

Quiz 3

GENOMICS

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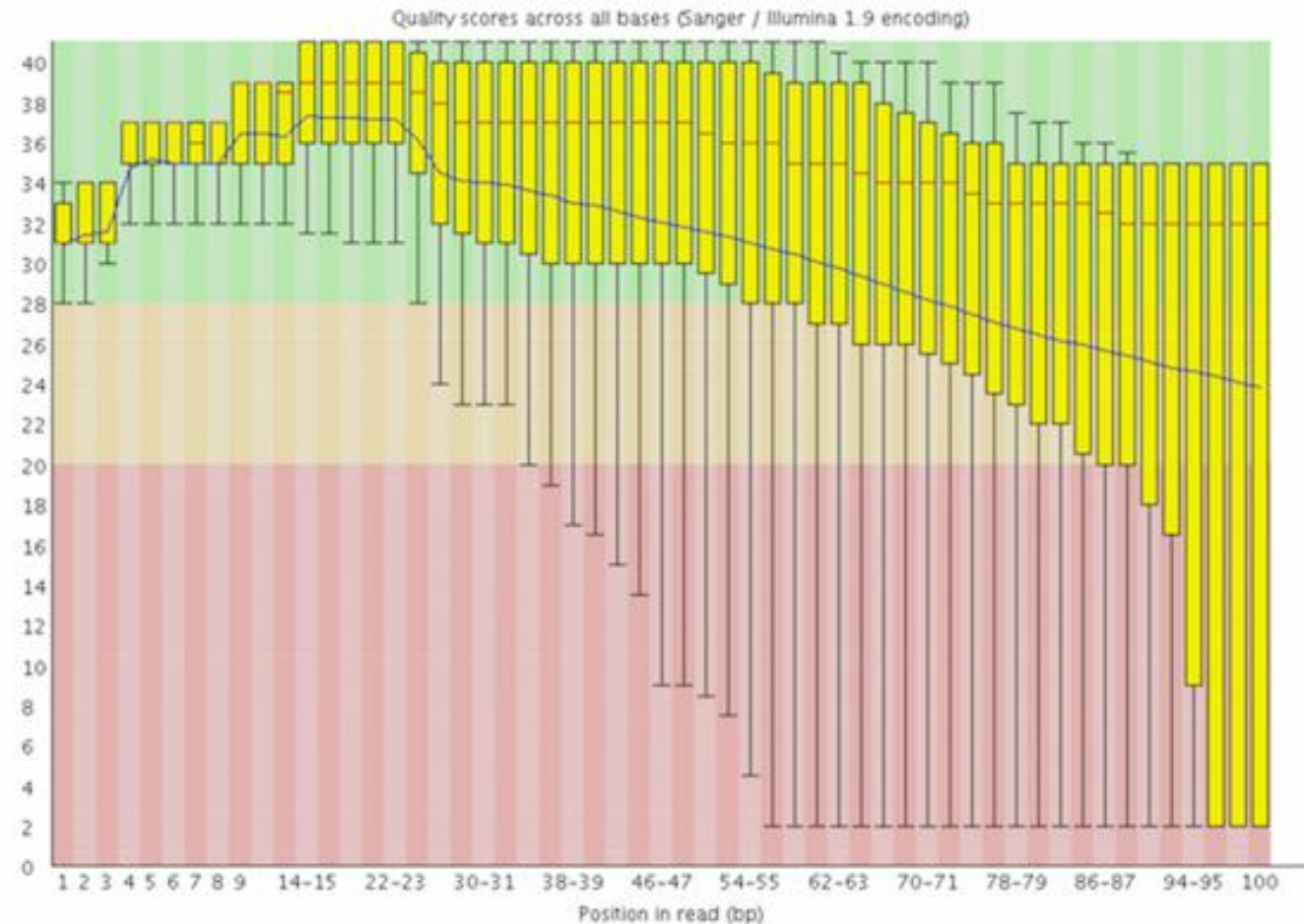
Question 1

You did your fastqc on a RNAseq illumina library, you observed this plot. What do you deduce ?

