Instituto Nacional de Saúde Doutor Ricardo Jorge

ecdc EUROPEAN CENTRE FOR DISEASE PREVENTION AND CONTROL

Virus Detection

From Reads to viral detection

Wrap Up - TELEVIR

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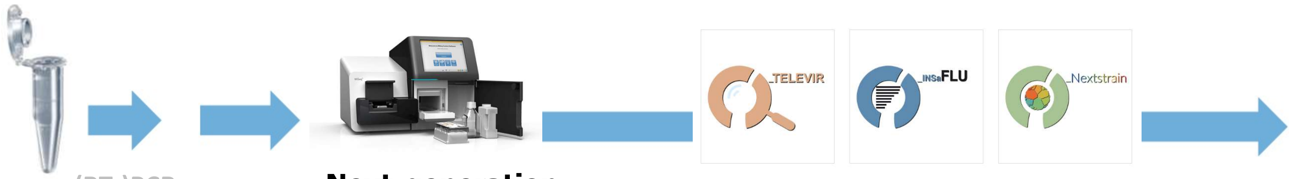
New era of <u>virus detection and surveillance</u>

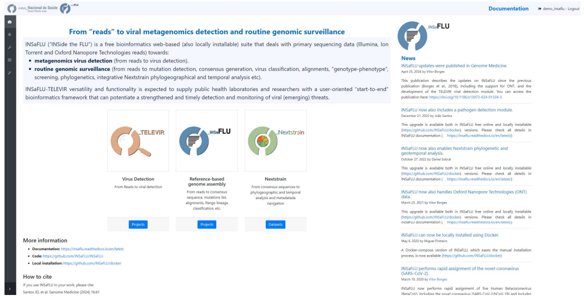
Metagenomics diagnosis and Whole-genome sequencing



(RT-)PCR

Next-generation Sequencing (NGS)

illumina
ion torrent
Oxford NANOPORE Technologies



From "reads" to viral metagenomics detection and routine genomic surveillance

INSaFLU ("INSide the FLU") is a free bioinformatics web-based (also locally installable) suite that deals with primary sequencing data (Illumina, Ion Torrent and Oxford Nanopore Technologies reads) towards:

- **metagenomics virus detection** (from reads to virus detection).
- **routine genomic surveillance** (from reads to mutation detection, consensus generation, virus classification, alignments, "genotype-phenotype", screening, phylogenetics, integrative Nextstrain phylogeographical and temporal analysis etc).

INSaFLU-TELEVIR versatility and functionality is expected to supply public health laboratories and researchers with a user-oriented "start-to-end" bioinformatics framework that can potentiate a strengthened and timely detection and monitoring of viral (emerging) threats.

Virus Detection
From Reads to viral detection

Reference-based genome assembly
From reads to consensus sequence, mutations list, alignments, Pango lineage classification, etc.

Nextstrain
From consensus sequences to phylogeographic and temporal analysis and metadatada navigation

More information

- **Documentation:** https://insaflu.readthedocs.io/en/latest
- **Code:** https://github.com/INSaFLU/INSaFLU
- **Local installation:** https://github.com/INSaFLU/docker

How to cite

If you use INSaFLU in your work, please cite:
Santos JD, et al. Genome Medicine (2024) 16:61

News

INSaFLU updates were published in Genome Medicine.
April 25, 2024 by Vitor Borges

This publication describes the updates on INSaFLU since the previous publication (Borges et al., 2018), including the support for ONT, and the development of the TELEVIR viral detection module. You can access the publication here: https://doi.org/10.1186/s13073-024-01334-3

INSaFLU now also includes a pathogen detection module.
December 21, 2022 by João Santos

This upgrade is available both in INSaFLU free online and locally installable (https://github.com/INSaFLU/docker) versions. Please check all details in INSaFLU documentation (https://insaflu.readthedocs.io/en/latest/)

INSaFLU now also enables Nextstrain phylogenetic and geotemporal analysis.
October 27, 2022 by Daniel Sobral

