

Genomics Platform NGS Unit accepts either nucleic acid samples (RNA or DNA) or ready sequencing libraries. We do not perform nucleic acid isolation from cells and tissues.

## Nucleic acid samples guidelines

### Nucleic acids isolation methods

- Generally, we have no preferences for RNA and DNA isolation methods.
- To sequence small RNA, take care to use a protocol which preserves small RNAs in total RNA (e.g. Qiagen AllPrep DNA/RNA/miRNA Universal Kit, #80224)

**RNA samples should be treated with DNase I to avoid DNA contamination of the sequencing reads.**

### Concentration measurement

- Concentration should be measured with Qubit. Nanodrop measures all nucleic acids, overestimating the concentration of your sample.
- Contact us if you do not have access to Qubit.

### Quality check

- Knowledge of the integrity state of DNA and RNA is crucial to choose the correct library preparation protocol and proper fragmentation parameters.
- Check RNA Integrity Number (RIN) using BioAnalyser or TapeStation.
- Determine the DNA size range using BioAnalyser or an agarose gel.
- For highly degraded DNA and RNA (e.g. originating from FFPE samples, or improperly stored tissue samples) – estimate the degradation level using PCR fragmentation assay (e.g. KAPA hgDNA Quantification and QC Kit, Roche, #KK4960)

**Contact us if you do not have access to BioAnalyser or TapeStation.**

### Input amounts for sequencing libraries preparation

- Input material recommendations for the offered library preparation methods are summarized in the table below - at least twice the amount should be provided.
- If concentration measurement/QC have to be performed by the Genomics Platforms, provide as much material as possible. Unused DNA and RNA will be properly stored and can be picked up within a year.

**If your samples do not fulfill the standard requirements, please contact us to find an optimal solution.**

Table 1. Input sample requirements for Illumina sequencing libraries

Library preparation protocol	Input material	Input amount range (according to the manufacturer)	Input amount range used so far in the facility*	maximal input material volume	Quality requirements
Illumina TruSeq stranded mRNA	total RNA	0.1–1 µg	100-500 ng	50 µl	RIN≥8
Illumina TruSeq stranded total RNA library prep with Ribo Zero Gold rRNA depletion	total RNA	0.1–1 µg	100-500 ng	10 µl	RIN≥8
Illumina TruSeq small RNA	total RNA	1 µg	1 µg	5 µl	RIN≥8
NEBNext Ultra II total RNA	total RNA	5 ng-1 µg	20-500 ng	12 µl	
TAKARA SMARTer® Stranded Total RNA-Seq Pico Input	total RNA	250 pg -10 ng	250 pg -10 ng	8 µl	
exome sequencing: SureSelect XT human All Exon v7	genomic DNA	10-200 ng	200 ng	50 µl (shearing by sonication) 7 µl (enzymatic shearing)	
Illumina ChIP-Seq	ChIP-enriched, fragmented DNA	5–10 ng	10 ng	50 µl	

\*Amount of material which performed well in our facility

### Shipment

- Please provide your samples in 1.5ml Safe Lock LoBind tubes, labeled readably with sample name (instructions for naming are in the [Sample\\_Transfer\\_Sheets](#) (link)) and date.
- RNA samples (in dry ice) and DNA samples (frozen or in dry ice) can be sent per post, MDC shuttle or just brought along (after making an appointment) to our laboratory in Berlin-Mitte:

***MDC-BIMSB, Genomics Platform, R 1.50, Hannoversche Str. 28, 10115 Berlin***