- A. You sequencing is fine, we usually have a loss of quality at the end of the reads.
- B. The experiment has failed, I have to redo it.
- C. You have some issues, at the end of the reads, may be due a signal issue at the end of the run.
- D. You have some issues, at the end of the reads, may be due phasing issues at the end of the run.



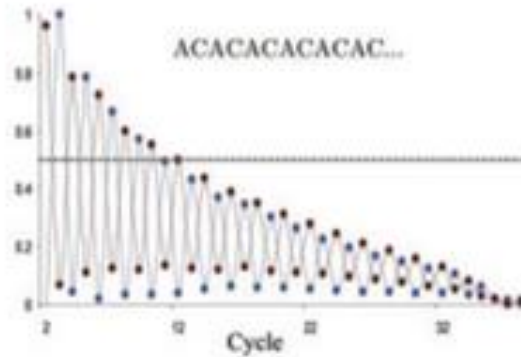
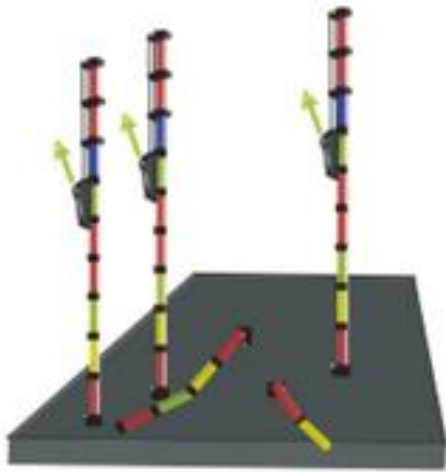
Per base sequence quality



Question 1

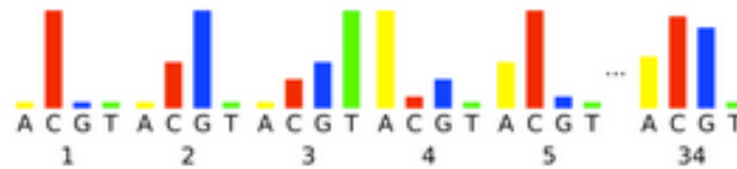
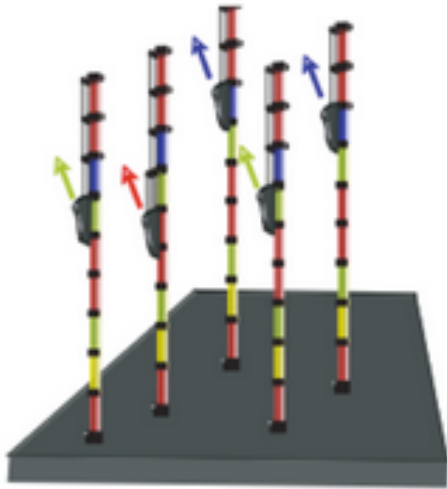
- **Signal decay:** As sequencing proceeds, the fluorescent signal intensity decays with each cycle, yielding decreasing quality scores at the **3' end** of the read. This is due to:
 1. Degrading fluorophores
 2. A proportion of the strands in the cluster not being elongated

Therefore, the proportion of signal being emitted continues to decrease with each cycle.



Question 1

- **Phasing:** As the number of cycles increases, the signal starts to blur as the cluster loses synchronicity, also yielding a decrease in quality scores at the **3' end** of the read. As the cycles progress, some strands get random failure of nucleotides to incorporate due to:
 1. Incomplete removal of the 3' terminators and fluorophores
 2. Incorporation of nucleotides without effective 3' terminators



