Introduction to sequencing technologies

Katarzyna Sikora

Overview

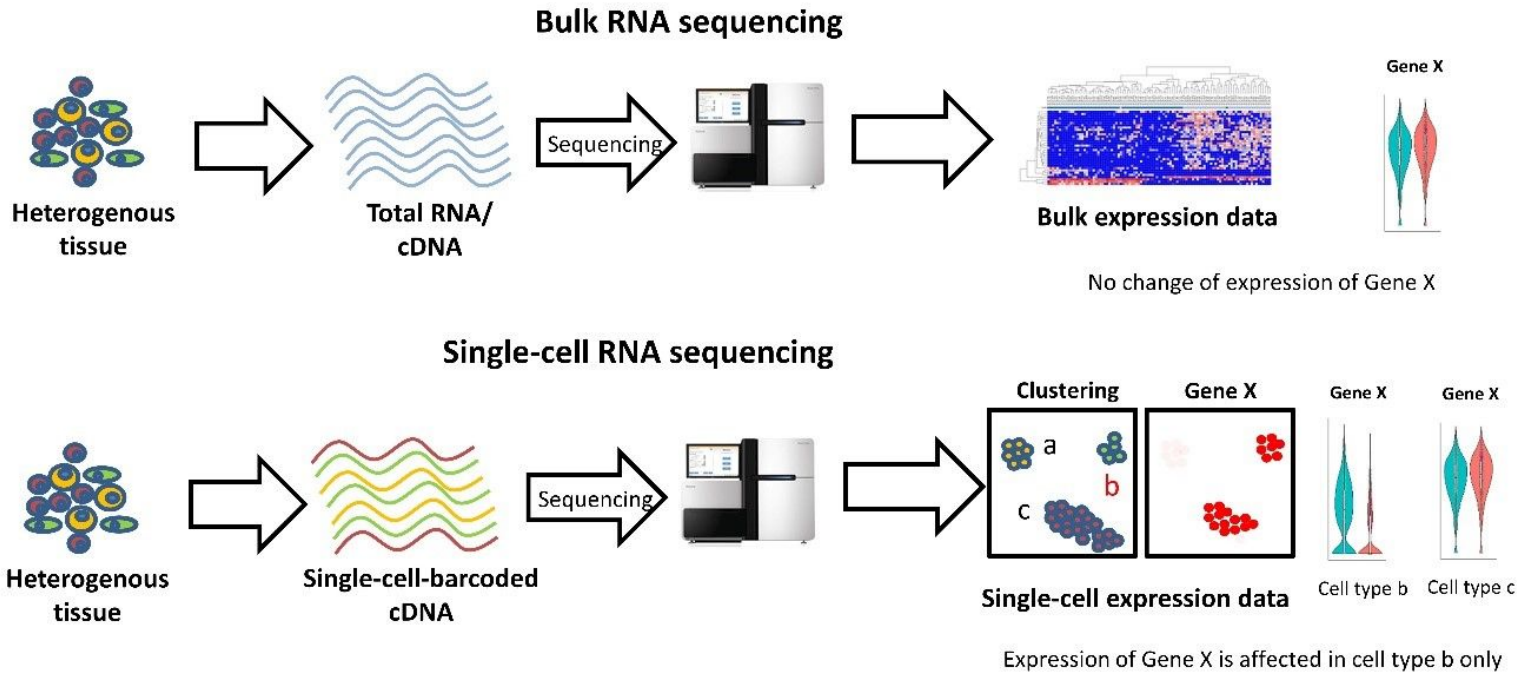
- advantages/disadvantages of scRNAseq
- modern single cell sequencing technologies
- balanced experimental design

Advantages and disadvantages of scRNAseq

- evaluate gene expression heterogeneity
- discover rare cell types
- calculate cell differentiation trajectories
- single cell omics
- spatial transcriptomics
- RNA splicing velocity
- whole organism single cell atlases

- low input material
- high dropout rates
- cell duplets
- computational artefacts in pseudotime, RNA velocity, UMAP projections, trajectory calculations etc.
- limited power to differentiate closely related cell types given only gene expression

Cell-type-specific gene expression



Modern single cell technologies

Platform Name	Separation Method	Amplification Method	Using UMI	Amplification Range	Advantages	Disadvantages	Release Date	References
VASA-seq	FANS	PCR	YES	All transcripts	Low cost and accurate dosing	/	2022	[10]
Smart-seq3	Microfluidics	PCR	YES	5′ end	High sensitivity	Time-consuming	2020	[11,12]
DNBelabC4	Microfluidics	PCR	YES	All transcripts	Precise quantification	/	2019	[13]
Seq-Well	Microfluidics	PCR	YES	3′ end	Low cost and precise quantification	Unsuitable for variable splicing and allelic expression	2017	[14]
MATQ-seq	FACS	PCR	YES	All transcripts	Precise quantification	Low cell throughput	2017	[15]
10× Genomics	Microfluidics	PCR	YES	3′ end	High cell capture efficiency, fast cycle time, high cell suitability, and reproducibility	Sequencing can be performed only for the 3′ end	2016	[16]

Wang et al., Int J Mol Sci. 2023

Balanced experimental design