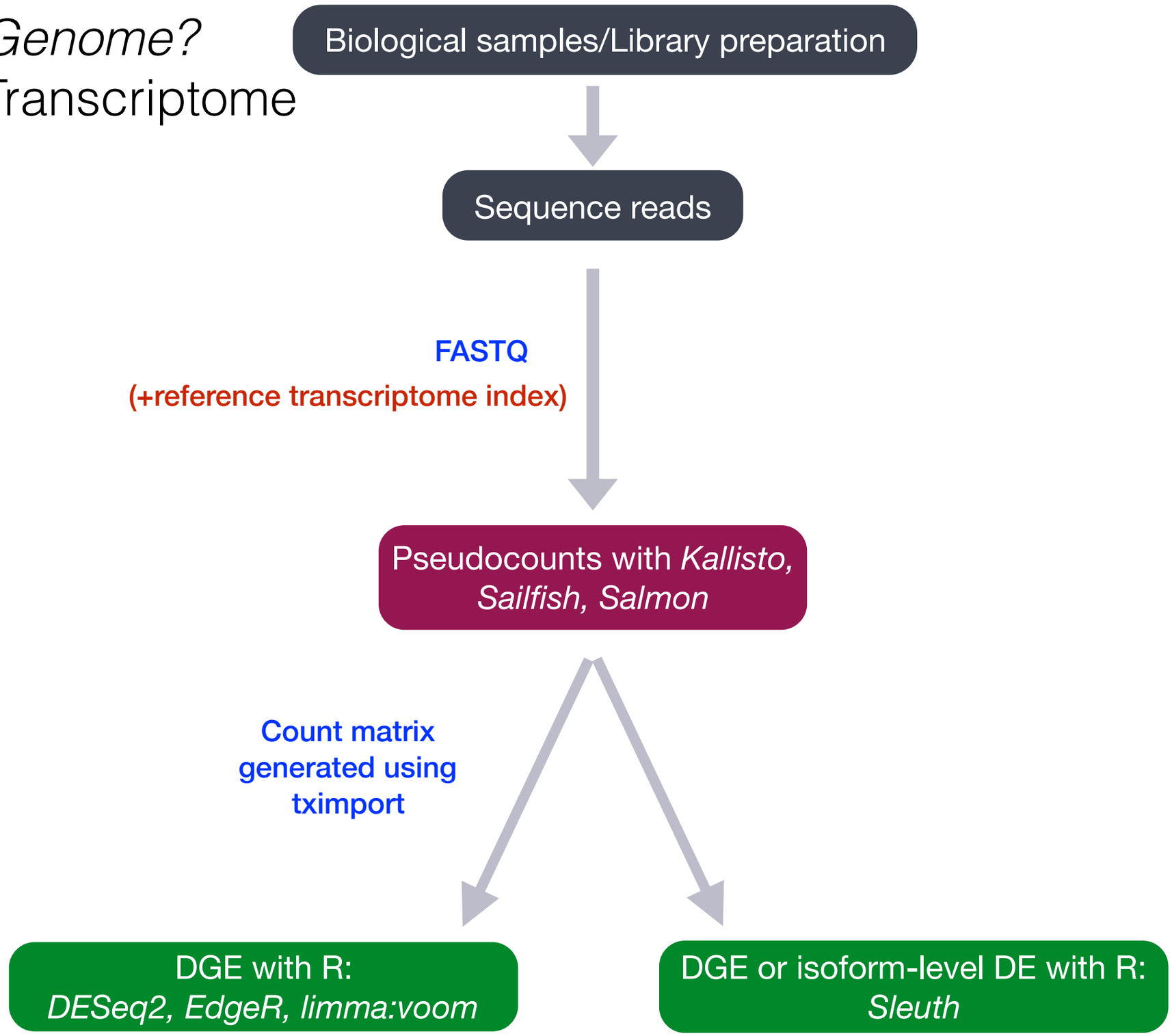
