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Robust quality control of Next Generation Sequencing alignment data

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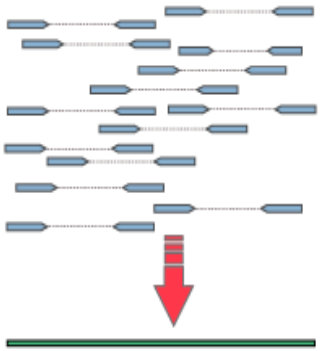
Next Generation Sequencing: amazing discovery tool

High speed & and deep coverage



Technologies:

- Whole genome or exome
- **Whole transcriptome (RNA-seq)**
- Histone modifications (ChIP-seq)
- Much more …

Various applications

Fast developing

But…

… there are some things to take into account

-Small read size vs repetetive genome

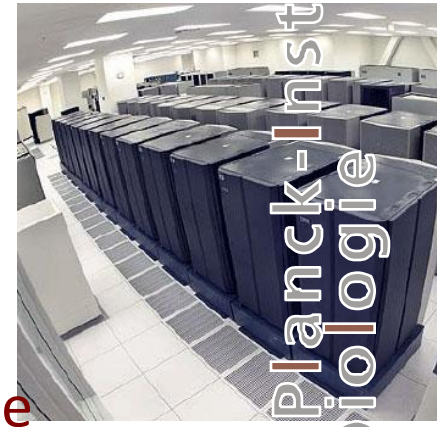
- High price (even now up to 3K $)

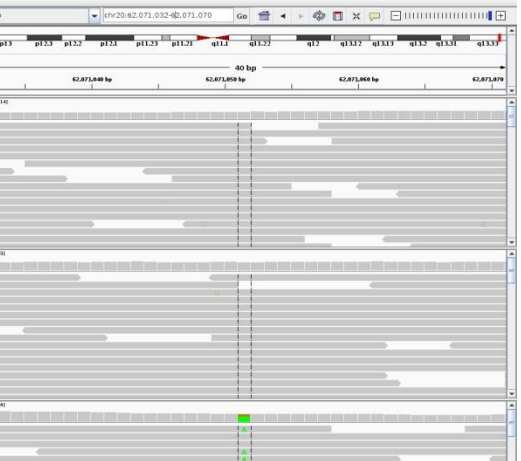
-Computational requirements:

  - Sophisticated bioinformatics analysis
  - Storage and computational performance

- **Platform specific and protocol errors**

  - Optical duplicates, PCR artifacts…
  - -Algorithm induced biases





Quality Control of NGS data

The systematic detection of the biases is **crucial ->** saves **time** and **$money**

Some packages exist:

- **FastQC**
- **Samtools**
- **Picard tools**
- **RNA-seq QC**

However there is room for improvement:
more comprehensive and user-friendly tool could be useful.

Quality Control of NGS data

Our solution:

QualiMap

A Java application, which allows computing statistics and presenting different graphs for the evaluation of NGS alignment data.

