

Quality Control



# Tapestation

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## Learning outcomes

You will learn:

1. How the TapeStation works
2. How to interpret Quality Control (QC) data

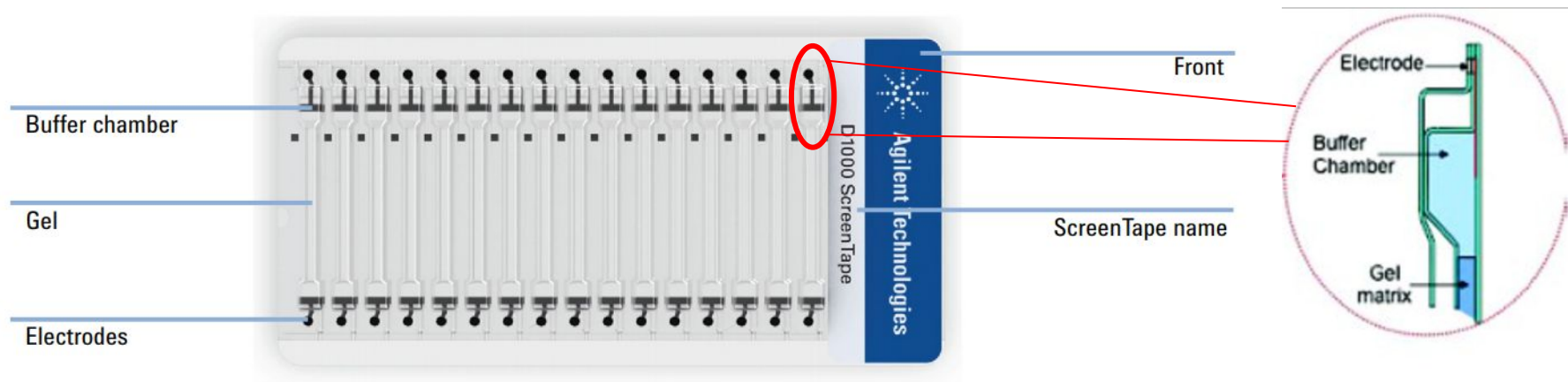


# Tapestation



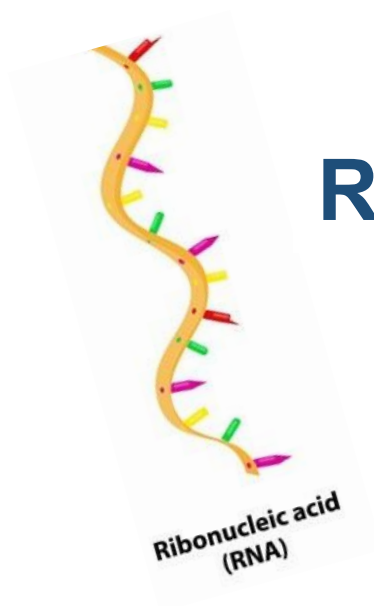
- Automated **electrophoresis** system which separates charged biomolecules in a fluid using a field electrically charged
- Analysis of **SIZE, CONCENTRATION** and **INTEGRITY**

## ScreenTape



- 16 channels: buffer chamber, gel, electrodes
- DNA and RNA (negative charged) are separated by size and weight



