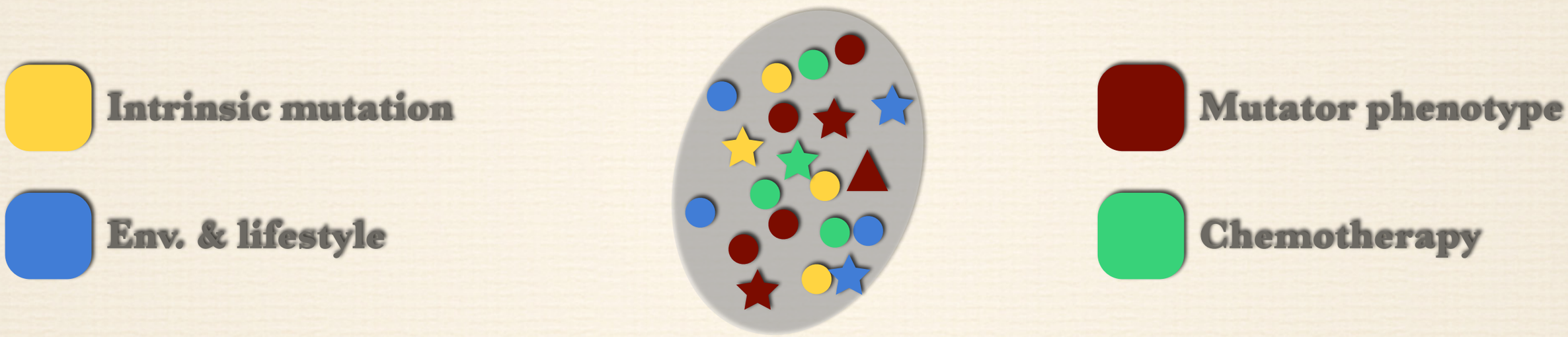
Tumor Heterogeneity Workshop

Part 1 - Concepts

Yi Qiao, 2018

Tumorigenesis

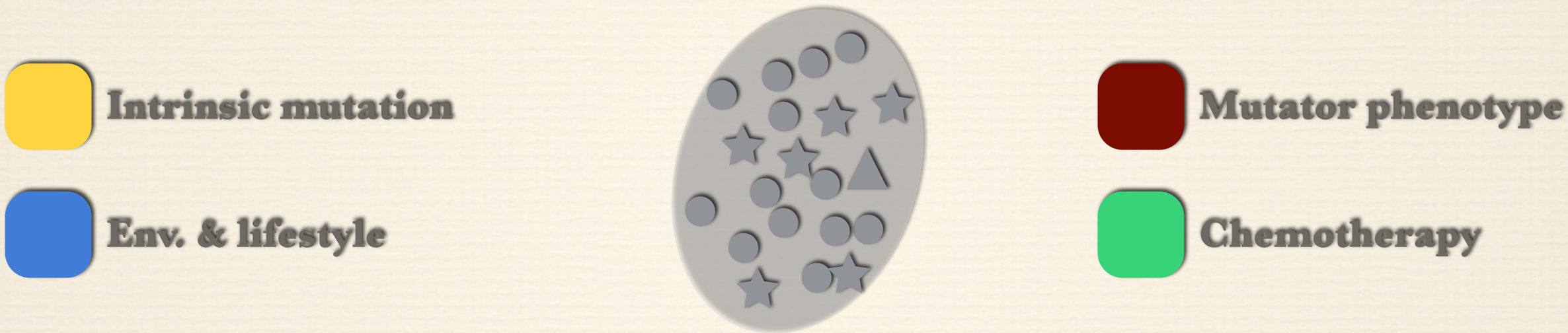
As a result of somatically acquired mutations



