MORU Tropical Health Network

wellcome

UNIVERSITY OF OXFORD

Sequence QC

Sequence QC

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- Basic checks **before** you run your analysis will save time later!
- Common issues to look for:
  - Poor quality reads
  - Adapter contamination
  - GC bias

- https://sequencing.qcfail.com/

FastQC

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- Performs basic quality checks fast
- Gives easily readable output
- Good for picking up problems but don't get too worried by the output!

Sequence problems

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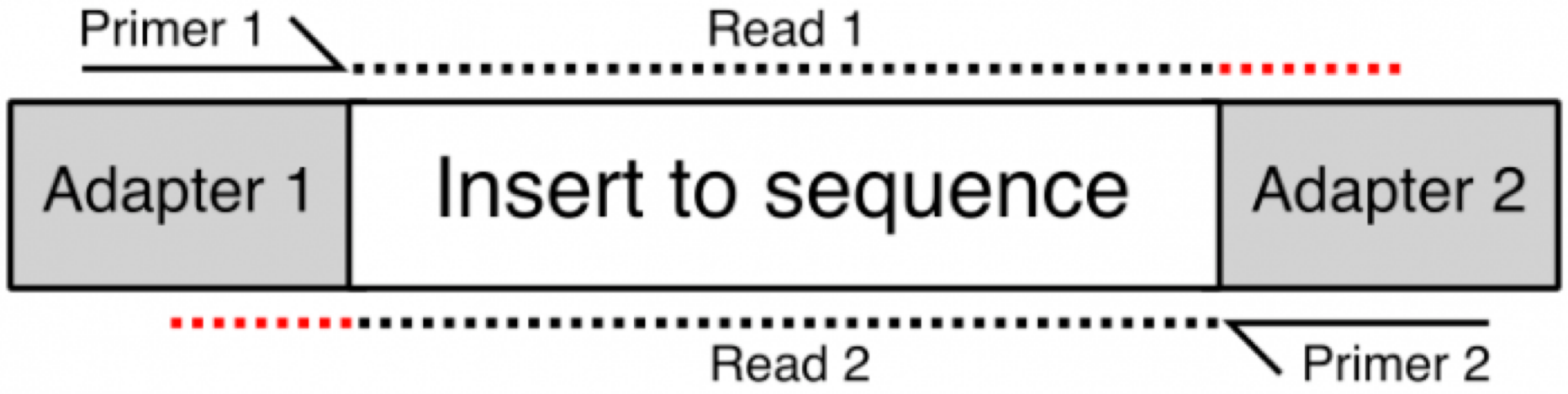
- Adaptor read-through
- When reads are longer than the insert size
- MiSeq will trim this automatically