Provides both GUI and command line interfaces

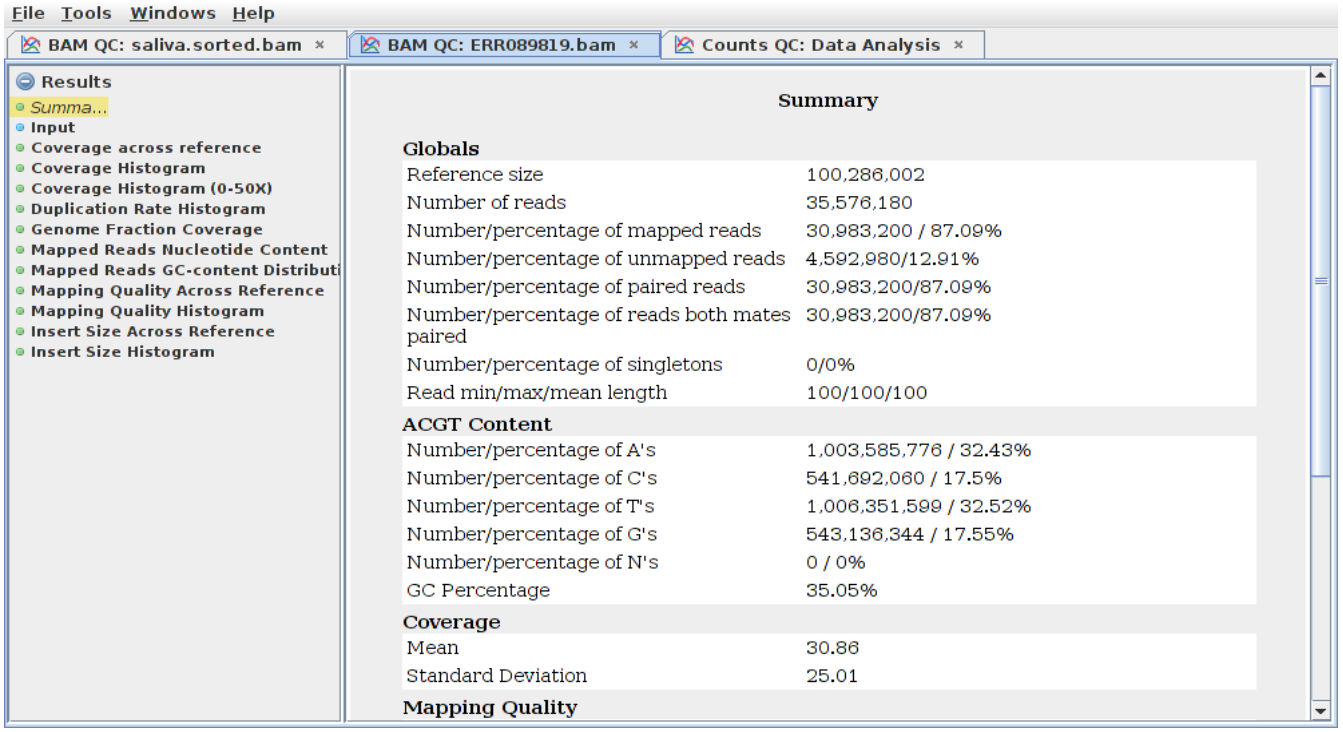
Qualimap features

- Supported types of experiments: WG-seq, RNA-seq, exome seq, methylation studies, …
- 3 modes of analysis: **BAM QC**, Counts QC, RNA-seq QC
- Analysis possible for whole alignment or for arbitrary regions
- Input:
  - BAM/SAM alignment
  - GFF/GTF/BED annotations
- Output:
  - Interactive visualization
  - PDF or HTML report