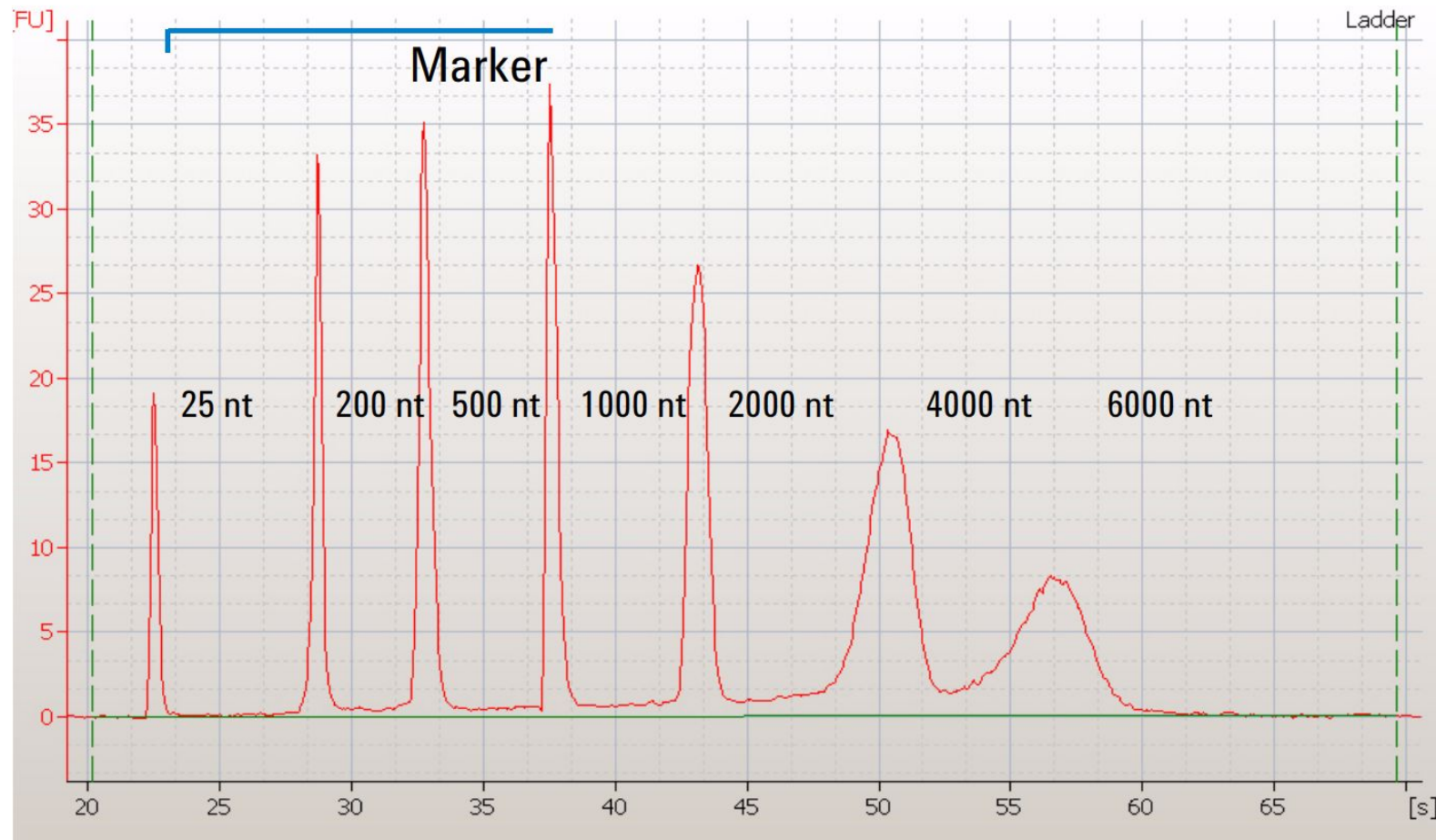


## RNA Tapestation analysis

1. RNA Ladder
2. RNA buffer
3. RNA sample
4. RNA screentape



# RNA Ladder

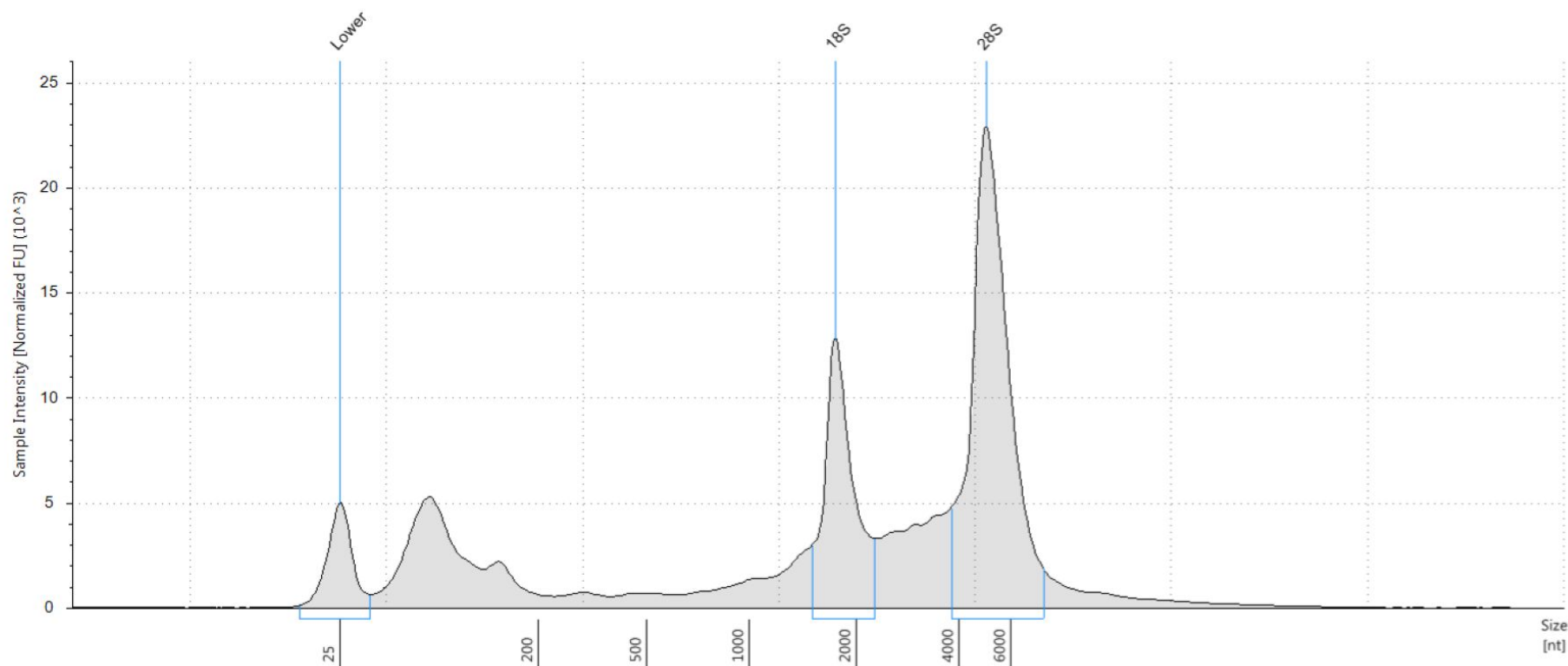


The RNA ladder is a set of different and known RNAs band size.

The RNA ladder is a reference for estimating **size** and **mass** of RNA sample.



