

# Quiz 3

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GENOMICS

BIGNAUD AMAURY

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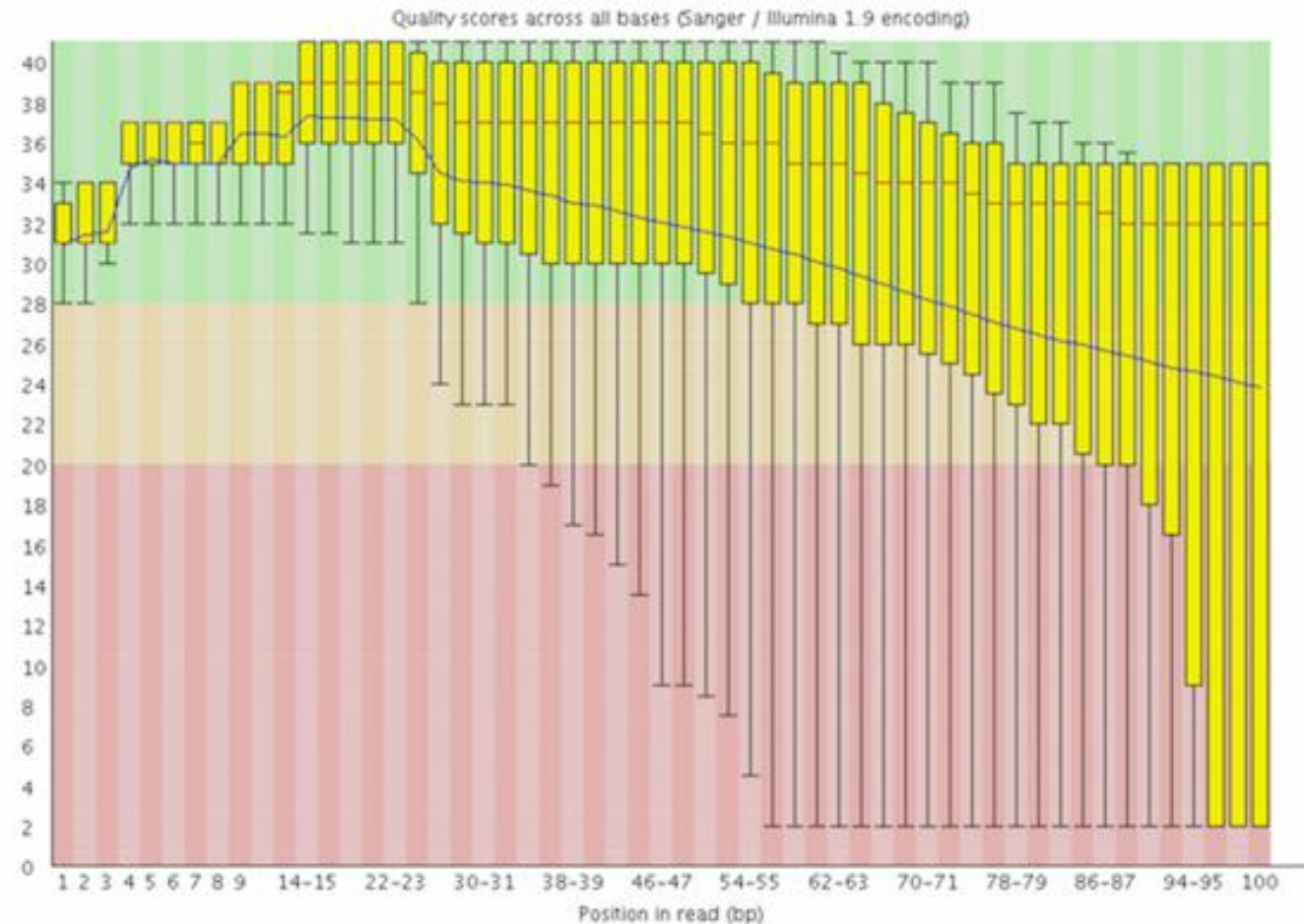
# Question 1

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.



