

YAVSAP

Versatile viral quasispecies analysis for veterinary samples

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Mitzel ² William Wilson ² Rachel Palinski ¹

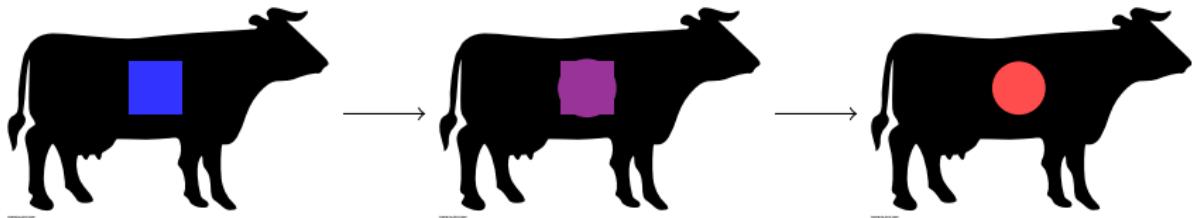
Kansas State University Veterinary Diagnostic Laboratory, Kansas State University,
Manhattan, KS

Foreign Arthropod Borne Animal Disease Research, National Bio- and Agro-Defence Facility,
United States Department of Agriculture Agricultural Research Service, Manhattan, KS

September 15, 2024

What is a quasispecies?

Consensus level



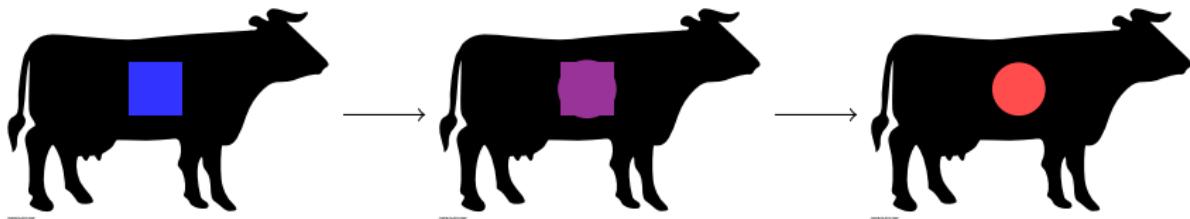
¹Domingo, Sheldon, and Perales, "Viral Quasispecies Evolution".

²Andino and Domingo, "Viral Quasispecies".

³Vignuzzi et al., "Quasispecies Diversity Determines Pathogenesis through Cooperative Interactions in a Viral Population".

What is a quasispecies?

Consensus level



Quasispecies model

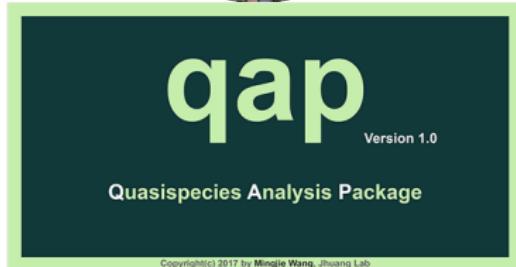
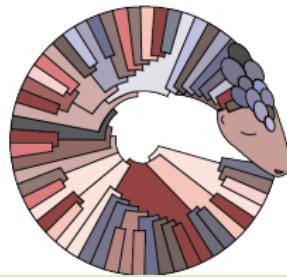


¹Domingo, Sheldon, and Perales, "Viral Quasispecies Evolution".

²Andino and Domingo, "Viral Quasispecies".

³Vignuzzi et al., "Quasispecies Diversity Determines Pathogenesis through Cooperative Interactions in a Viral Population".

Existing quasispecies pipelines



⁴Harshil Patel et al., *Nf-Core/Viralrecon*.

⁵O'Toole et al., "Assignment of Epidemiological Lineages in an Emerging Pandemic Using the Pangolin Tool".

⁶Hadfield et al., "Nextstrain".

⁷Wang et al., "An Integrated Software for Virus Community Sequencing Data Analysis".