# RNA Quantification

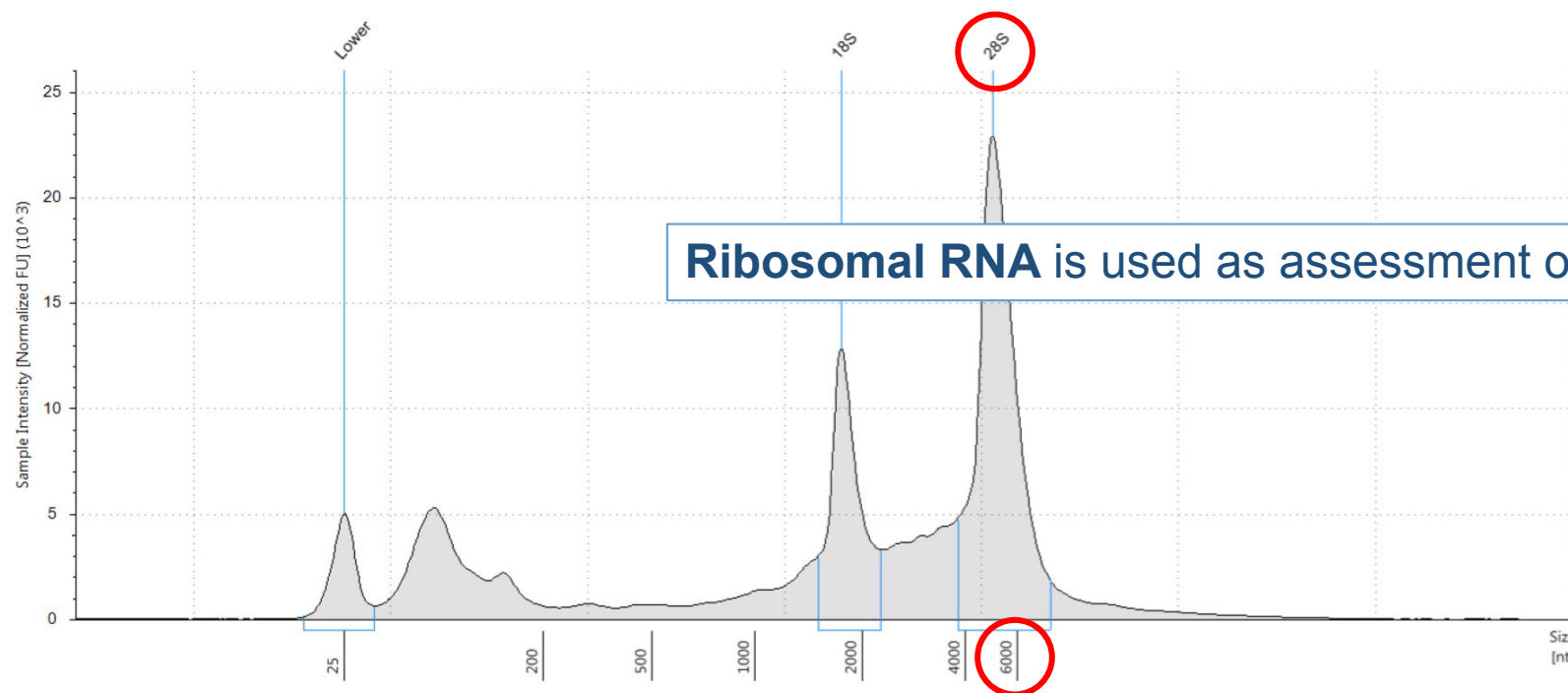


- **Lower marker:** 25nt fragment, run with each of the samples, internal marker used to align all samples to the ladder data.
- **18S** and **28S** peaks of Eukaryote total RNA:
  - 80% of tot RNA
  - used to assess the **quantity & quality**.

**Concentration** is calculated considering the area under the **18S** and **28S** peaks compared to the area under the ladder.



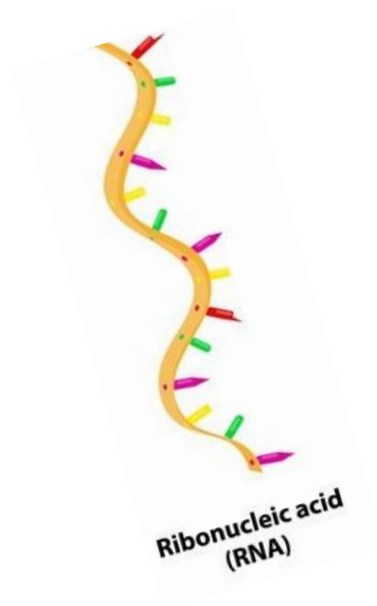
# RNA Integrity



The **RNA integrity number (RIN)**:

- Numerical value to indicate the RNA integrity  
**Scale 1-10 (10 being perfect)**  
 Higher the number, better the quality of RNA sample is!
- The **28S/18S ratio** gives an indication on the RNA integrity



