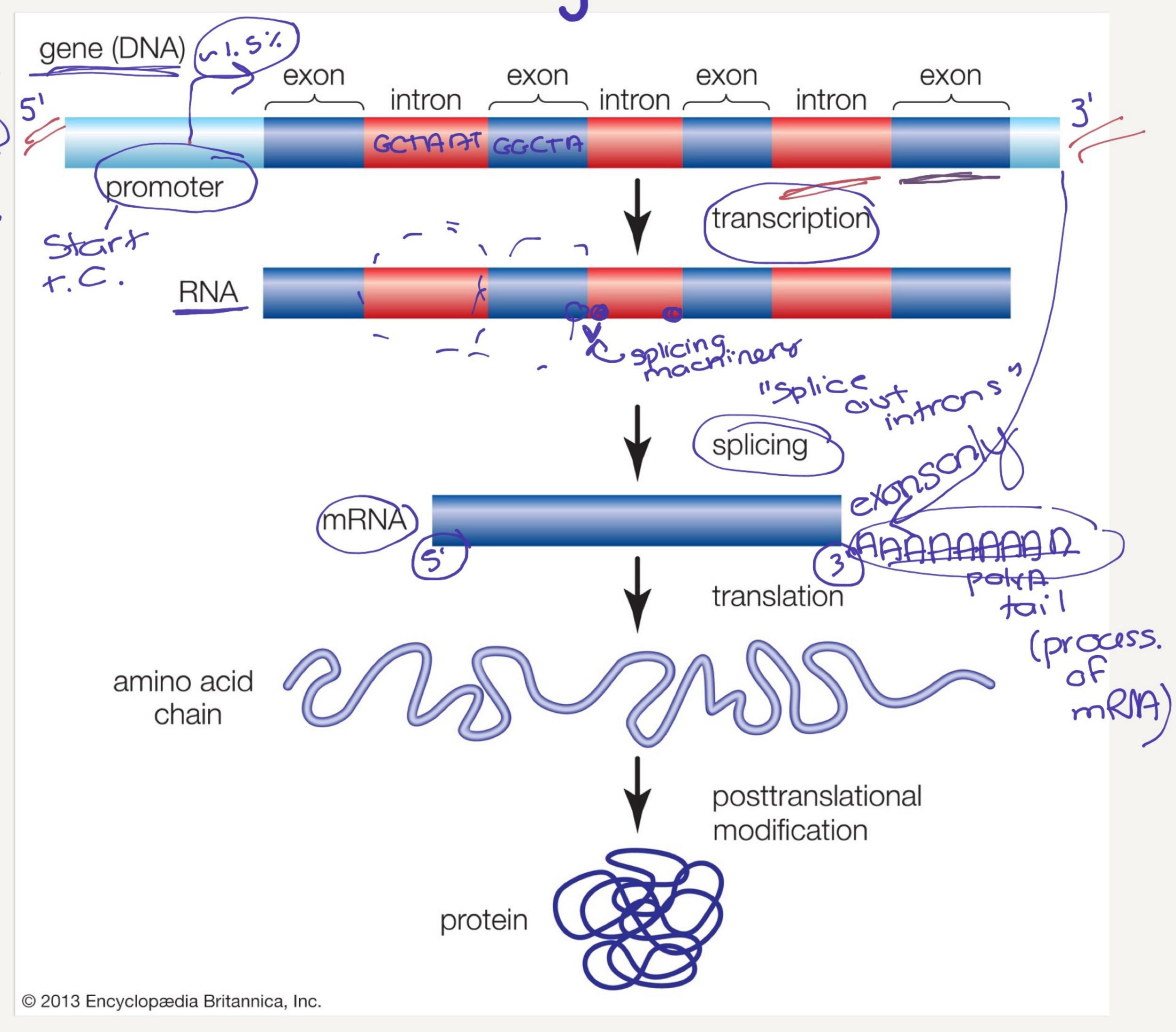
The structure of the gene





# RNA. Seq: the whole process

- Designing project
- RNA extraction
- Purification and enriching mRNA
- cDNA synthesis
- Fragmentation
- Adaptor ligation and amplification
- ·cDNA libraries to be sequenced