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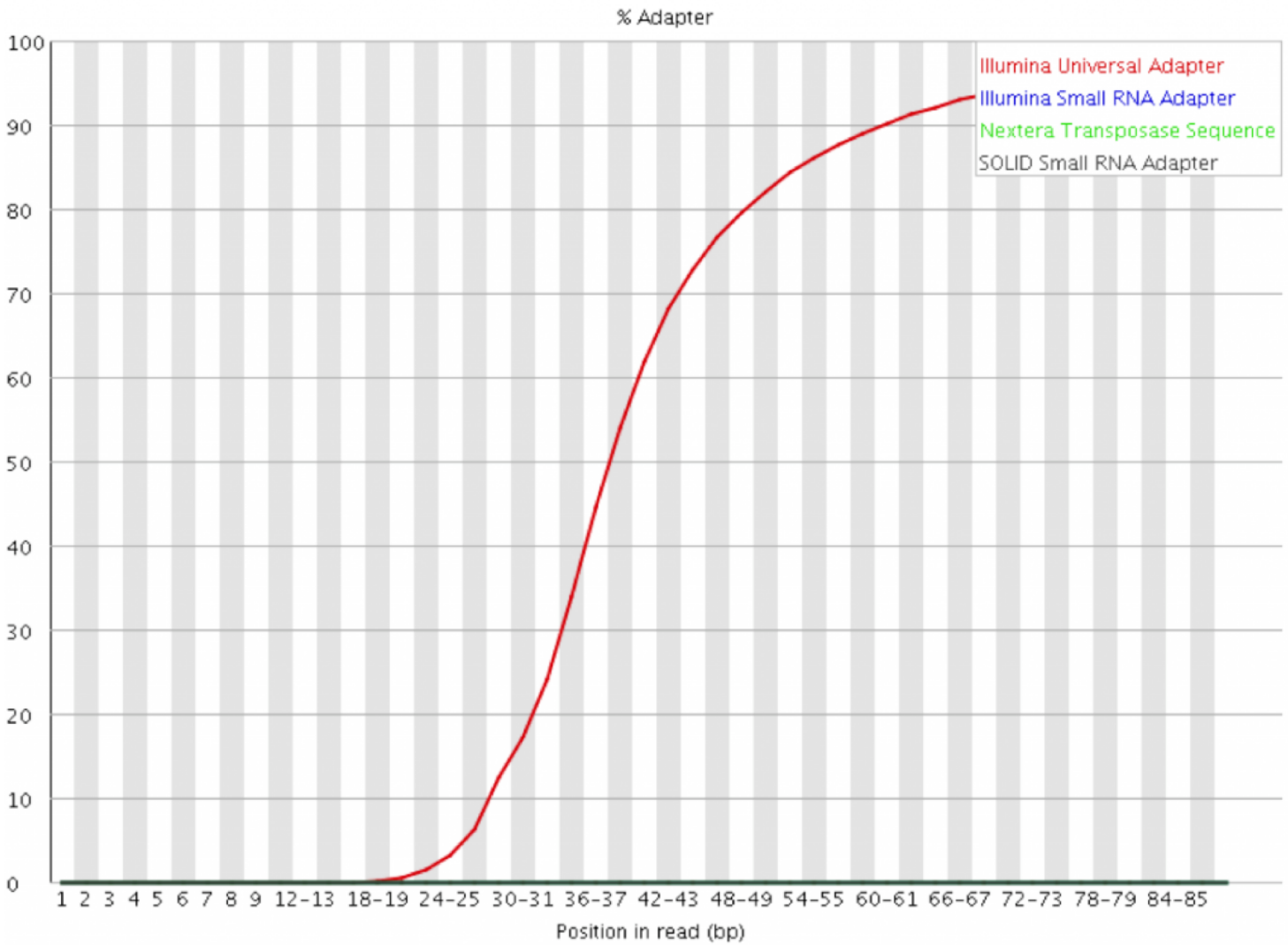
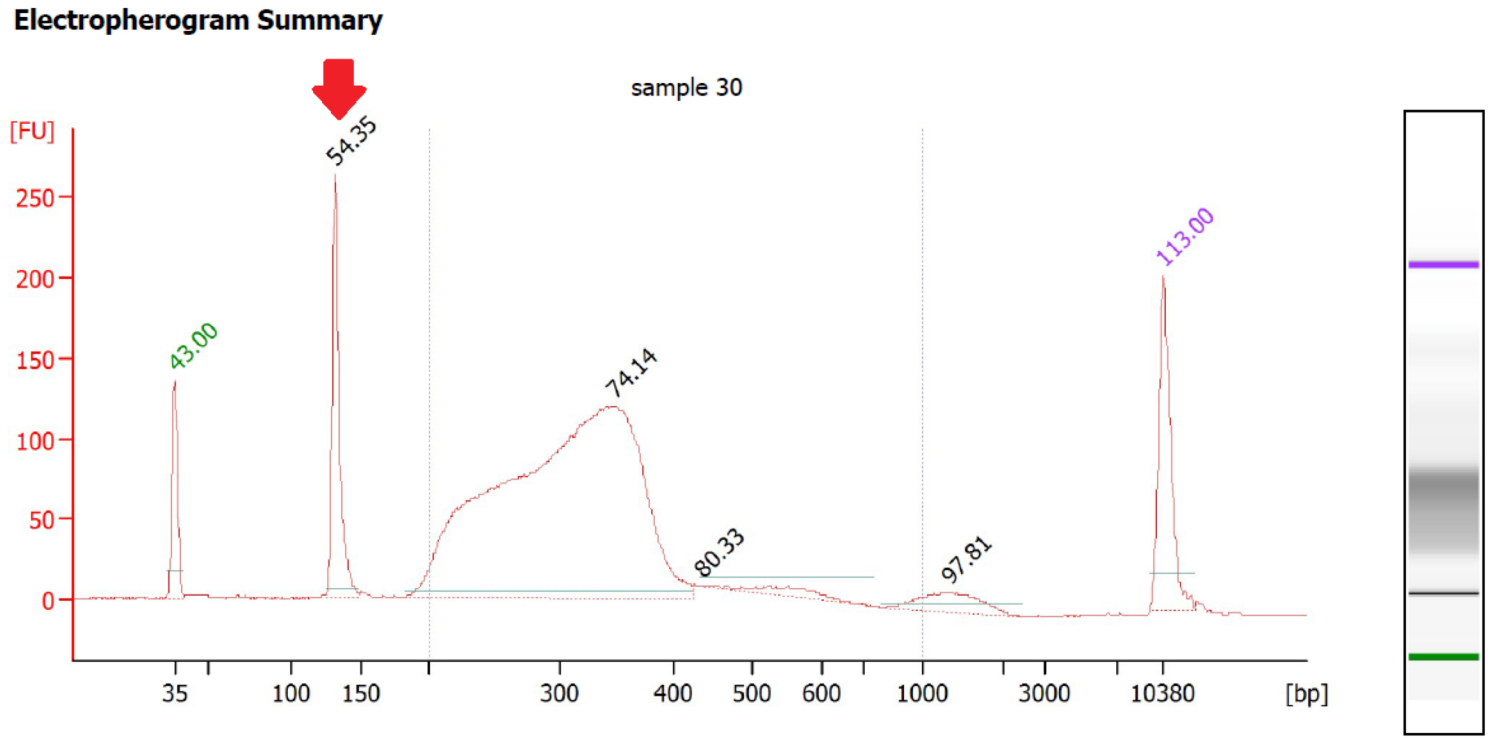