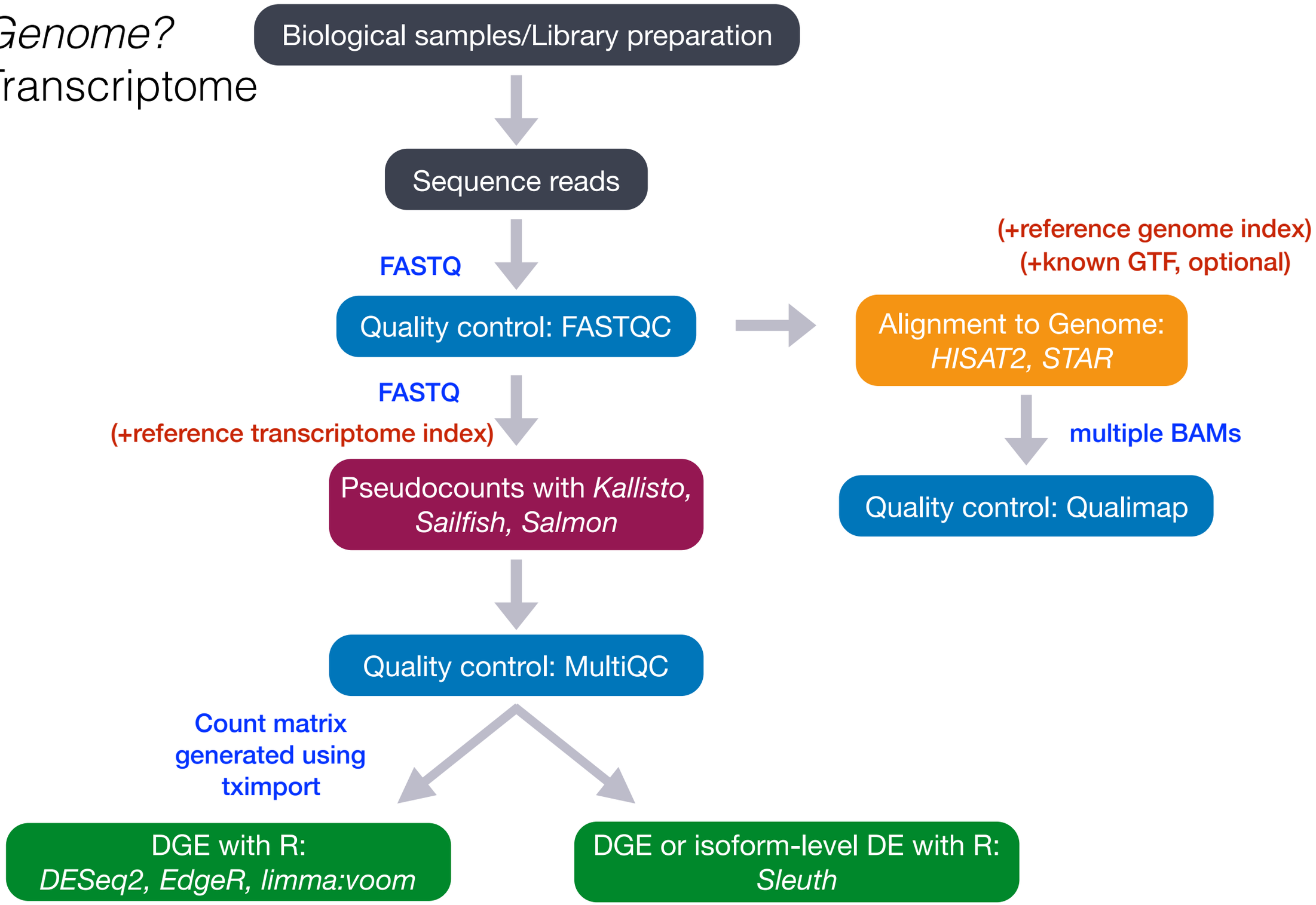
RNA-seq: Analysis options

