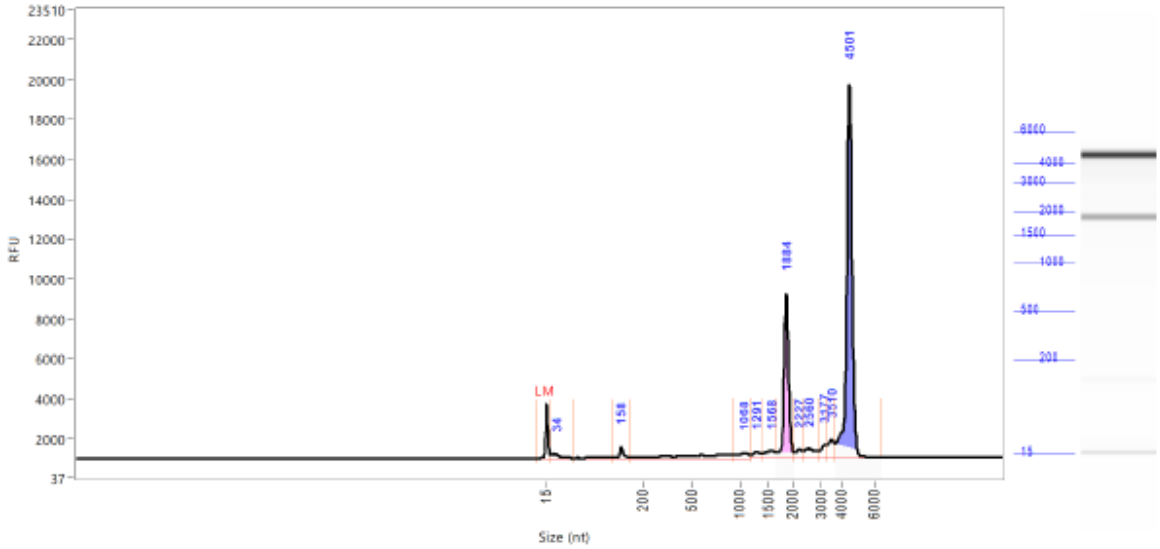
RNA and DNA quality check

RNA quality control via Fragment Analyzer:

- a 2.5 µL aliquot of each sample is needed
- please provide the samples in 0.2 mL – 1.5 mL tubes, larger amounts ≥ 8 samples have to be provided in 0.2 mL PCR tube strips
- store samples in the dedicated box in the -80°C freezer (room 3.30, sequencing rack, pink "fragment analyzer" box)
- please label your samples clearly with sample ID, your name and workgroup
- please send an Excel-table with the respectively measured concentrations (NanoDrop) to the lab managers

After analysis you'll receive a pdf-report per email.
Example:



Peak	Size (nt)	Conc. (ng/uL)	From (nt)	To (nt)	RFU
1	15 (LM)	0.3527	0	23	2791
2	34	1.0007	23	65	225
3	158	1.1725	143	176	568
4	1068	1.2130	936	1190	248
5	1291	0.9396	1190	1385	309
6	1568	1.4660	1385	1667	334
7	1884	14.8329	1667	2068	8198
8	2227	1.0530	2068	2374	395
9	2560	1.9676	2374	2933	451
10	3177	1.3061	2933	3275	655
11	3510	2.0543	3275	3687	897
12	4501	36.1444	3687	6472	18728

TIC:	63.1502	ng/uL
TIM:	179.2299	nmole/L
Total Conc.:	67.0617	ng/uL
28S/18S:	2.4	
RQN	9.8	

Key:

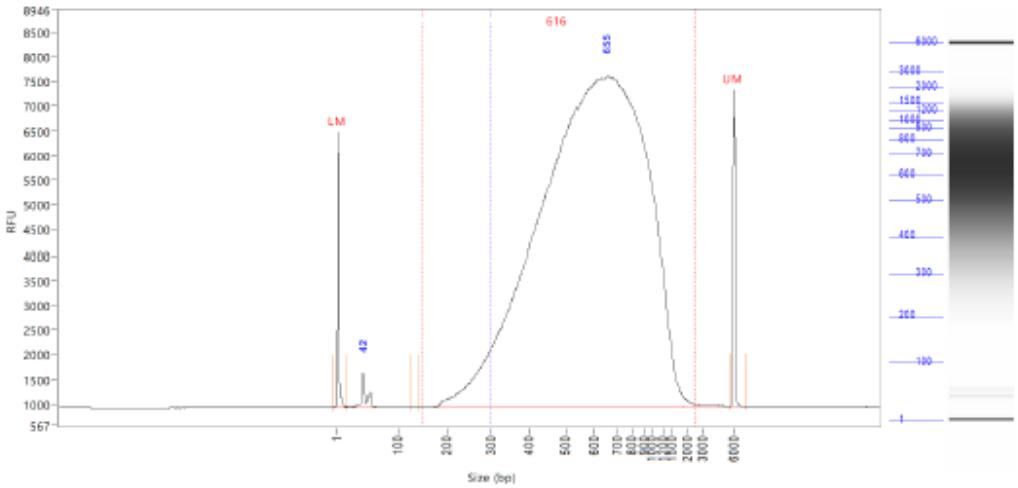
RQN	RNA quality number (should be ≥ 8 for sequencing samples)
28S/18S ratio	should be ≥ 2 for sequencing samples (intact, high quality RNA)
Total conc.	this concentration does not include any dilution factors of pre-dilutions

DNA quality control via Fragment Analyzer:

NOTE: This analysis is only intended for the quality checks of sequencing libraries!

- a 2.5 µL aliquot of each sample is needed; in case a pre-dilution of the sample is required (according to library prep. protocol), please prepare the dilution for the final aliquot!
- please provide the samples in 0.2 mL – 1.5 mL tubes, larger amounts ≥ 8 samples have to be provided in 0.2 mL PCR tube strips
- store samples in the dedicated box in the -20°C freezer (room 3.05, top most drawer, yellow "fragment analyzer" box)
- please label your samples clearly with sample ID, your name and workgroup
- send an email with all essential information (sample amount, type of fragment analyzer kit, fragment size range that should be used to determine the average fragment size (& quantification) to the lab managers

Example (final Nextera XT library)



Peak	Size (bp)	Conc. (ng/uL)	From (bp)	To (bp)	Avg. Size (bp)	CV%	RFU	Corr. Peak Area
1	1 (LM)	0.0223	0	15	1	148.24	5537	30.299
2	42	0.0998	15	128	45	21.23	705	11.313
3	655	14.2476	141	5593	619	41.44	6669	1615.735
4	6000 (UM)	0.0163	5593	7061	5982	1.82	6378	22.158

TIC:	14.3474	ng/uL
TIM:	41.5073	nmole/L
Total Conc.:	14.3474	ng/uL
DQN:	9.6	
Threshold:	300	

Smear Analysis	150 bp to 2500 bp	14.2335 ng/ul	99.2 %Total	38.0042 nmole/L	616 Avg. Size (bp)	38.47 %CV

Key:

DQN DNA quality number (good quality: ≥ 8)
Total conc. this concentration does not include any dilution factors of pre-dilutions

- red lines in the graph mark the borders of the smear analysis, which were used to determine the average fragment size
- smear analysis: data (range, conc.[ng/µL and in mM/L], average fragment size...) are listed below the sample integration table (red values). These values are used for final library calculations (sample normalisation...).