Chemotherapy-resistant recurrence

Tumorigenesis

As a result of somatically acquired mutations

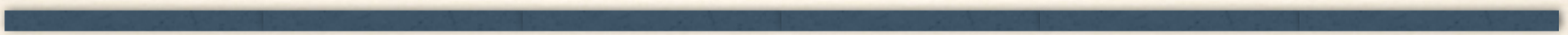


Chemotherapy-resistant recurrence

Cancer Genomics

Massive Parallel NGS

Genomic DNA



Cancer Genomics

Massive Parallel NGS

Cancer Genomics

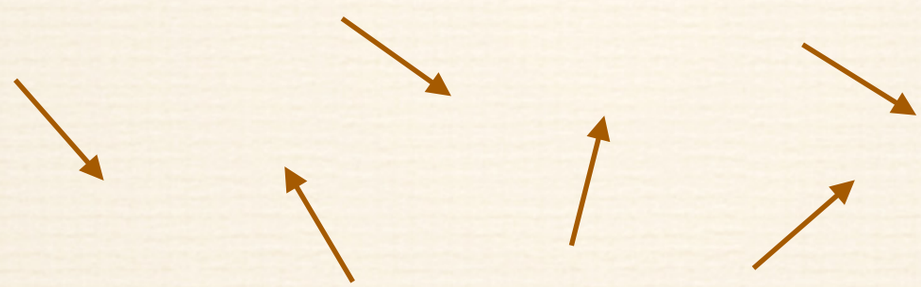
Massive Parallel NGS

Sequence in Parallel

Cancer Genomics

Massive Parallel NGS

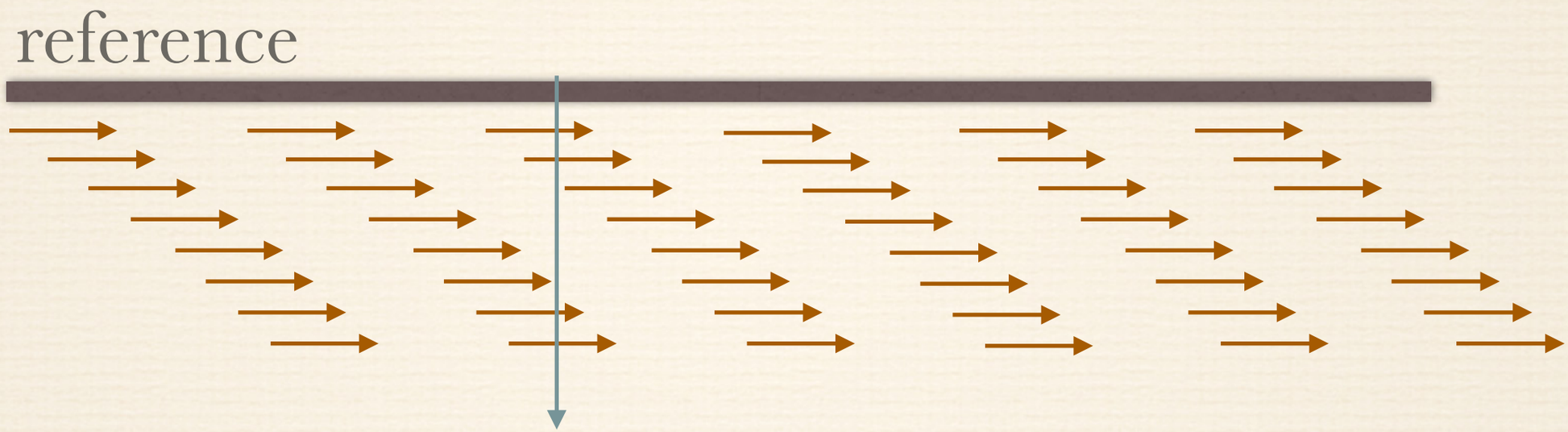
Short Sequencing Reads



Cancer Genomics

Massive Parallel NGS

Alignment

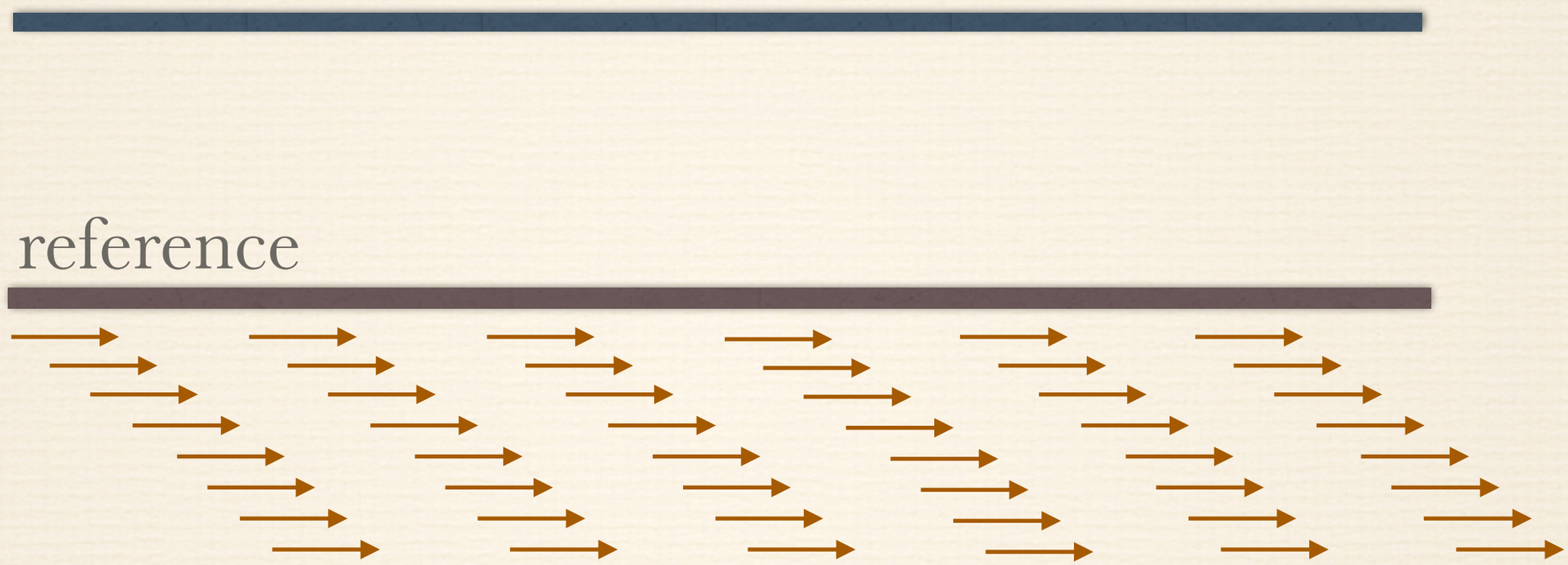