- ✓ *Genome?*
- ✓ Transcriptome



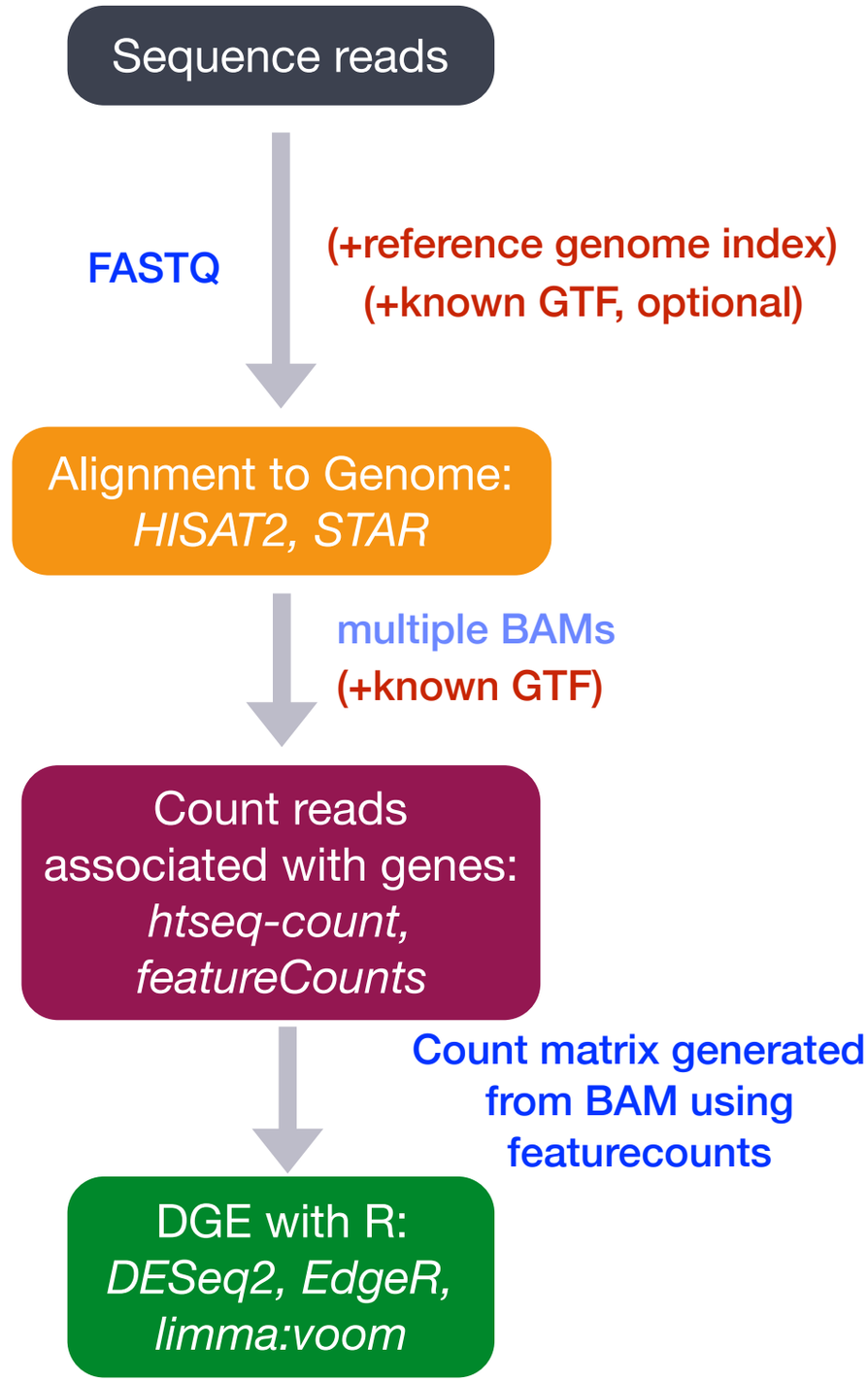
Differential Expression Analysis Workflow #1

