Sarek

Lab Meeting

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Karolinska Institutet

Barntumörbanken

SciLifeLab

GANATIONALCTAC ATCAGENOMICSGT INFRASTRUCTURE

NBIS

What is Sarek?

Sarek

http://sarek.scilifelab.se/

- Analysis germline and somatic workflow

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NATIONAL GENOMICS INFRASTRUCTURE

NBIS

What is Sarek?

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- Analysis germline and somatic workflow
- Whole genome or targeted sequencing
- Developed with NGI and NBIS
- Support from The Swedish Childhood Tumor Biobank

Powered by

https://www.nextflow.io/

https://www.sylabs.io/singularity/

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Data-driven workflow language

https://www.sylabs.io/singularity/

HPC specific container engine

Conda

BIOCONDA®

https://bioconda.github.io/

- Virtual environment management system

Sarek exists in multiple flavors

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Sarek

Sarek
Exome

Sarek exists in multiple flavors

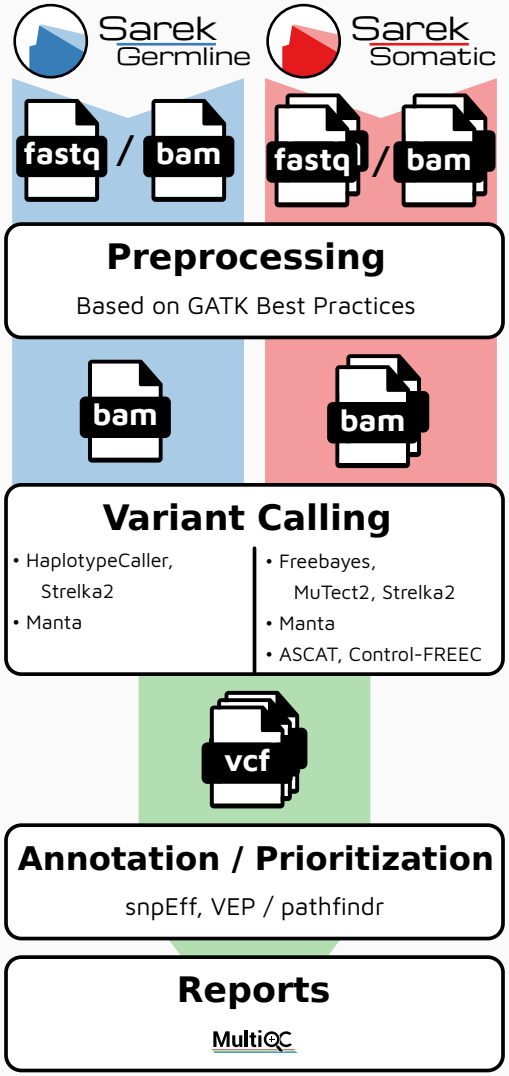
Sarek

Sarek Exome

Sarek Germline

Sarek Somatic

Data and files workflow



AWS iGenomes

https://ewels.github.io/AWS-iGenomes/

- Human `GRCh37` from the GATK Resource Bundle
- Human `GRCh38` from the GATK Resource Bundle

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- Dog `CanFam3.1`
- Mouse `GRCm38`

Preprocessing

https://software.broadinstitute.org/gatk/best-practices/

Based on GATK Best Practices (GATK 4.0)

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- Reads mapped to reference genome with `bwa mem`
  - FASTQs or BAMs

Preprocessing

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- Reads mapped to reference genome with `bwa mem`
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- Duplicates marked with `picard MarkDuplicates`

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Based on GATK Best Practices (GATK 4.0)

- Reads mapped to reference genome with `bwa mem`
  - FASTQs or BAMs
- Duplicates marked with `picard MarkDuplicates`
- Recalibrate with `GATK BaseRecalibrator`

Variant Calling

- SNVs and small indels:

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  - Freebayes
  - HaplotypeCaller
  - MuTect2
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  - ASCAT
  - Control-FREEC

Annotation

- VEP and SnpEff
- 🗄 ClinVar, COSMIC, dbSNP, GENCODE, gnomAD, polyphen, sift, etc.