Coverage: 4x

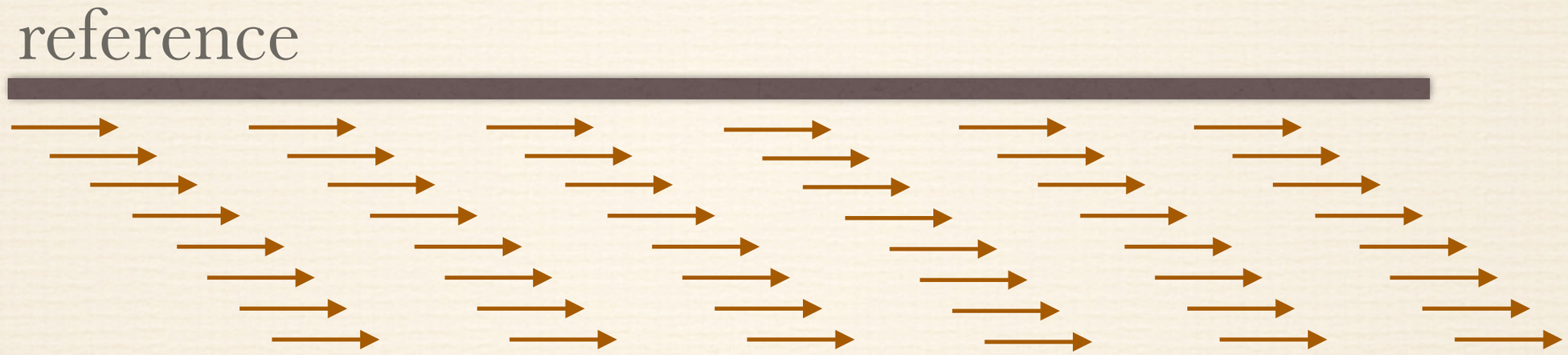
Normal Contamination

- Normal genomes that went into sequencing together with the tumor samples
- Signal: Lack of somatic mutations
- Entry Point: Copy number variation

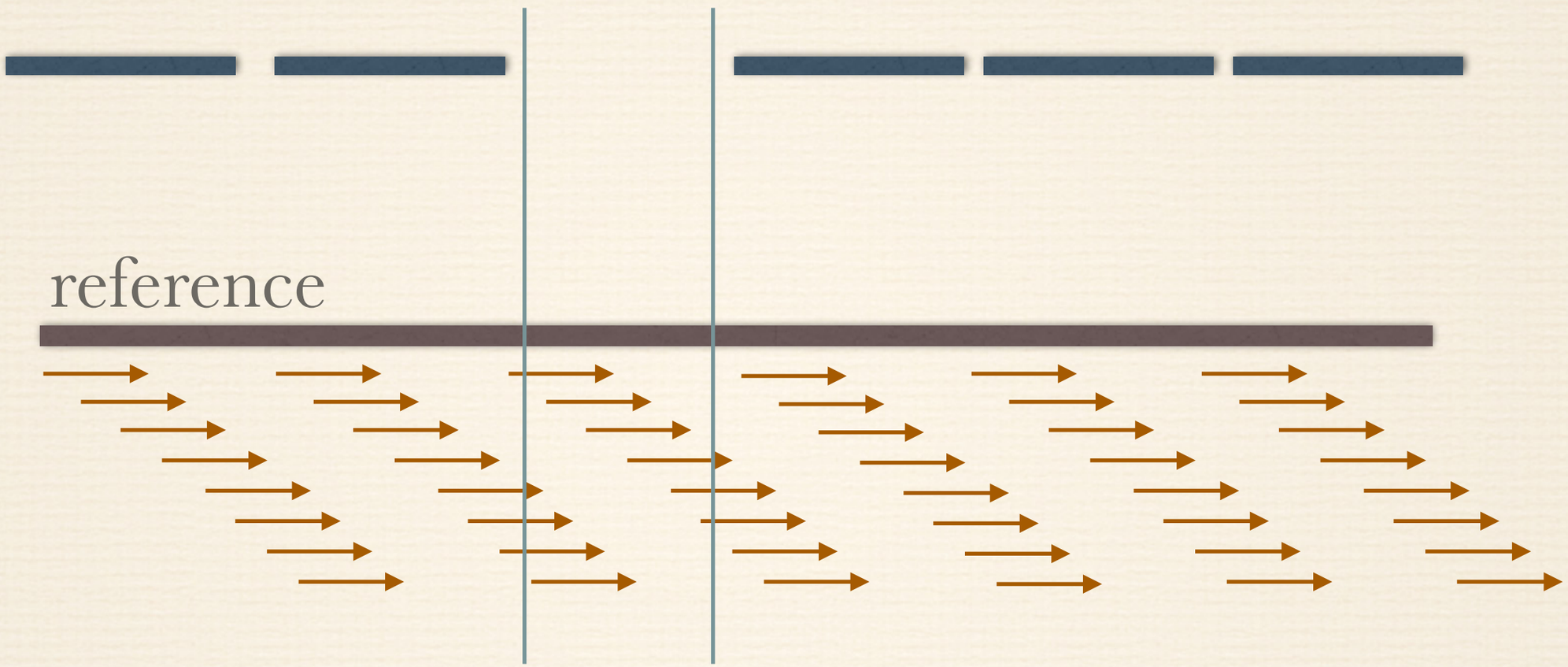
Somatic Copy Number Variation



Somatic Copy Number Variation



Somatic Copy Number Variation



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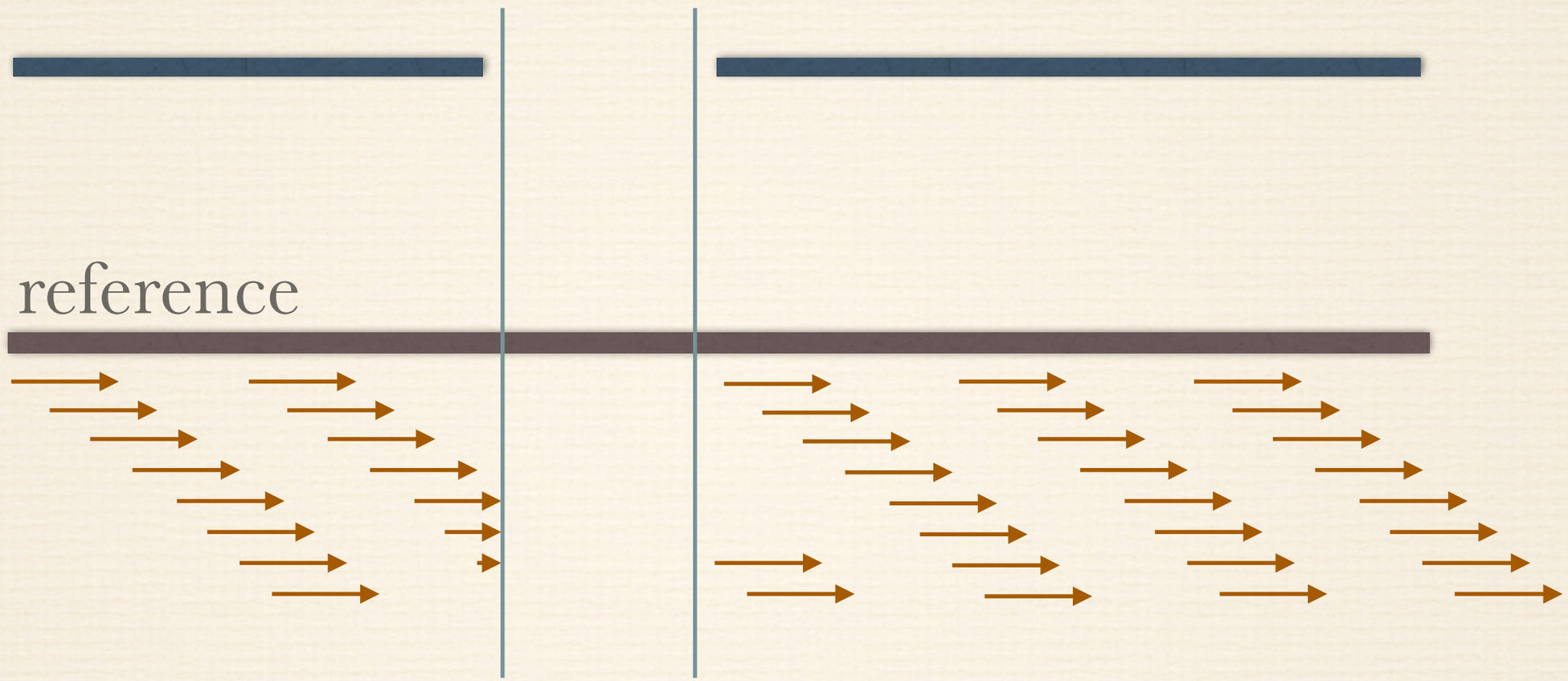
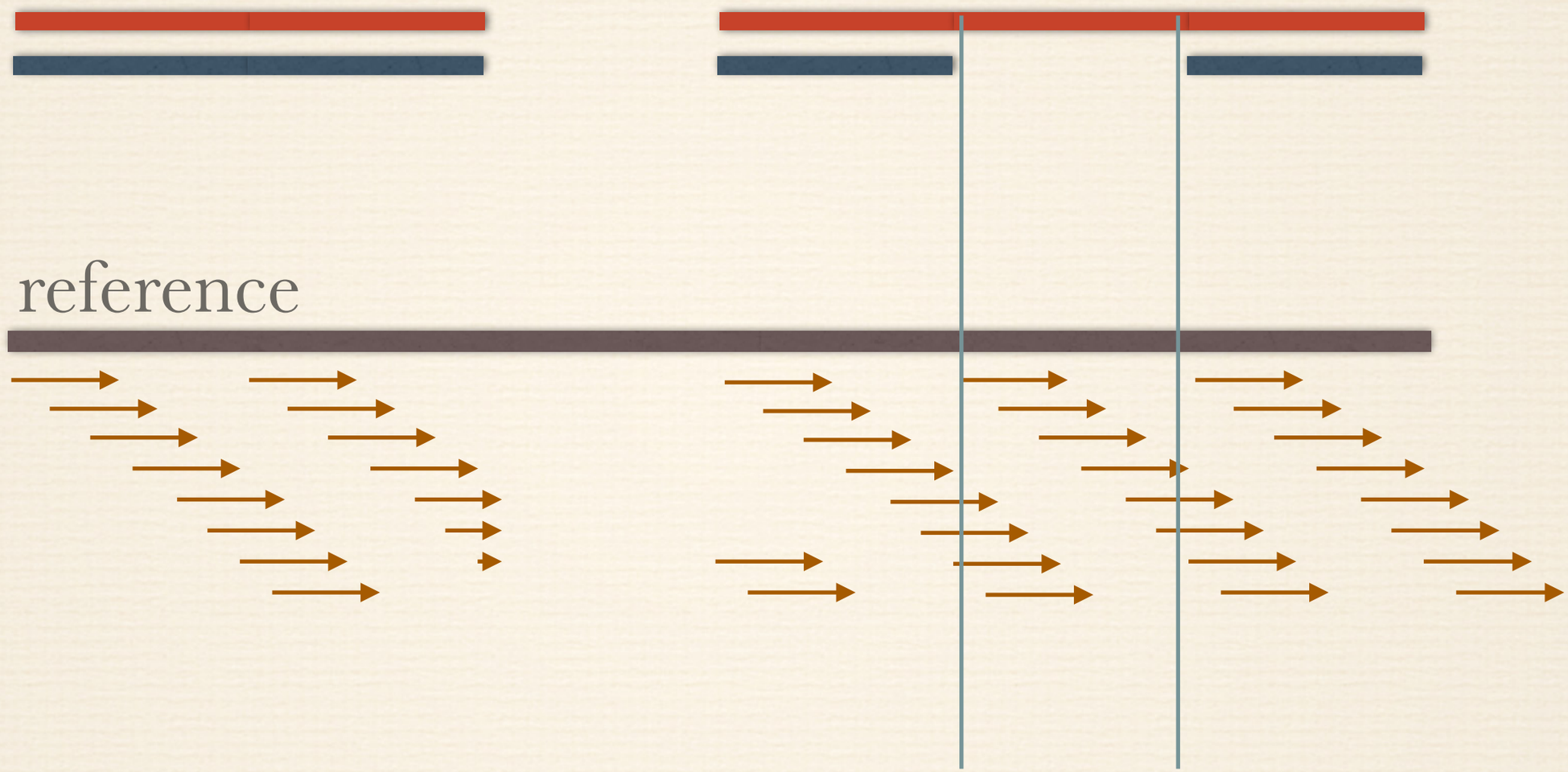
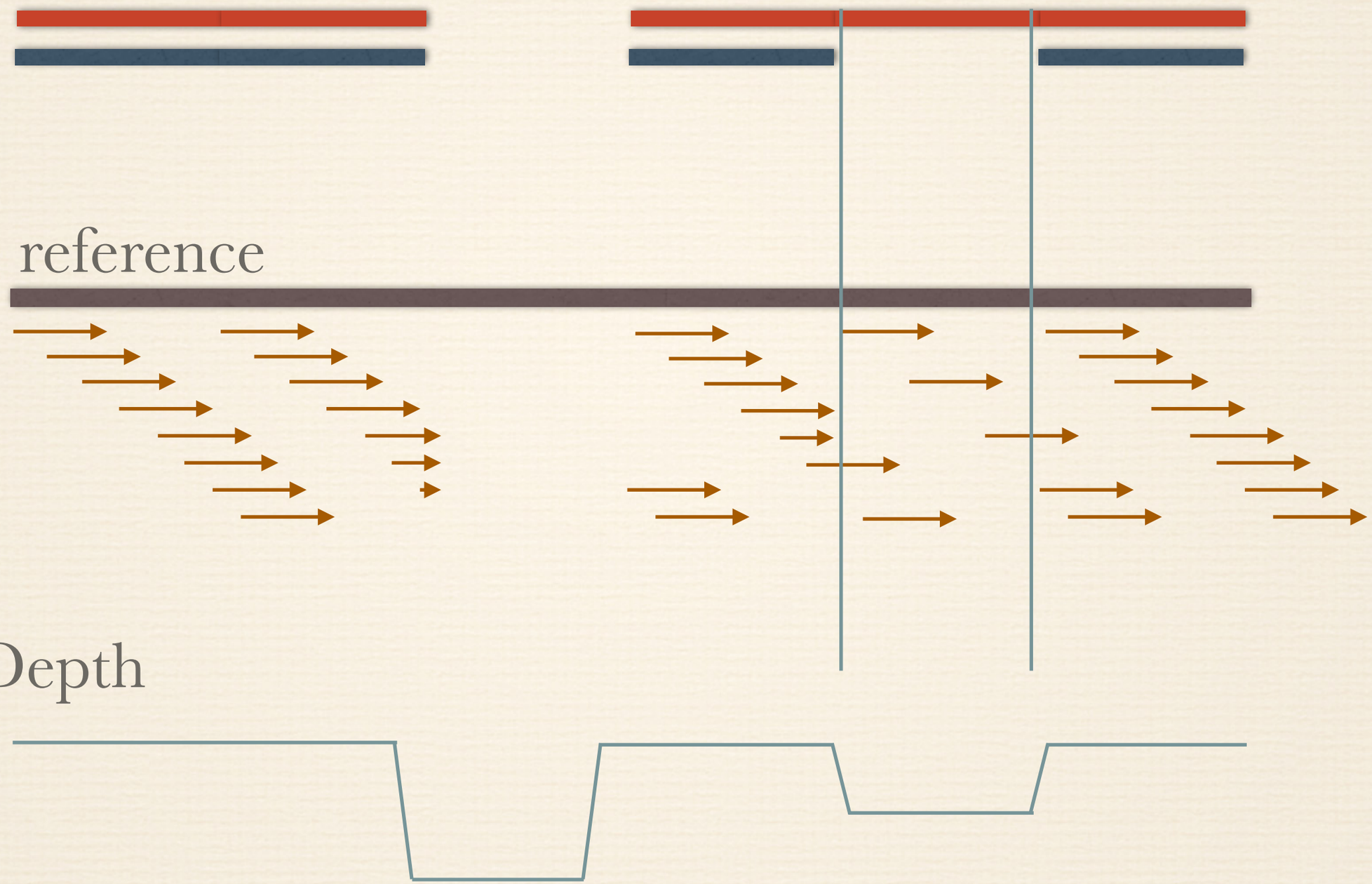


