Comparative multi-tissue virus-host transcriptomics with Nextflow

Marie Lataretu

14.02.2020

Friedrich-Schiller-Universität Jena
RNA Bioinformatics and High-Throughput Analysis
Hölzer Lab

Previously in virus-host transcriptomics …

Special Issue

Mechanisms of Extracellular Immunomodulation Mediated by Infectious Agents

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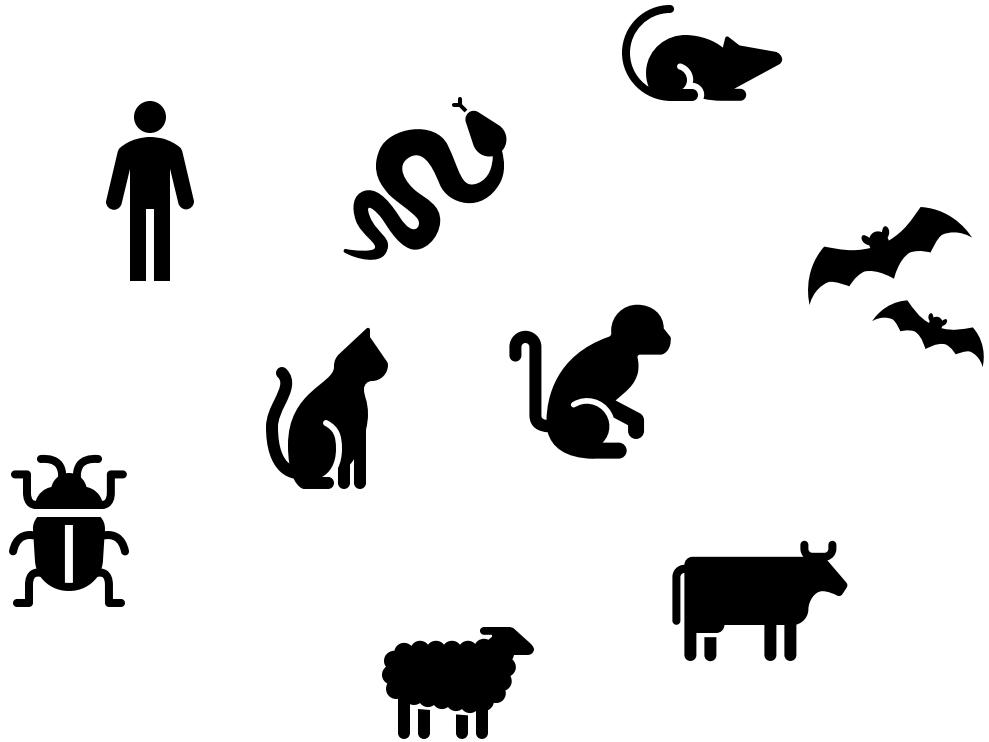
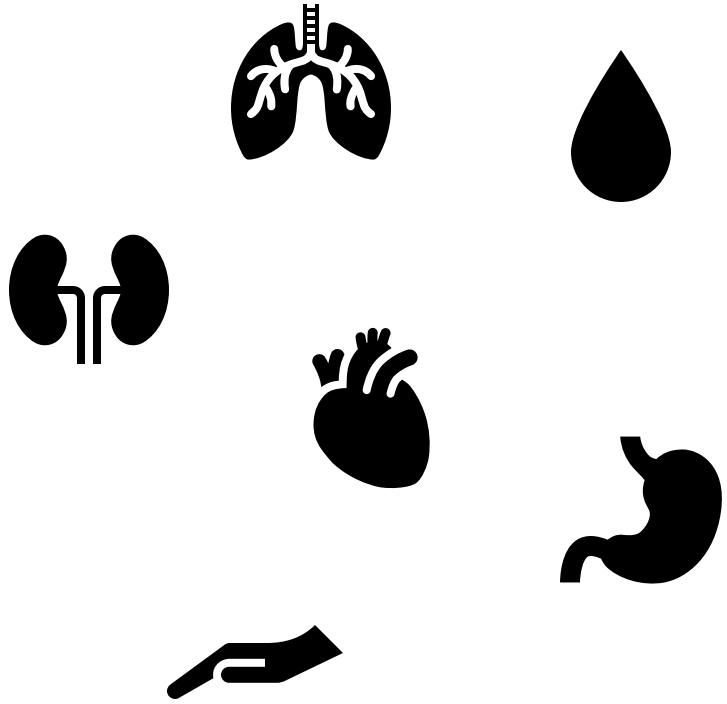
But, what about …

- Different hosts



But, what about …

- Different hosts
- Different host tissues



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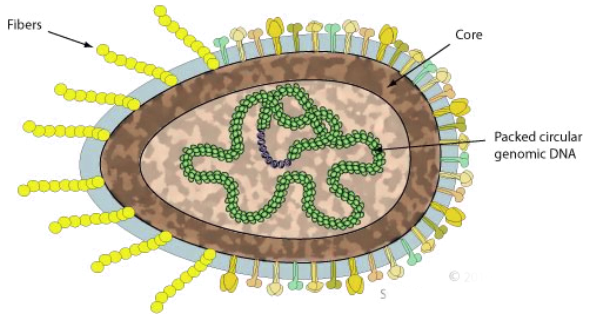
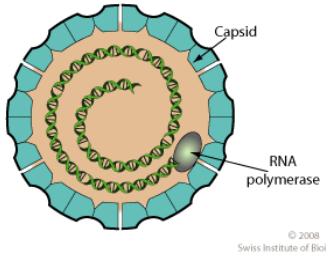
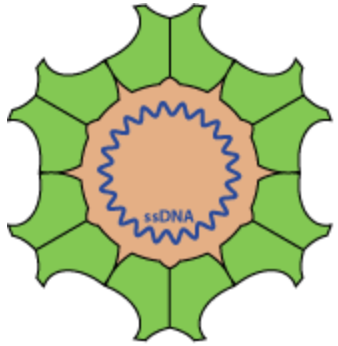
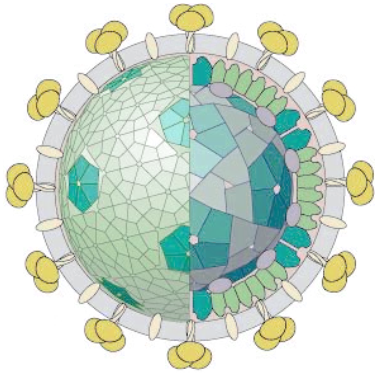
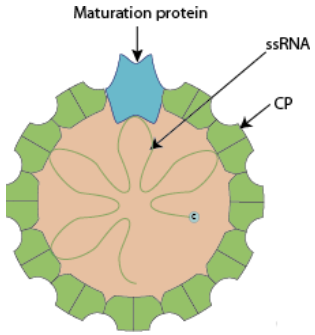






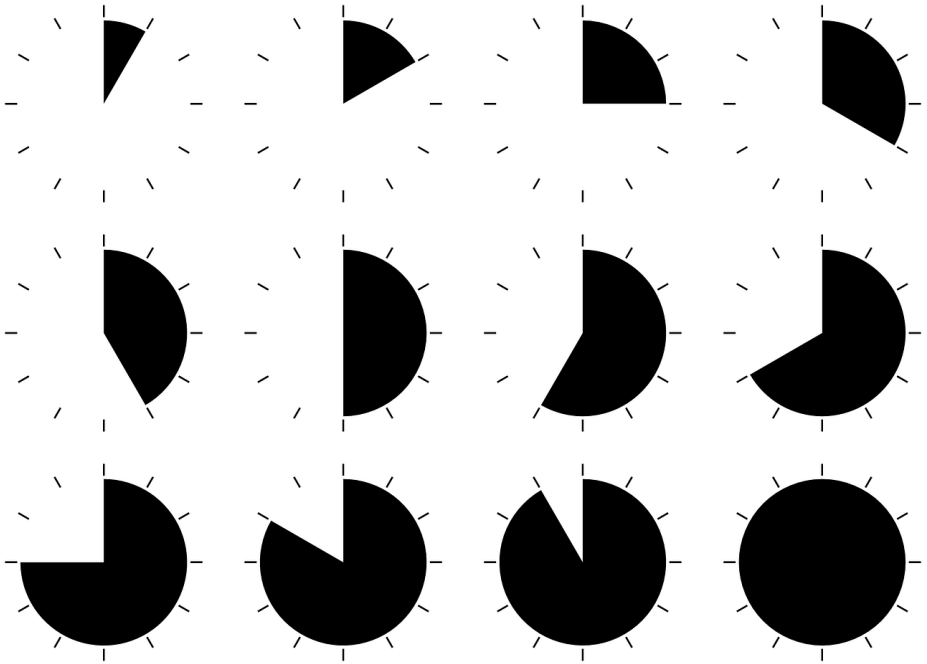




Hulo, Chantal, et al. "ViralZone: a knowledge resource to understand virus diversity." *Nucleic acids research* 39 (2011)

But, what about …

- Different hosts
- Different host tissues
- Different viruses
- Different timepoints