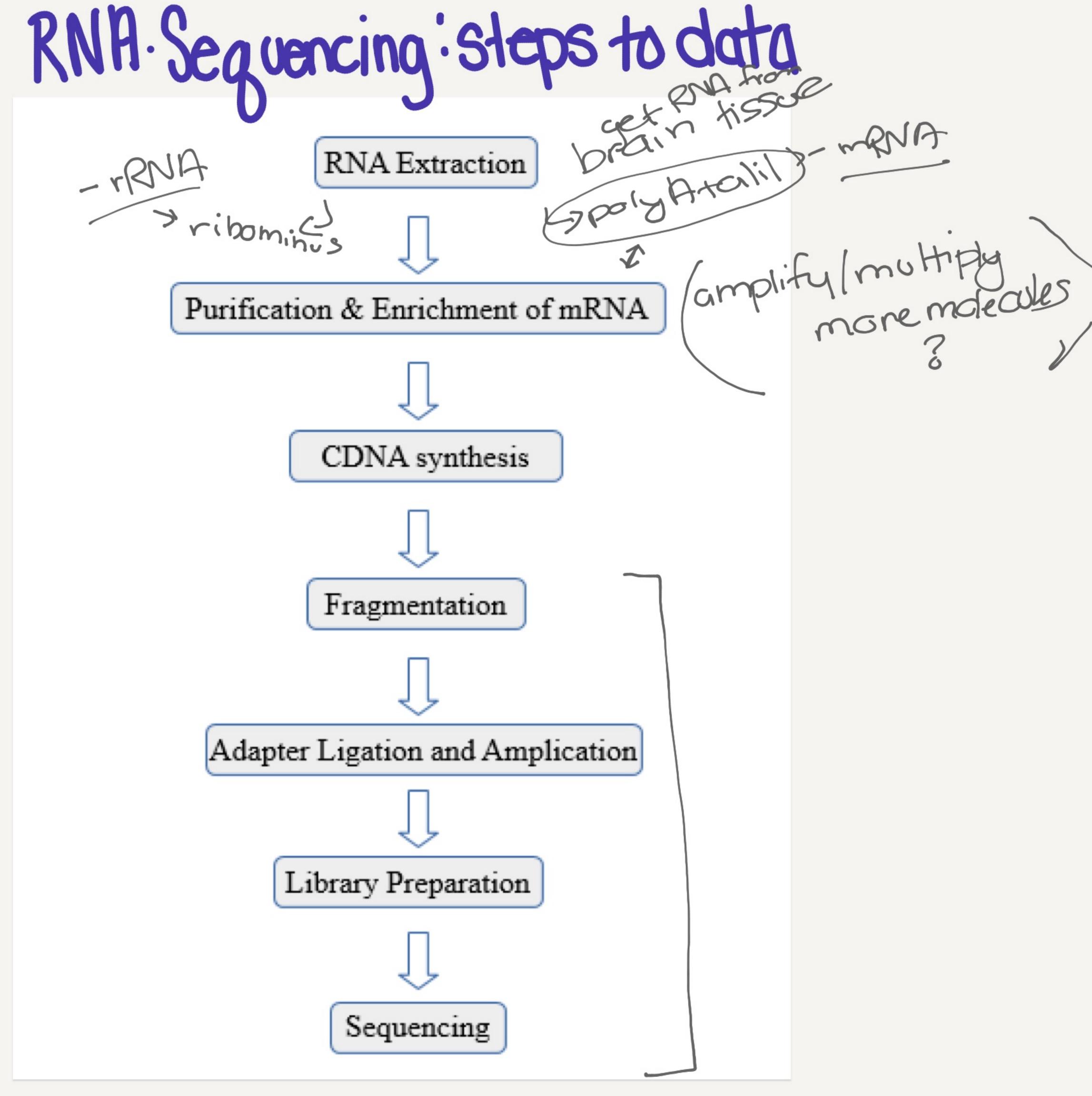
### wet-lab

- · Choosing suitable platform
- · Designing how to run samples
- Sequencing
- · Files including sequences to be processed and analyzed

## in equipo

- Quality control
- Cleaning and triming
- Alignment
- Mapping
- Annotation
- Further analyses like differential expression, phylogenetics, genetic variation, SNPs, etc.