This upgrade is available both in INSaFLU free online and locally installable (https://github.com/INSaFLU/docker) versions. Please check all details in INSaFLU documentation (https://insaflu.readthedocs.io/en/latest/)

INSaFLU now also handles Oxford Nanopore Technologies (ONT) data.
March 25, 2021 by Vitor Borges

This upgrade is available both in INSaFLU free online and locally installable (https://github.com/INSaFLU/docker) versions. Please check all details in INSaFLU documentation (https://insaflu.readthedocs.io/en/latest/)

INSaFLU can now be locally installed using Docker.
May 6, 2020 by Miguel Pinheiro

A Docker-compose version of INSaFLU, which eases the manual installation process, is now available (https://github.com/INSaFLU/docker)

INSaFLU performs rapid assignment of the novel coronavirus (SARS-CoV-2).
March 10, 2020 by Vitor Borges

INSaFLU now performs rapid assignment of five Human Betacoronavirus (BetaCoV) including the novel coronavirus (SARS-CoV-2/hCoV-19) and includes

https://insaflu.insa.pt

QC **QUALITY CONTROL AND CLASSIFICATION**

INSaFLU **GENOMIC SURVEILLANCE (*INSaFLU*)**

Nextstrain **PHYLOGEOGRAPHY (*Nextstrain*)**

TELEVIR **VIRUS DETECTION (*TELEVIR*)**

Dourado Santos J et al., (2024) Genome Medicine; 16:61
https://doi.org/10.1186/s13073-024-01334-3

<u>GOAL</u>: To supply public health laboratories and researchers around the world with an **open and user-friendly bioinformatics framework** for viral metagenomic detection and routine genomic surveillance

INSaFLU-TELEVIR https://insaflu.insa.pt/

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_TELEVIR

illumina

iontorrent

Oxford NANOPORE Technologies

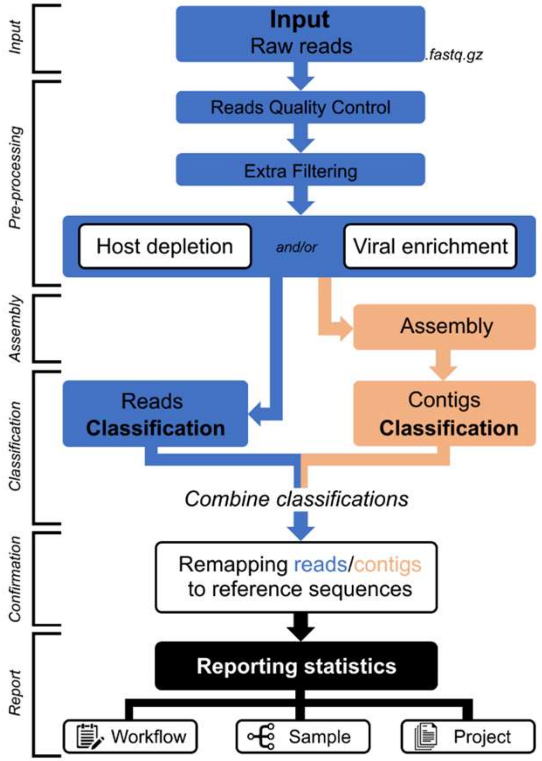
Viral metagenomic detection

✓ **Hypothesis-free virus detection**

- Multiple Classification methods
- Multiple Viral databases
- Multiple Host sequences for Depletion
- Innovative False Positive Discovery