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- Possibility to use cache directories

Prioritization

What we need

- Adapt settings where necessary, and ensure they give good results

Markus Mayrhofer

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- Coherent overview to allow critical assessment of sequence and variant quality

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What we need

- Adapt settings where necessary, and ensure they give good results
- Coherent overview to allow critical assessment of sequence and variant quality
- Tables of variants with probable relevance for the disease

Markus Mayrhofer

Prioritization with pathfindr

Our solution

- Parse all results into R environment

Prioritization with pathfindr

Our solution

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- Rank variants based on evidence for being a driver mutation

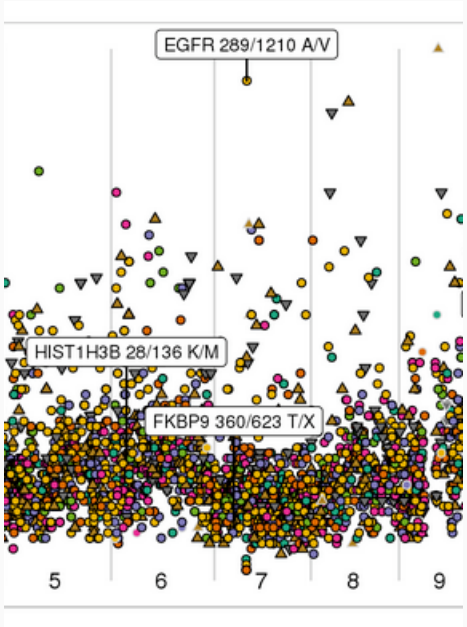
Prioritization with pathfindr

Our solution

- Parse all results into R environment
- Rank variants based on evidence for being a driver mutation
- Visualize in portable html report for easy browsing

Tables and visualization

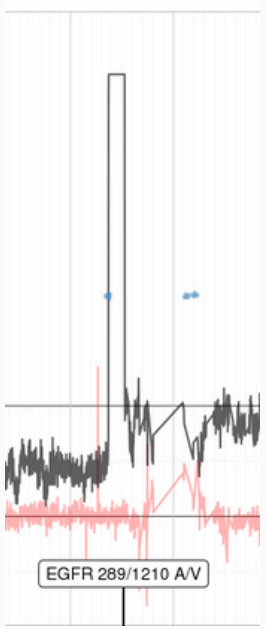
Gene	Mutation	Rank score	Rank Terms	Allele_ratio
EGFR	289/1210 A/V	10	T1_gene moderate_impact clinvar polyphen/SIFT hotspot cosmic_>50	0.94
TP53		9	T1_gene high_impact high+TSG clinvar cosmic_>50	0.86
HIST1H3B	28/136 K/M	7	T1_gene	0.23



SNVs, Indels

Tables and visualization

Gene	Mutation	Rank score	Rank Terms
EGFR	gain	5	T1_gene focal high_amp
ETNK1	gain	5	T1_gene focal high_amp
KRAS	gain	5	T1_gene focal high_amp