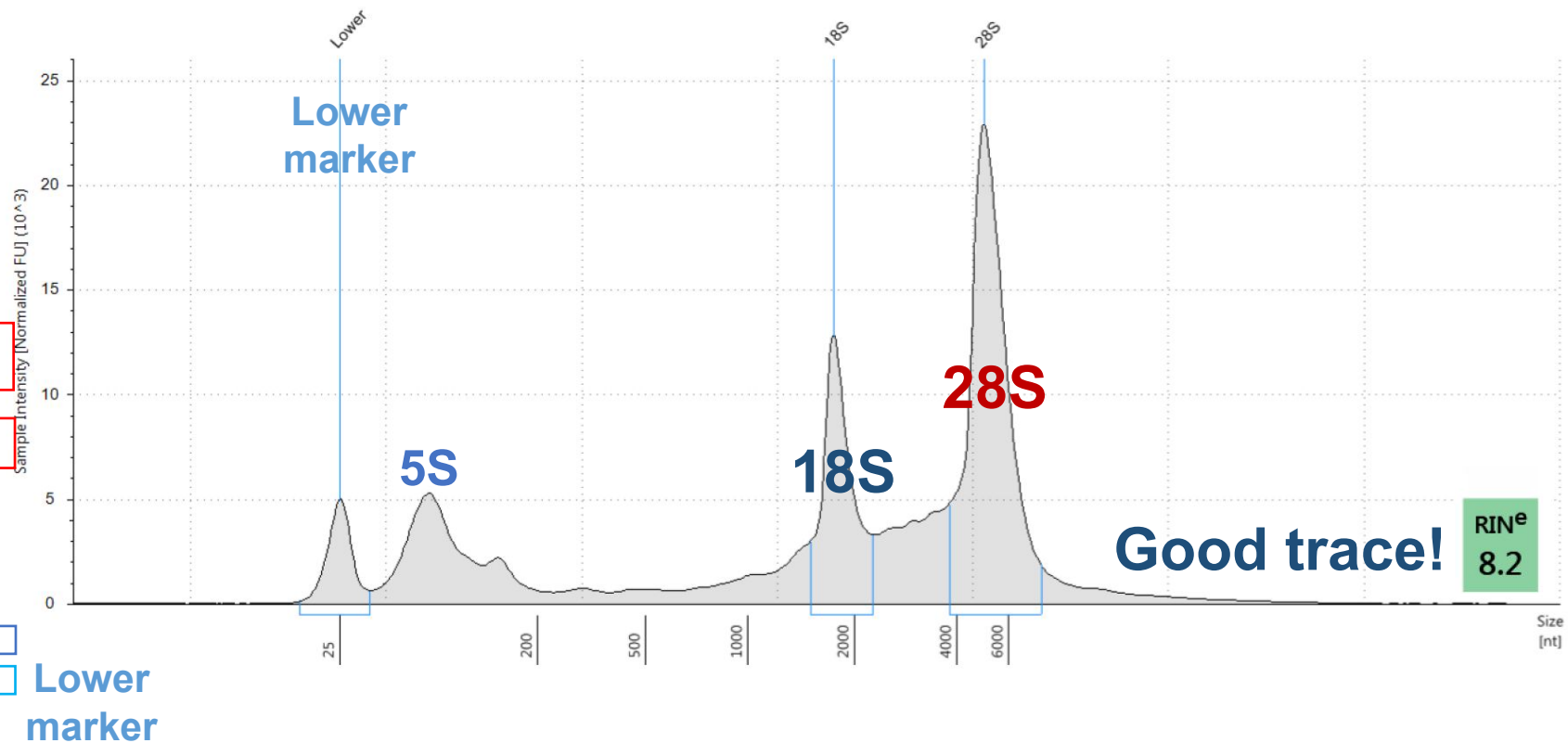
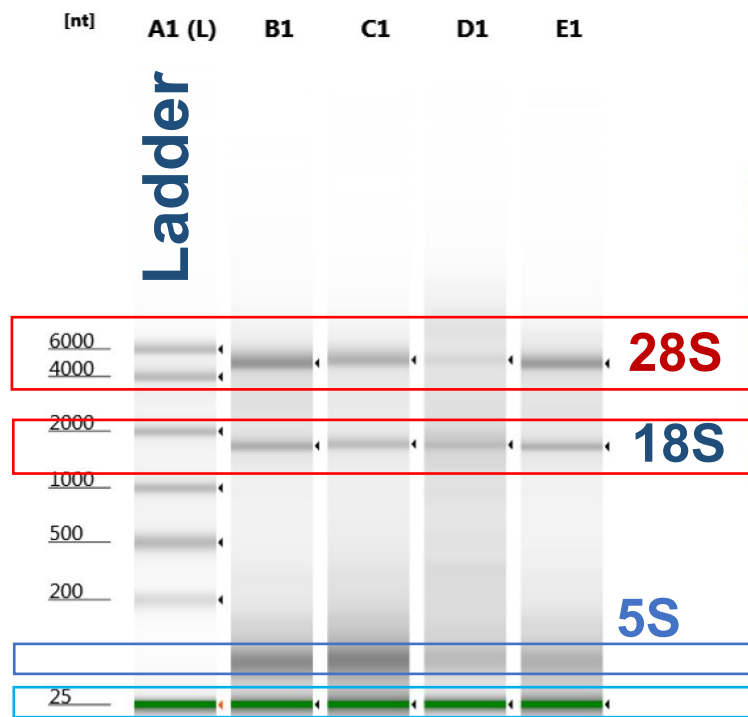