But, what about …

- Different hosts
- Different host tissues
- Different viruses
- Different timepoints
- Different RNA-Seq libraries

rRNA-

sRNA

Multidimensional approach

CPXV

EBOV 1

EBOV 2

MARV

RVFV

Clone 13

IFN

Multidimensional approach

CPXV | EBOV 1 | EBOV 2 | MARV | RVFV | Clone 13 | IFN

Rattus norvegicus

Rousettus aegyptiacus

Myotis daubentonii

Homo sapiens

Multidimensional approach

CPXV | EBOV 1 | EBOV 2 | MARV | RVFV | Clone 13 | IFN

Lung | Blood | Skin

Rattus norvegicus

Embryonic*

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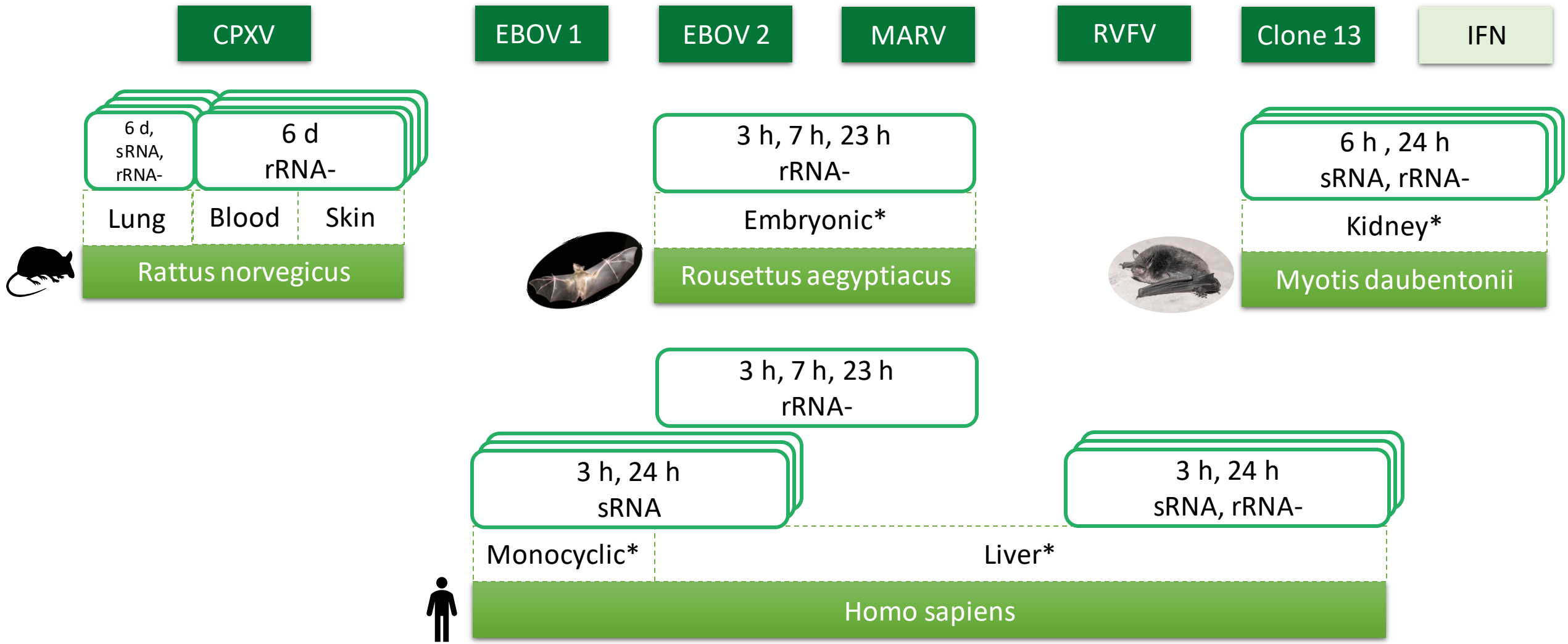
Monocyclic* | Liver*

Homo sapiens

* ⟶ cell line

Multidimensional approach



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Multidimensional approach

CPXV | EBOV 1 | EBOV 2 | MARV | RVFV | Clone 13 | IFN

6 d, sRNA, rRNA-

6 d rRNA-

Lung | Blood | Skin

Rattus norvegicus

3 h, 7 h, 23 h rRNA-

Embryonic*

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6 h , 24 h sRNA, rRNA-

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Myotis daubentonii

Σ 158

3 h, 7 h, 23 h rRNA-

3 h, 24 h sRNA

3 h, 24 h sRNA, rRNA-

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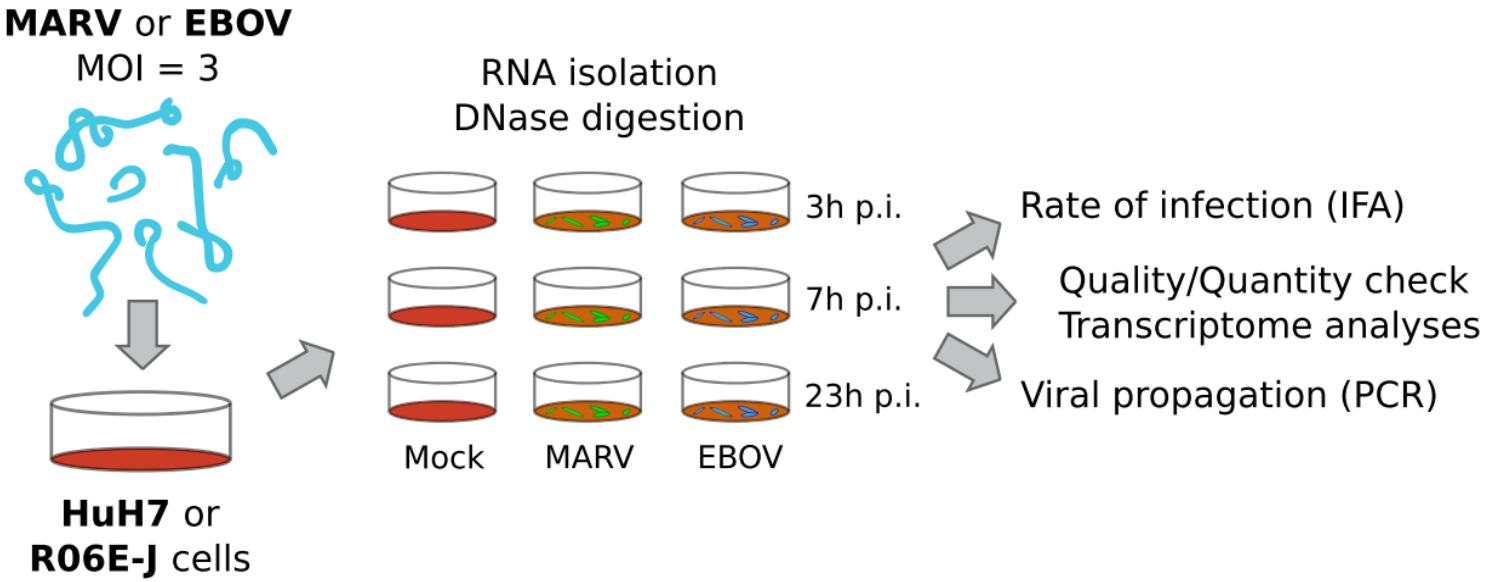

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Author Notes

NAR Genomics and Bioinformatics, Volume 2, Issue 1, March 2020, lqz006, https://doi.org/10.1093/nargab/lqz006

Published: 30 September 2019 **Article history** ▾

6 h , 24 h
sRNA
3x

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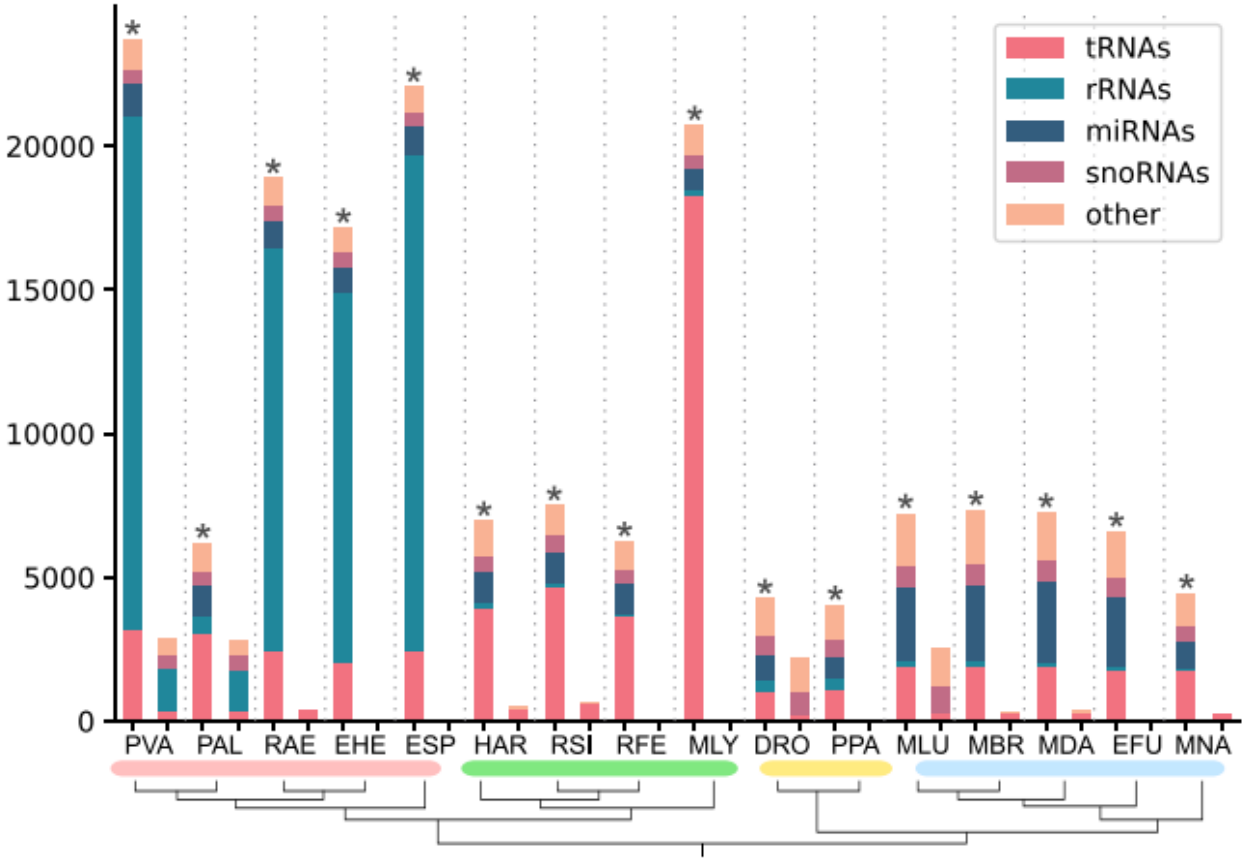