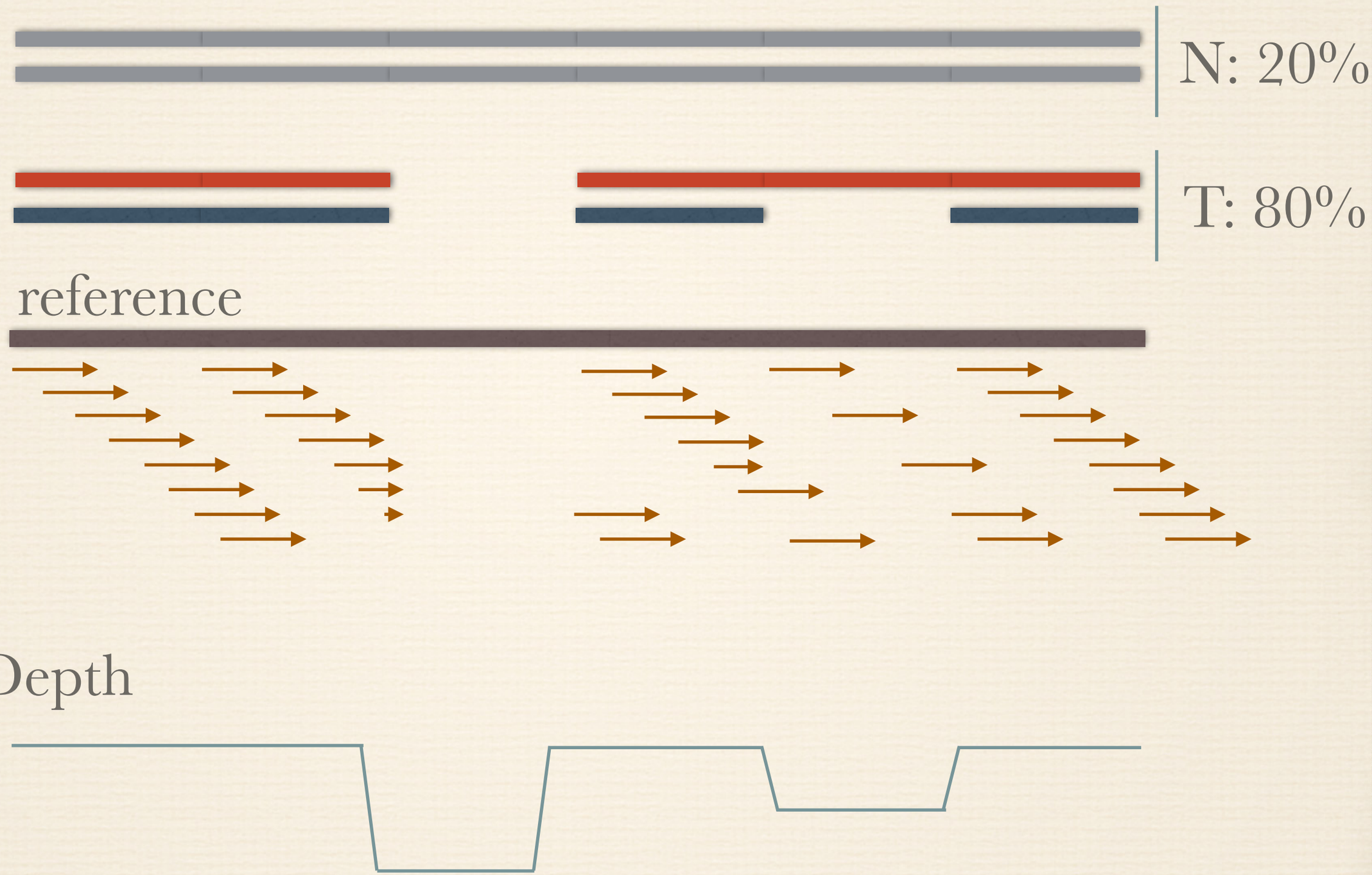
Somatic Copy Number Variation



Somatic Copy Number Variation



Somatic Copy Number Variation



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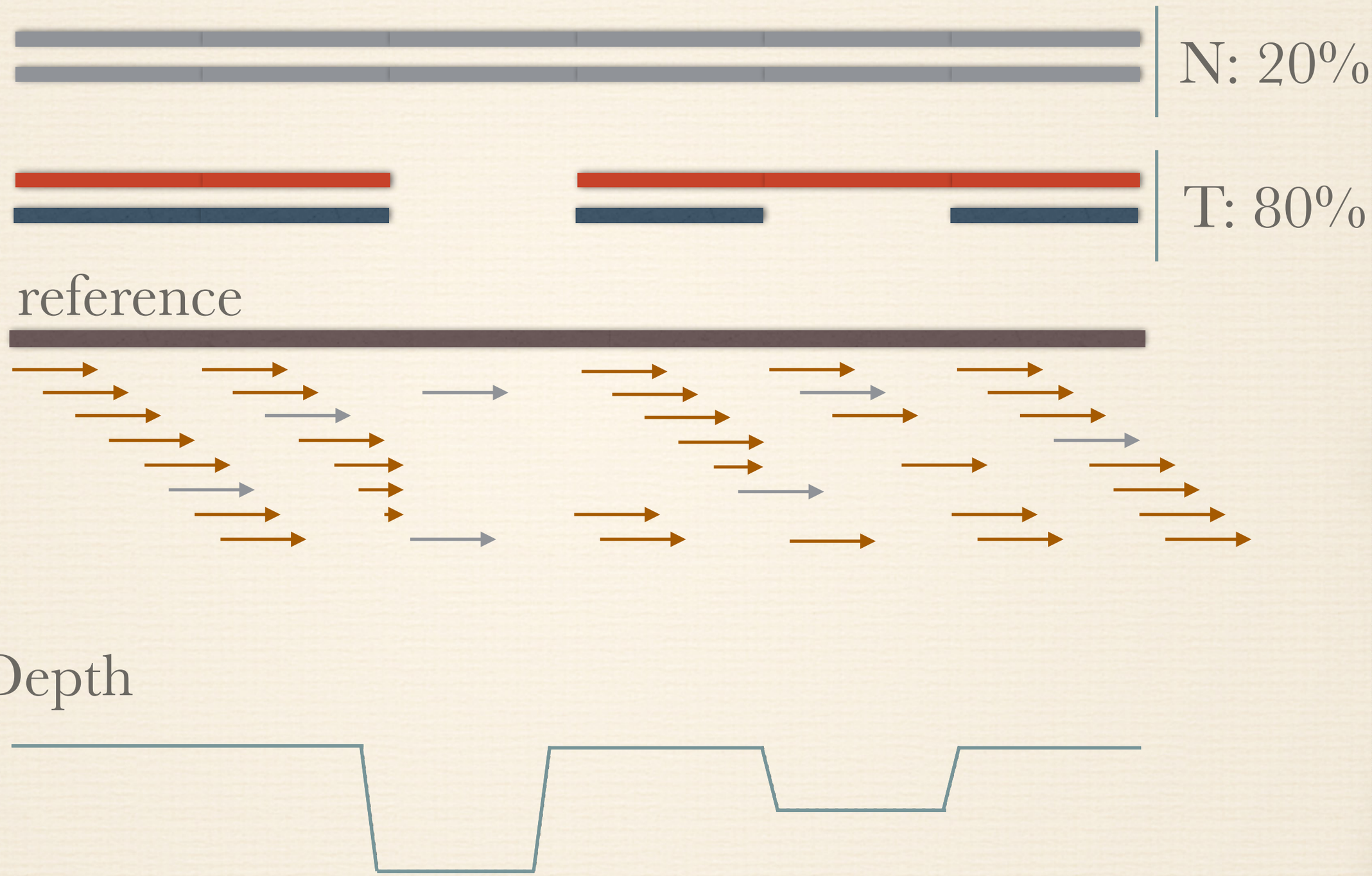
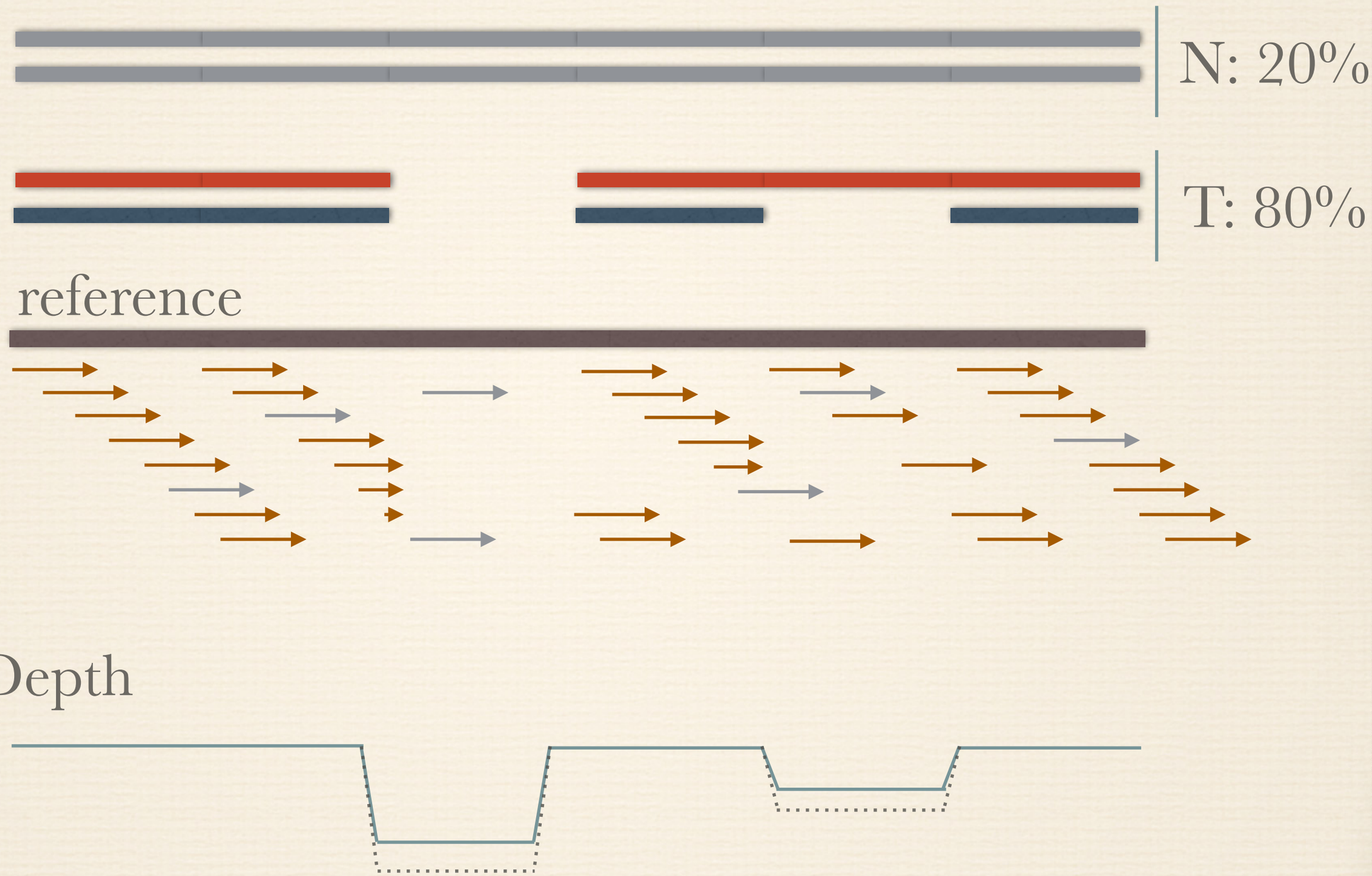
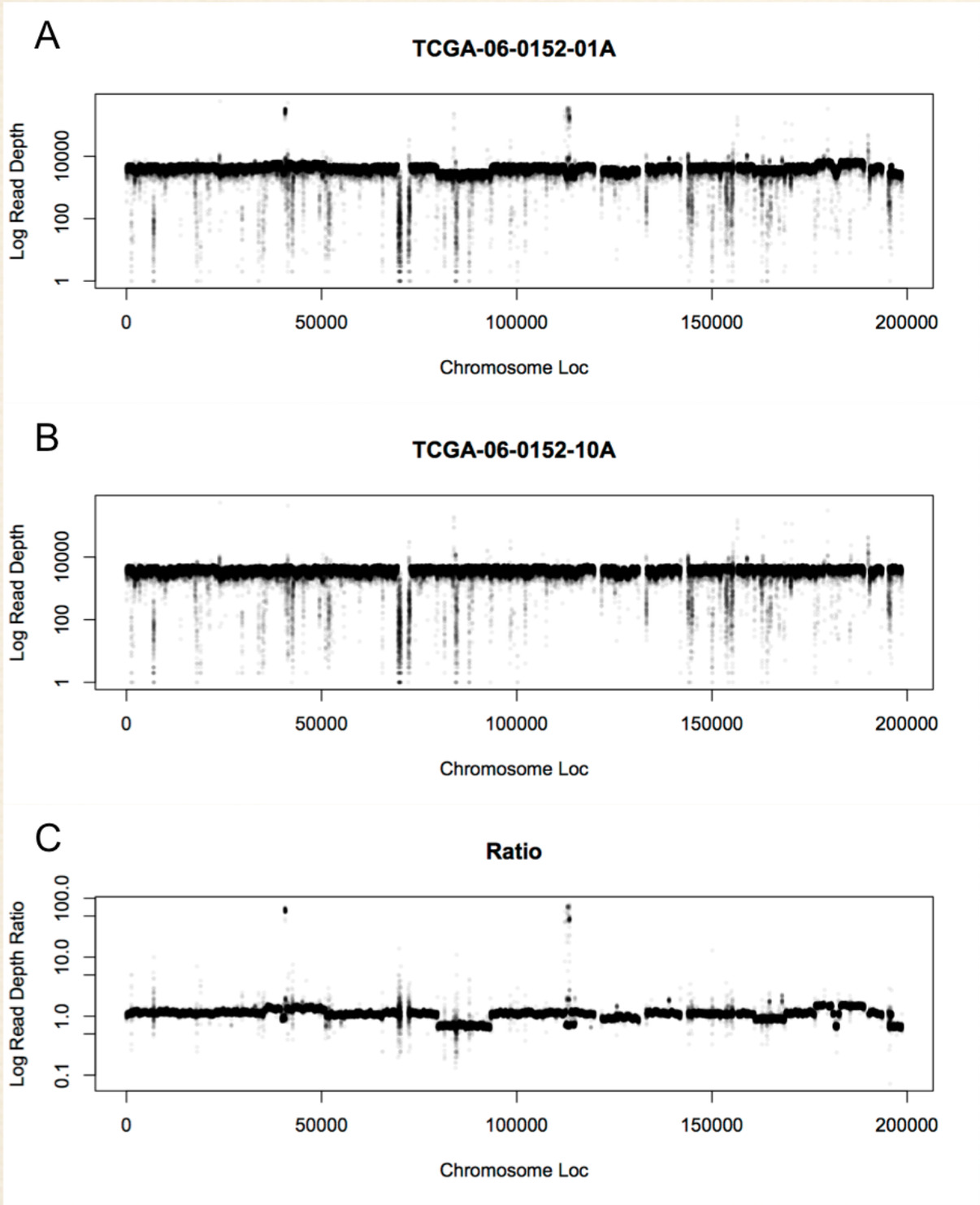