Sequence problems

- Adaptor dimer
- When you ligate adapters together with no input sequence!



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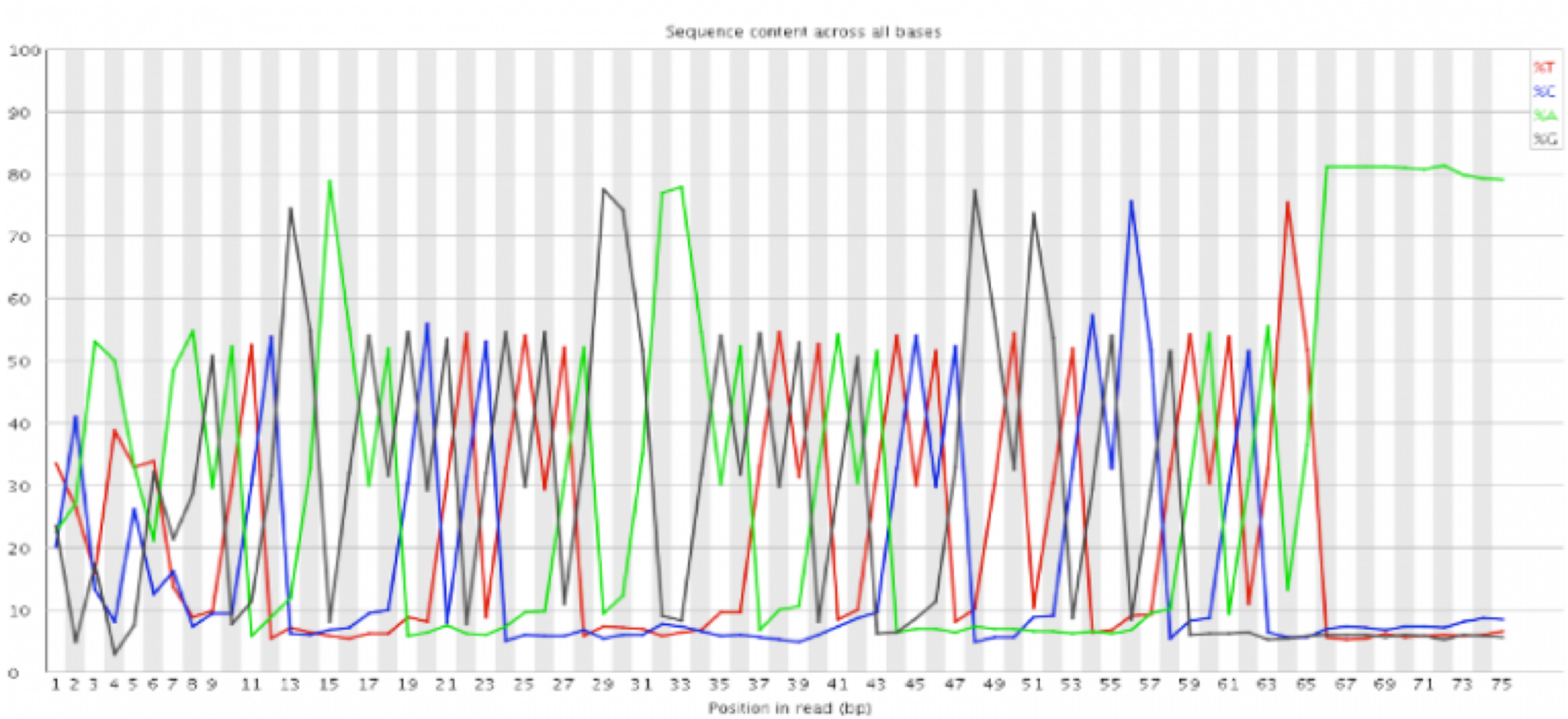

