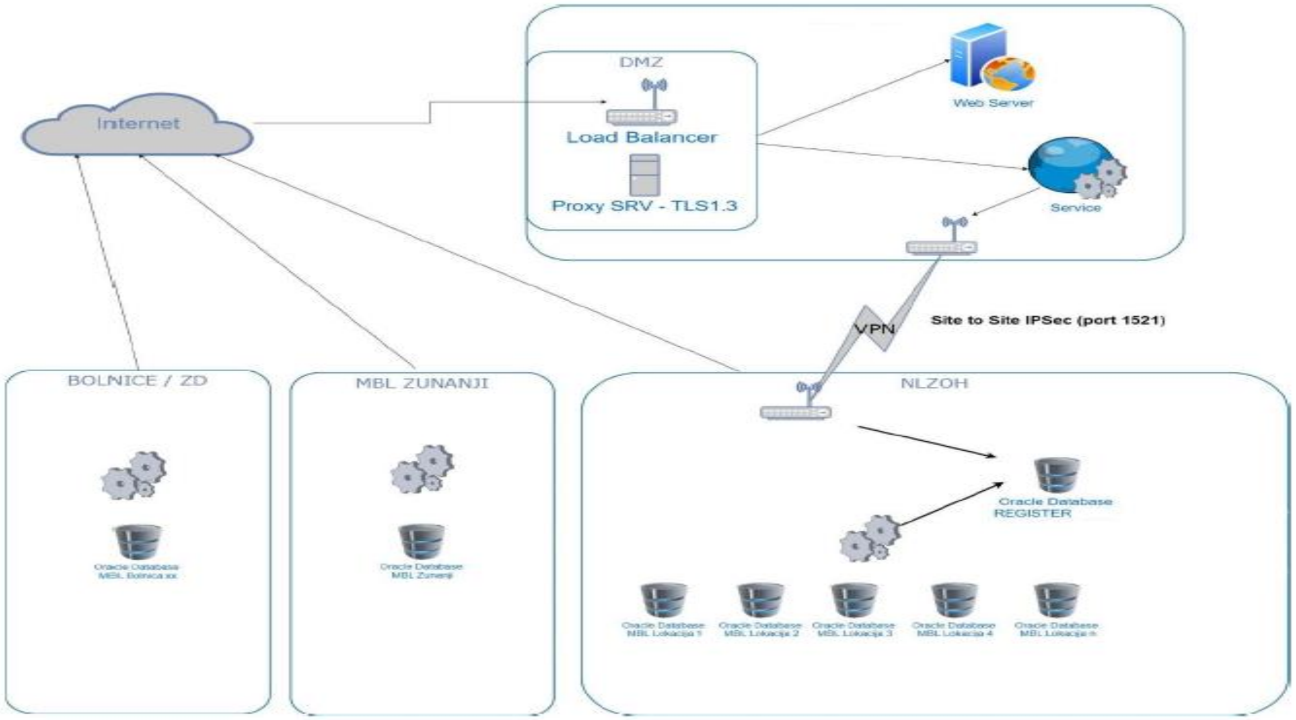
Action plan to establish the National Coordination Centre (NCC) for AMR