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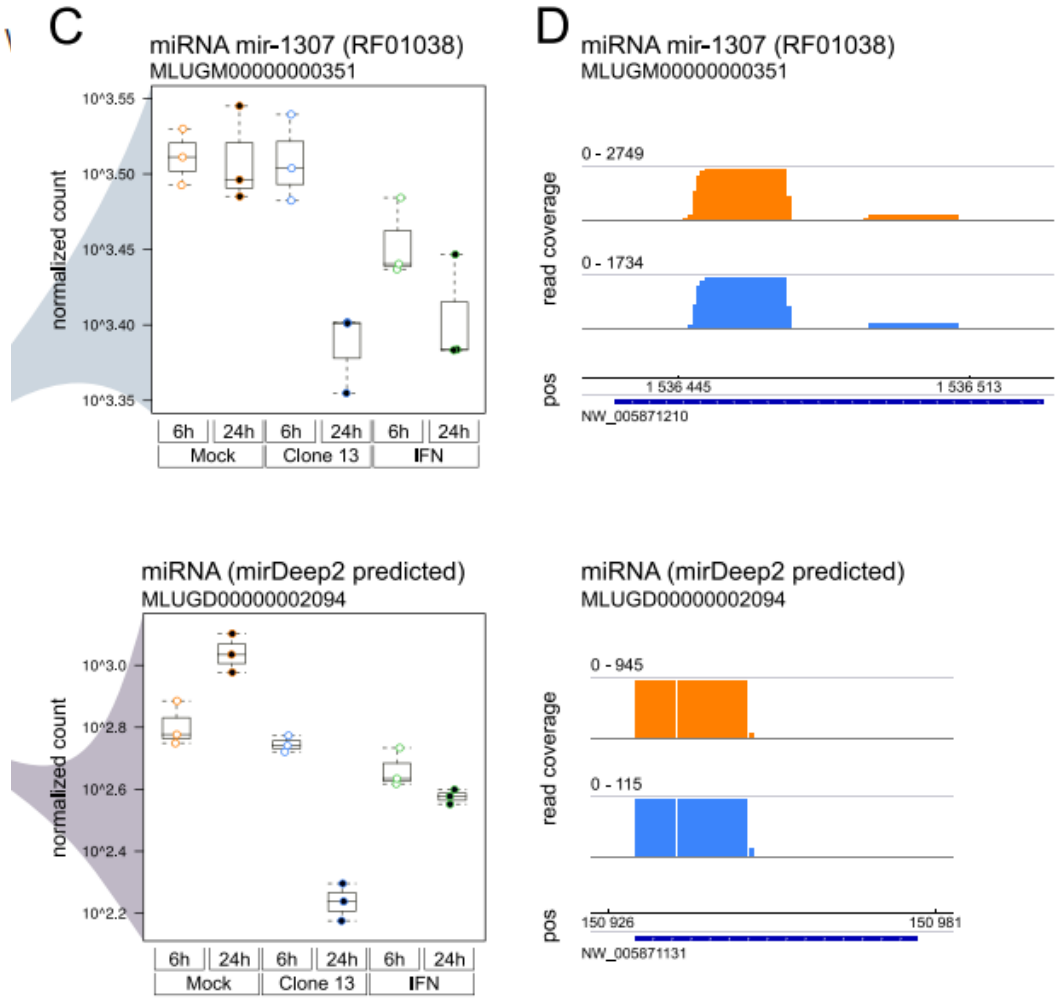
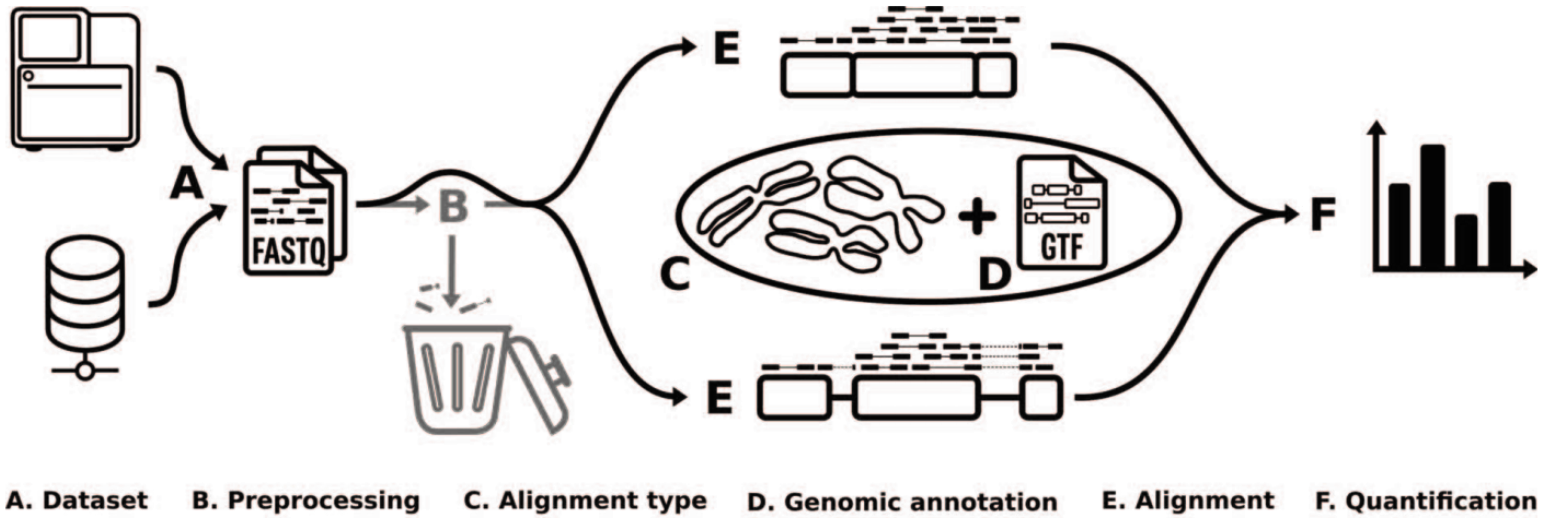


Battle plan



Simoneau, Joël, et al. "Current RNA-seq methodology reporting limits reproducibility." *Briefings in Bioinformatics* (2019)

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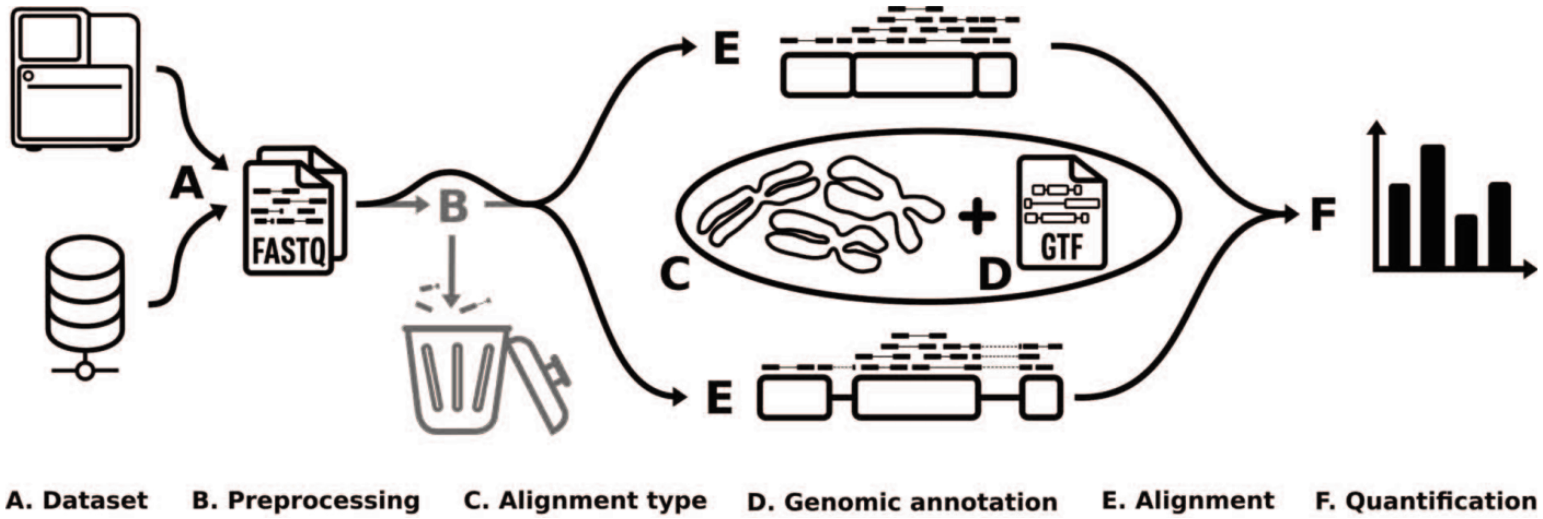

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- Homology mapping
- Which samples should be processed together?
- Which comparisons to do?

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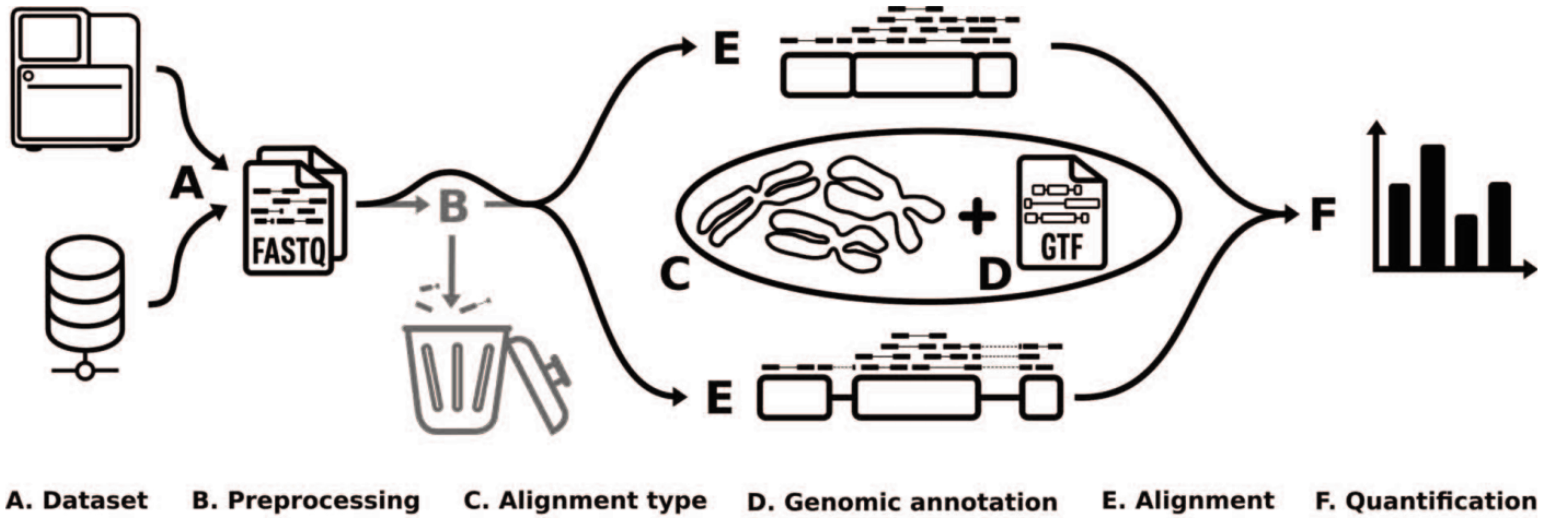

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Implementation

- Git
- Conda
- Nextflow

Why use a workflow management system?