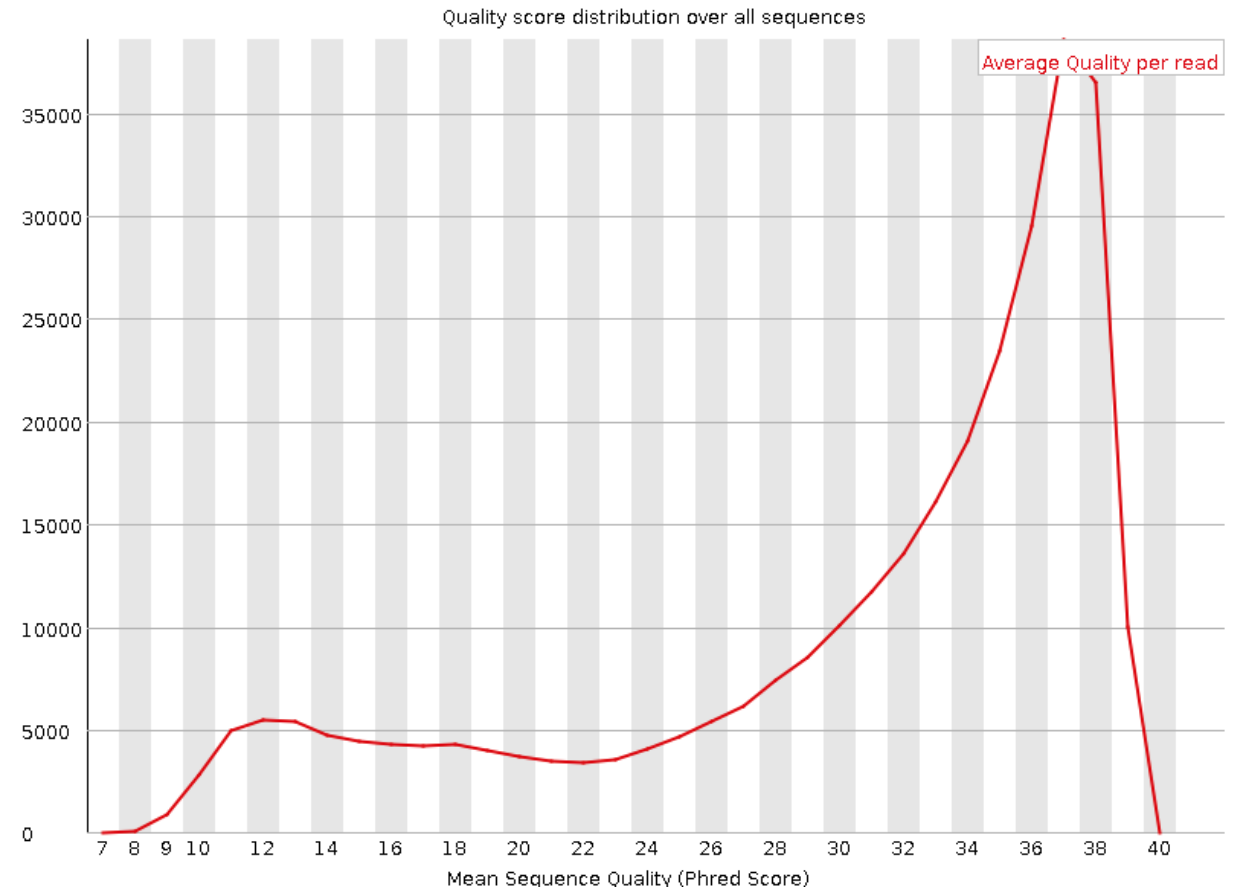
Question 2

You did your fastqc on a RNAseq illumina library, you observed this plot. What do you deduce ?