- ✓ *Genome?*
- ✓ Transcriptome



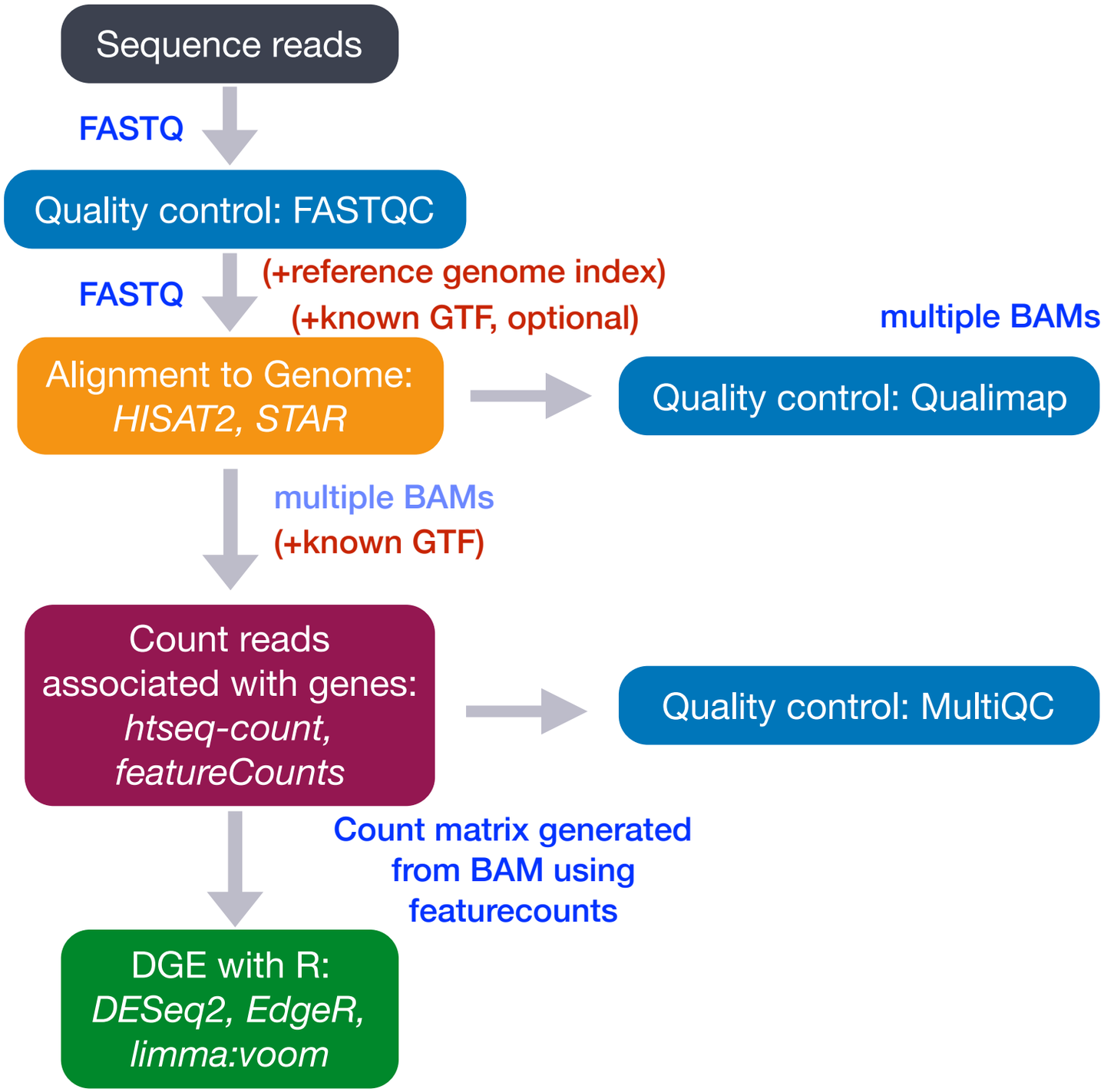
Differential Expression Analysis Workflow #1

- ✓ Genome
- ✓ GTF annotation file (transcriptome)