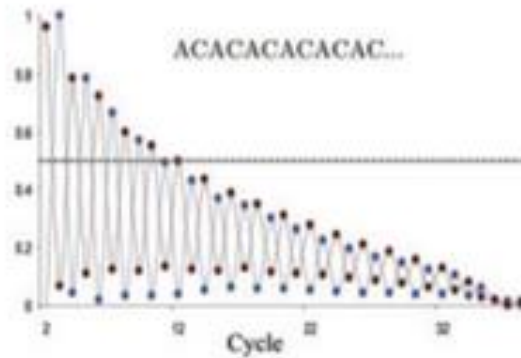
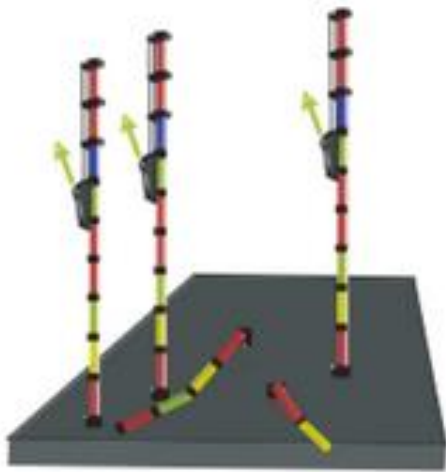
## Per base sequence quality



# Question 1

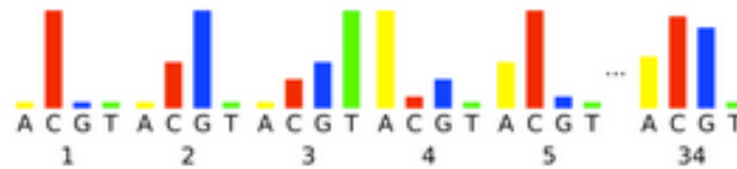
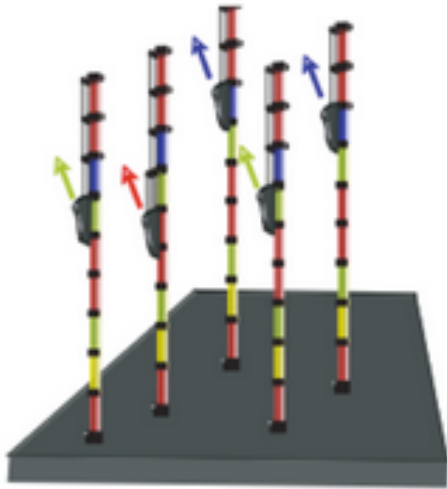
- **Signal decay:** As sequencing proceeds, the fluorescent signal intensity decays with each cycle, yielding decreasing quality scores at the **3' end** of the read. This is due to:
  1. Degrading fluorophores
  2. A proportion of the strands in the cluster not being elongated

