Dataset

SEQUINS - Mix A

SIRVs - set 4

Flow cell & Library kit

- R9 + LSK109
- R9 + RNA002
- R10 + LSK114
- RP4 + RNA004

- R10 + LSK114

Molecules

cDNA
dRNA
cDNA
dRNA

cDNA

Bioinformatic steps

Raw Reads

Adapter trimming (cDNA)

Alignment

Genomic ref

Assembly

Transcriptomic ref

Assembled Transcriptome
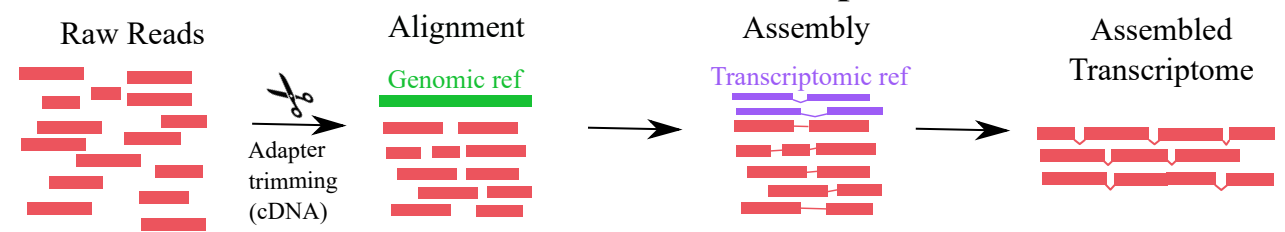
Assembling strategies & tools

Guided

Bambu ; Flair ; FLAMES ; isoQuant ; Mandalorion ; Stringtie2 ; Talon

De novo

Bambu-noRef ; Flair-noRef ; isoQuant-noRef ; Stringtie2-noRef

Ab initio

isONclust ; isONclust 2 ; RATTLE ; RNAbloom ; RNAbloom 2
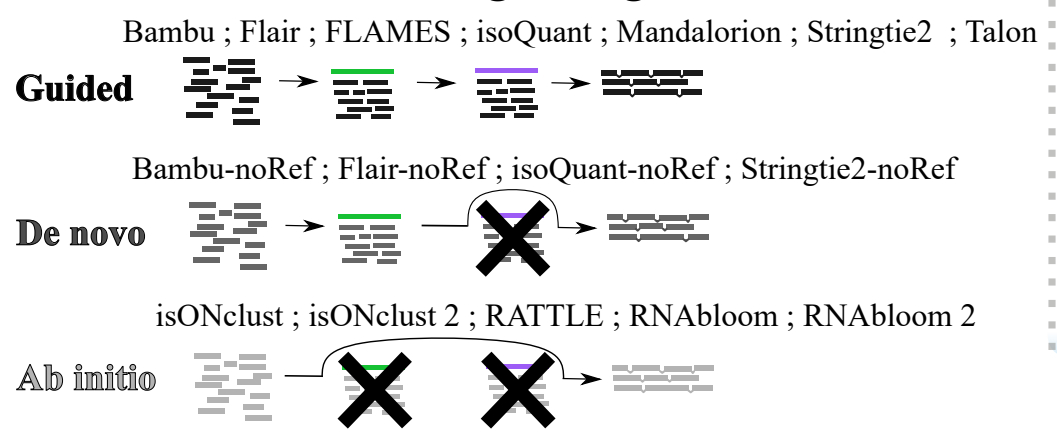
Evaluators

GFFcompare

SQANTI3

Transcript Classification	Reference Transcript	**GFFcompare Class codes**	**SQANTI3 Classification**
Novel transcript		u (or unknown)	Intergenic
True positive (TP)		= (or complete)	Full Splice Match (FSM)
		c (or contained in ref.)	Incomplete Splice Match (ISM) (except intron retention)
False positive (FP)		Other class-codes / classification titles	