

# Isoform Sequencing (Full-length Transcript Sequencing)

## 1. Sample Requirements

Library Type	Sample Type	Amount	Volume	Concentration	RIN (Agilent 2100)	Purity (Nanodrop™/Agarose Gel)
PacBio sequel II/ Ile RNA Library	Total RNA	≥ 800 ng	≥ 50 μL	≥ 60ng/μL	≥ 6.5	A260/280=1.8-2.2; A260/230=1.3-2.5; NC/QC ≤ 2.5

\*RIN: RNA Integrity Number

## 2. Sequencing Parameters

Platform	PacBio Sequel system
Recommended sequencing depth	≥ 15 G bases pair per sample
**Turnaround time	Within 7 weeks from project verification to data releasing without bioinformatic analysis

\*\*Turnaround time varies depending on the project volume.

## 3. Data Analysis Contents

Standard Analysis (for Species with Reference)	
(1) Data quality control	
(2) Identification, clustering and correction of full-length transcripts	
(3) Mapping reads to reference genome	
(4) Prediction and annotation (GO, Swiss-Prot) of novel transcripts	
(5) Structural Analysis of Isoforms	Fusion transcript analysis
	Alternative splicing
	Alternative polyadenylation
	TTS & TSS prediction
(6) Transcription factor analysis	
(7) lncRNA prediction	
(8) Differential expression analysis (Only for Compared Groupings)	Isoform quantification and differential expression analysis
	GO enrichment
	KEGG enrichment

Standard analysis (for Species without Reference)	
(1) Data quality control	
(2) Identification, cluster and correction of full-length transcripts	
(3) Simple Sequence Repeat (SSR) analysis	
(4) Functional annotation (via Nr, Nt, KEGG, GO, KOG, Swiss-Prot and Pfam databases)	
(5) Transcription factor analysis (only for plants and animals species)	
(6) Differential expression analysis (Only for Compared Groupings)	Isoform quantification and differential expression analysis
	GO enrichment
	KEGG enrichment