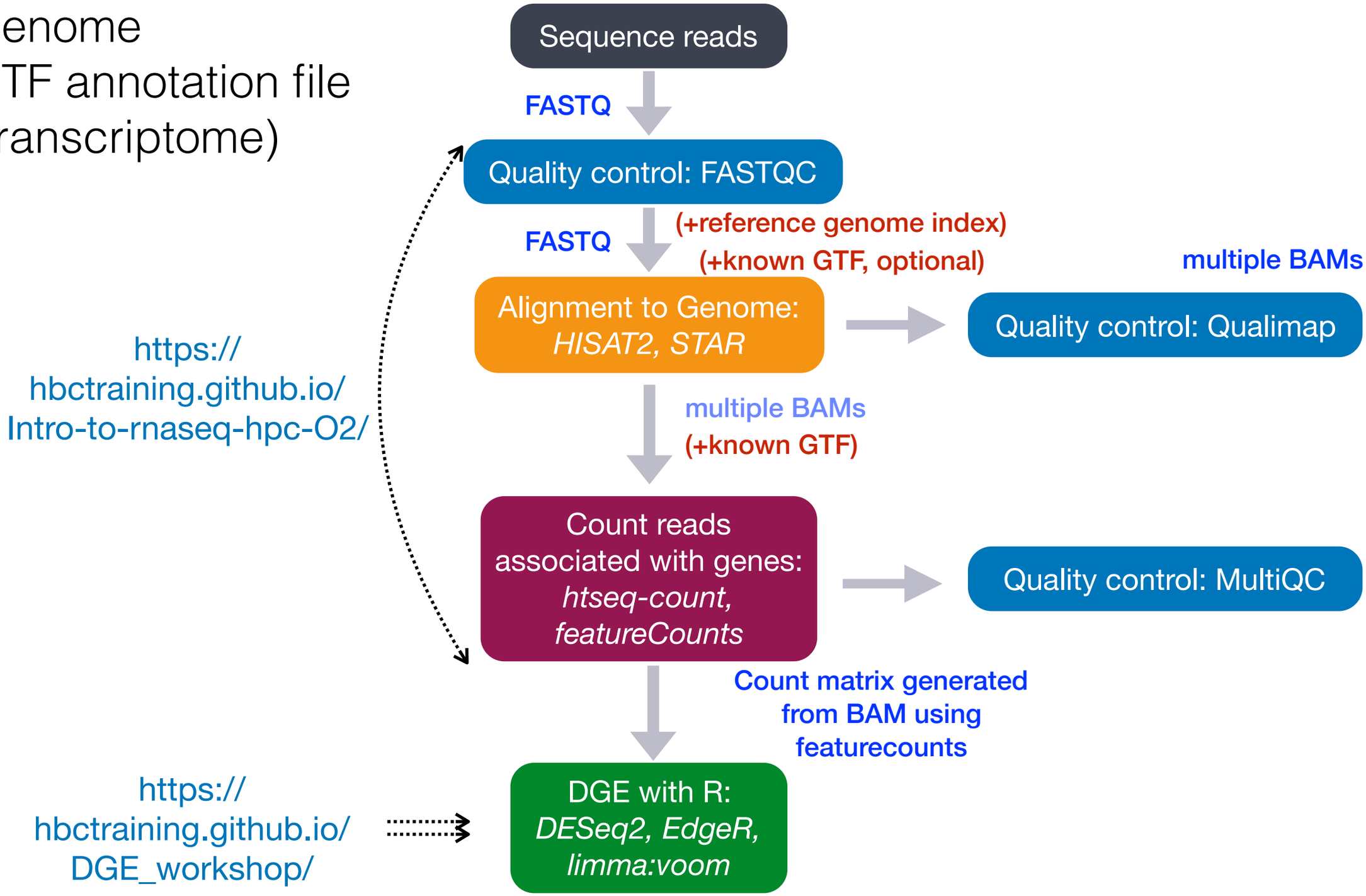
Differential Expression Analysis Workflow #2

- ✓ Genome
- ✓ GTF annotation file (transcriptome)



Differential Expression Analysis Workflow #2

- ✓ Genome
- ✓ GTF annotation file (transcriptome)



https://hbctraining.github.io/Intro-to-rnaseq-hpc-O2/

https://hbctraining.github.io/DGE_workshop/

Differential Expression Analysis Workflow #2

Alternative methods: transcriptome assembly

Reference-based assembly

- Genome is known

Alternative methods: transcriptome assembly

Reference-based assembly

- Genome is known
- Transcriptome not available or is not good enough

Alternative methods: transcriptome assembly

Reference-based assembly

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- Cufflinks and Scripture are two reference-based transcriptome assemblers

Alternative methods: transcriptome assembly

Reference-based assembly

- Genome is known
- Transcriptome not available or is not good enough
- Cufflinks and Scripture are two reference-based transcriptome assemblers
- Additional annotation of any newly-discovered genes or isoforms will need to be generated