- A. Nothing, this plot is useless.
- B. The experiment has failed, I have to redo it.
- C. **We have a small portion of low-quality reads that we should removed before processing.**
- D. **We have a small portion of low-quality reads that we can keep before processing.**



Per sequence quality scores

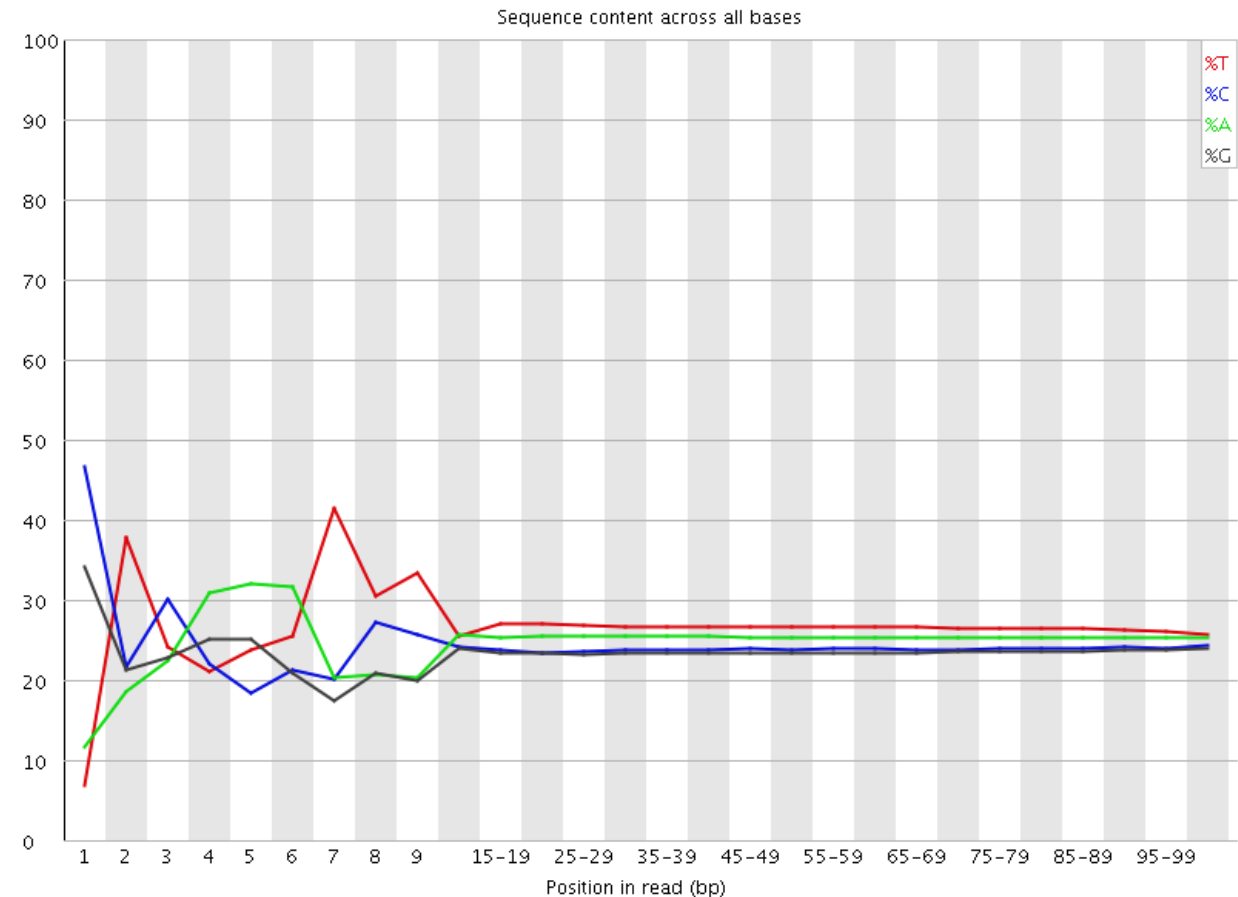


Question 3

You did your fastqc on a RNAseq illumina library, you observed this plot. What do you deduce ?

- A. We have an adapters contamination.
- B. It may just be an artifact due to 'random' hexamer which are not so random.
- C. We have DNA contamination from another organism.
- D. The line at the end should be all equals. I have an issue about my library.

