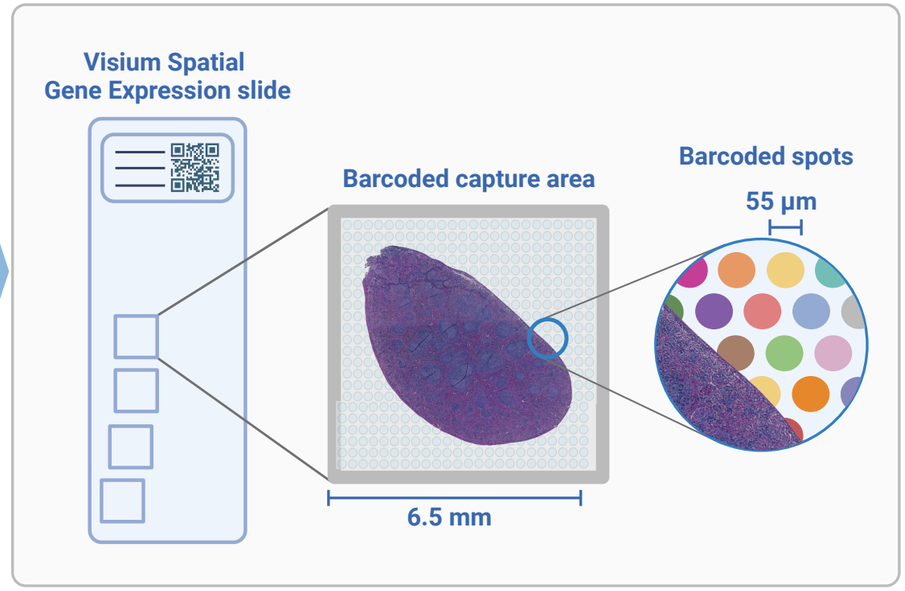
Experimental design

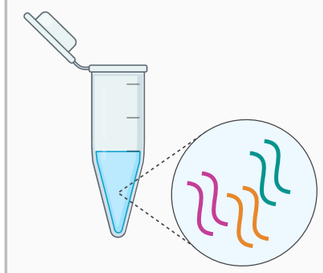
Male	CTL	173 174			
	KO	167 168			
Female	WT	544 545 708 709	460 462 463 708* 709* 710 713*	709 713	709 713
		OCT	FFPE	OCT CA	FFPE CA
		Poly-A	Probe		

SpatialBench

Sample preparation: hybridisation, ligation and barcoding

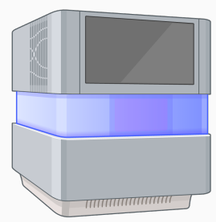


Library construction



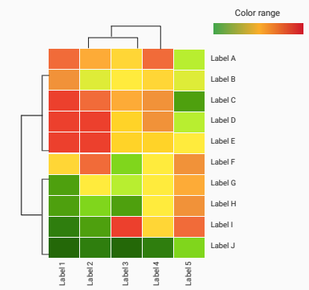
Sequencing

Illumina
NextSeq 2000



Data analysis

Space Ranger 2.0.0



1. Quality control

- Keep spots only over tissue
- Calculate per-spot QC metrics and identify outliers
- Visualise spots to be discarded
- Remove low quality spots
- Normalisation

scater scran

2. Feature selection

- Identify highly variable genes (HVGs)
- Identify spatially variable genes (SVGs)

scran nnSVG

3. Dimensionality reduction

- PCA
- UMAP

scater

4. Downstream analysis

- Clustering
- Marker gene detection
- Cell type deconvolution
- Visualisation

scran spacexr iSC.MEB