Gap in veterinary coverage: viralrecon

```
N E X T F L O W ~ version 23.04.2
Pulling nf-core/viralrecon ...
Already-up-to-date
Launching 'https://github.com/nf-core/viralrecon' [nostalgic_curran] DSL2 - revision: 3731dd3a32 [master]

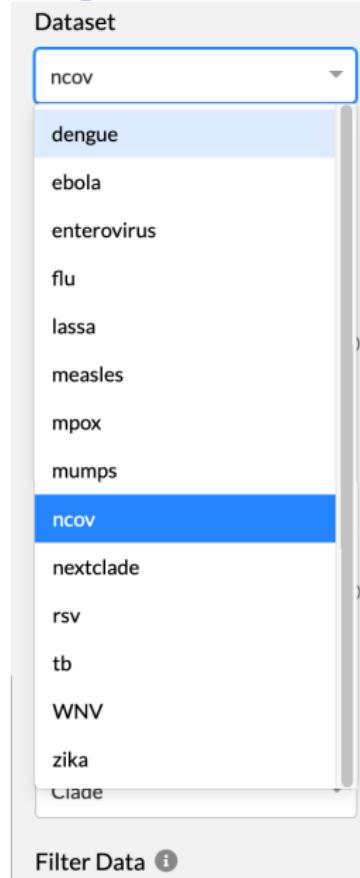
ERROR ~ -----
Genome 'genome.fasta' not found in any config files provided to the pipeline.
Currently, the available genome keys are:
NC_045512.2, MN908947.3, NC_063383.1, ON563414.3, MT903344.1
-----
-- Check '.nextflow.log' file for details
```

Gap in veterinary coverage: Nextstrain

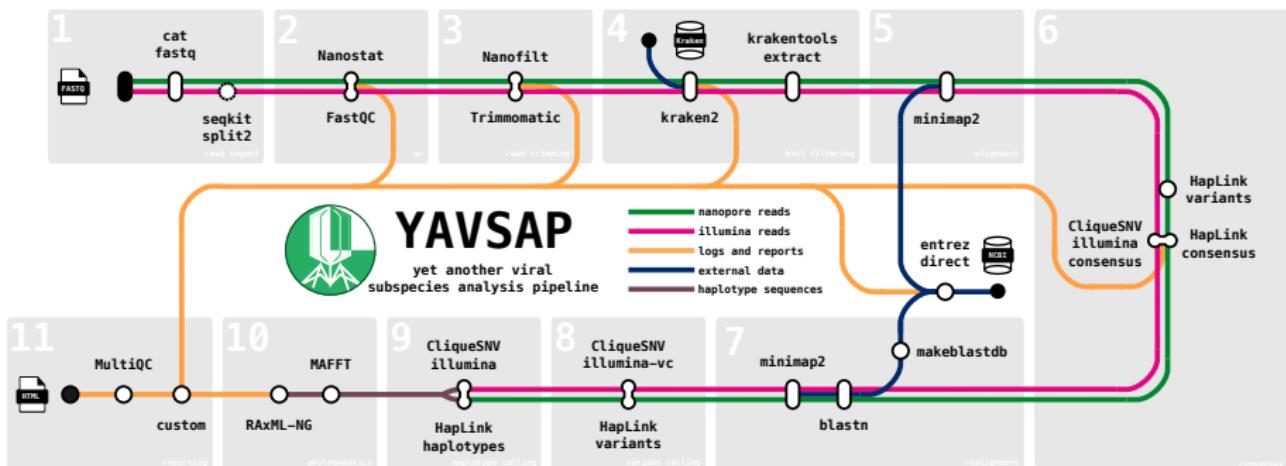
Dataset

- ncov
- dengue
- ebola
- enterovirus
- flu
- lassa
-)
- measles
- mpox
- mumps
- ncov
- nextclade
-)
- rsv
- tb
- WNV
- zika
- Clade