Somatic Copy Number Variation



Normal Cont. Estimate

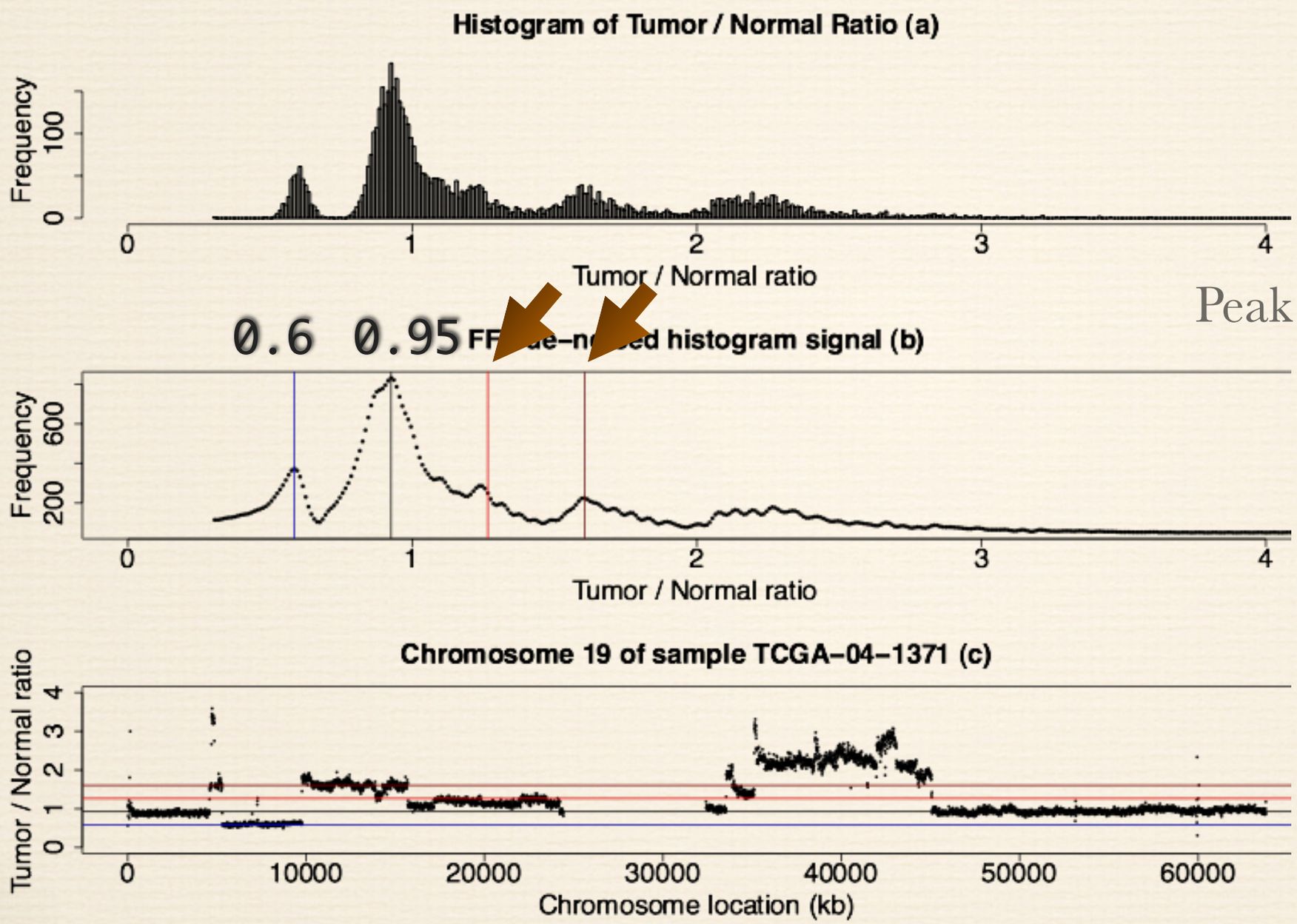


Tumor Read Depth

Normal Read Depth

Tumor to Normal ratio

Normal Cont. Estimate



Let $\alpha \in (0, 1)$ be the normal component

$$\text{Peak1} = \frac{1*(1-\alpha) + 2*\alpha}{2}$$

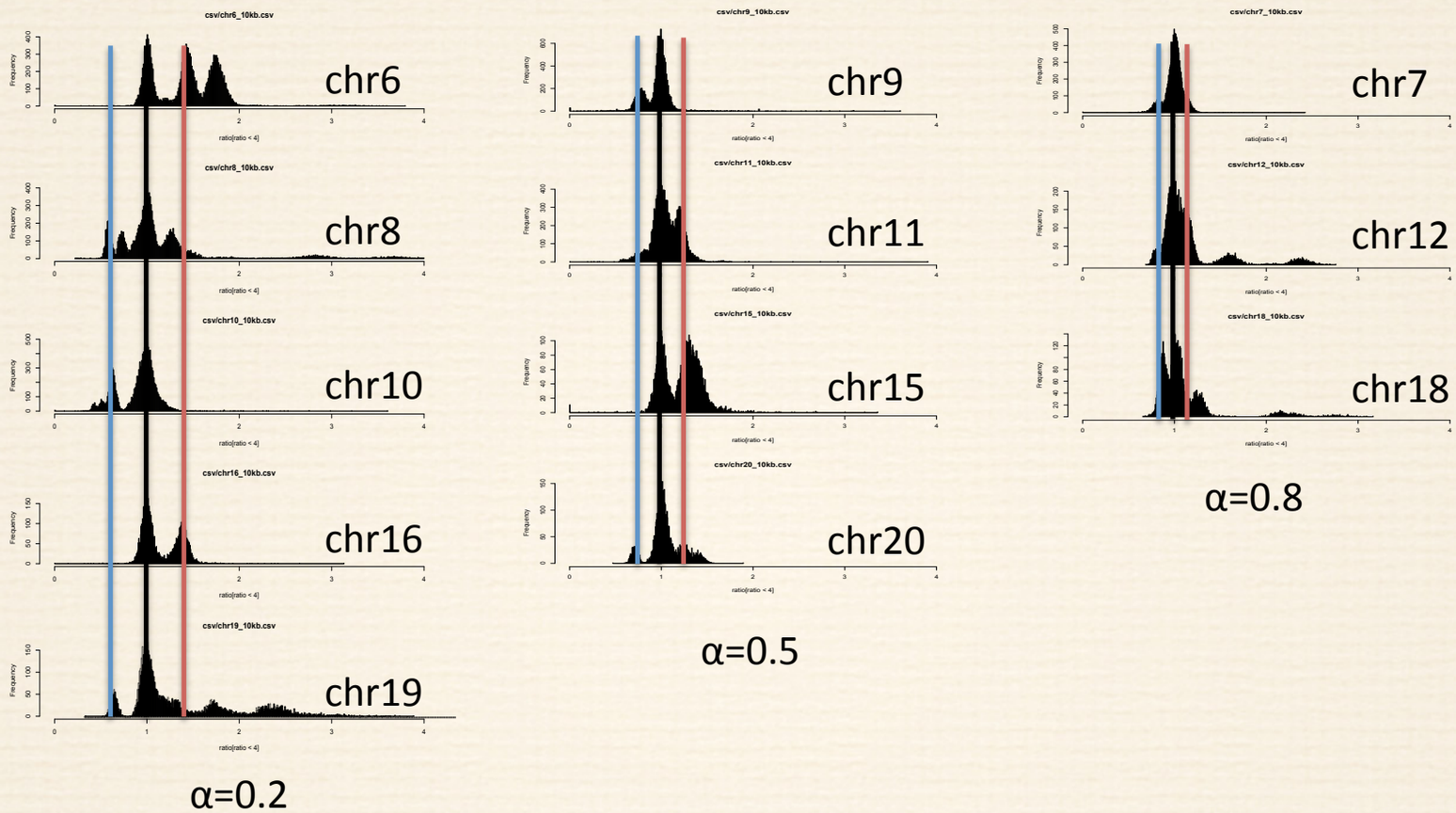
$$= 0.6 / 0.95$$

$$\alpha = 2*(0.6/0.95) - 1$$

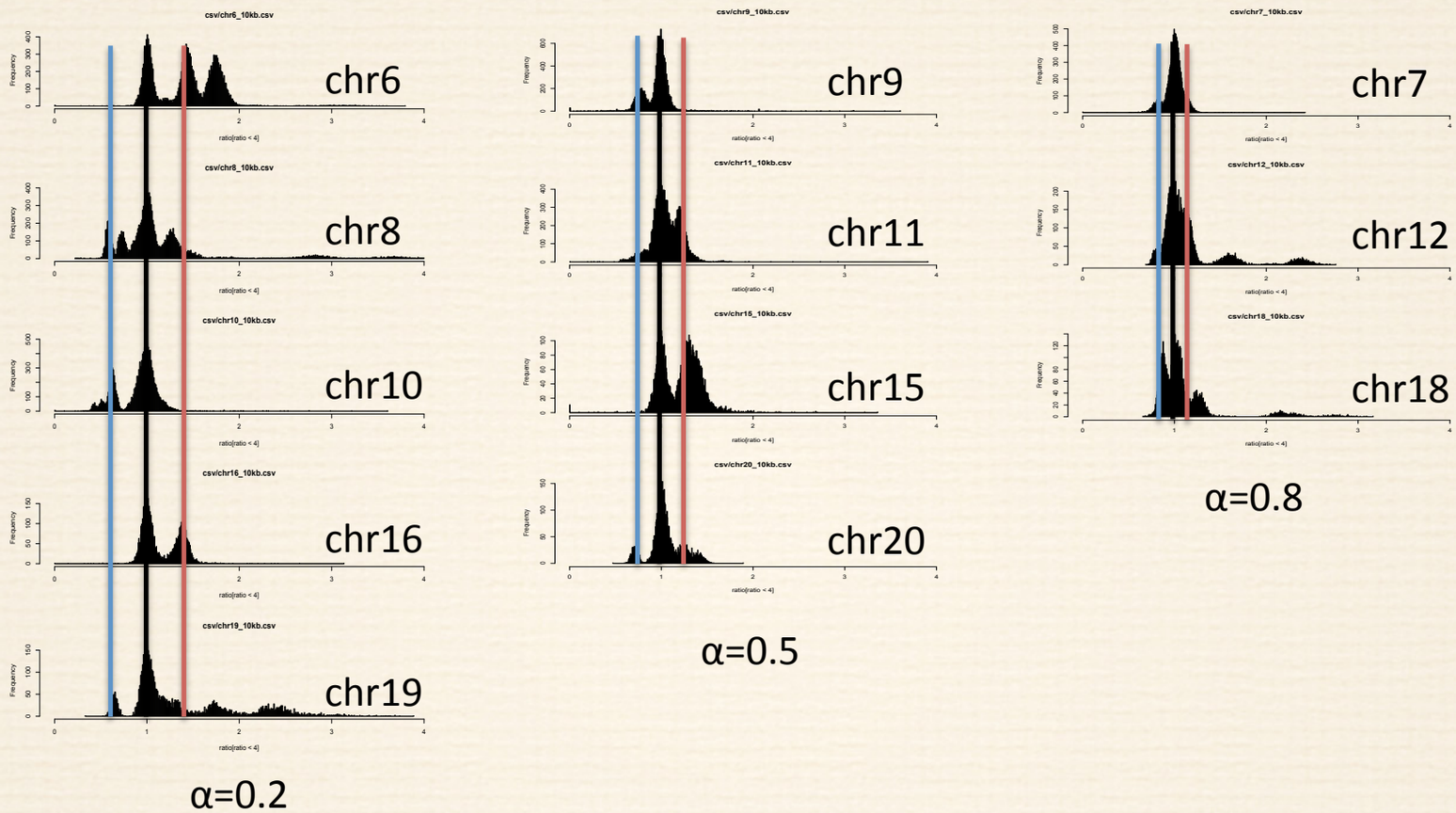