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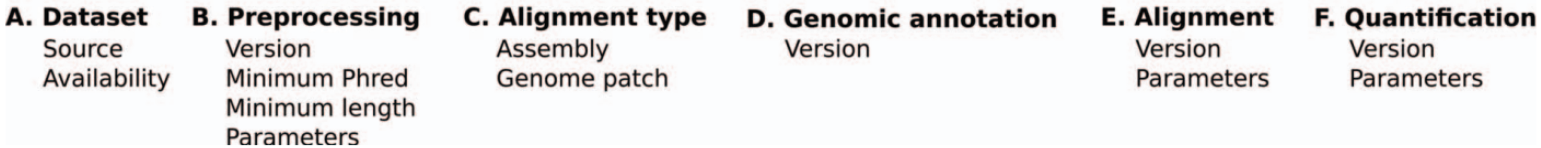

n = 465

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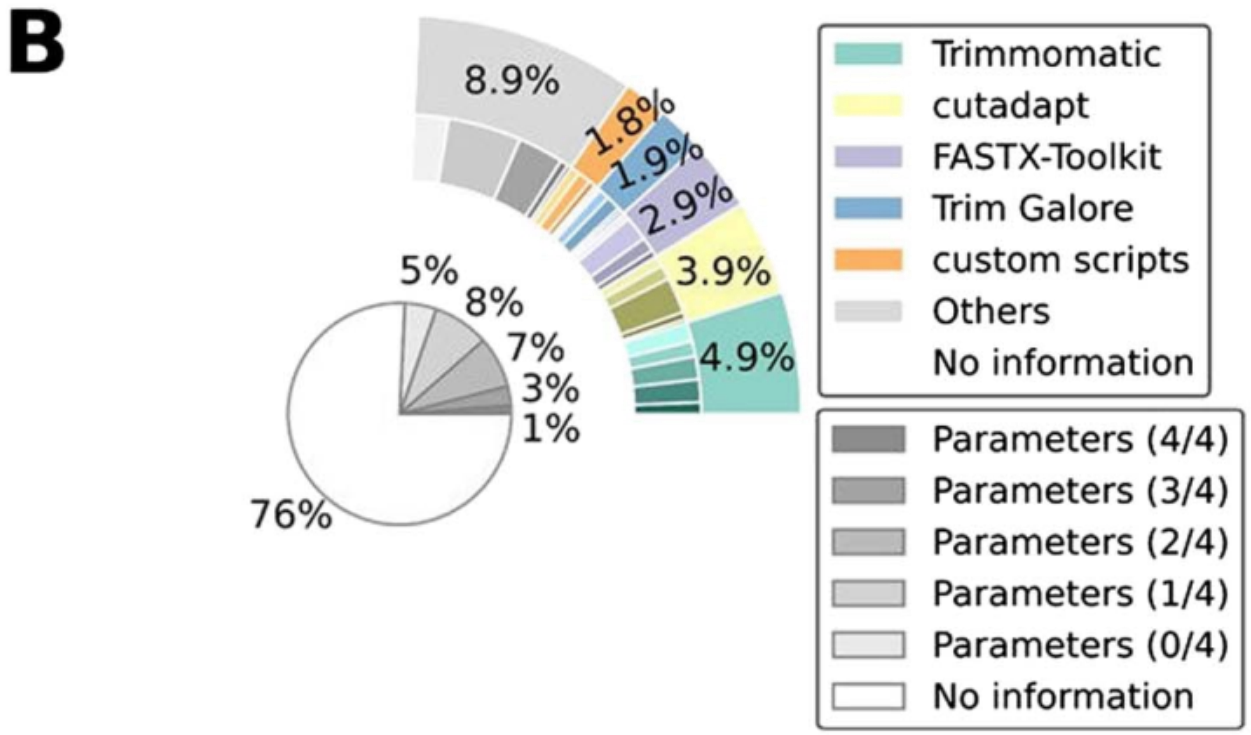

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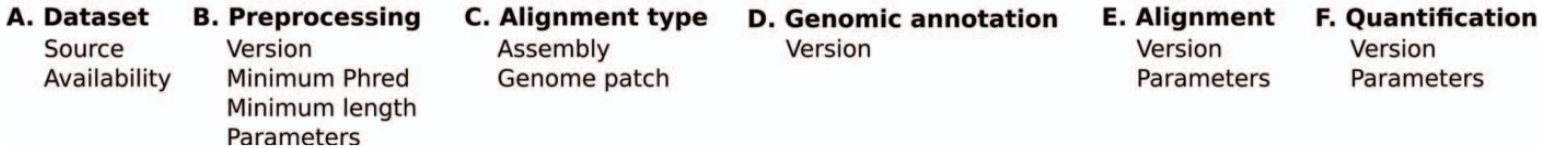

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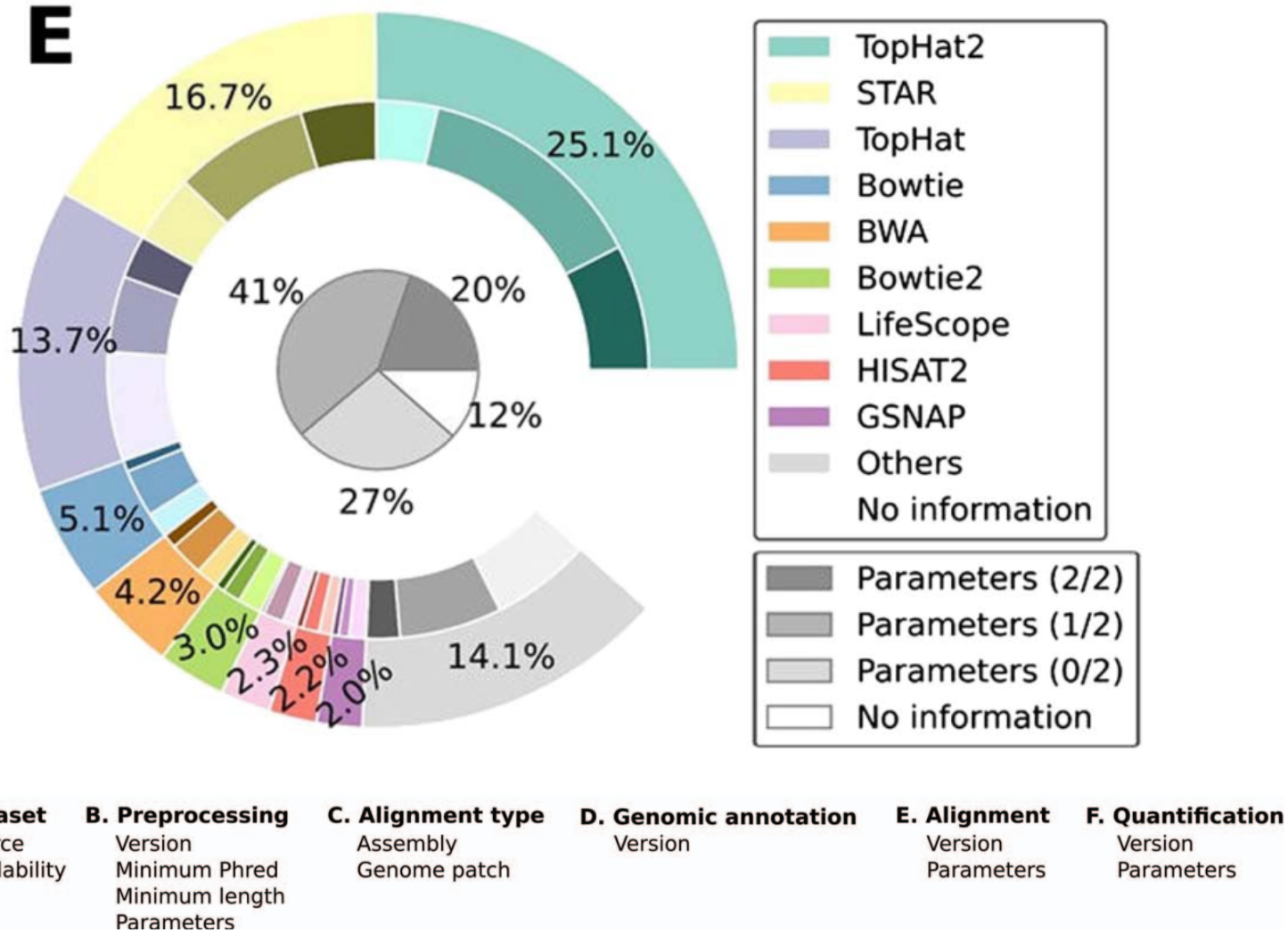