❌ Per base sequence content



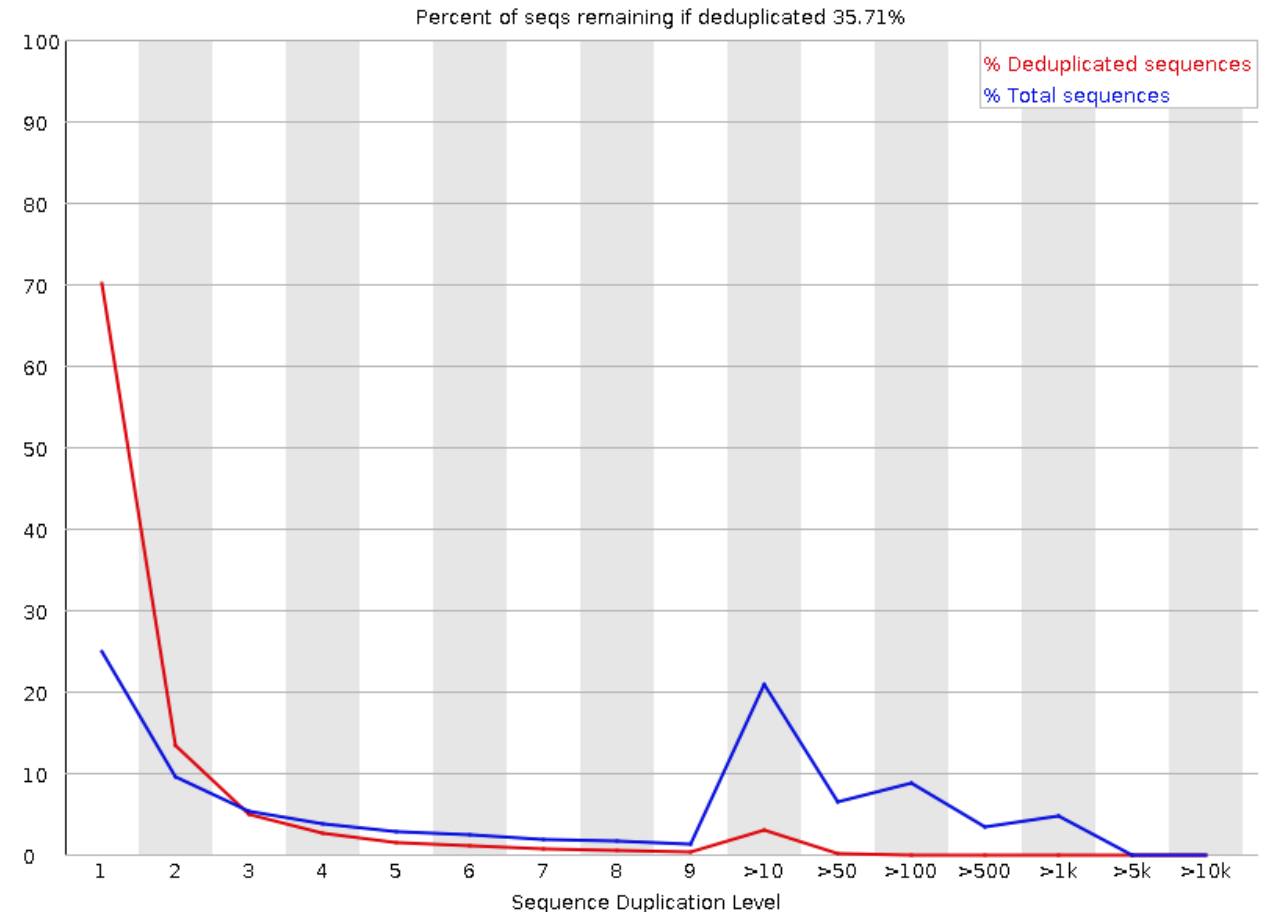
Question 4

You did your fastqc on a RNAseq illumina library, you observed this plot. What do you deduce ?