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



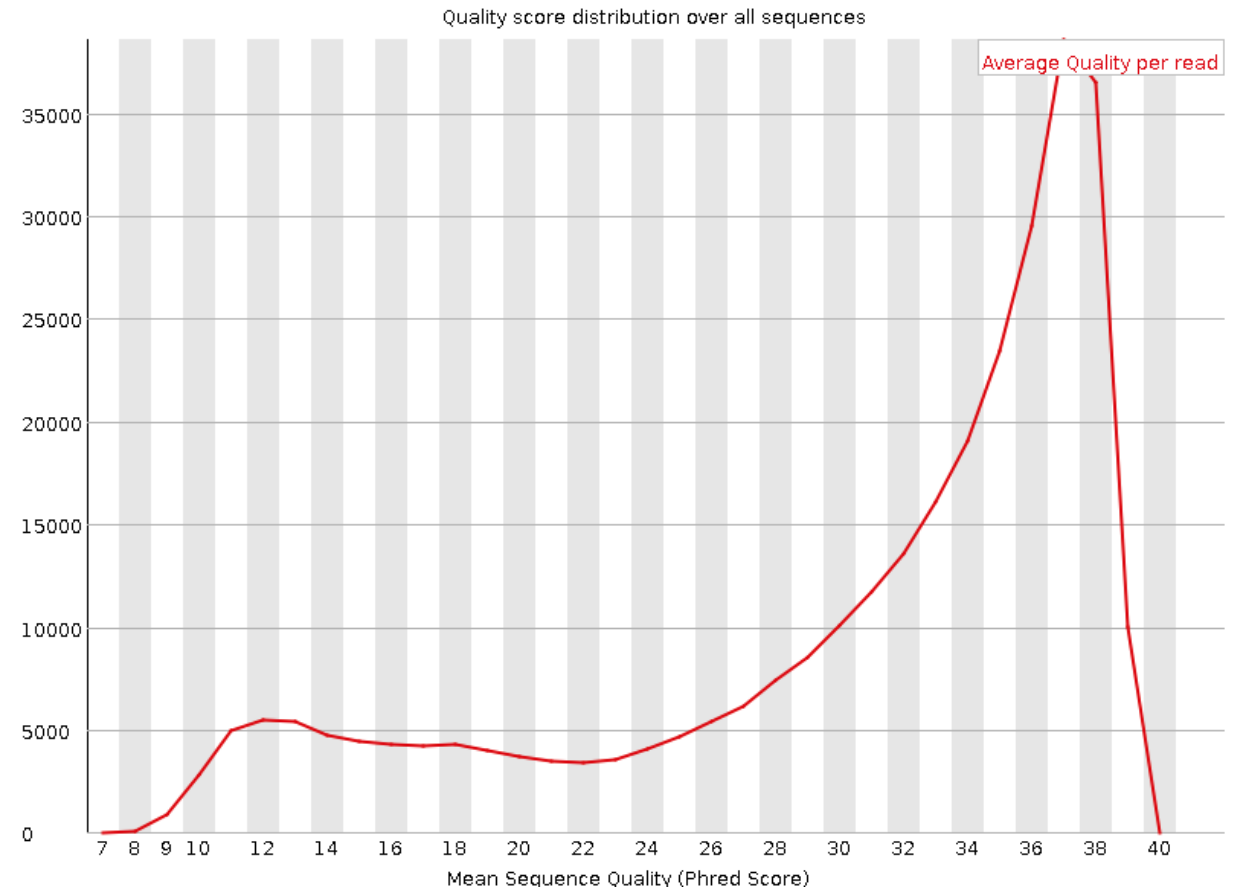
# Question 2

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

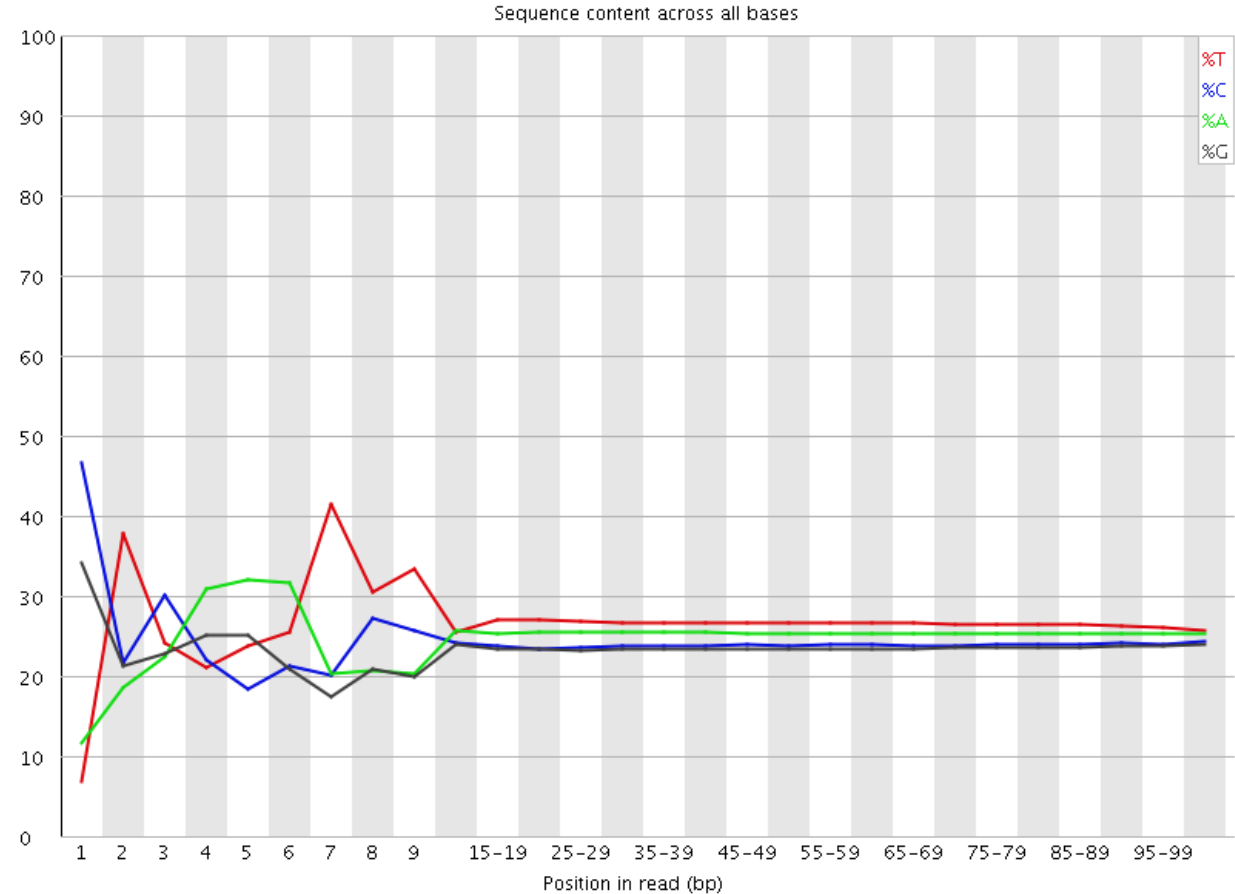