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nextflow

- Bioinformatics workflow manager

  ⟶ Portable and reproducible workflow

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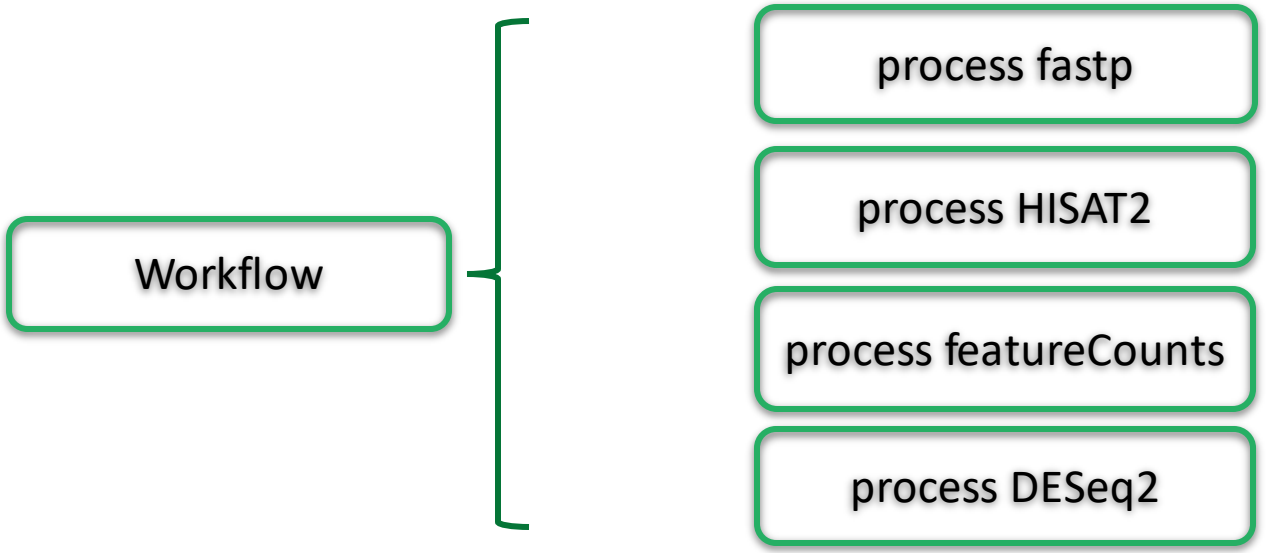

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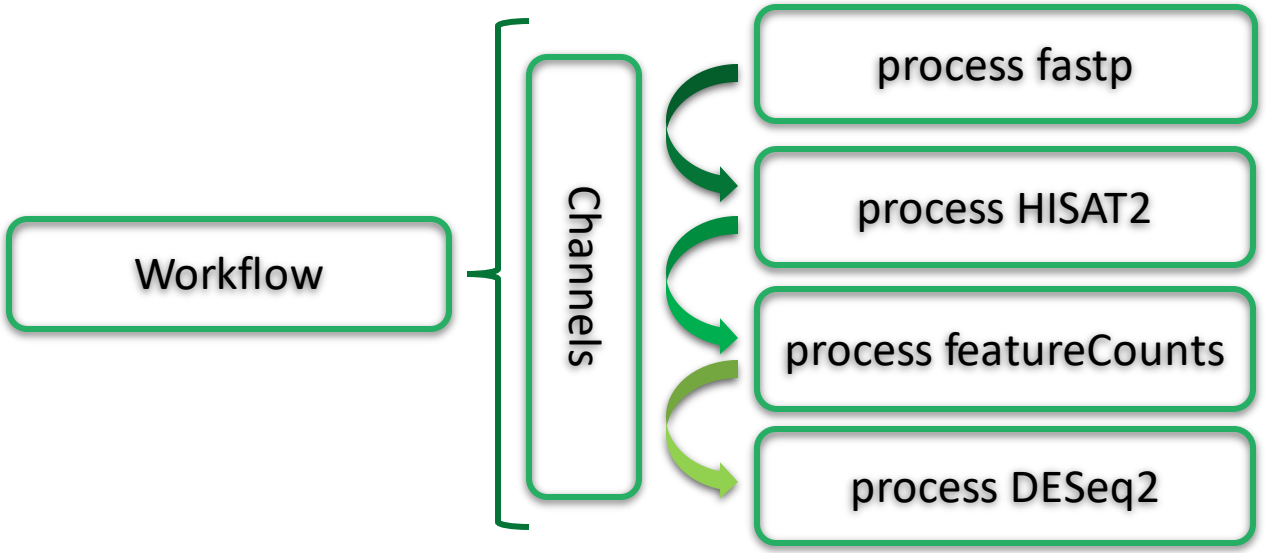

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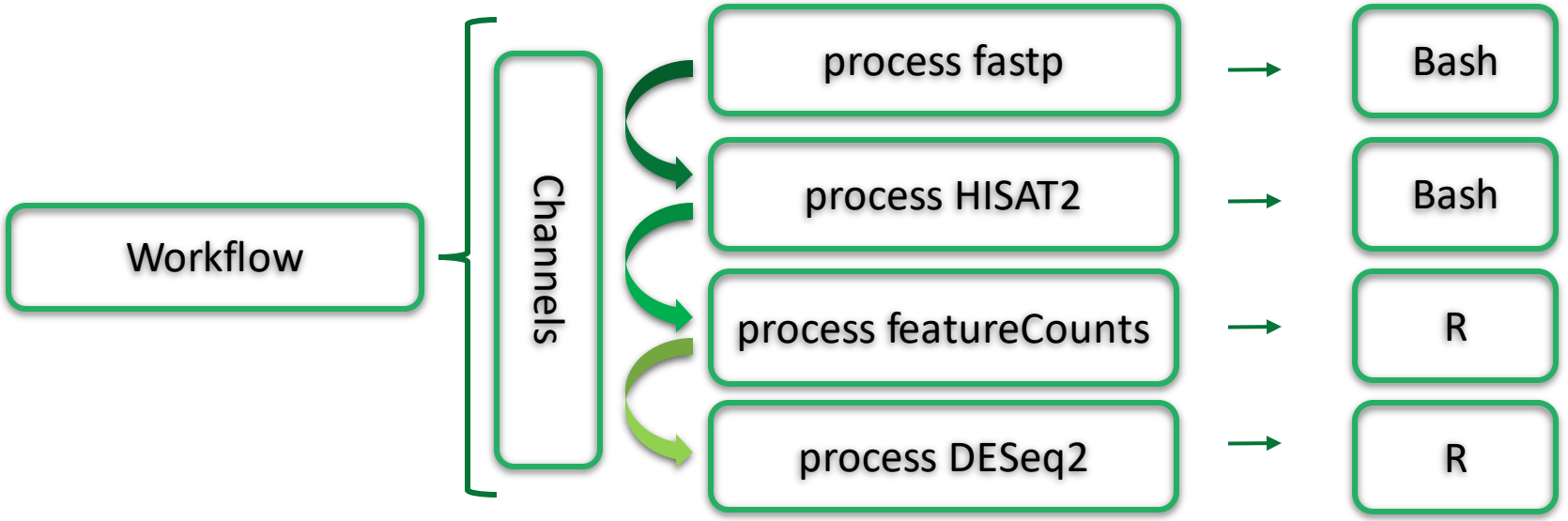

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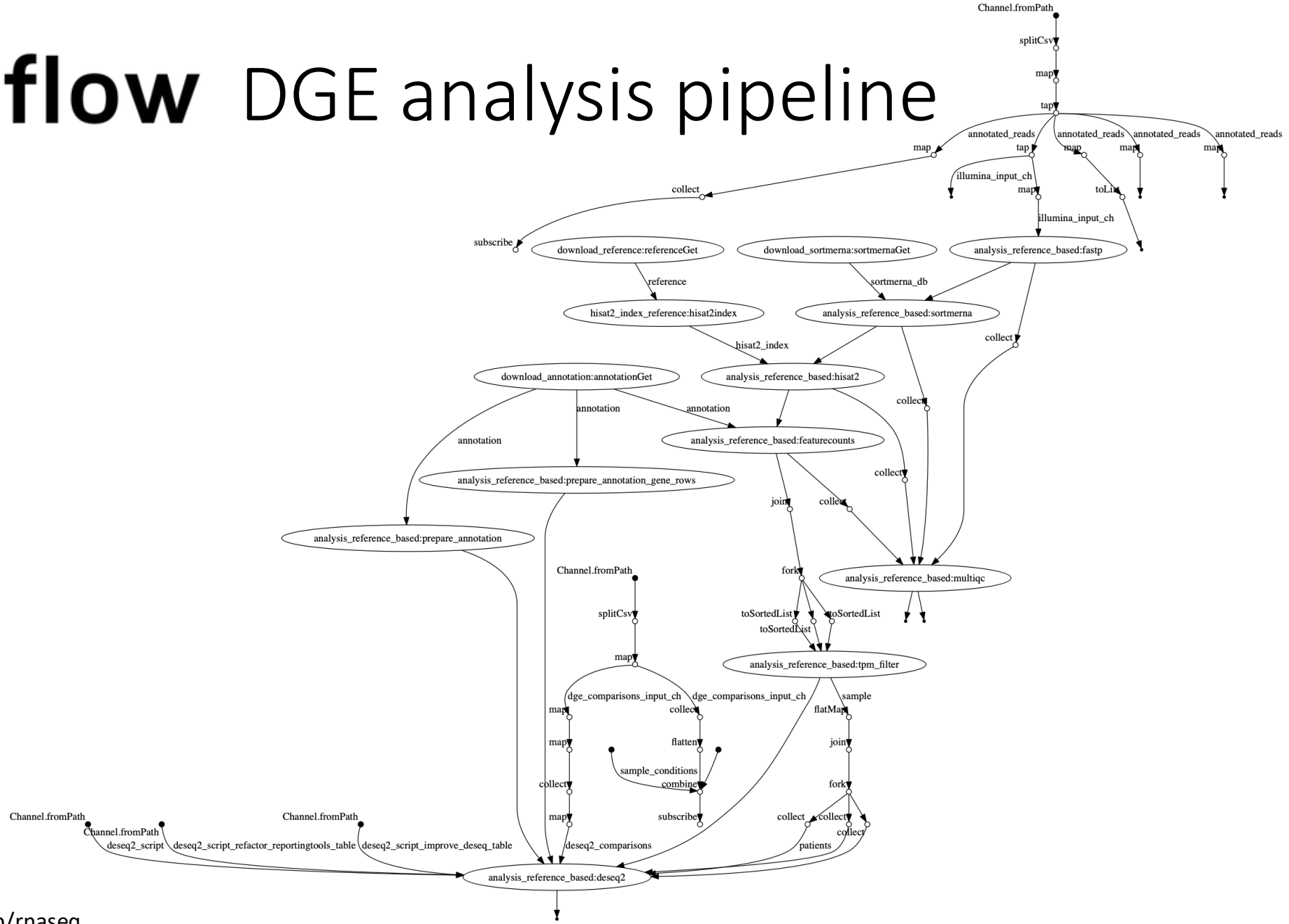
nextflow DGE analysis pipeline



