# Cornell Notes – RNA-Seq Lecture (Prof. Jun Wang, 2025/2026)

This document follows the Cornell note-taking format with three sections: Cues/Keywords, Notes, and Summary. Based on uploaded lecture slides.

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| Cues / Keywords | Notes |
| RNA-seq definition | Sequencing of mRNA to study transcriptome; powerful NGS tool. |
| Bulk RNA-seq vs scRNA-seq | Bulk: average expression across all cells. scRNA-seq: captures heterogeneity and microenvironment at single-cell resolution. |
| Poly-A selection vs ribosomal depletion | Poly-A: captures coding RNAs. Ribosomal depletion: retains lncRNAs and non-coding RNAs. |
| Applications of RNA-seq | Differential expression, alternative splicing, gene fusion, novel transcripts, RNA-level variants, allelic expression, co-expression networks. |
| Analysis methods | Sequence alignment, expression analysis, PCA/clustering, pathway analysis (DAVID, GSEA), IGVtools visualization. |
| Pathways (example) | Cell cycle downregulated; oxidative phosphorylation upregulated under estrogen treatment in breast cancer cells. |
| lncRNAs | Long noncoding RNAs >200nt, low expression, regulatory roles; example MALAT1 linked to metastasis and poor prognosis. |
| scRNA-seq applications | Study tumour microenvironment (immune/stroma), tumour heterogeneity, resistant clones, trajectory analysis. |
| Challenges | Expensive, preparation-sensitive, requires optimization. |

## Summary

RNA-seq enables comprehensive analysis of the transcriptome, identifying gene expression changes, splicing events, fusions, variants, and noncoding RNAs. Bulk RNA-seq provides averaged data, while scRNA-seq uncovers cellular heterogeneity and tumour microenvironment composition. Applications include cancer research, pathway analysis, and biomarker discovery.