✓ **Targeted virus screening**

- > 17000 viral TAXIDs available
- Easy reference panel creation



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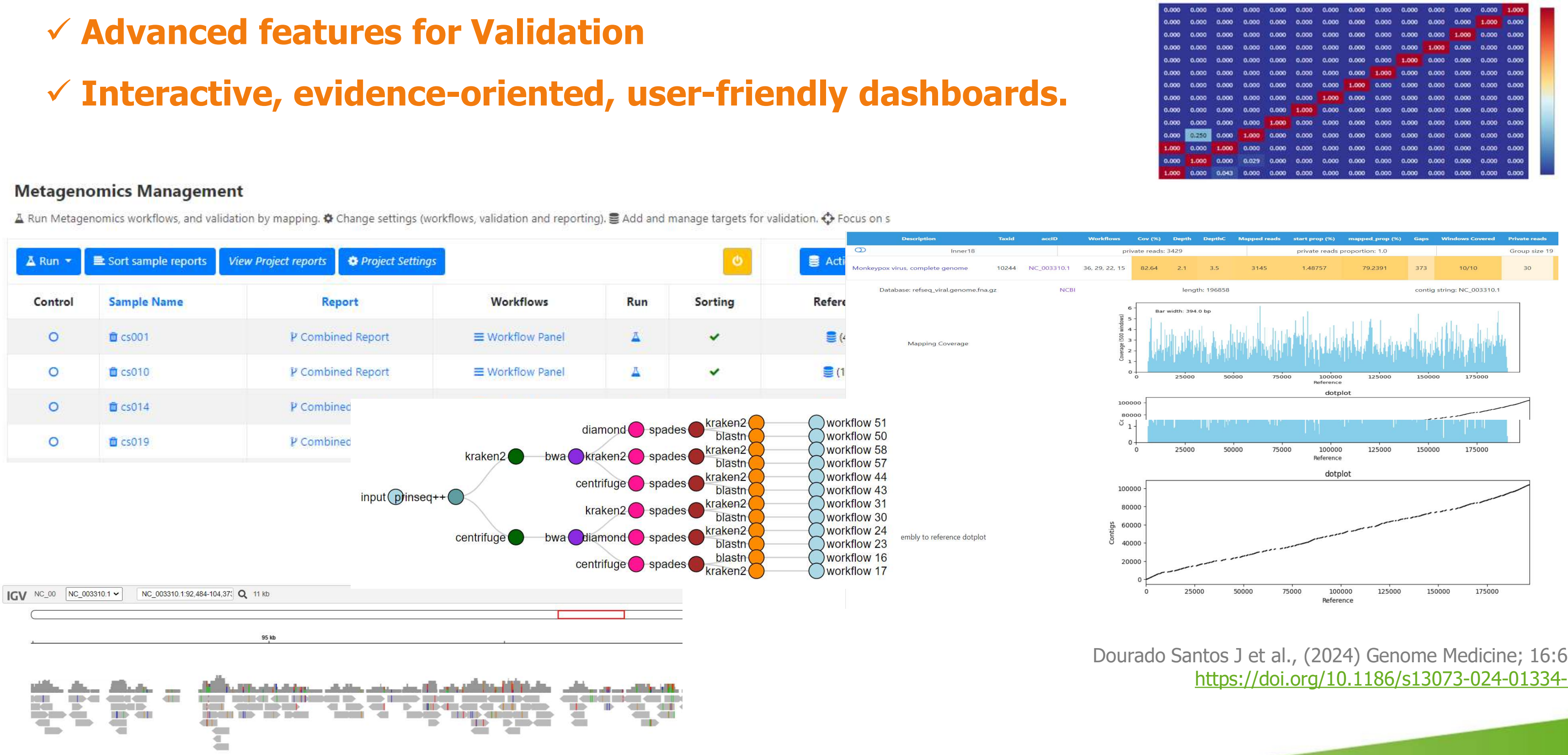
Viral metagenomic detection

- ✓ **Advanced features for Validation**
- ✓ **Interactive, evidence-oriented, user-friendly dashboards.**



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1.000	0.000	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
0.000	1.000	0.000	0.029	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
1.000	0.000	0.043	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000

Metagenomics Management

Run Metagenomics workflows, and validation by mapping. Change settings (workflows, validation and reporting). Add and manage targets for validation. Focus on s

Control	Sample Name	Report	Workflows	Run	Sorting	Refere
○	cs001	Combined Report	Workflow Panel		✓	(4
○	cs010	Combined Report	Workflow Panel		✓	(1
○	cs014	Combined				
○	cs019	Combined				