# RNA Tapestation *examples*





# RNA gel

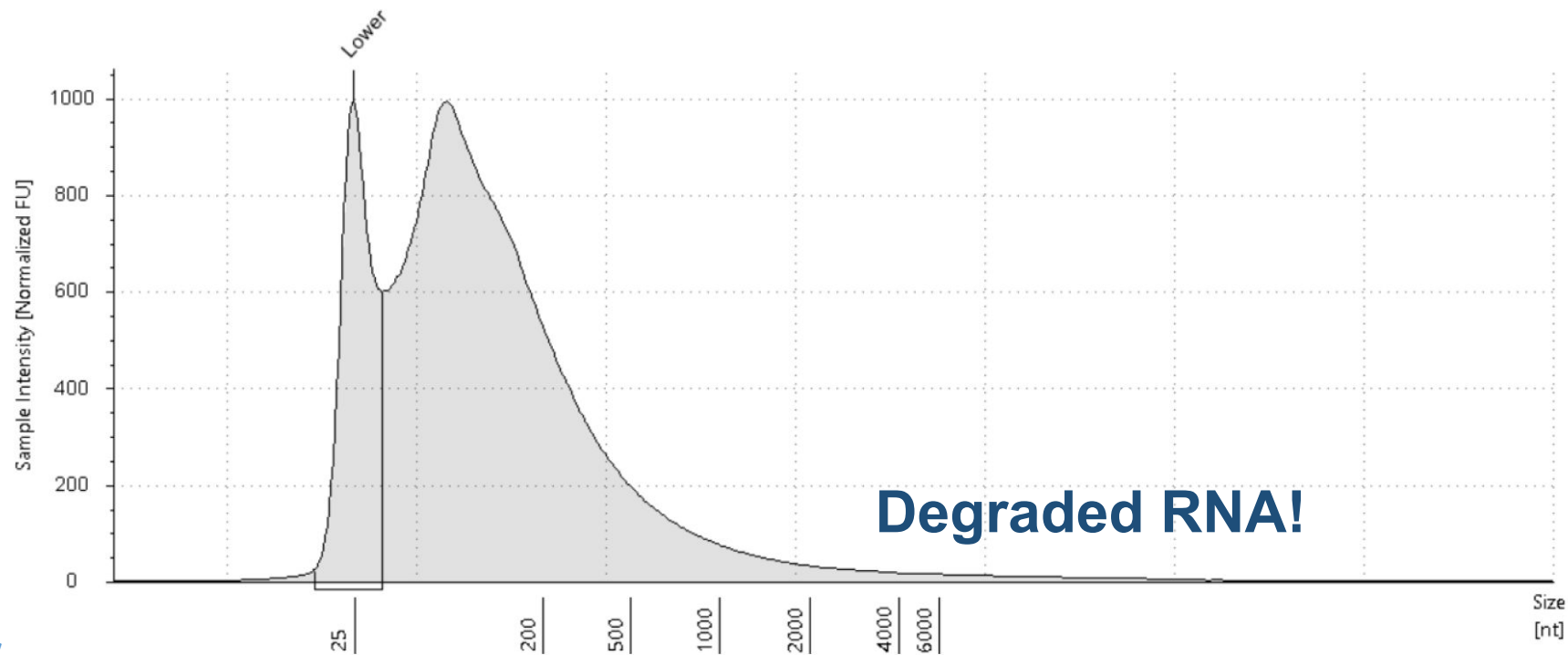
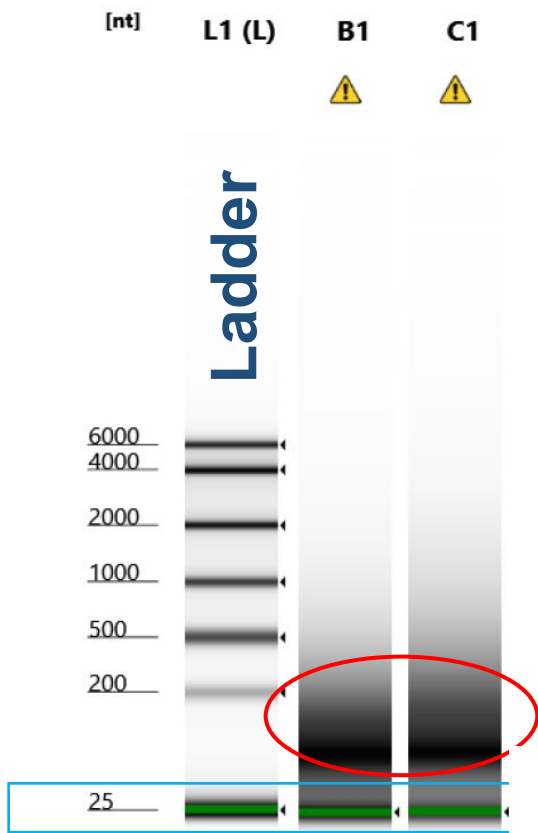


**RNA Integrity Number (RIN)**

RIN <sup>e</sup> 8.0	RIN <sup>e</sup> 7.5	RIN <sup>e</sup> 5.3	RIN <sup>e</sup> 8.2
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RNA with RIN equal or > 7 is suitable for downstream applications!