https://github.com/hoelzer-lab/rnaseq

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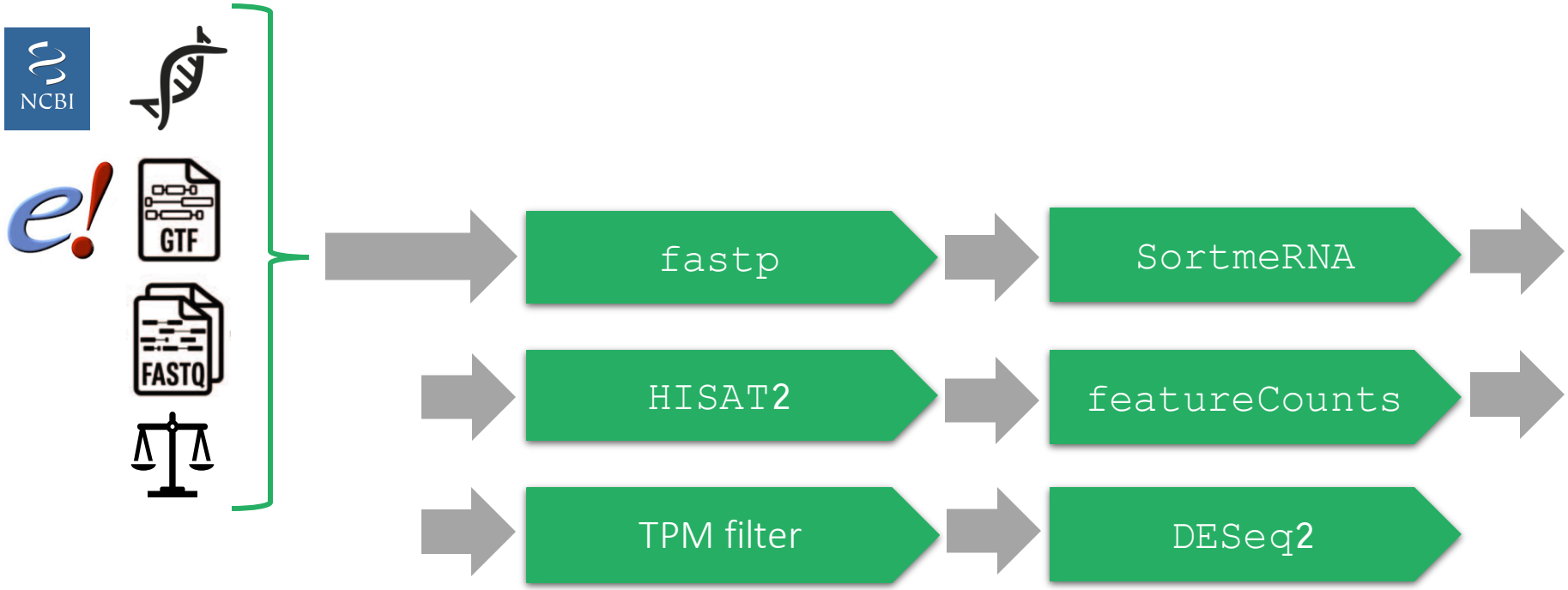

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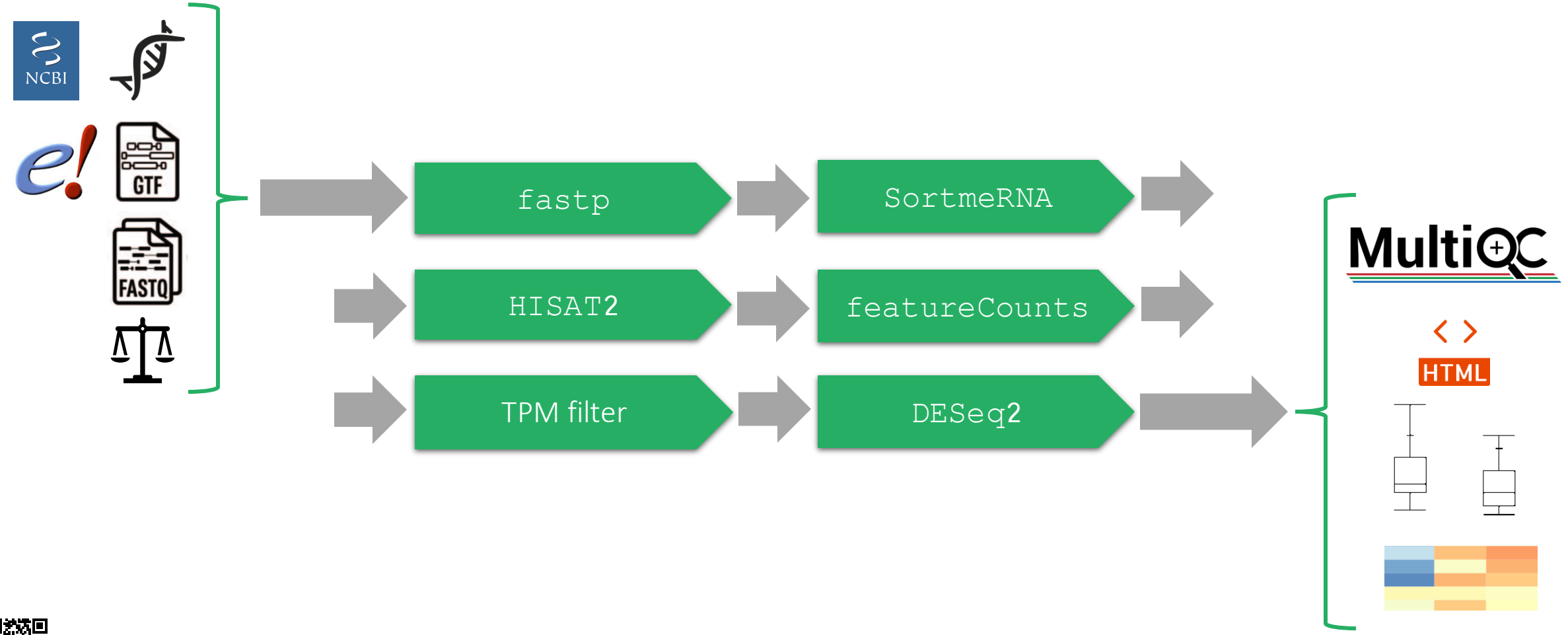


https://github.com/hoelzer-lab/rnaseq

nextflow DGE analysis pipeline - Output

RNA-seq analysis of differential expression using DESeq2, pvalue cutoff 1

10 records per page

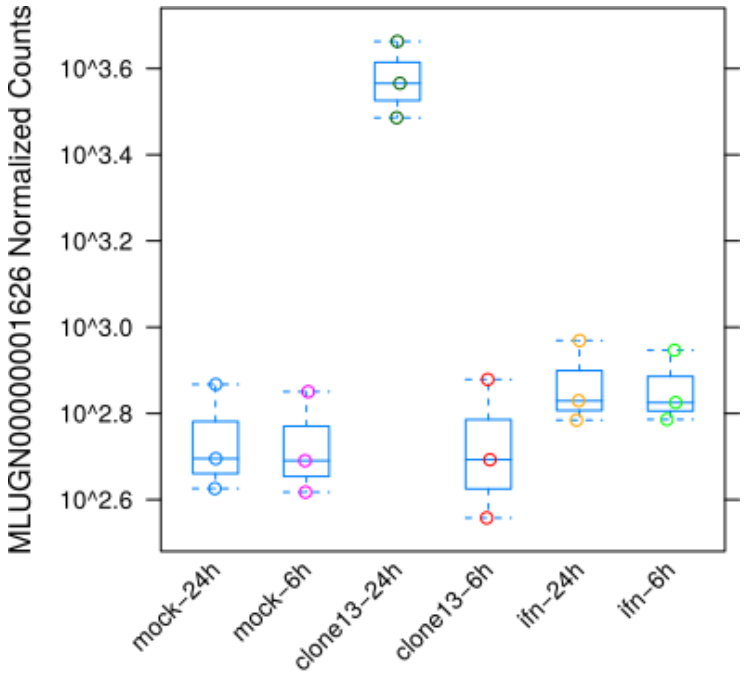
Search all columns:

ID	Name	Type	Position	Image	logFC (From / to)	p-Value (From / to)	Adjusted p-Value (From / to)
ER3413_1193	hlyE	protein_coding	Chromosome:1211292-1212203 (-)		-2.99000	6.01e-09	1.31e-06
ER3413_2342	elaB	protein_coding	Chromosome:2371605-2371910 (-)		-2.83000	1.17e-08	2.12e-06
ER3413_1529	gadB	protein_coding	Chromosome:1551074-1552474 (-)		-2.68000	8.21e-07	9.92e-05
ER3413_3604	yhiD	protein_coding	Chromosome:3624934-3625581 (-)		-2.67000	8.78e-13	3.18e-10
ER3413_3605	hdeB	protein_coding	Chromosome:3625645-3625971 (-)		-2.41000	4.33e-13	2.35e-10
ER3413_3608	gadE	protein_coding	Chromosome:3628045-3628572 (+)		-2.37000	2.58e-08	4.01e-06
ER3413_3606	hdeA	protein_coding	Chromosome:3626087-3626419 (-)		-2.34000	6.23e-14	6.78e-11
ER3413_1026	yccJ	protein_coding	Chromosome:1063696-1063923 (-)		-2.16000	7.20e-08	9.78e-06
ER3413_1528	gadC	protein_coding	Chromosome:1549383-1550918 (-)		-2.08000	9.81e-06	9.29e-04
ER3413_3603	dctR	protein_coding	Chromosome:3624362-3624892 (+)		-2.07000	3.12e-05	2.42e-03