$$= 0.26$$

Tumor to Normal ratio

Normal Cont. Estimate

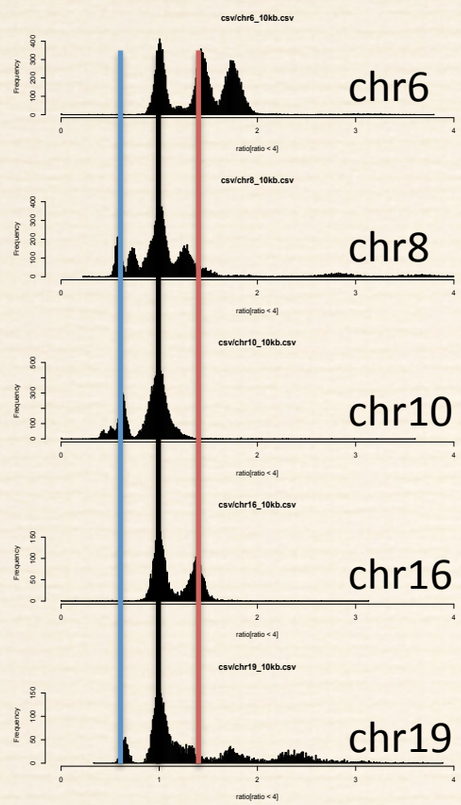


Normal Cont. Estimate

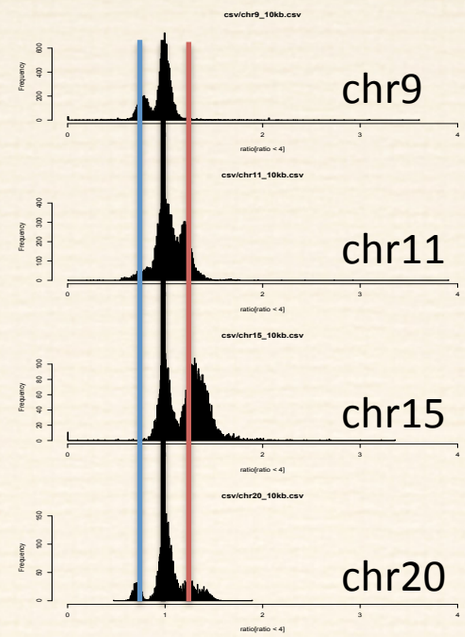


Normal

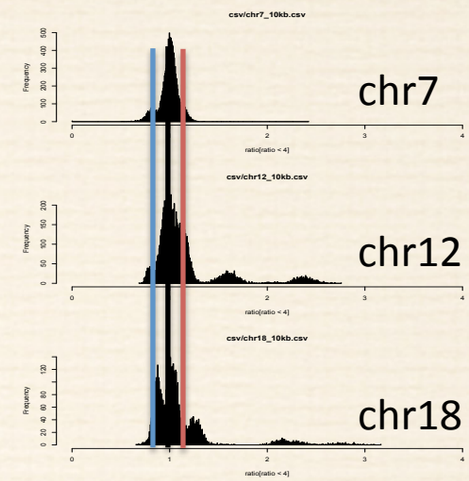
Normal Cont. Estimate



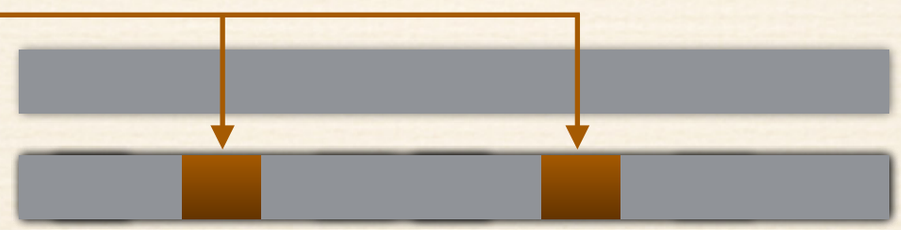
α=0.2



α=0.5



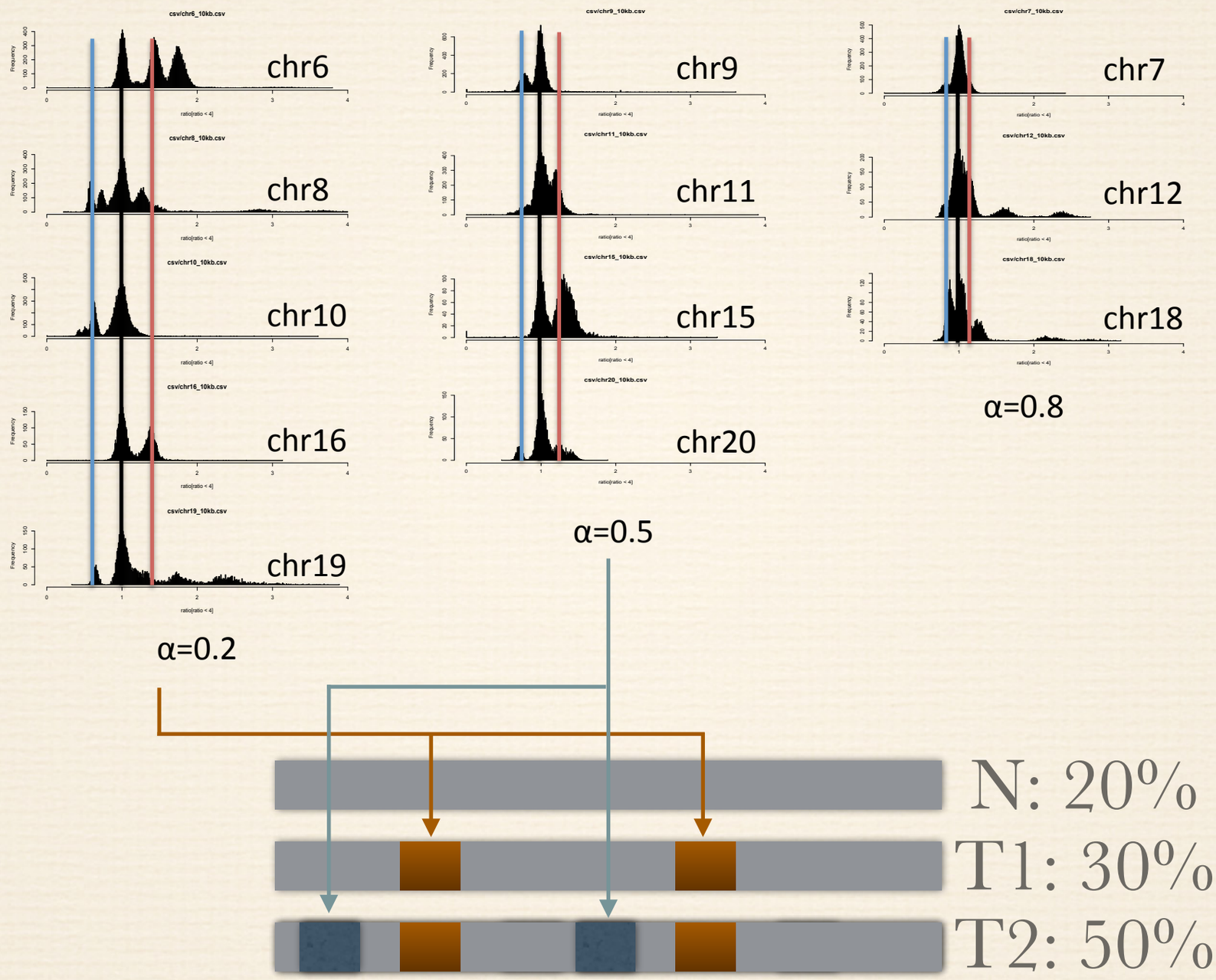
α=0.8



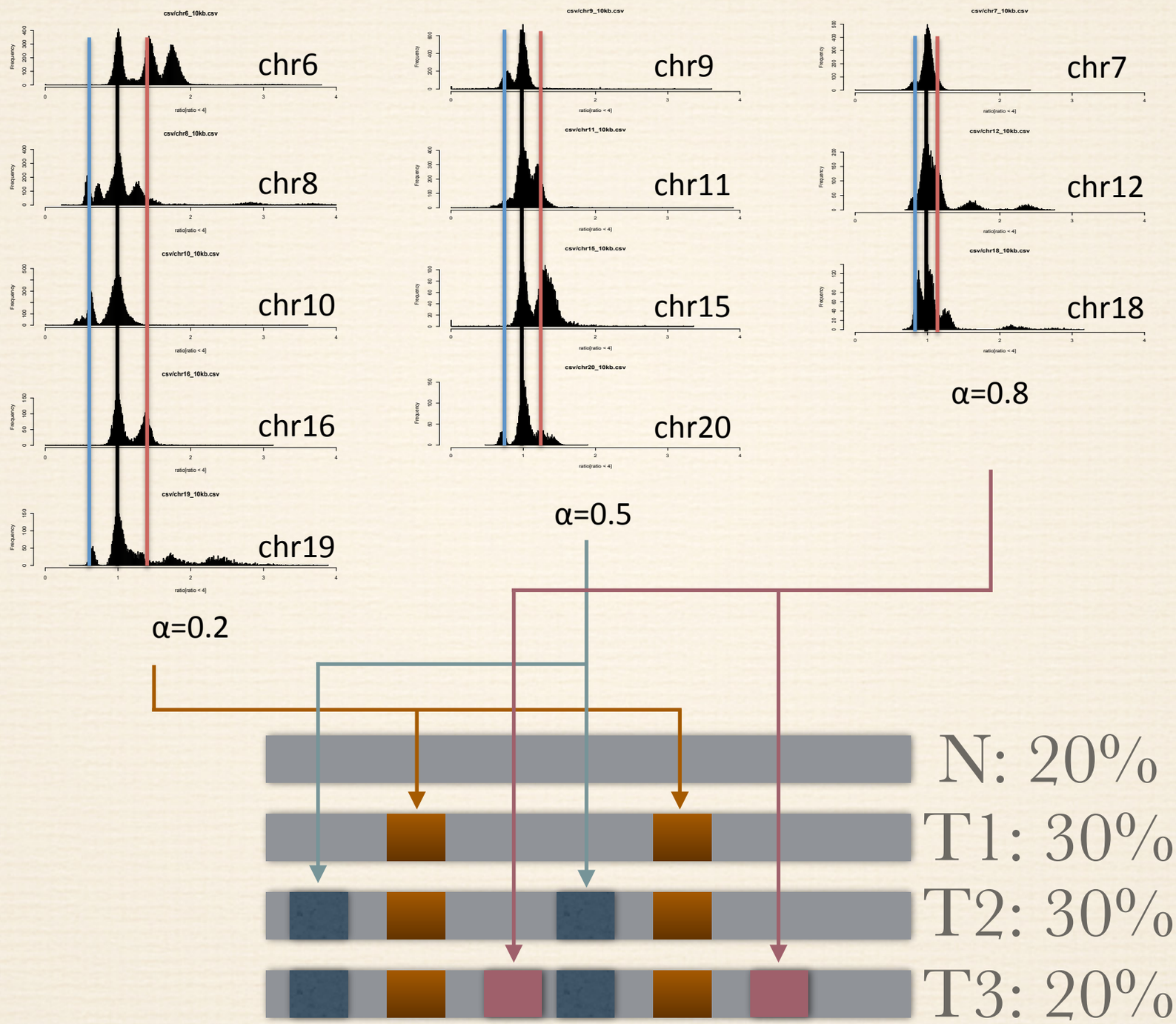