Alternative methods: transcriptome assembly

De novo assembly

- Genome is not known, or is of poor quality

Alternative methods: transcriptome assembly

De novo assembly

- Genome is not known, or is of poor quality
- Amount of data needed is greater than for a reference-based assembly

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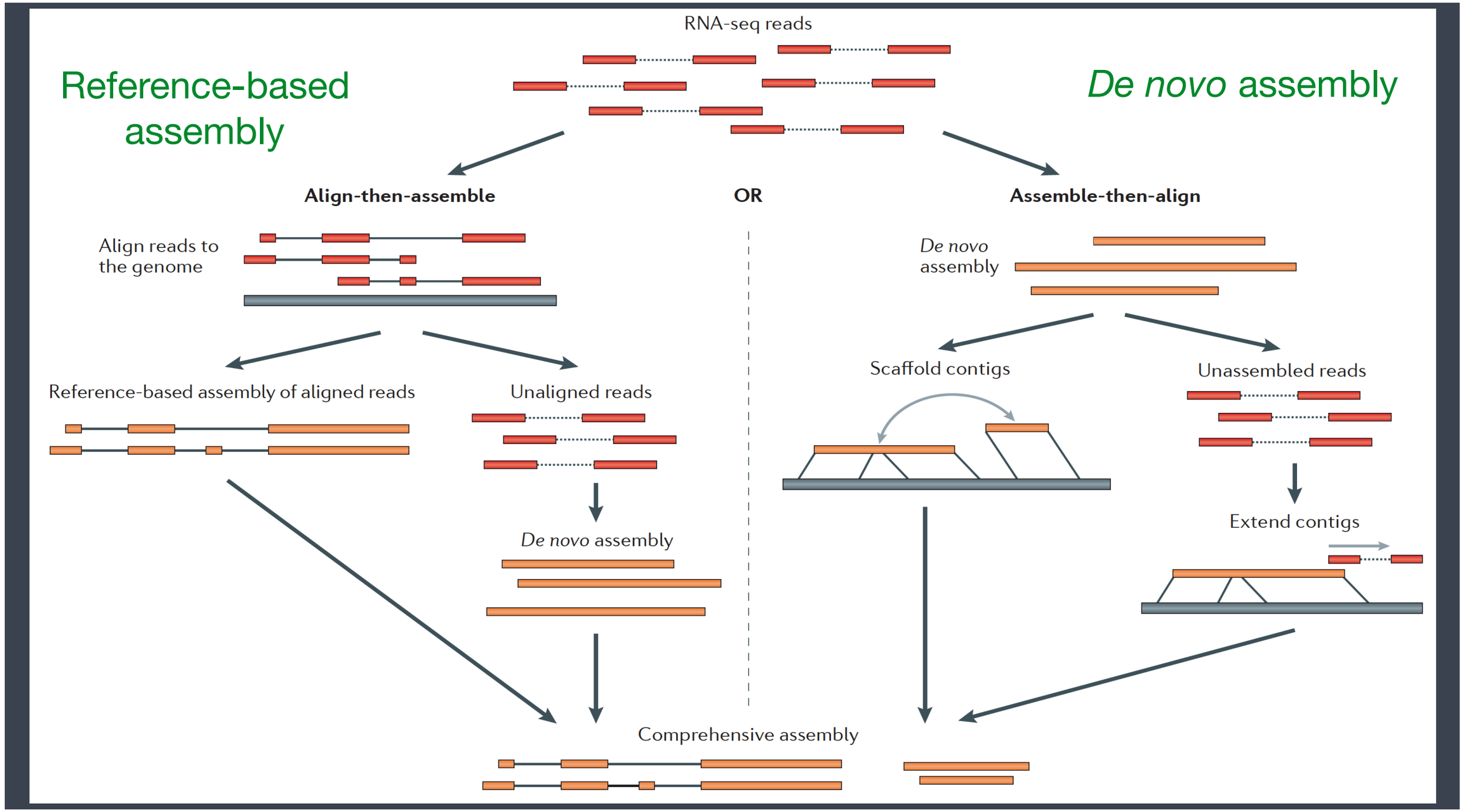
- Oases, TransABySS, Trinity are examples of well-regarded transcriptome assemblers, **especially Trinity**

