

## **Technical specification - Transcriptome Analysis (RNAseq) - 320 samples**

- RNAseq analysis of delivered samples of Norway spruce (total RNA), 320 samples
- RNA samples delivered to the Contractor will meet the following parameters: RIN  $\geq 6.0$ , amount RNA  $\geq 0.4 \mu\text{g}$ , volume  $\geq 20 \mu\text{L}$ , concentration  $\geq 20\text{ng}/\mu\text{L}$ , purity: no contamination
- After delivery, if some of the samples will not be assessed by the Contractor fulfilled minimal quality and parameters, the Contractor agrees to proceed pursuant to Art. 3.2 of this Contract

### **Requirements for the methodological procedure:**

Quality control of the delivered RNA samples before the analysis commencement

mRNA fraction enrichment (polyA enrichment) before cDNA library preparation

cDNA library preparation (random-primed library type)

- cDNA library prepared with the directional RNA kit

genotyping platform: Illumina – NovaSeq

150 bp pair-end reads (bilateral reading in the section length 150 bp)

Minimum 30 million good quality reads (reading frames) per sample (9Gb raw data)

Data quality requirement: Phred value at least Q30 for at least 85% of the reads (reading frames) and at the same time Phred value at least Q20 for at least 90% of the reads (reading frames)

De-multiplexing, removing of adaptors, data quality control (QC report)

Availability of the sequencing results (data delivery) electronically via server/shared disc in the format fastq and fasta format

the Client requires a description of the methodology of the library preparation

the data must be generally compatible with the reference genome databases regardless of the used technology of sequencing.

### **Consultations:**

- 15 (fifteen) consulting hours (each 60 min) with a bioinformatician for 2 people determined by the Client, contact-based meetings (up to 15 one-hour meetings according to Client requirements, on Client's registered seat
- Content of consultations: „secondary bioinformatics on sequencing results“ – software guidance, mapping to the reference genome, coverage QC, statistics and annotation