Description	Taxid	accID	Workflows	Cov (%)	Depth	DepthC	Mapped reads	start prop (%)	mapped_prop (%)	Gaps	Windows Covered	Private reads
Inner18			private reads: 3429					private reads proportion: 1.0				Group size 19
Monkeypox virus, complete genome	10244	NC_003310.1	36, 29, 22, 15	82.64	2.1	3.5	3145	1.48757	79.2391	373	10/10	30

Database: refseq_viral.genome.fna.gz NCBI length: 196858 contig string: NC_003310.1

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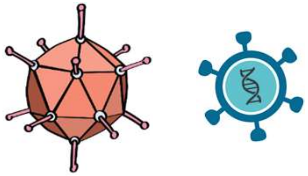
Viral metagenomic detection



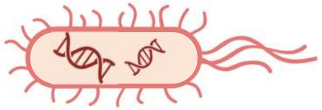
illumina

iontorrent

Oxford NANOPORE Technologies

- ✓ **Available online**
- ✓ **Free to Use**

- ✓ **Open Source**
- ✓ **Locally installable**



GitHub

When installed locally, TELEVIR can be expanded to encompass **bacterial databases** and indices that allow the deployment of the whole suite for bacterial metagenomic identification.

Wrap Up - Global

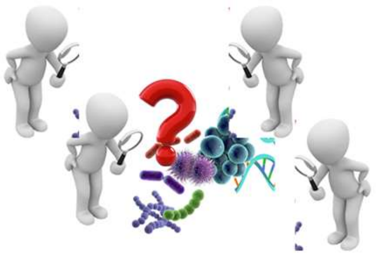
Clinical metagenomic NGS (mNGS)

mNGS does not replace conventional tests entirely but serves as a powerful complementary tool.

Keys to Unlock the full impact of clinical mNGS

…

- Continuous **advances** in **bioinformatics**
- **Stronger integration** and **communication** between **laboratories** and **clinical teams**



Clinical metagenomic NGS (mNGS)

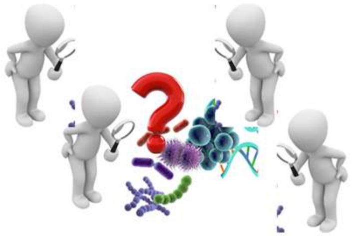
Future bioinformatics directions

- **Preparedness for complexity** (recombinants/reassortments, novel and divergent viruses) and keep **mitigating false-positive signals** through continued developments.
- **Incorporate Artificial Intelligence:**
  - **Improve Taxonomy, Harmonize Databases**
  - Prediction of **phenotypically relevant traits** (AMR, immune evasion mutations, etc)
  - **Advanced reports** with sample history, patient data, literature, advanced metrics.
- Streamline **inter-laboratory comparability and multi-center validation**

etc

Clinical metagenomic NGS (mNGS)

Clinical Integration Strategies



- **Multidisciplinary collaboration:**
    - Clinicians, microbiologists, and bioinformaticians can jointly decide when to order mNGS, select best samples, etc.
    - Microbial sequencing boards (similar to tumor boards) to enable case-based interpretation and discussion of results.
- **Structured training programs** to increase clinician familiarity and confidence in mNGS.
- **Continuous feedback loops between labs and clinicians** improves accuracy and utility (e.g., surveys on diagnostic impact).

Clinical Metagenomics and Bioinformatics

From Complex Data to Meaningful Diagnosis

Key messages:

1. Robust bioinformatics workflows are not a luxury but a necessity
2. No one-size-fits-all tool: diversity of tools = flexibility + challenge
3. Tools need to match context, resources, and goals
4. Think beyond your lab: Tool and Experience Sharing, Harmonized metrics and Comparability are crucial
5. Robust Reporting and Communication with clinicians is crucial for full impact

📌 The ultimate goal: data → decisions → action