Showing 1 to 10 of 1,087 entries

← Previous 1 2 3 4 5 Next →

nextflow DGE analysis pipeline - Output



Thanks to

Hölzer Bioinformatics Team

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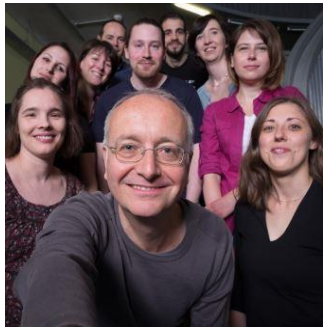

Friedemann Weber

Martin Beer

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FRIEDRICH-LOEFFLER-INSTITUT
FLI
Bundesforschungsinstitut für Tiergesundheit
Federal Research Institute for Animal Health

u b
UNIVERSITÄT BERN

Philipps Universität Marburg

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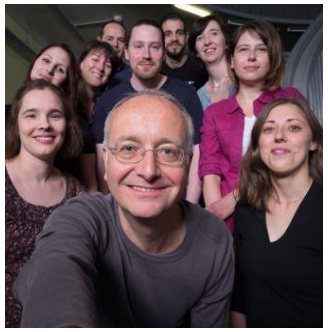

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RNA BIOINFORMATICS & HIGH-THROUGHPUT ANALYSIS

JUSTUS-LIEBIG-UNIVERSITÄT GIESSEN

FRIEDRICH-LOEFFLER-INSTITUT
FLI
Bundesforschungsinstitut für Tiergesundheit
Federal Research Institute for Animal Health

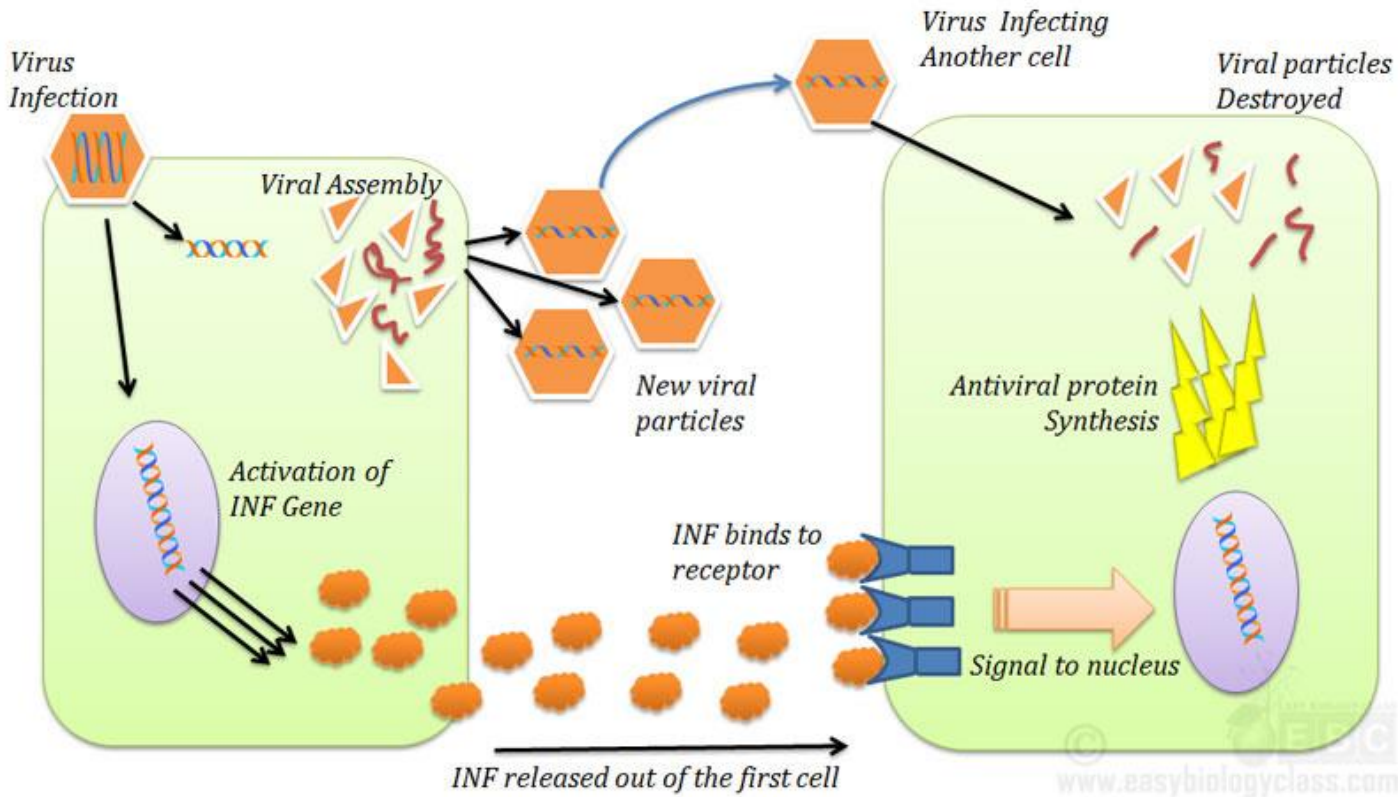
u b
UNIVERSITÄT BERN

Philipps Universität Marburg

nextflow

```
nextflow run hoelzer-lab/rnaseq
--reads input.csv
--dge input.dge_comparison.csv
--species eco
--cores 10
--profile slurm
```

ANTIVIRAL ACTION OF INTERFERON (INF)



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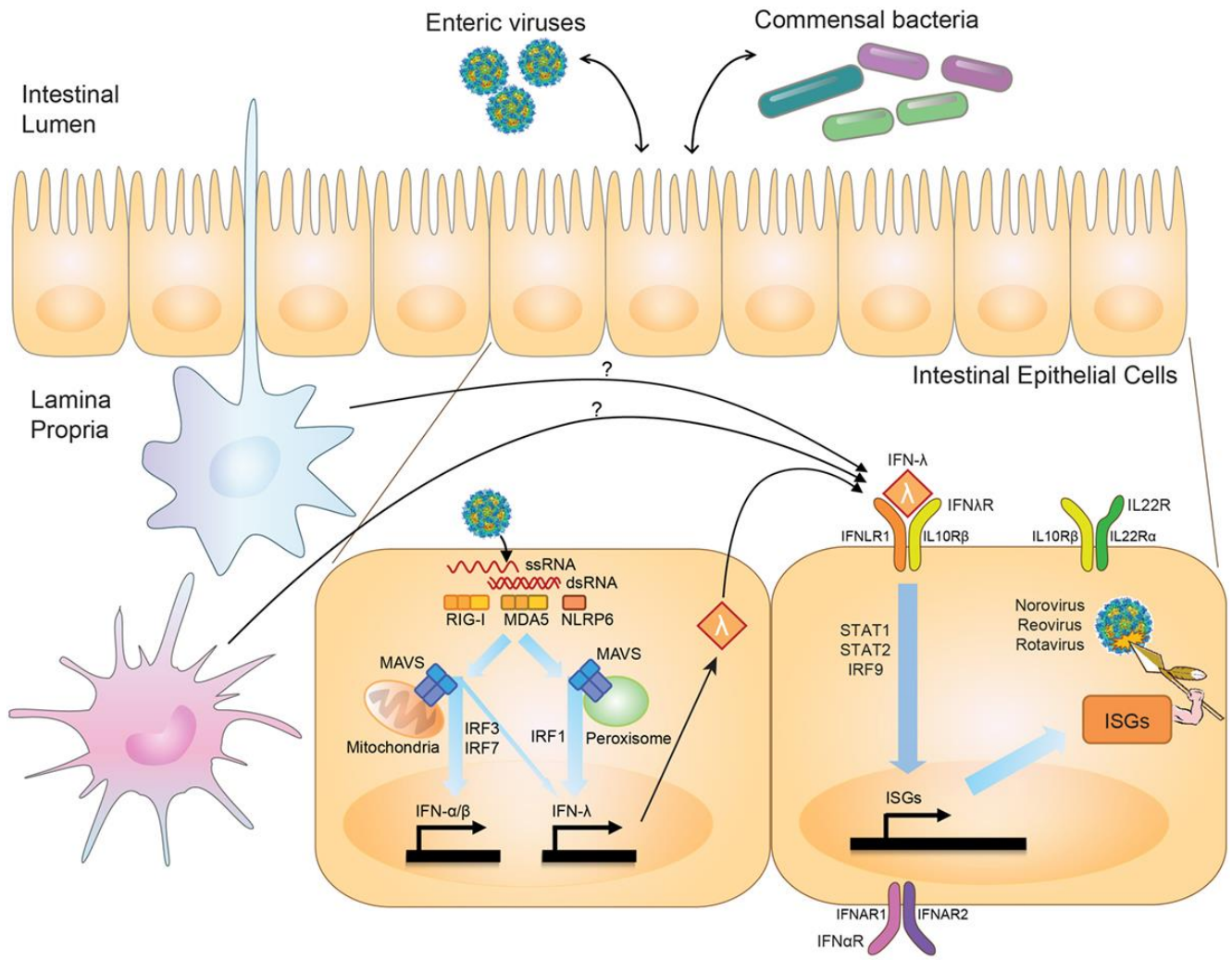

Enteric viruses
Commensal bacteria
Intestinal Lumen
Intestinal Epithelial Cells
Lamina Propria
?
?
IFN-λ
IFNλR
IFNLR1
IL10Rβ
IL22R
IL10Rβ
IL22Rα
ssRNA
dsRNA
RIG-I
MDA5
NLRP6
MAVS
MAVS
Mitochondria
IRF3
IRF7
IRF1
Peroxisome
IFN-α/β
IFN-λ
STAT1
STAT2
IRF9
Norovirus
Reovirus
Rotavirus
ISGs
ISGs
IFNAR1
IFNAR2
IFNαR