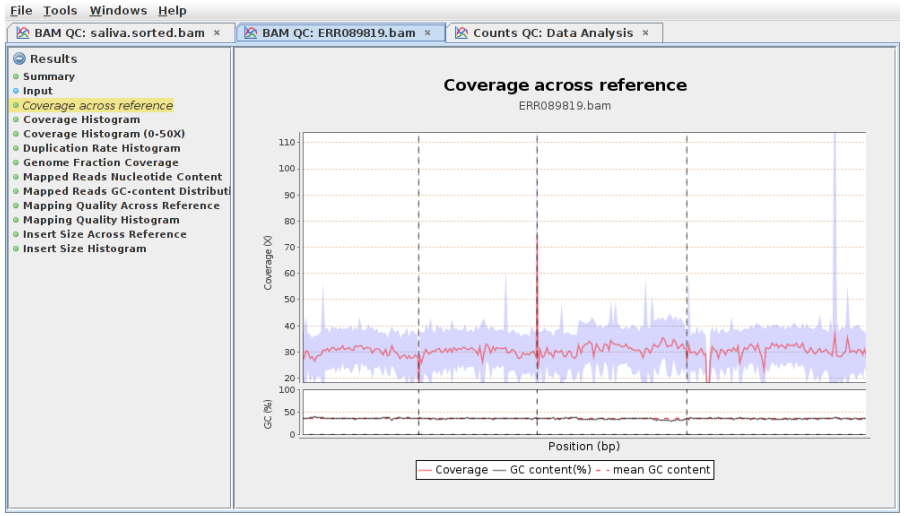
Qualimap features: BAM QC

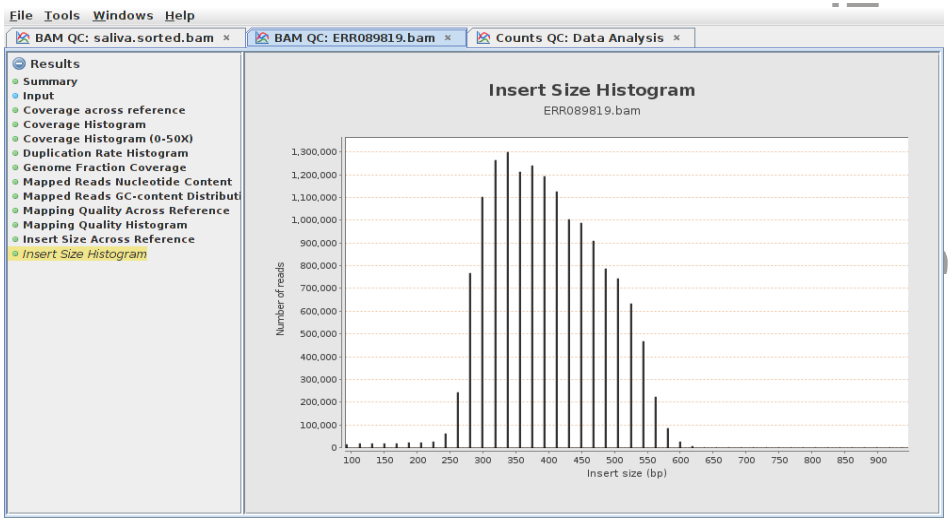
Summary:

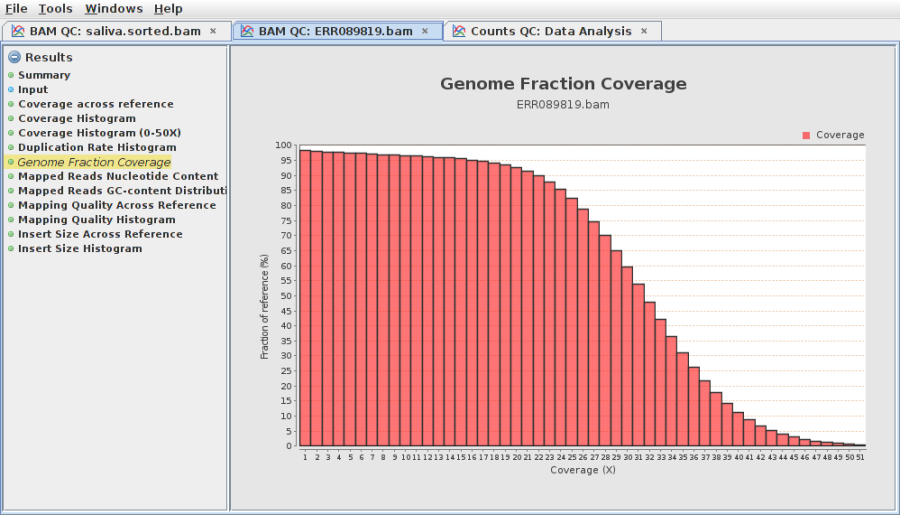
- Global data (reference size, number of reads)
- Coverage (mapped, paired, per chromosome)
- Reads info (insert size, quality, homopolymers, duplication rate)