Copy number

Sarek at work

- 50 tumor/normal pairs with GRCh37 reference

Sarek at work

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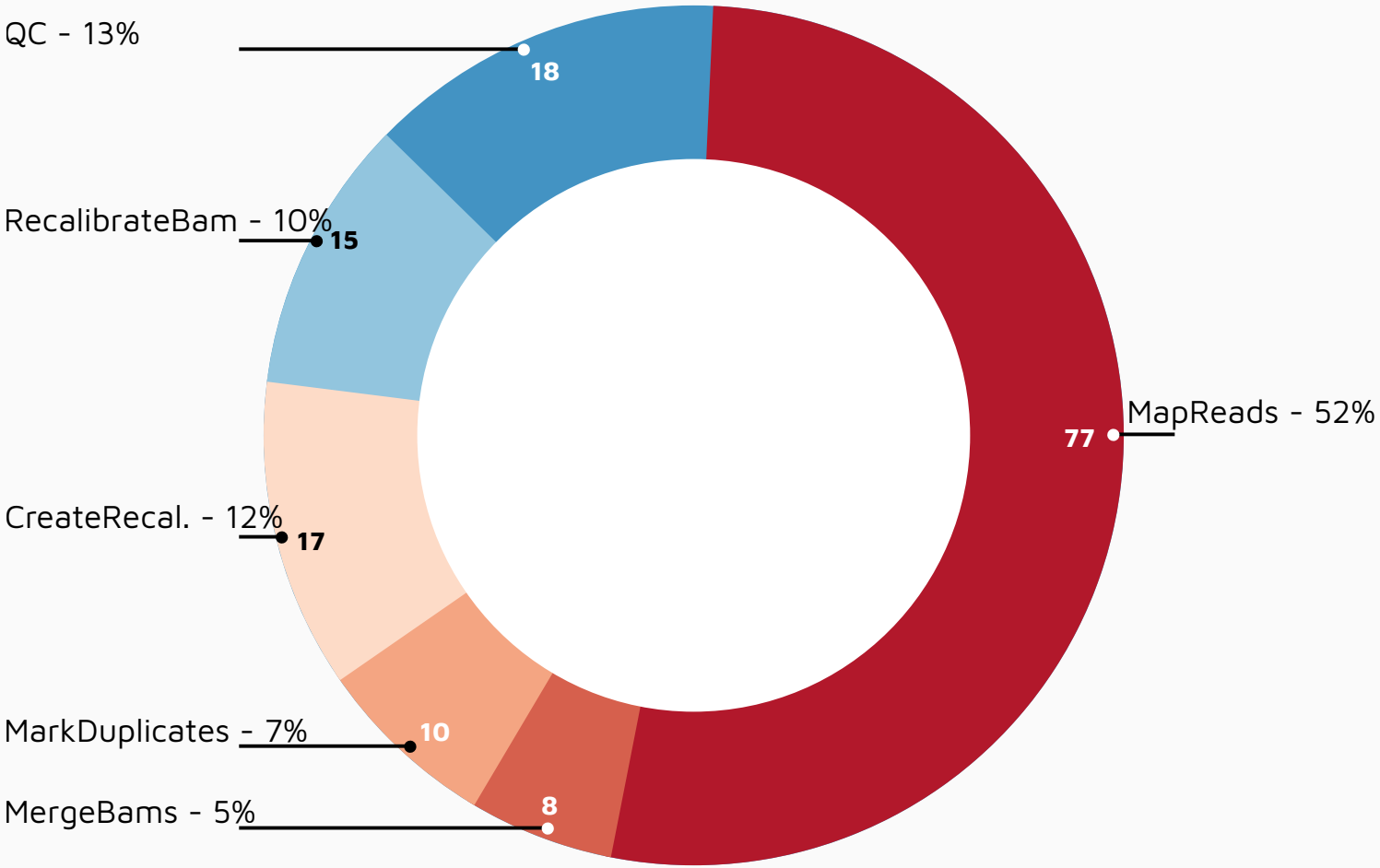
- The whole SweGen dataset with GRCh38 reference
  - 1 000 samples in germline settings
- Clinical samples with Genomic Medicine Sweden initiative
- Used at NGI
  - 200 samples
  - testing it in production
  - plans for validation