Alternative methods: transcriptome assembly

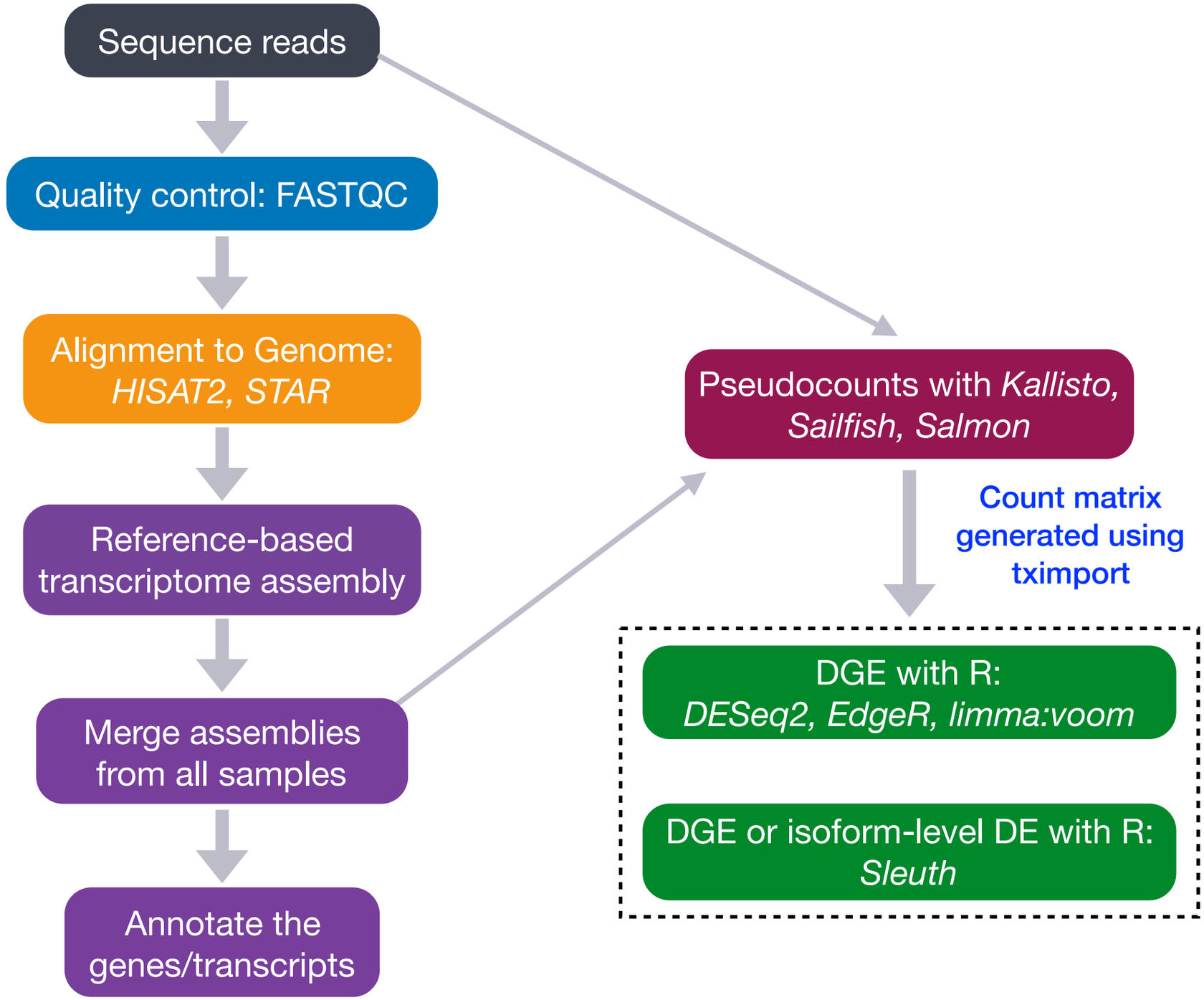
De novo assembly

- Genome is not known, or is of poor quality
- Amount of data needed is greater than for a reference-based assembly
- Oases, TransABySS, Trinity are examples of well-regarded transcriptome assemblers, **especially Trinity**
- Newly-discovered genes or isoforms will need to be annotated using homolog-based and other methodologies