N: 20%

T: 80%

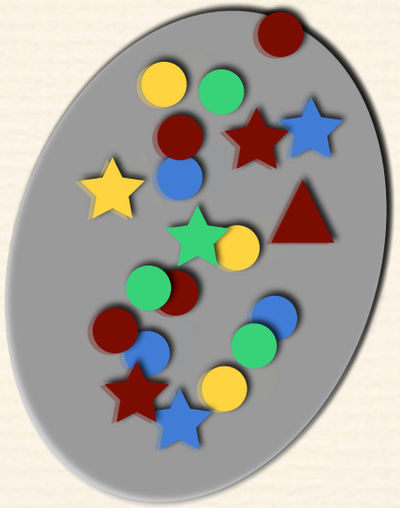
Normal Cont. Estimate



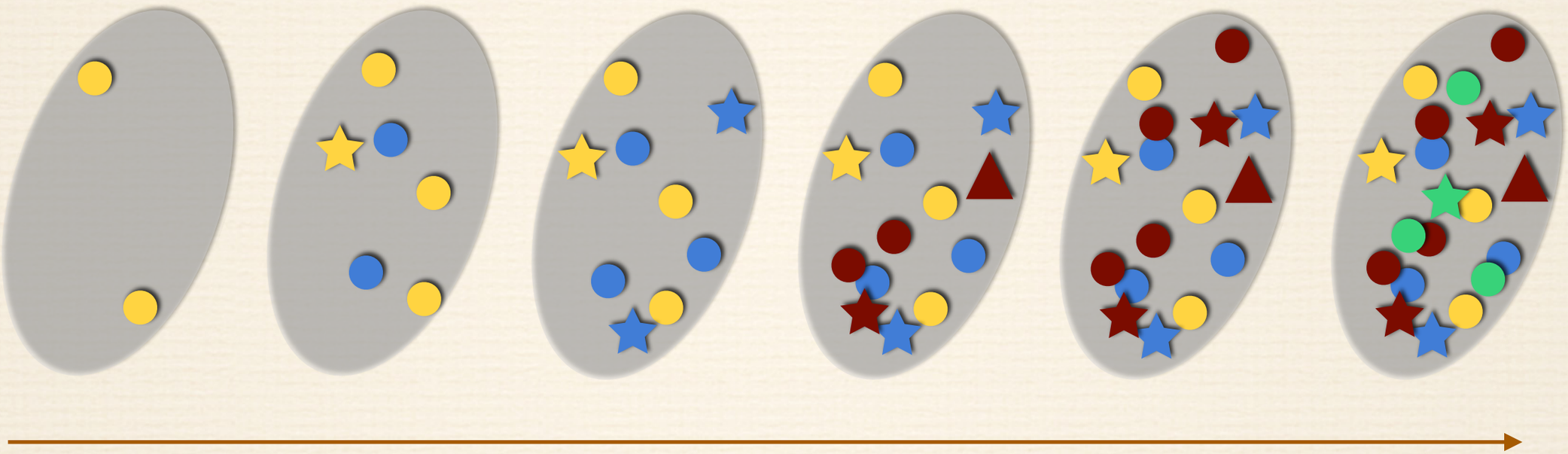
Normal Cont. Estimate



Limitations

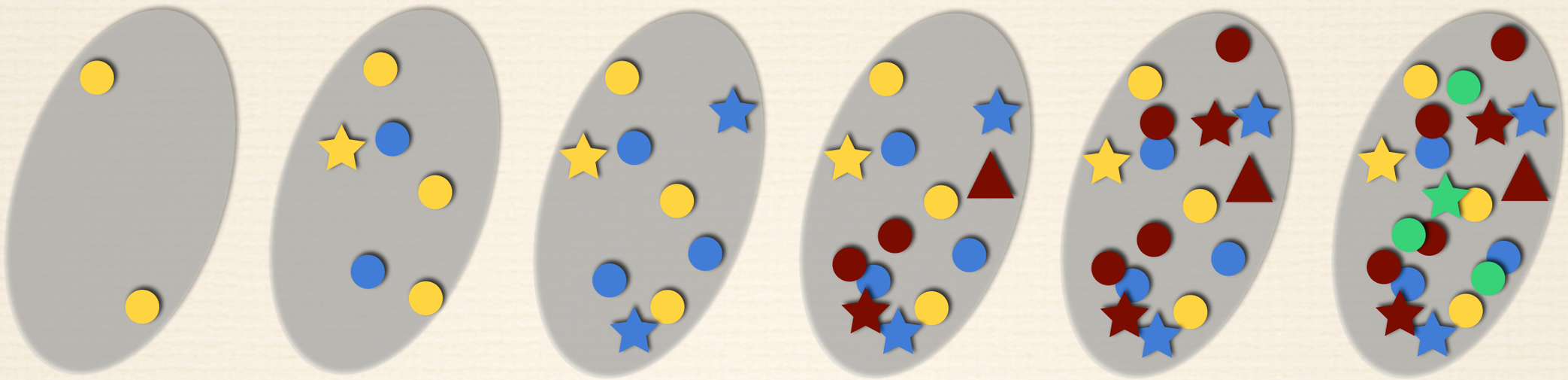


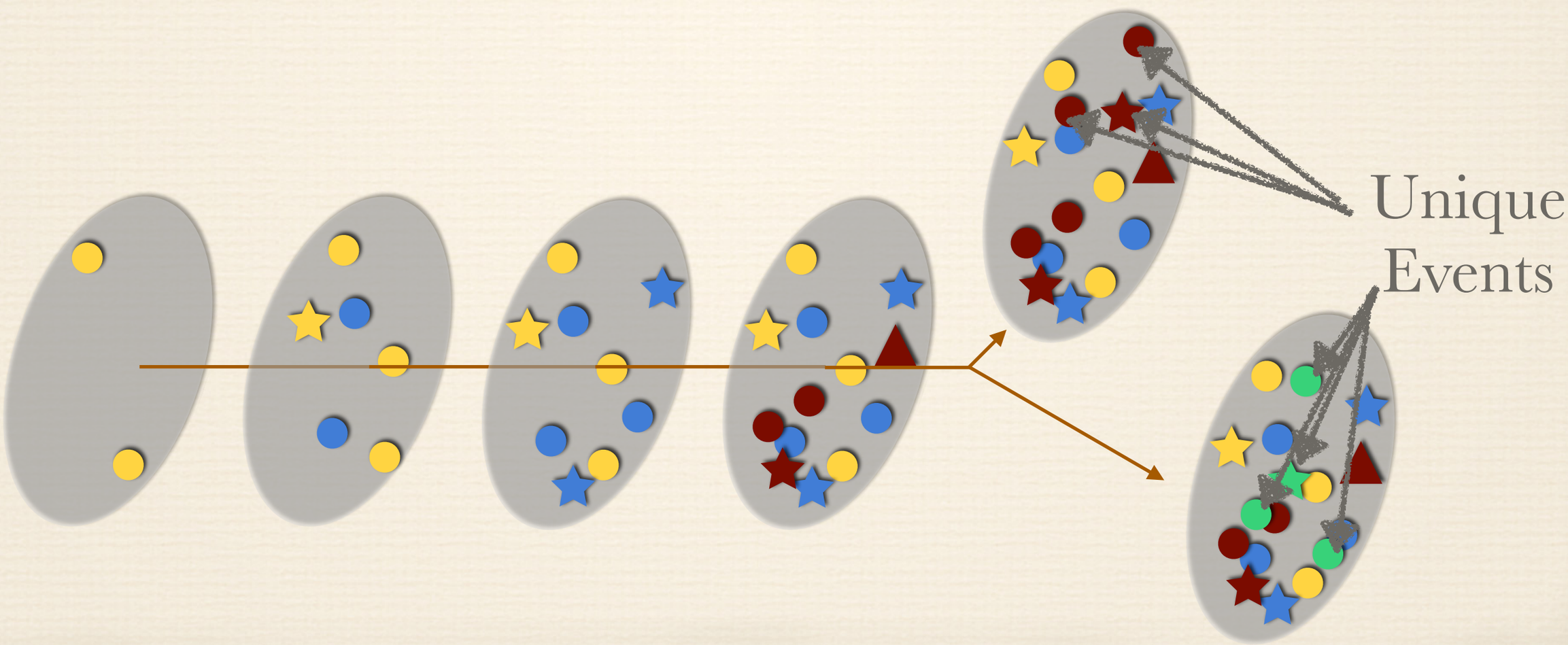
Limitations



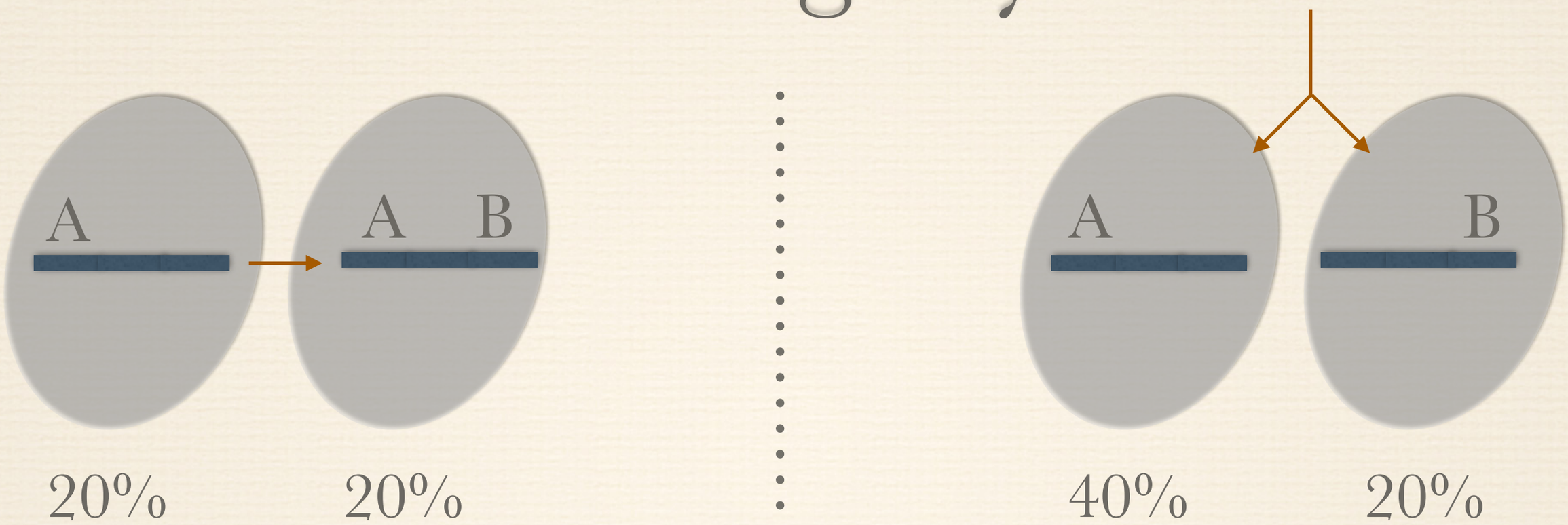
Linear Progression

Limitations





Ambiguity



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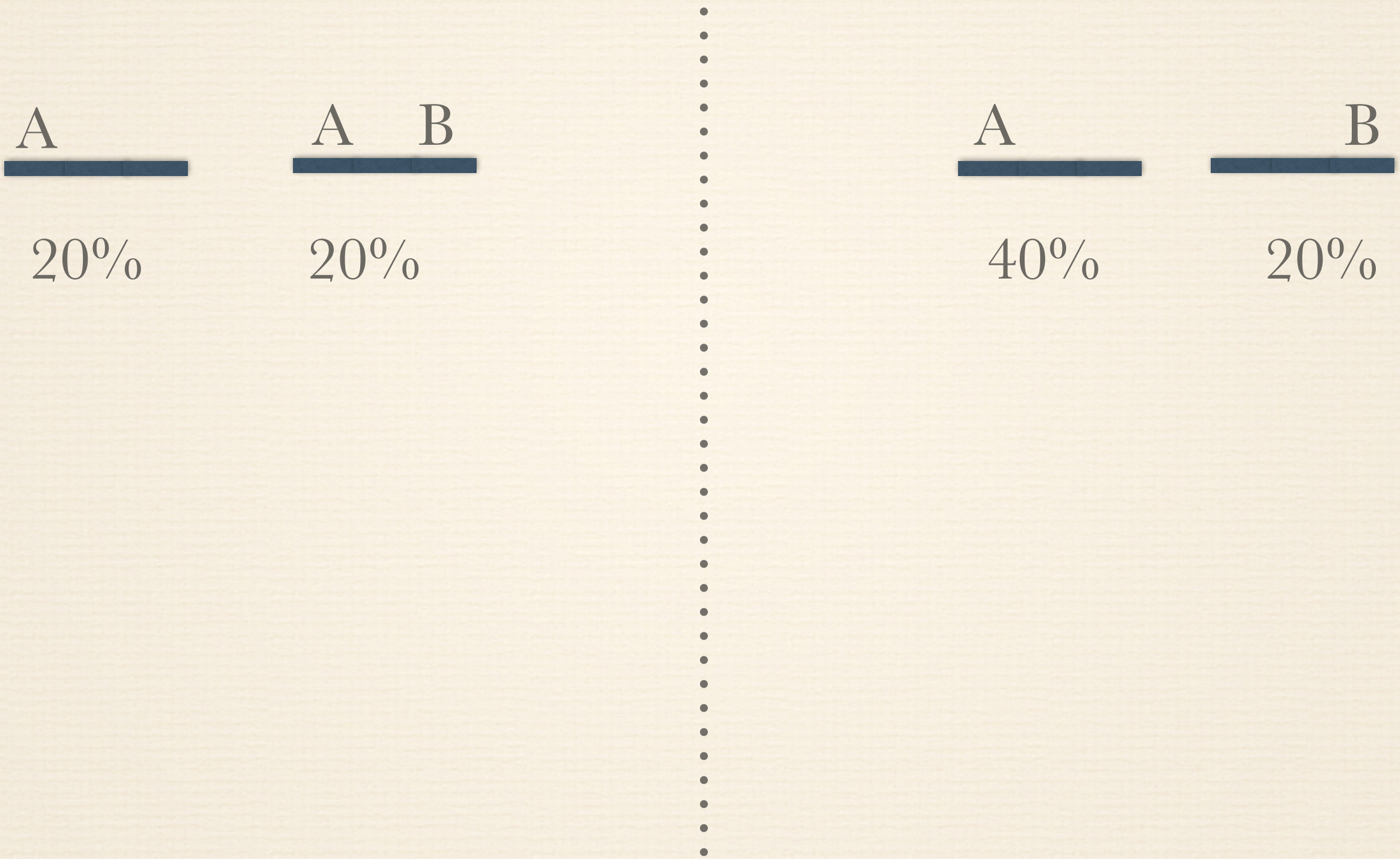