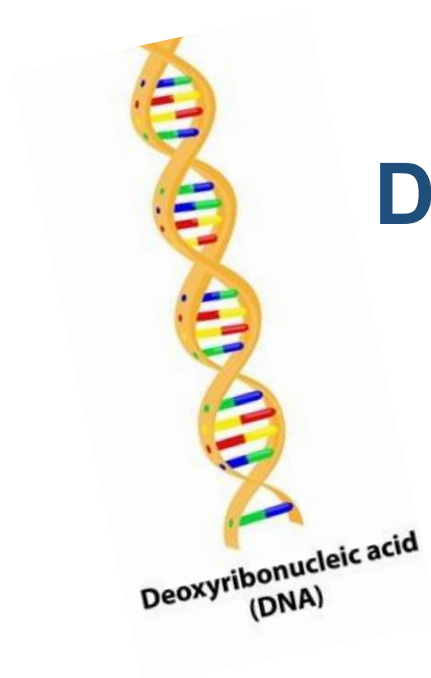




**RNA  
Integrity  
Number  
(RIN)**

RIN <sup>e</sup>	RIN <sup>e</sup>
1.2	1.3



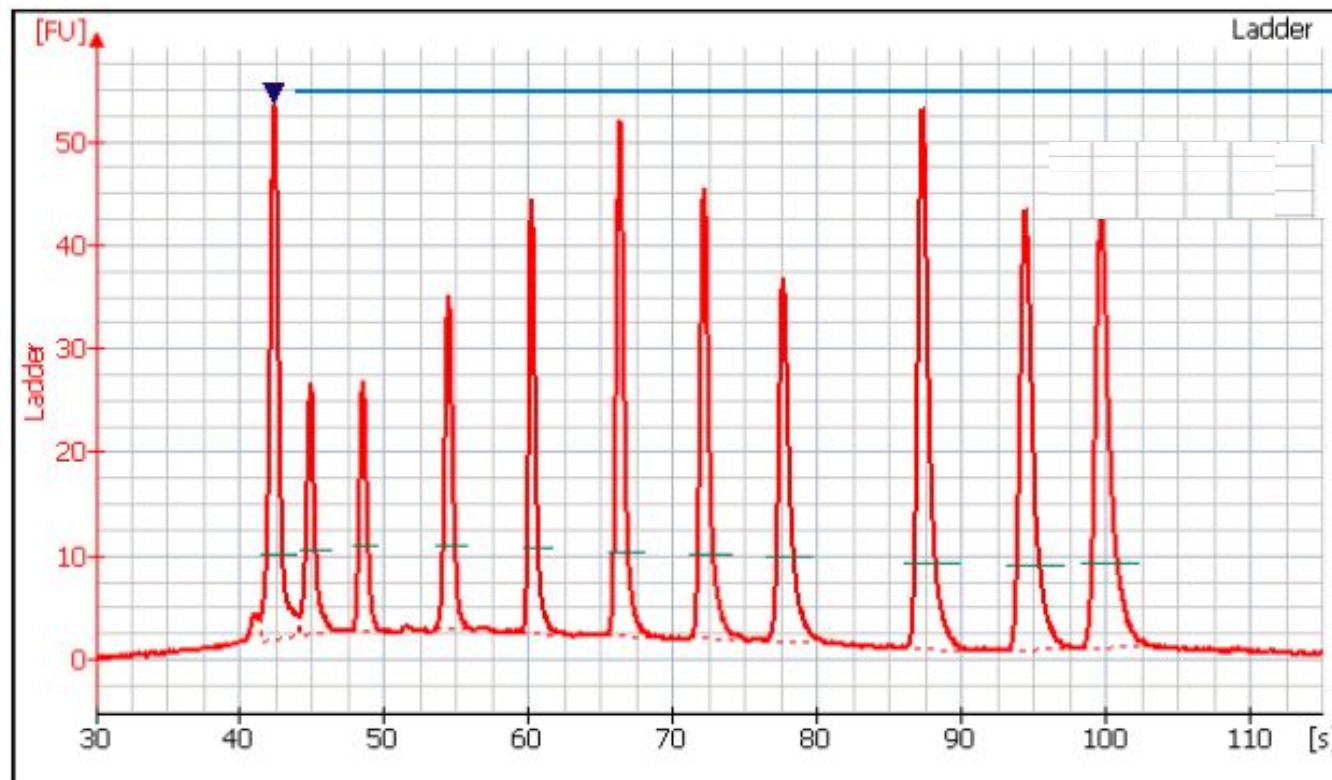


## DNA Tapestation analysis

1. DNA Ladder
2. DNA buffer
3. DNA sample
4. DNA screentape



# DNA Ladder



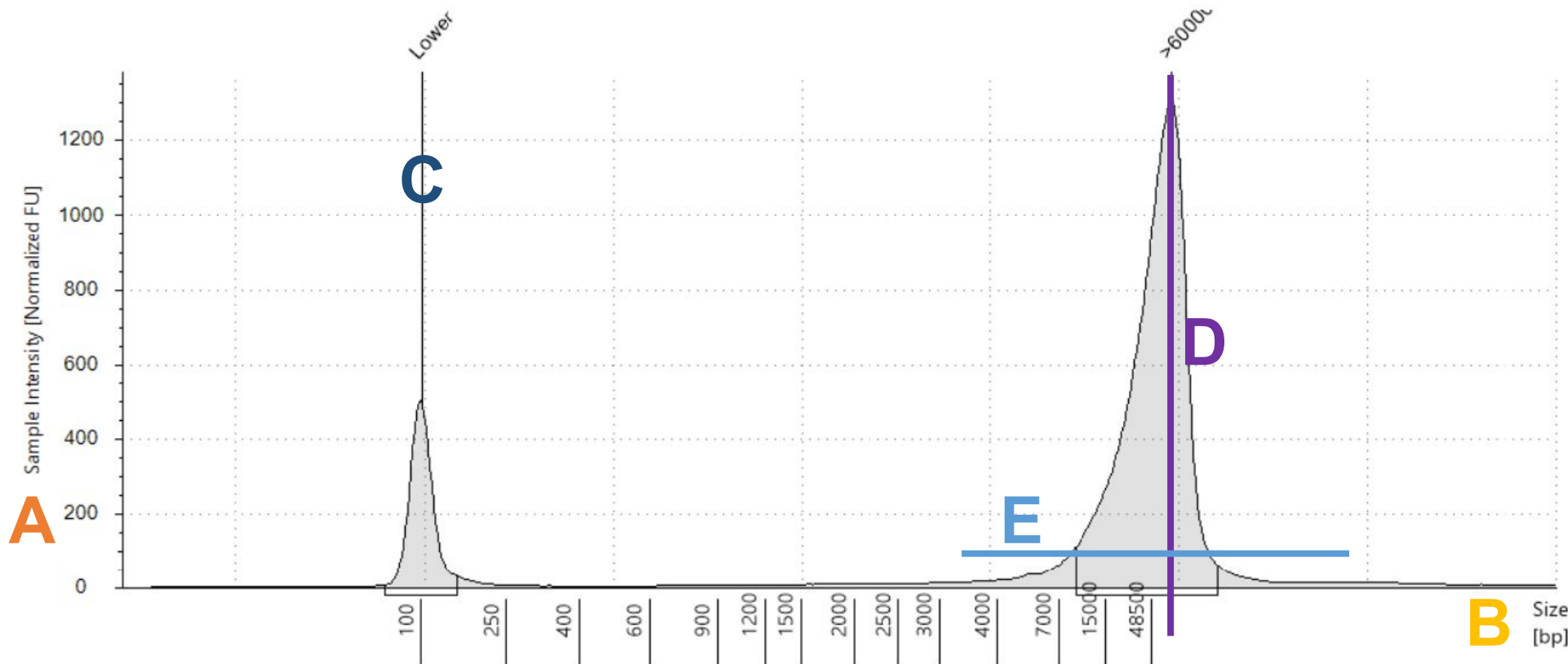
Lower marker

The **DNA ladder** is a set of DNAs of known size, important to estimate size and mass of DNA sample.

**Lower marker** is an internal standard used to align the ladder data to all samples.



# DNA Quantification & Integrity



Sample Table

Well	DIN	Conc. [ng/μl]	Sample Description	Alert	Observations
B4	9.4	56.5			

- A** (*y axis*) the fluorescence intensity
- B** (*x axis*) size in basepair
- C** Lower marker

- D** concentration based on the height of the peak: **Quantity!**
  - E** size of fragments based on the width of the peak: **Integrity!**
- DNA Integrity Number (DIN)*



# Tapestation



## Advantage:

- The best method to assess integrity of DNA and RNA
- RNA Integrity Number (RIN) or DNA Integrity Number (DIN)
- Information about size, quantity and quality (integrity)

## Limitation:

- No information about purity (use the NanoDrop instead!!!)

Combine more methods to have an accurate and complete picture of your sample quality!