Sequence problems

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- Nextera transposase signature
- Due to sequence preference of the Tn5 transposon
- Unavoidable in Nextera sequencing

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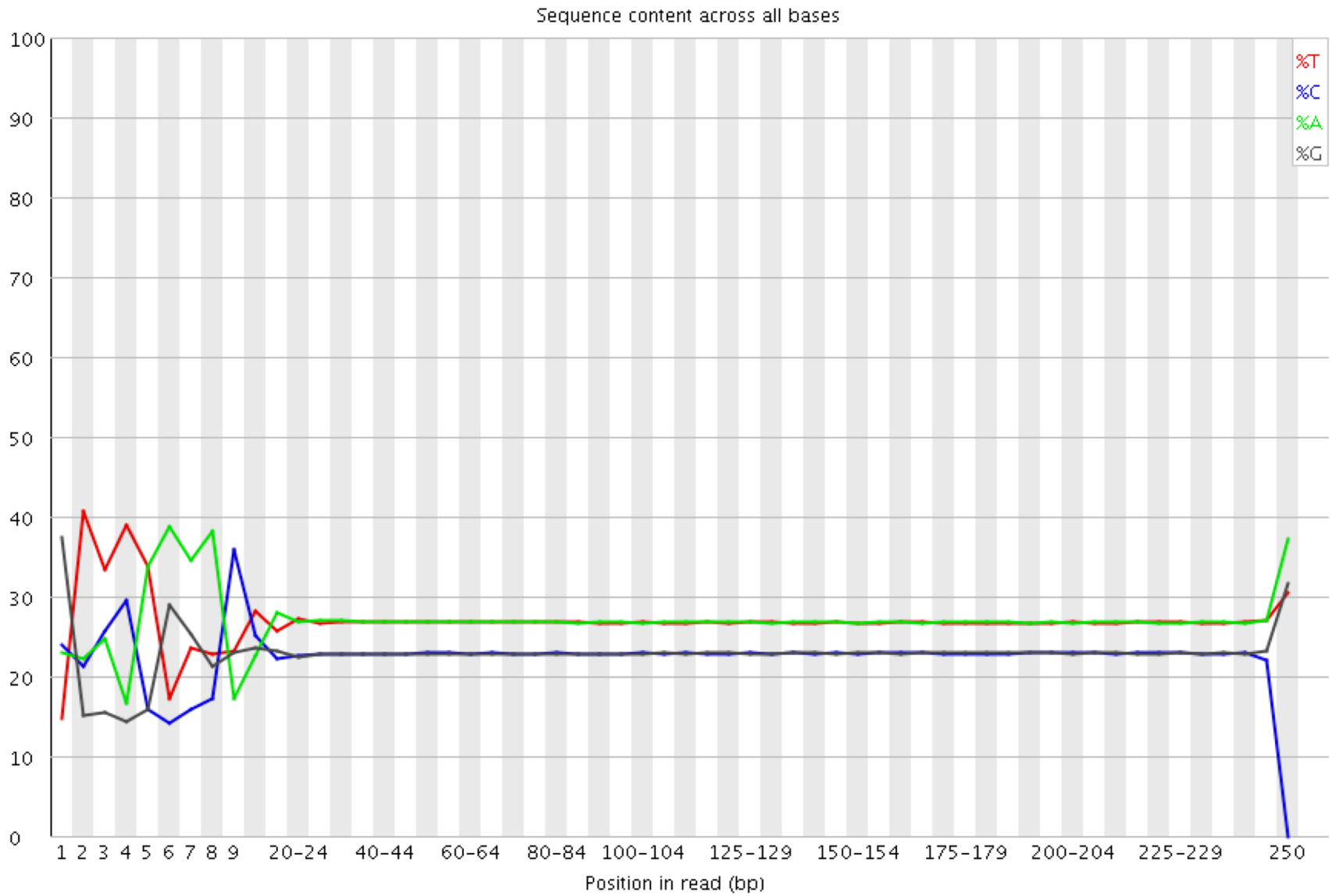

GC bias

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- Regions of high and low GC in the genome may be underrepresented in sequencing
- By looking at the number of reads mapped to the different regions of the genome, we can determine the level of GC bias

MultiQC

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- Takes the results of multiple different QC programs and produces an interactive report
- (MultiQC demo)