# Question 3

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

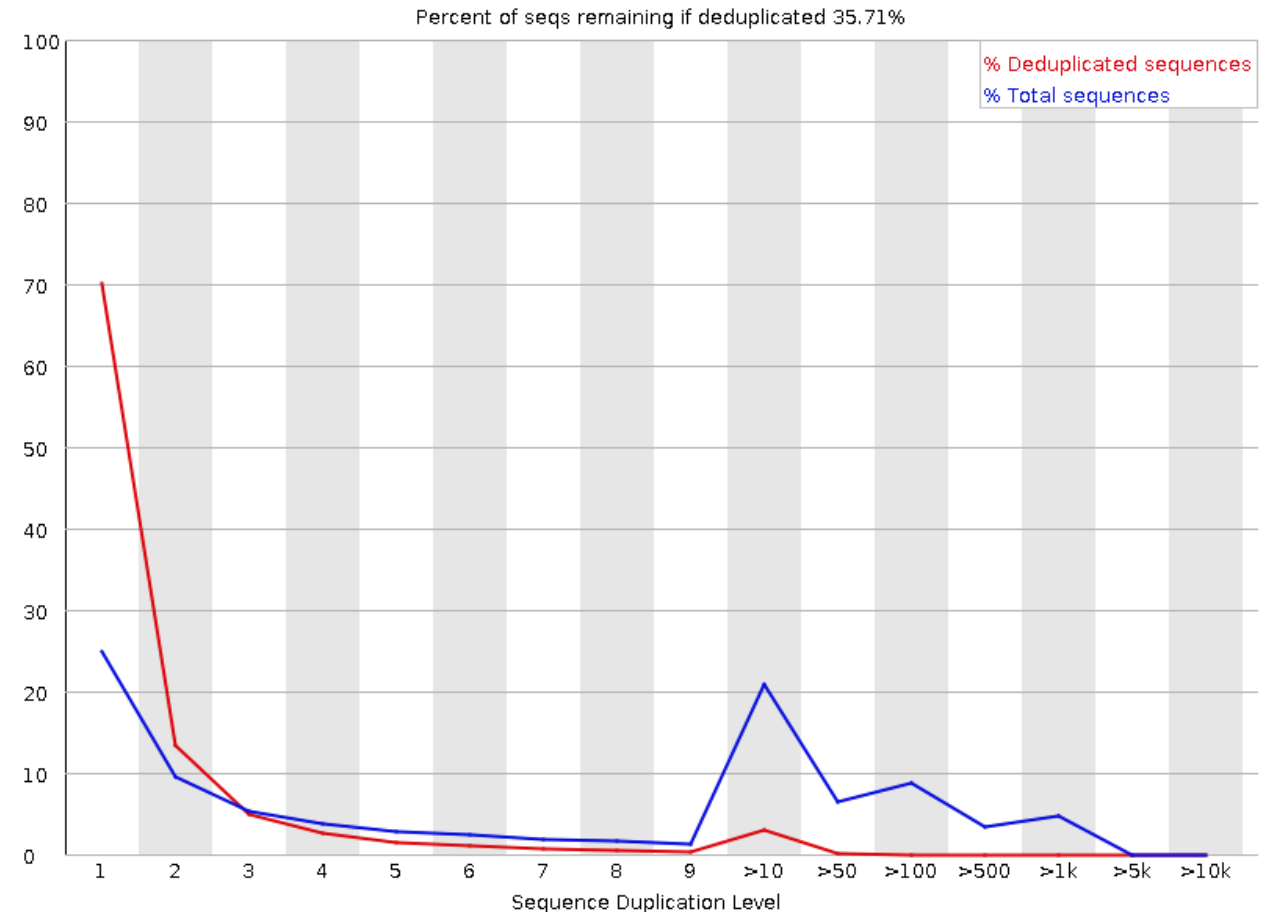


# Question 4

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.