Filter Data ⓘ



Introducing YAVSAP



Sequencing technologies

Illumina



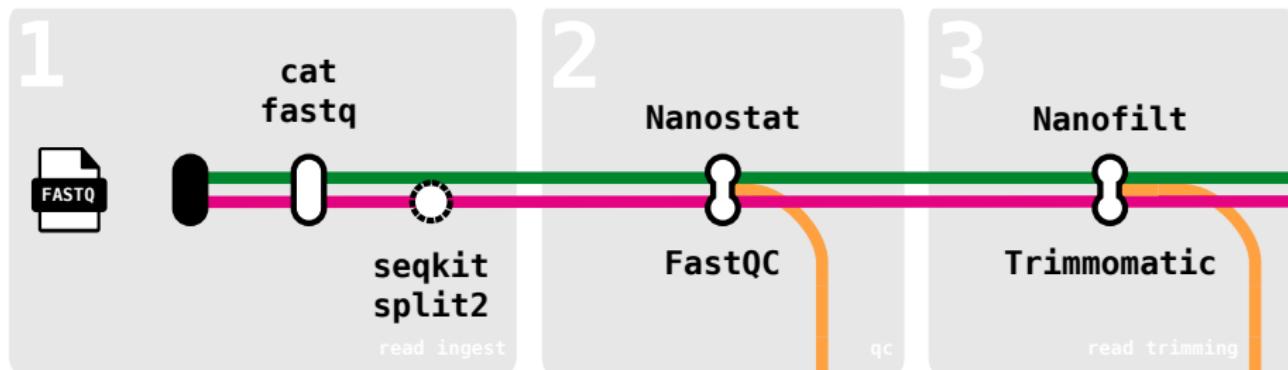
- short reads
- high quality
- reads come in pairs

