

RNA sample preparation for long non-coding RNA and whole transcriptome sequencing

1. Isolation of total RNA

The quality of the RNA to be used for RNA-seq is very important, we therefore highly recommend the Qiagen portfolio kit for total RNA extraction and purification.

2. RNA quantification, quality and preparation

When starting from **cell, tissue or whole blood (Human, mouse, rat only)** we would require total RNA matching these conditions:

- 10 ng minimum (recommended >100ng);
- 15 µl maximum volume in RNase-free water;
- RIN>7;
- Ratio of spectrophotometry measurement A260/A280>1.6 and A260/A230>1.6
- DNA-free RNA (treated with DNase and purified)

Ideally, each freshly prepared sample should be split into **two tubes**:

- 1- One tube with 10ng of RNA to be used for initial QC
- 2- One tube with > 10ng of RNA to be used for library preparation