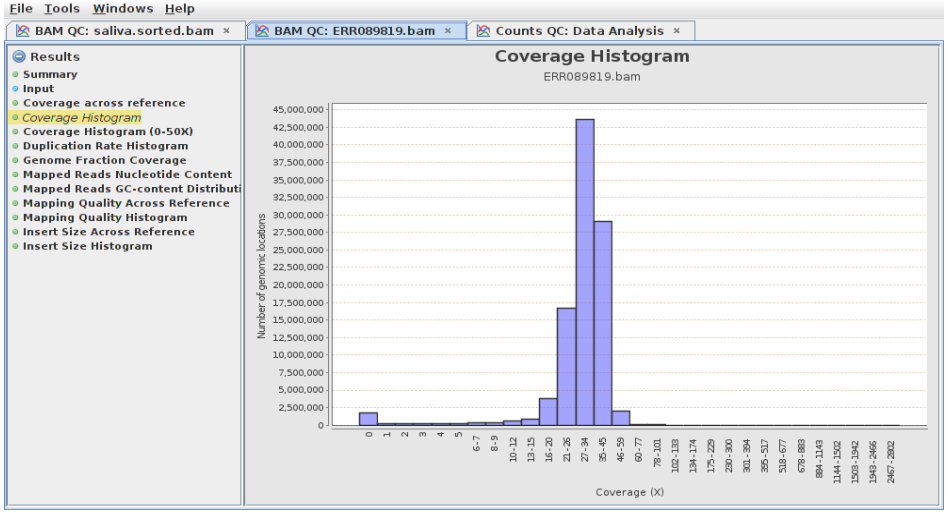


Qualimap features: BAM QC









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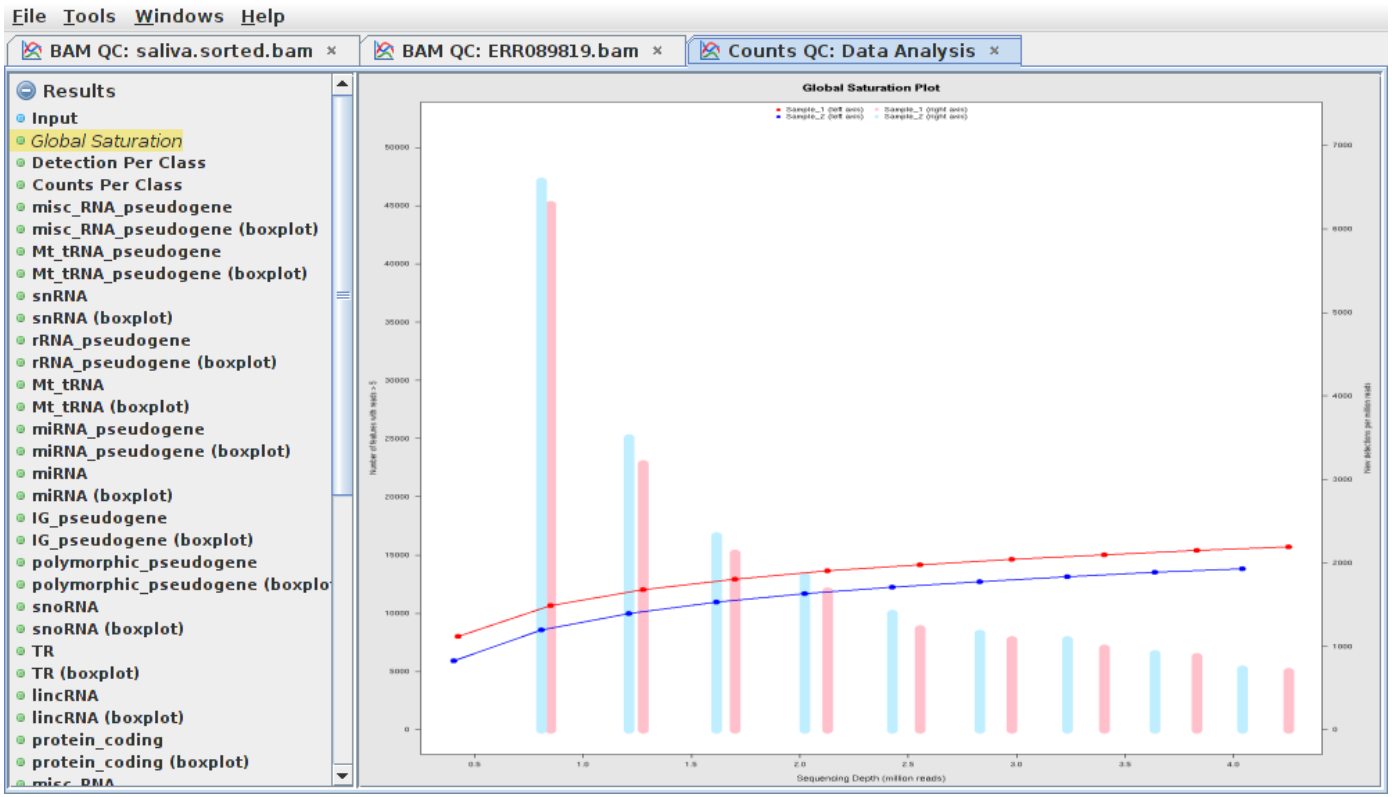
Qualimap features: Counts QC

- 2 samples comparison
- Sequencing saturation
- Feature by biotype classification