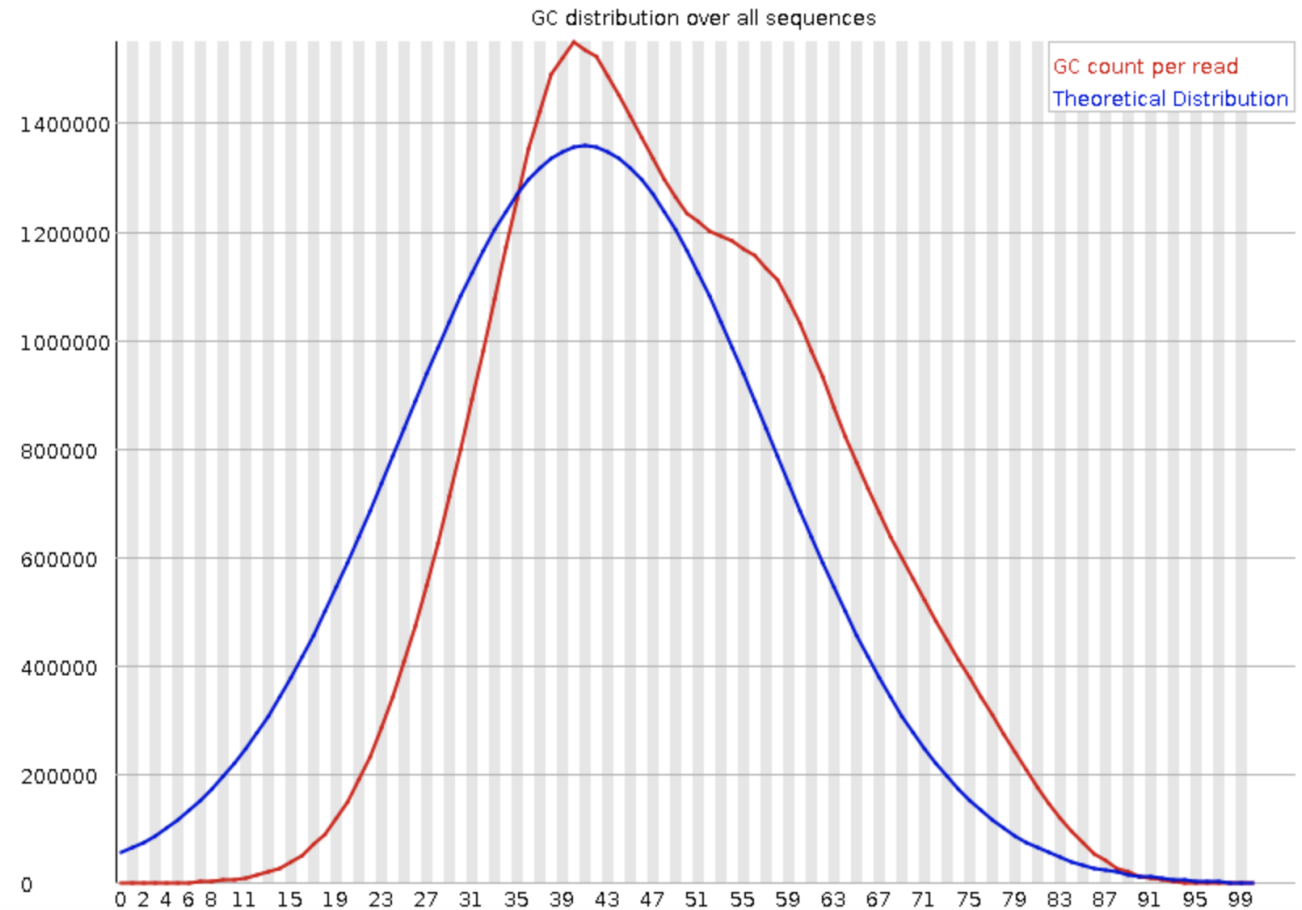
## ❌ Sequence Duplication Levels



# Question 5

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

## ✖ Per sequence GC content

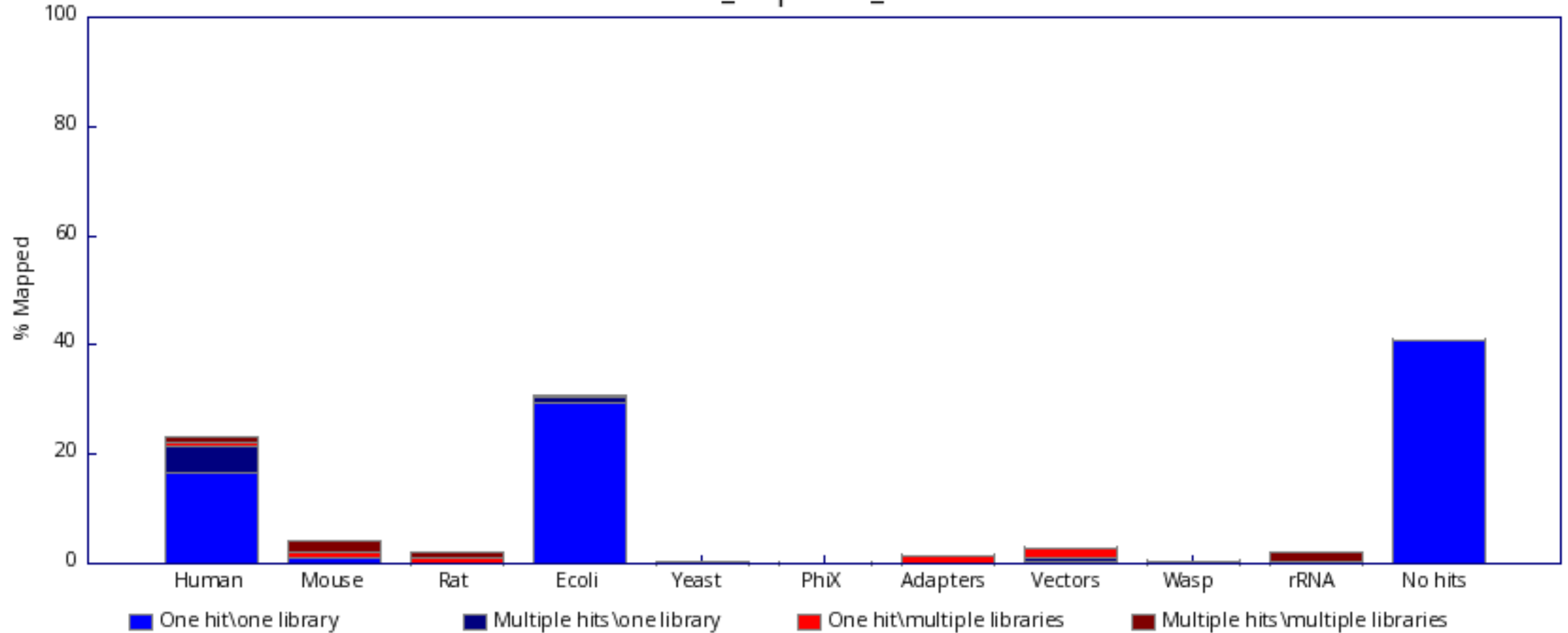


- A. We have a contamination from another species.
- B. We have an adapters contamination.
- C. We have a rDNA contamination.
- D. I can't know the origin of the contamination, I will blast my reads or/and do a fastq-screen.



# Question 5

bad\_sequence\_screen



*Escherichia coli* contamination of a human library.