- A. There are always a lot of duplication event in RNAseq experiment.
- B. We have an adapters contamination.
- C. We have a rDNA contamination.
- D. I have a low complexity library, that I have sequence way too much.



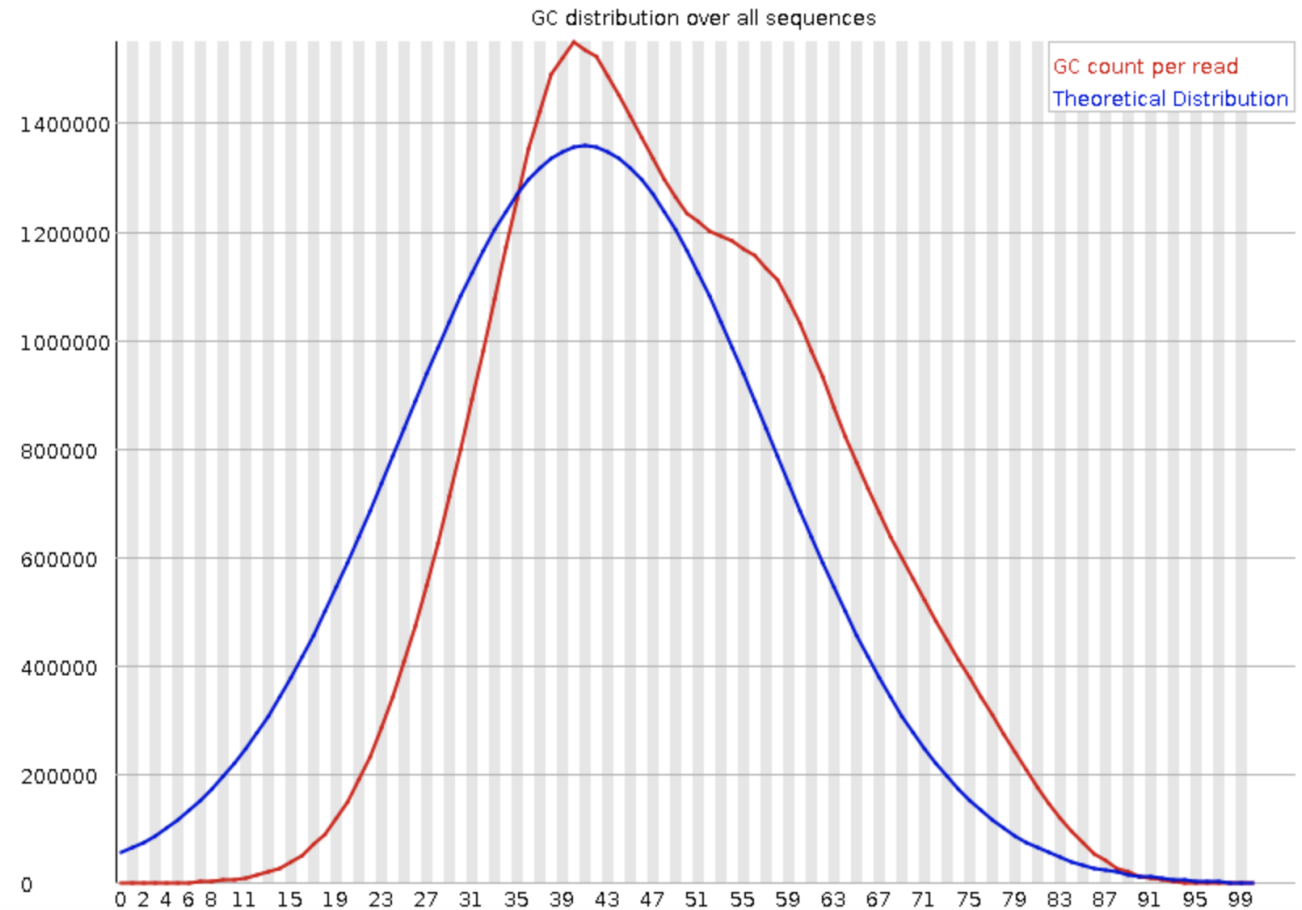
Sequence Duplication Levels



Question 5

You did your fastqc on a RNAseq illumina library, you observed this plot. What do you deduce ?