Oxford Nanopore



- long reads
- low quality
- single reads

Ingestion, quality control, and filtering



Host filtering

4

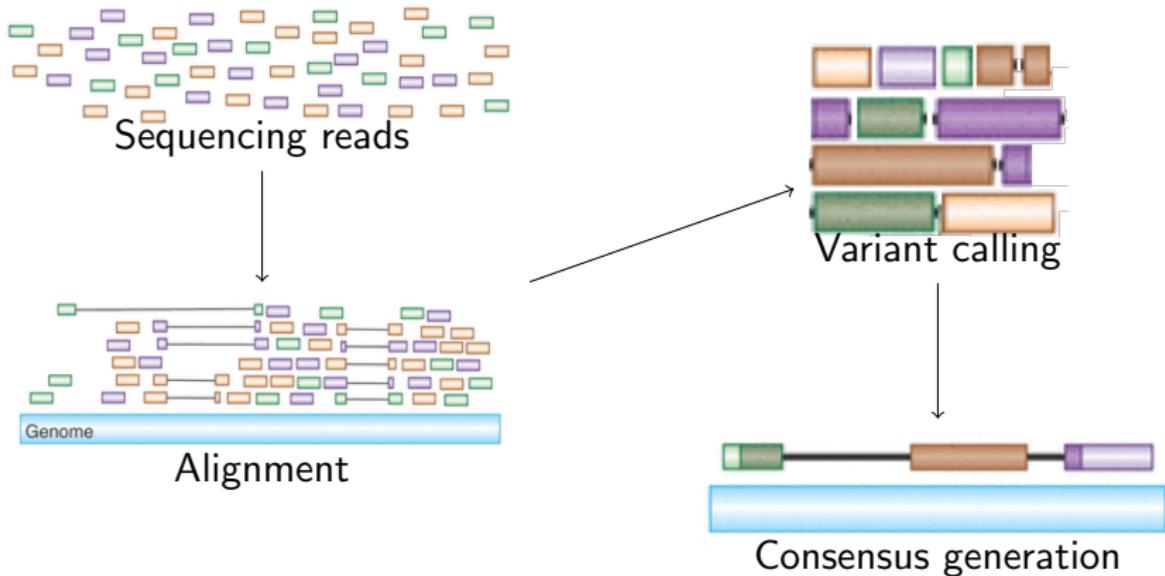


**krakentools
extract**

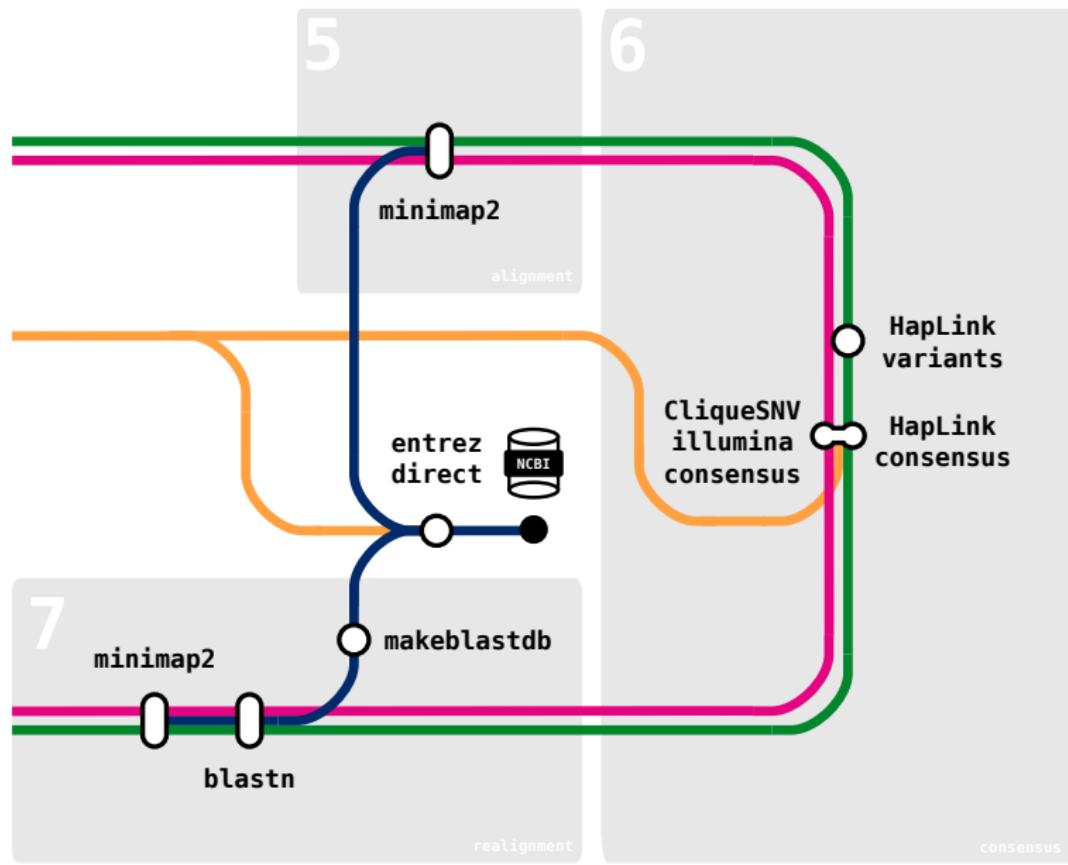
kraken2

host filtering

Alignment: process overview

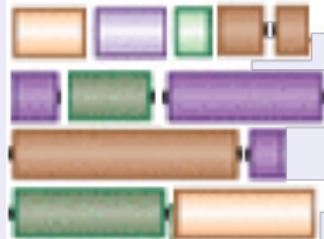
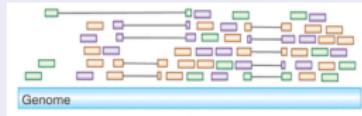