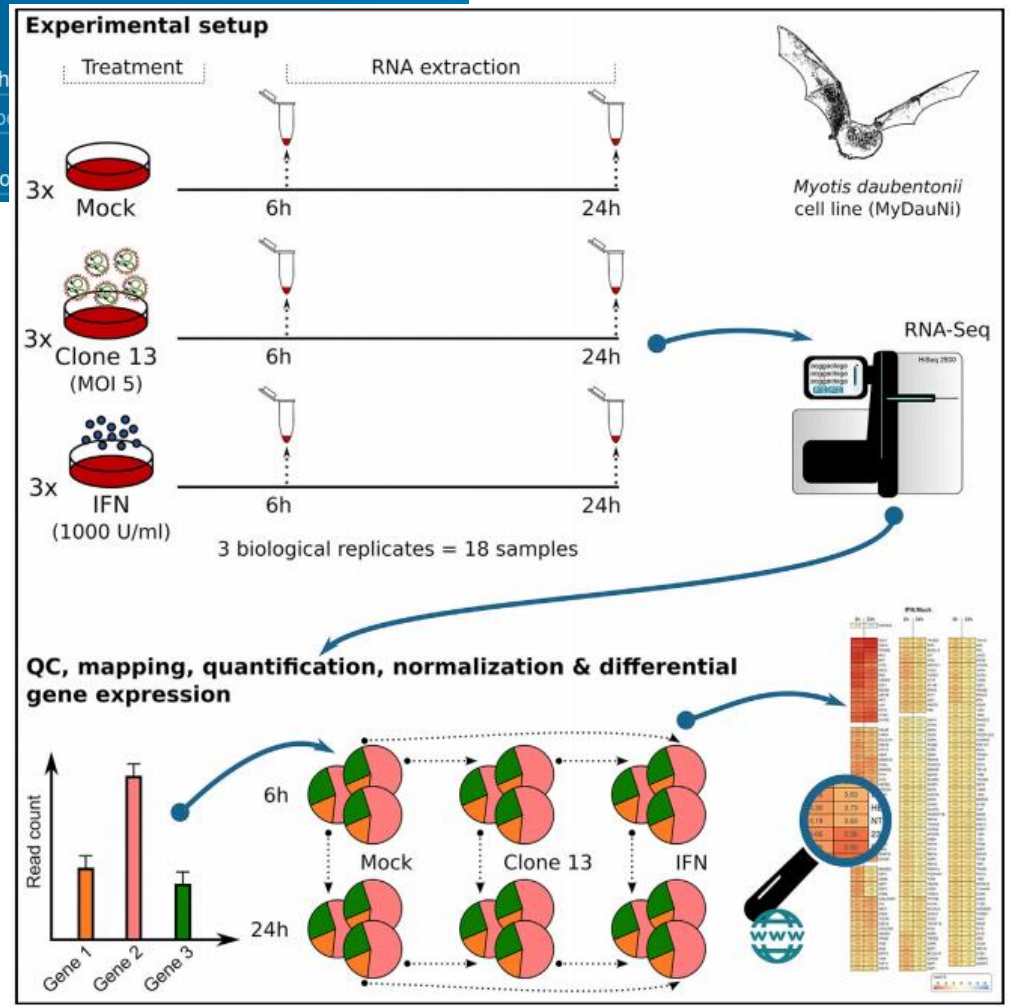
iScience

ARTICLE | VOLUME 19, P647-661, SEPTEMBER 27, 2019

Virus- and Interferon Alpha-Induced Transcriptomes of Cells from the Microbat *Myotis daubentonii*

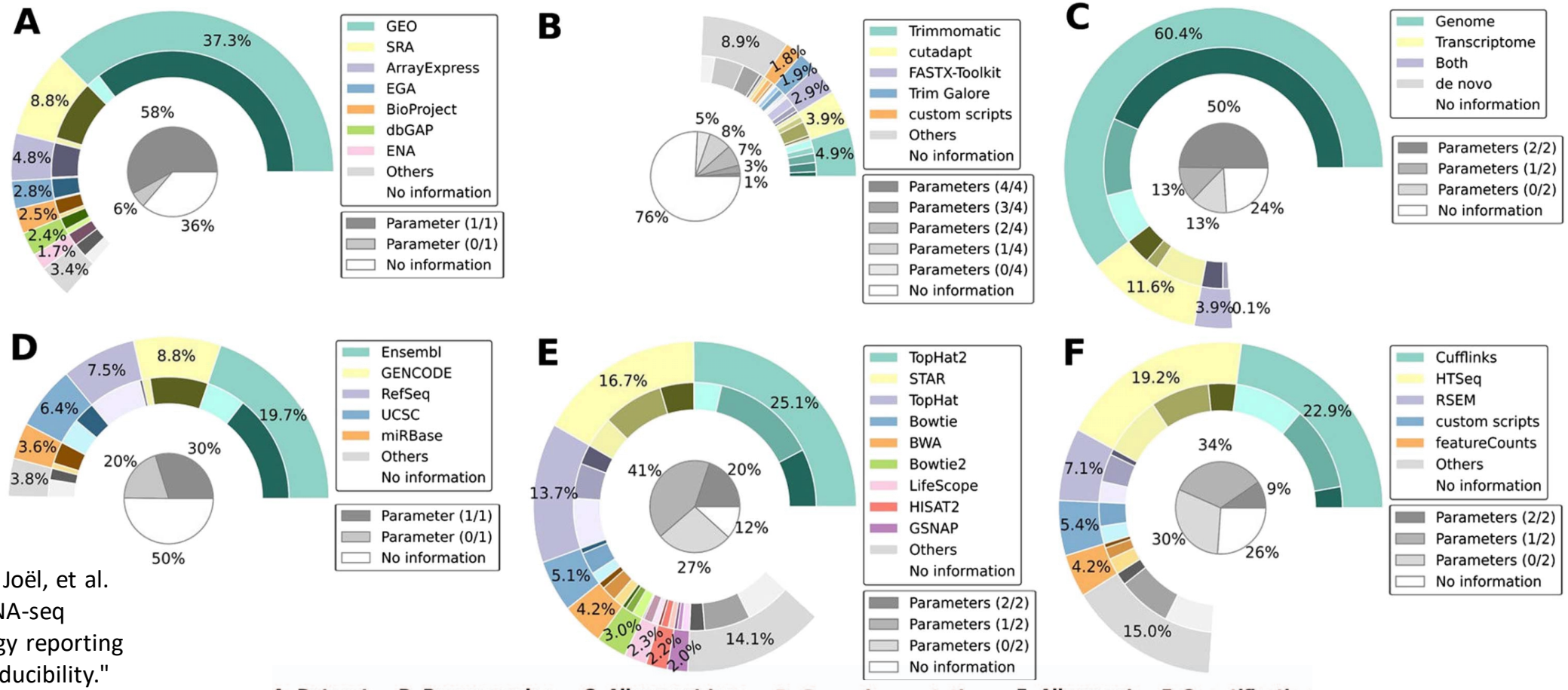
Martin Hölzer [8] • Andreas Schoen [8] • Julia Wulle • ... Ch
Friedemann Weber [9] • Show all authors • Show fo

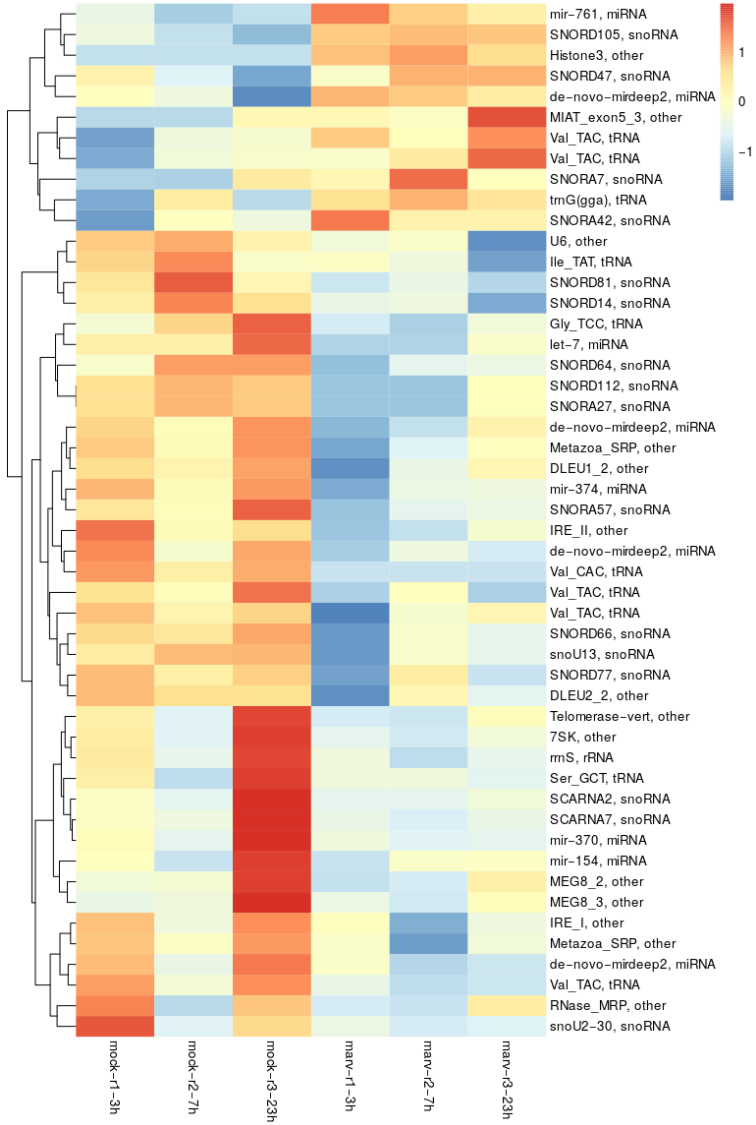
Open Access • Published: August 09, 2019 • DOI: https://do



6 h , 24 h
rRNA-
3x

Why use a workflow management system?



Simoneau, Joël, et al. "Current RNA-seq methodology reporting limits reproducibility." *Briefings in Bioinformatics* (2019)

nextflow DGE analysis pipeline - Output