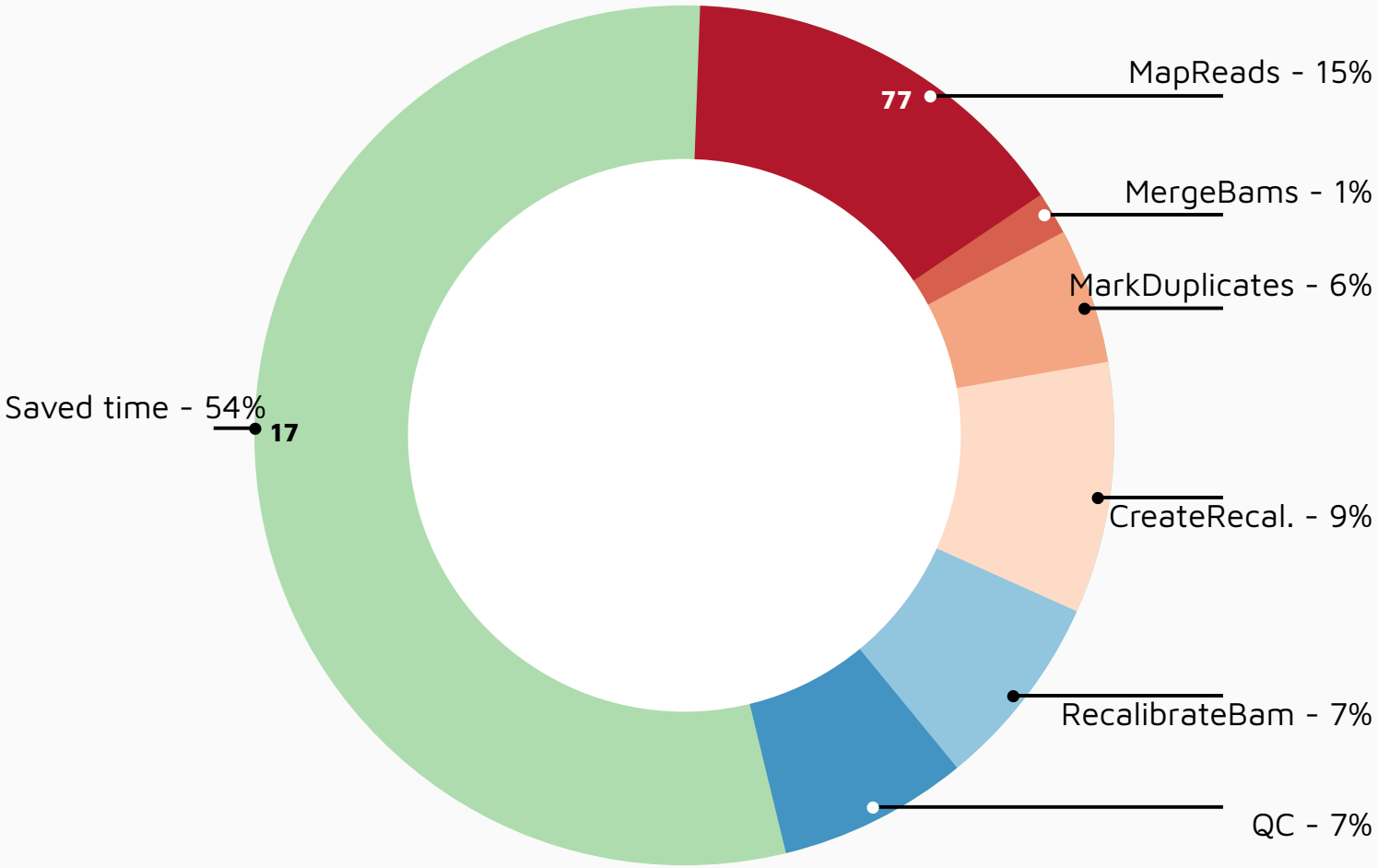
Preprocessing time



On a research settings

On UPPMAX secure cluster Bianca

Preprocessing time



On a clinical settings

On our own secure server munin

AWS

https://aws.amazon.com/

Johannes Alneberg

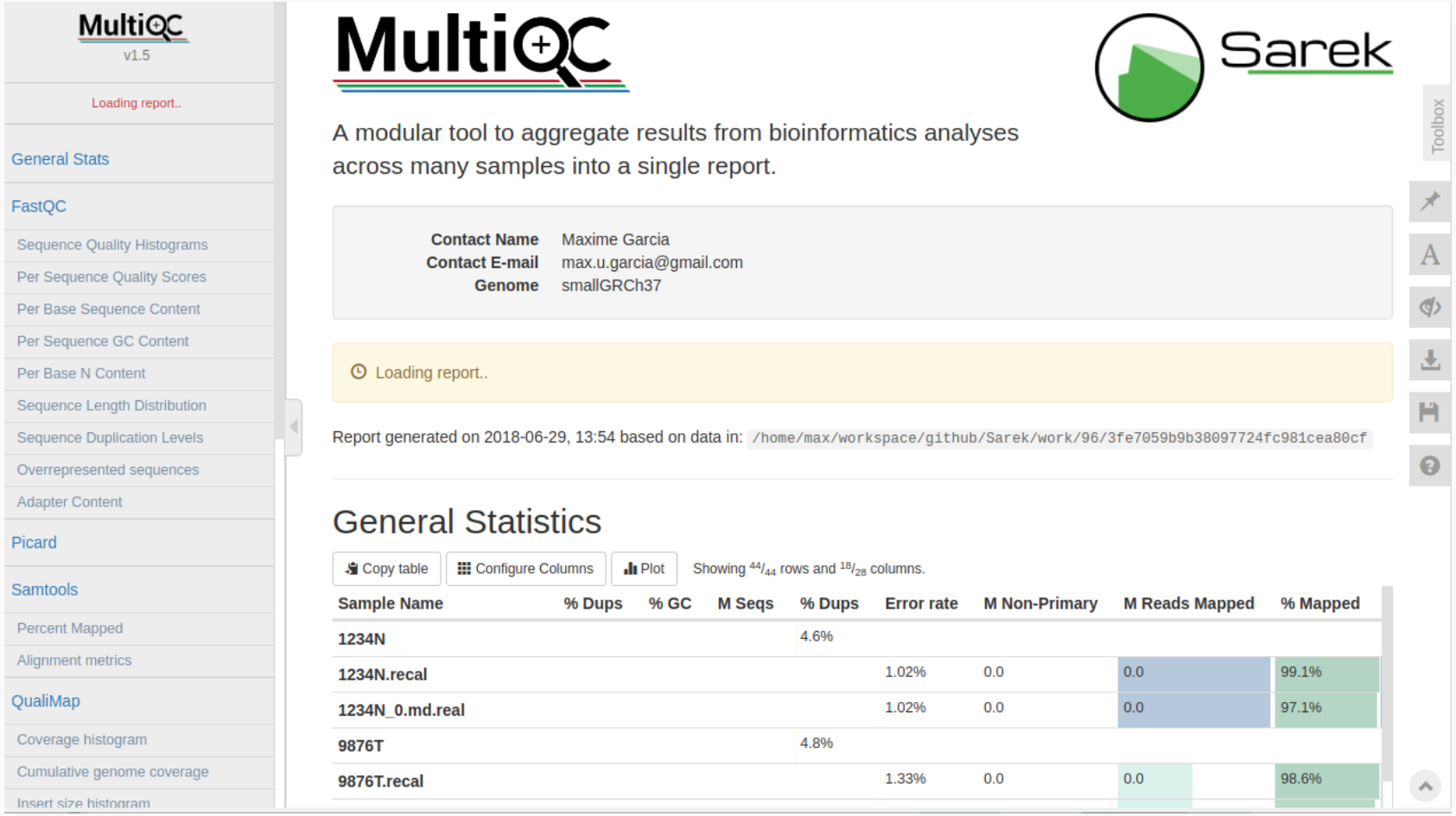
AWS

https://aws.amazon.com/

- Improved AWS usage

Johannes Alneberg

QC reports



http://multiqc.info/

munin

The CMM server room

munin

munin

Acknowledgments

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Any questions?

https://github.com/SciLifeLab/Sarek/

https://gitter.im/SciLifeLab/Sarek/

http://sarek.scilifelab.se/