Alignment, consensus generation, and realignment

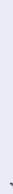


Variant and haplotype calling: process overview

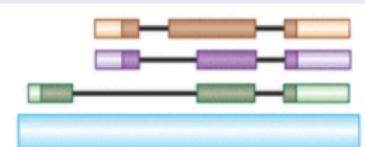
Variant calling



Consensus calling



Haplotype calling



Variant and haplotype calling

9

CliqueSNV
illumina



HapLink
haplotypes

haplotype calling

8

CliqueSNV
illumina–vc



HapLink
variants

variant calling

Phylogenetics

10

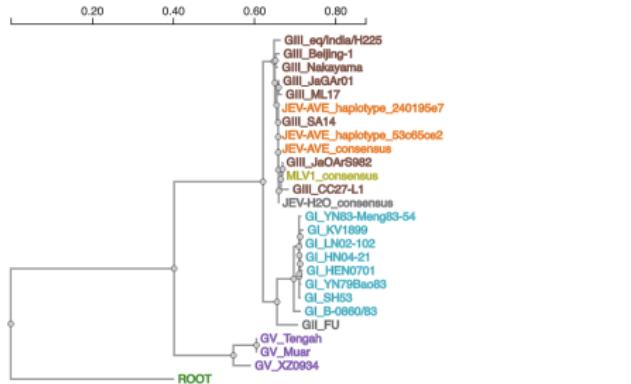
MAFFT



RAxML-NG

phylogenetics

```
>ROOT
ggcataaggcagaggatgtcactactattaac
>pool01_consensus
ggcataaggcagaggatgtcactactattaac
>sample03_haplotype_8637fdf8
ggcataaggcagaggatgtcactactattaac
```



Report

MultiQC
v1.12

General Stats
NanoStat
Fastq stats
Reads by quality
Kraken
HapLink.jl (Haplotypes)
RAxML-NG
yavsap Software Versions
yavsap Workflow Summary

MultiQC

A modular tool to aggregate results from bioinformatics analyses across many samples into a single report.

This report has been generated by the [yavsap](#) analysis pipeline. For information about how to interpret these results, please see the [documentation](#).

Report generated on 2022-09-17, 22:32

Welcome! Not sure where to start?

[Watch a tutorial video](#) (6:06)

don't show again

General Statistics

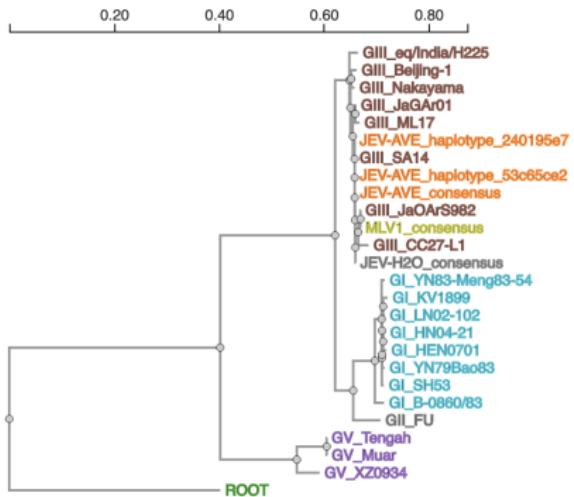
[Copy table](#) [Configure Columns](#) [Plot](#) Showing 3/3 rows and 3/3 columns.

Sample Name	% Mesocricetus auratus	% Top 5 Species	% Unclassified
JEV-AVE	18.2%	49.2%	4.7%
JEV-H2O	17.3%	44.0%	6.1%
MLV1	0.7%	26.3%	17.5%



Viral analyses

- Japanese Encephalitis Virus
- Influenza D Virus
- Bovine Coronavirus
- SARS CoV2
- Rift Valley Fever Virus
- Porcine Reproductive and Respiratory Syndrome Virus



Next steps

Internal

- Novel virus discovery
- Multi-strand virus improvements
- Protein transcript prediction

External

- Identify JEV “bottleneck” genomes
- Model transmission of JEV and RVFV
- Surveillance and monitoring of enteric and respiratory viruses
- Vaccine development

Acknowledgments

Molecular NGS Department

- Rachel Palinski
- Steven Stanic
- Andrea Lu
- Tyler Doerksen
- Jayme Souza-Neto

The nf-core community

- Harshil Patel
- Philip Ewels
- James Fellows Yates
- Edmund Miller

KSVDL

- Nanyan Lu
- Gregg Hanzlicek

The BioJulia community

- Jakob Nissen
- Ciaran O'Mara
- Kevin Bonham

USDA

- William Wilson
- Dana Mitzel
- Steven Lakin
- SCINet High-performance computing center

Funding

This material is based upon work supported by the U.S. Department of Agriculture, Agricultural Research Service, under agreement No. 58-3022-0-001.