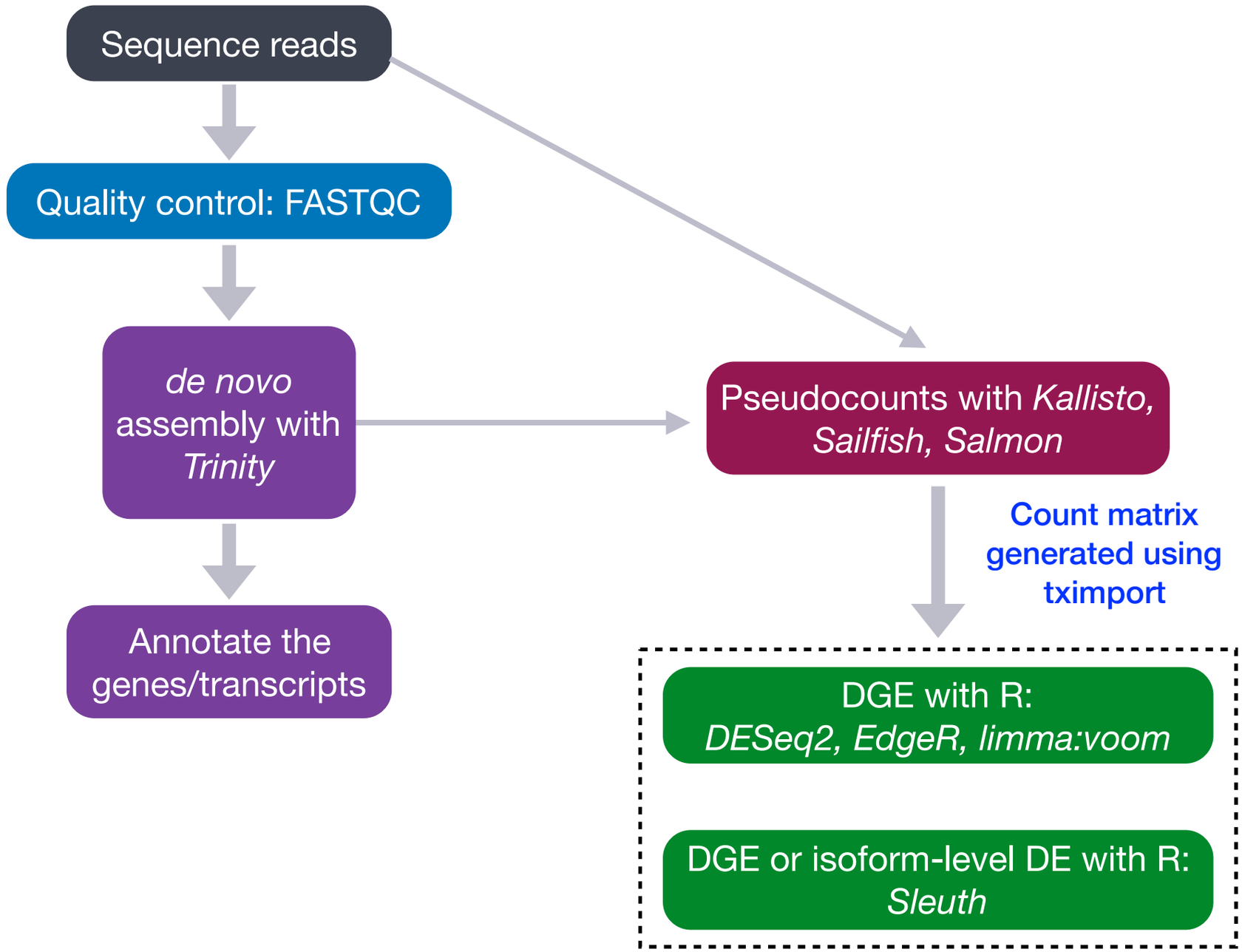
Transcriptome Assembly



Martin J.A. and Wang Z., Nat. Rev. Genet. (2011) 12:671–682



Differential Expression Analysis Workflow #3



Differential Expression Analysis Workflow #4