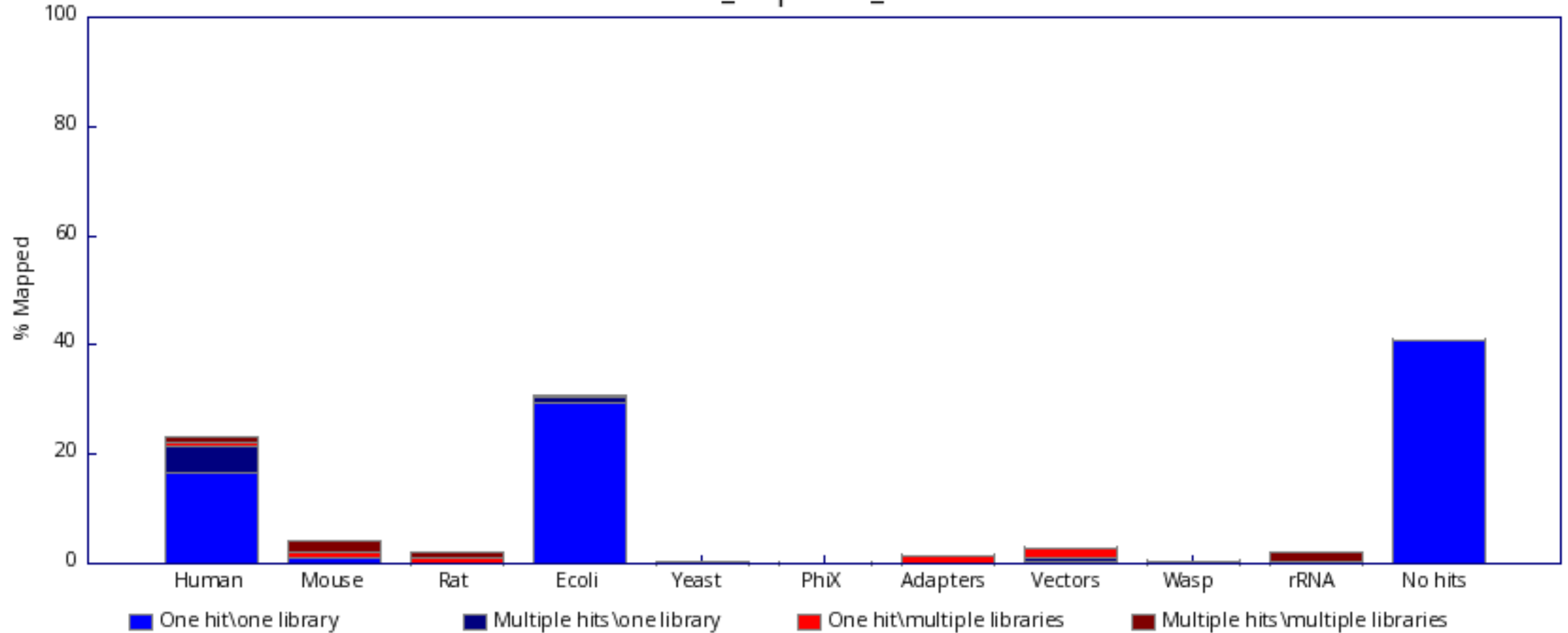
✖ Per sequence GC content



- A. We have a contamination from another species.**
- B. We have an adapters contamination.
- C. We have a rDNA contamination.
- D. I can't know the origin of the contamination, I will blast my reads or/and do a fastq-screen.

Question 5

bad_sequence_screen



Escherichia coli contamination of a human library.