- Financial and operational plan for national AMR database
- SOP for NCC-AMR

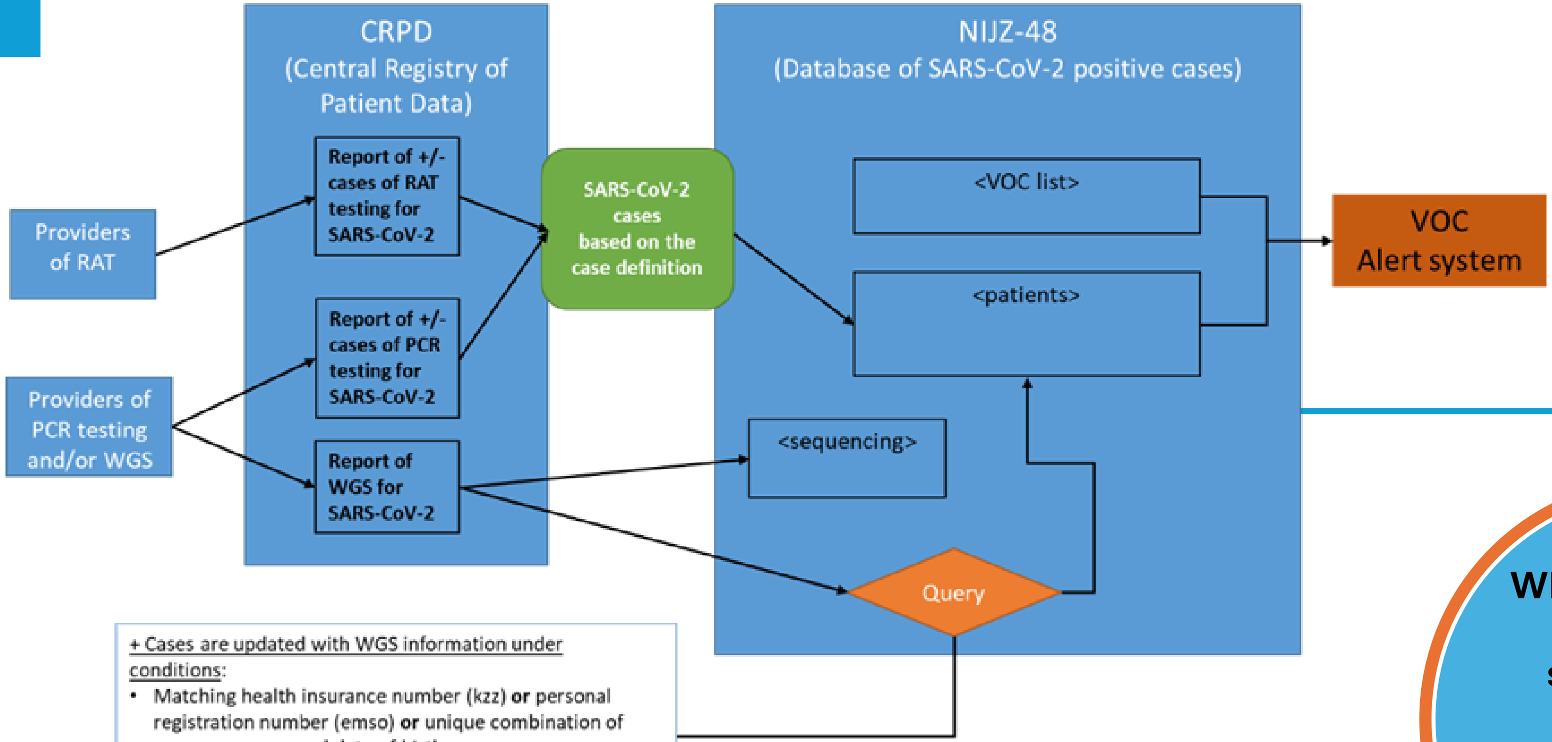
AMR

- Approved by the Ministry of Health
- Consensus within all diagnostic microbiology laboratories to connect and share the data
- Inclusion of all relevant professional comissions and boards in the field of AMR challenge

National AMR database centre



Dashboard
- Data depth according to the user level (public, professionals…)
- National/local (institutional)
- Patient tracking, trends of AMR strains and resistance mechanisms, real-time data for efficient action and spread prevention



CRPD
(Central Registry of Patient Data)

Providers of RAT

Providers of PCR testing and/or WGS

Report of +/- cases of RAT testing for SARS-CoV-2

Report of +/- cases of PCR testing for SARS-CoV-2

Report of WGS for SARS-CoV-2

SARS-CoV-2 cases based on the case definition

NIJZ-48
(Database of SARS-CoV-2 positive cases)

<VOC list>

<patients>

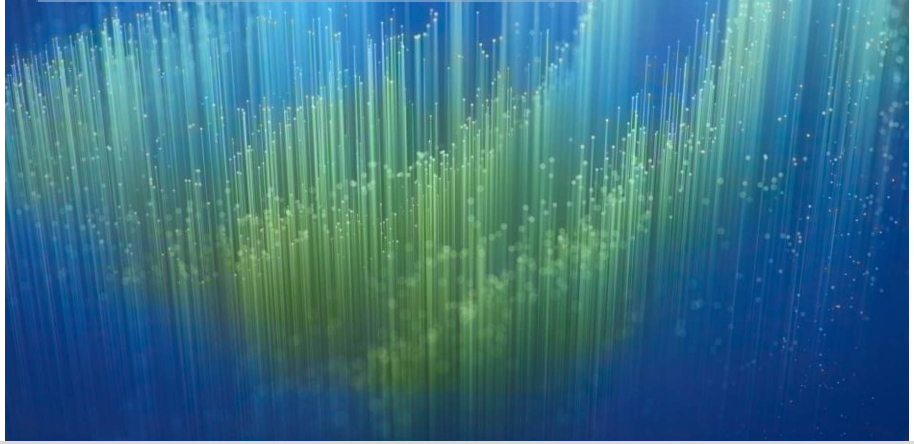
<sequencing>

Query

VOC
Alert system

+ Cases are updated with WGS information under conditions:

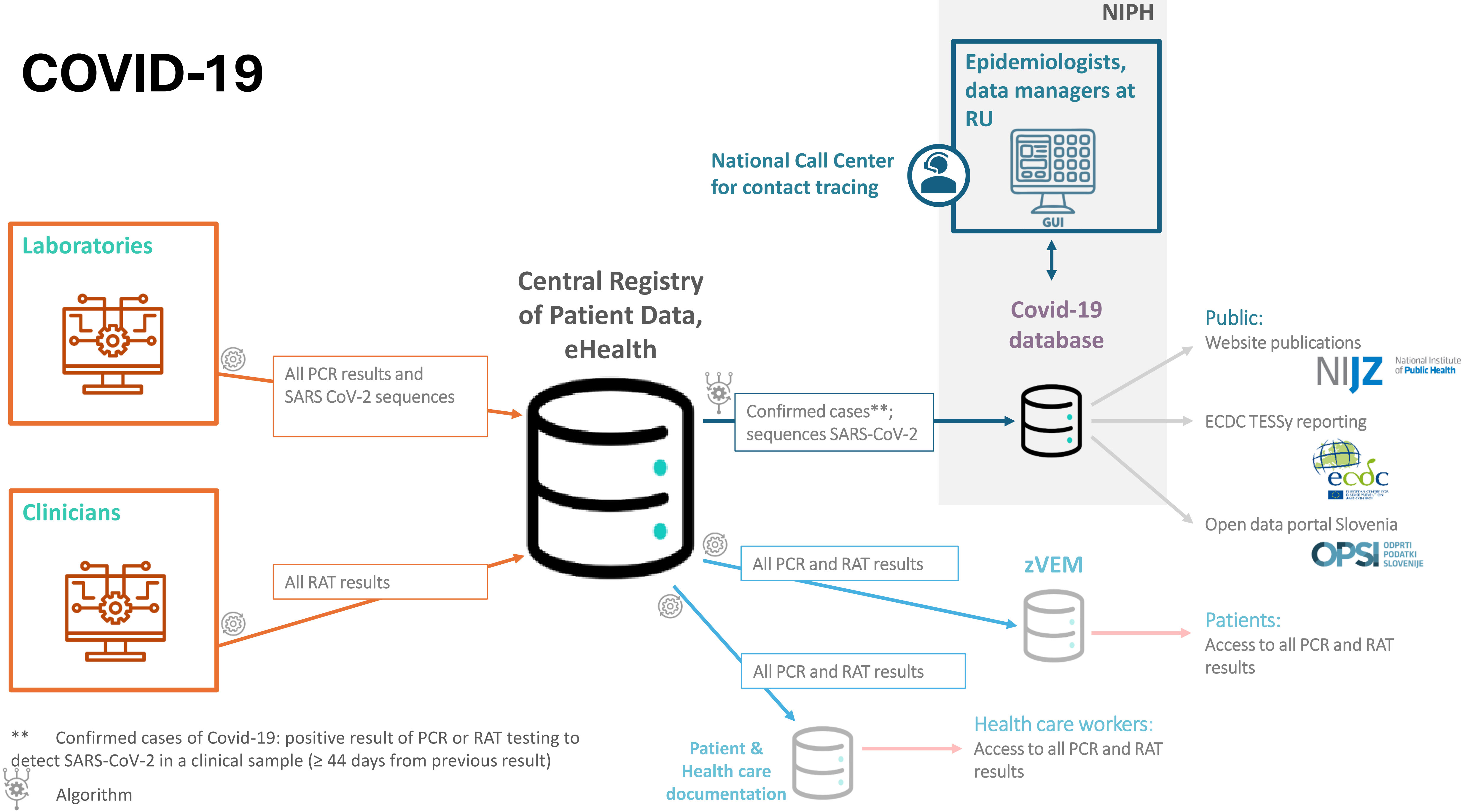
- Matching health insurance number (kzz) **or** personal registration number (emso) **or** unique combination of name, surname and date of birth.
- Matching date of PCR result **or** date of PCR result after RAT.
- Matching laboratory that has implemented PCR analysis.



WP6: Upgrade of the digital solutions for SARS-CoV-2 genomic surveillance and response

- Digital alert system for SARS-CoV-2 VOC reporting
- Lab reporting – NIPH database – alert system to regional epi teams
- As a pilot system for upgrade to other epi emergent reporting systems

COVID-19



Laboratories

Clinicians

All PCR results and SARS CoV-2 sequences

All RAT results

Central Registry of Patient Data, eHealth

Confirmed cases**; sequences SARS-CoV-2

All PCR and RAT results

All PCR and RAT results

NIPH

Epidemiologists, data managers at RU

GUI

National Call Center for contact tracing

Covid-19 database

Public:
Website publications

ECDC TESSy reporting

Open data portal Slovenia

zVEM

Patients:
Access to all PCR and RAT results

Patient & Health care documentation

Health care workers:
Access to all PCR and RAT results

** Confirmed cases of Covid-19: positive result of PCR or RAT testing to detect SARS-CoV-2 in a clinical sample (≥ 44 days from previous result)

Algorithm

Automated data transfer