### in silico

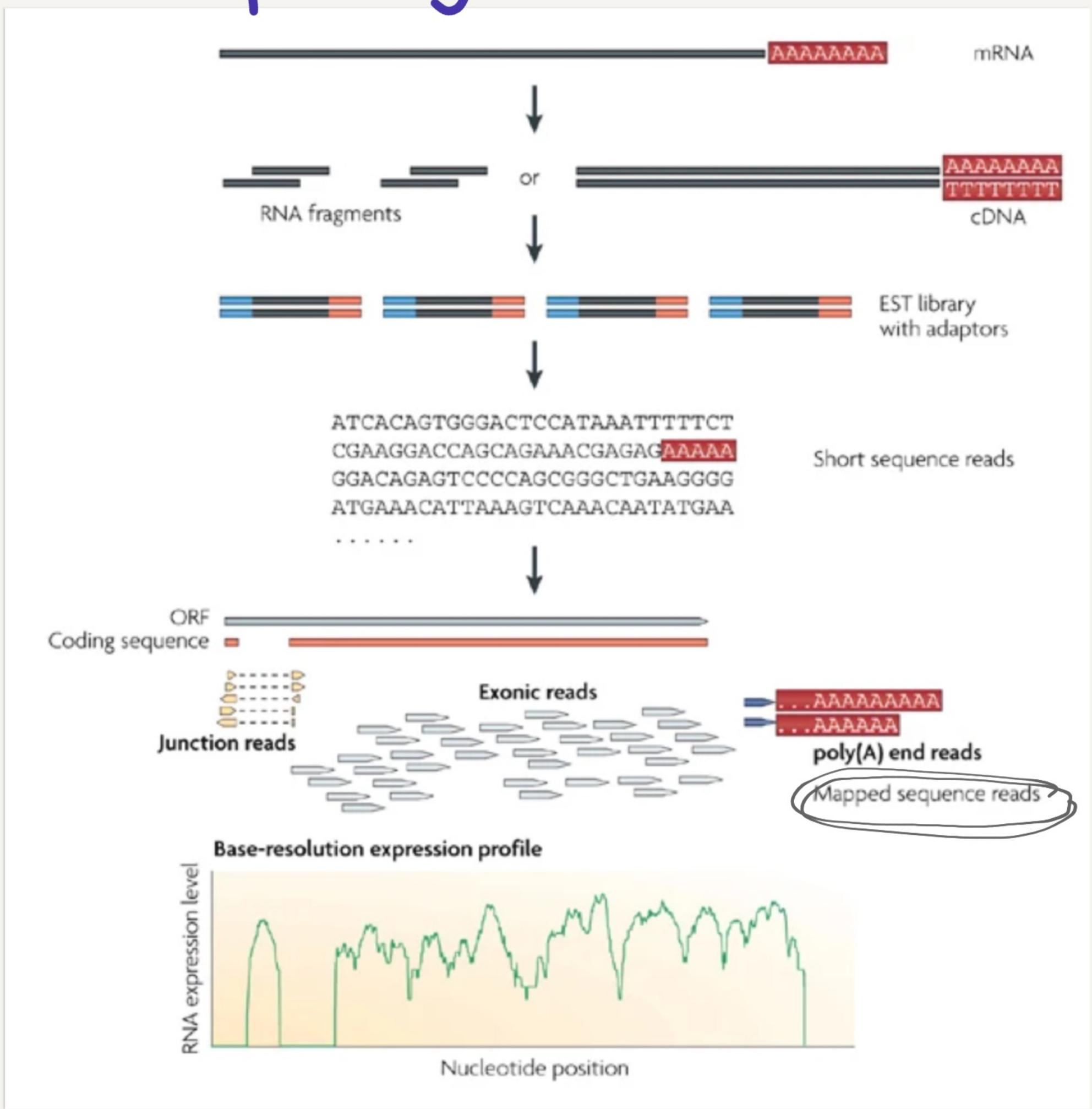


gene A:10 reacts

gene B: 100 reads

- profiting transcriptome
- profiting transcriptome
- quantity "expression"
levels - mRNA levels in

RNA Sequencing



Sequence

Sequence

Sequence

AGG CCC ATT... n=pa

I/ou Os > millions
Seq reads/ind

Alignment/Mapping -> sequences we already have? Human Genome Project (2001) - which fragrumts some from 100,00t chr2 expression in general # reak - higher cont. - if equal: set aside

# Terminology expression levels

Poly A - tags

RNA fragmentation

single:end

paired end Seq. deep sequencing

- depth
- coverage \$\$

clepth overage how much
of transc.

"background"