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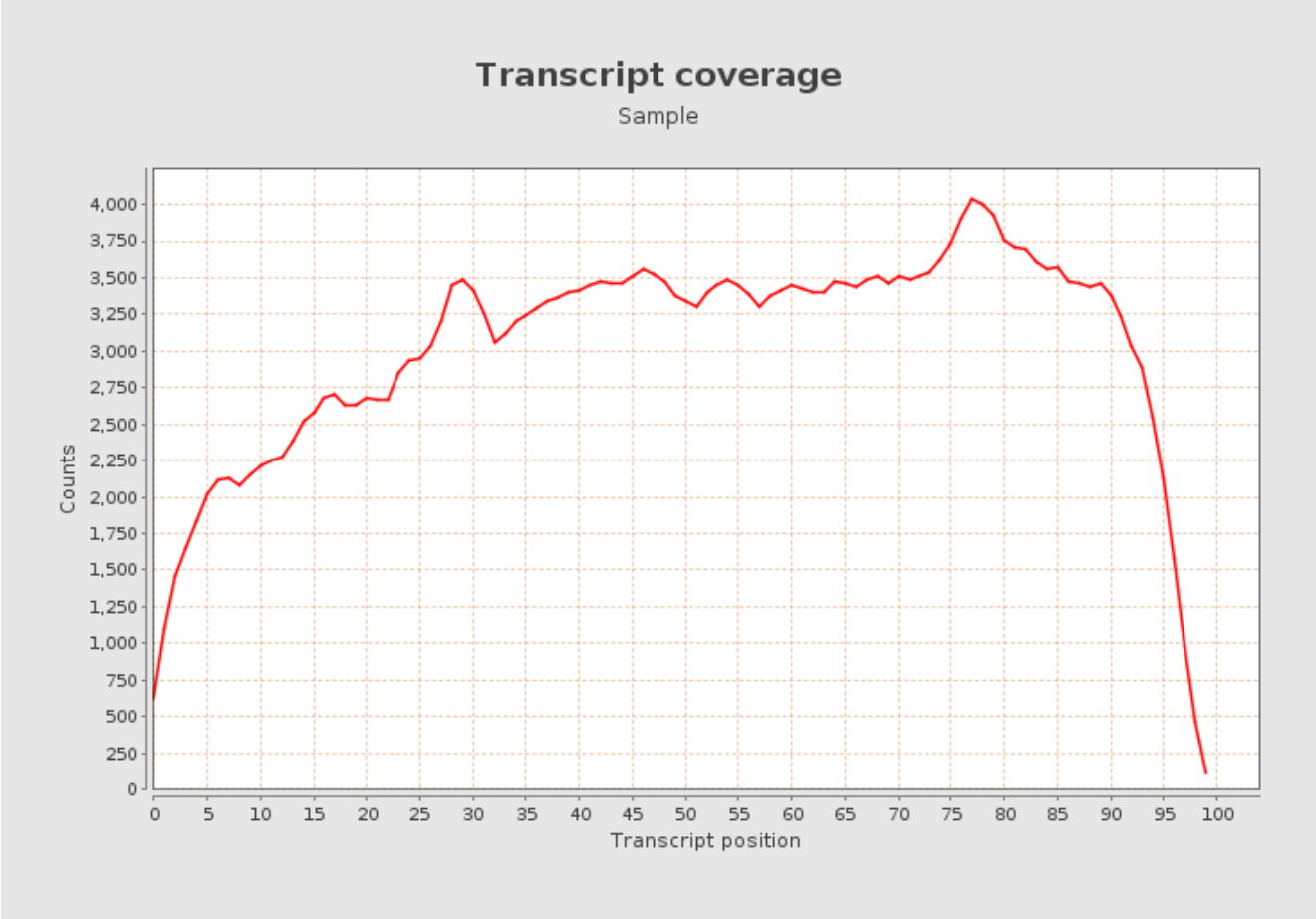
Qualimap features: RNA-seq QC

- Transcript coverage
- 5‘-3‘ bias calculation



Some more features

- Tools: counts computation, epigenetic clustering
- Command line interface: easy integration
- Performance: runs in parallel on multicore systems
- Manuscript:

  García-Alcalde F, Okonechnikov K, Carbonell J, Cruz L, Götz S, Tarazona S, Dopazo J, Meyer T, Conesa A. "Qualimap: evaluating next-generation sequencing alignment data." Bioinformatics 28, no. 20 (2012): 2678-2679.

Further development

- New features are suggested by users
- Discussion forum: google-groups
- Source code on bitbucket
- Early builds are available as snapshots
- There is a *Galaxy* wrapper developed by **Joachim Jacob** available from the Galaxy Tool Shed

Thank you for attention!
Please provide your questions.

Useful links:

- Web-site: **http://qualimap.bioinfo.cipf.es/**
- Bitbucket: *https://bitbucket.org/kokonech/qualimap*
- Google-groups: *http://groups.google.com/group/qualimap*
- Galaxy repo: *http://toolshed.g2.bx.psu.edu/repos/joachim-jacob/qualimap_suite*

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