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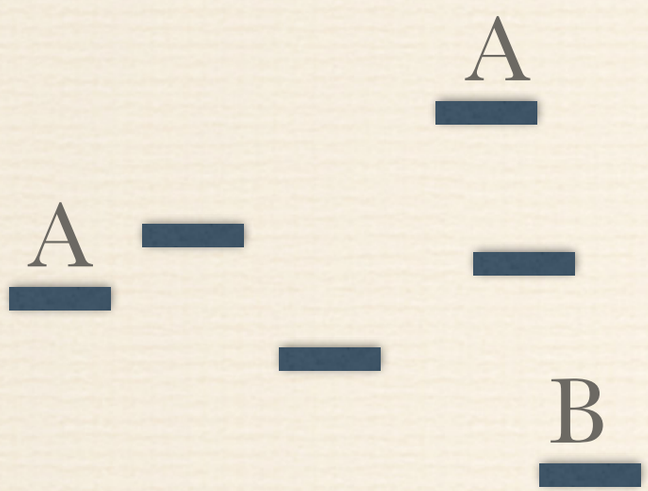

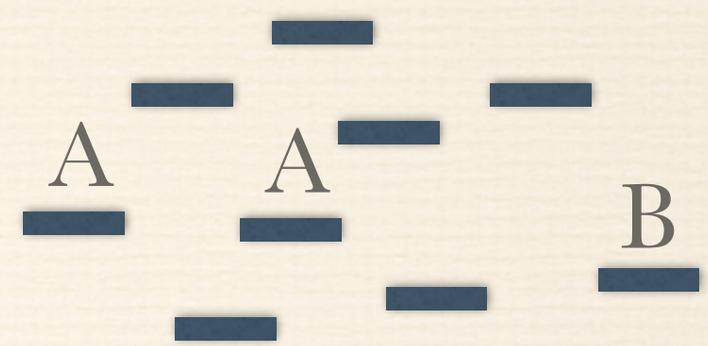

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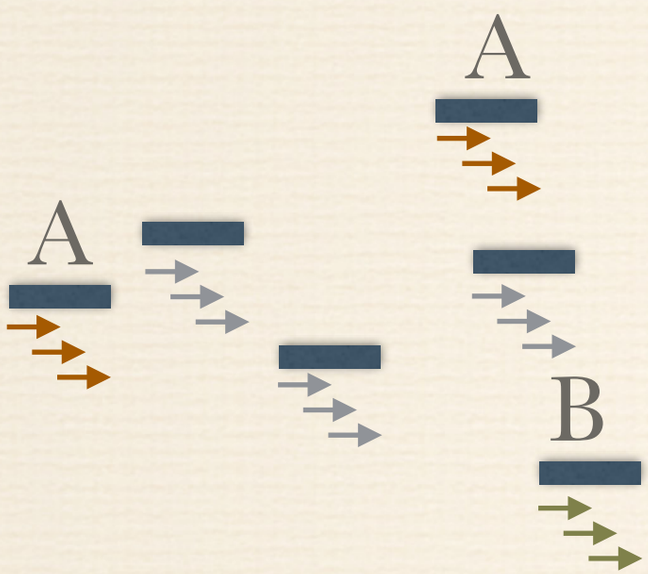

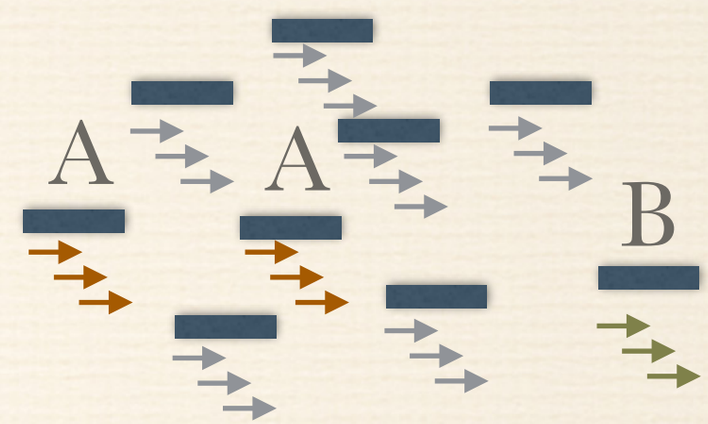

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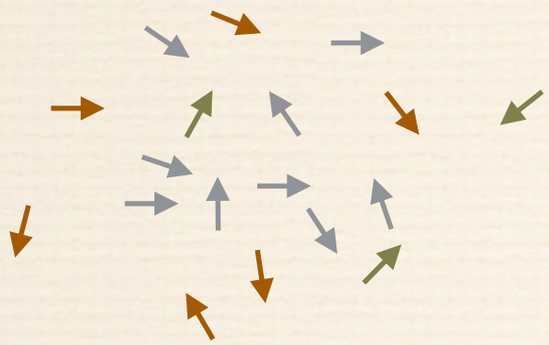
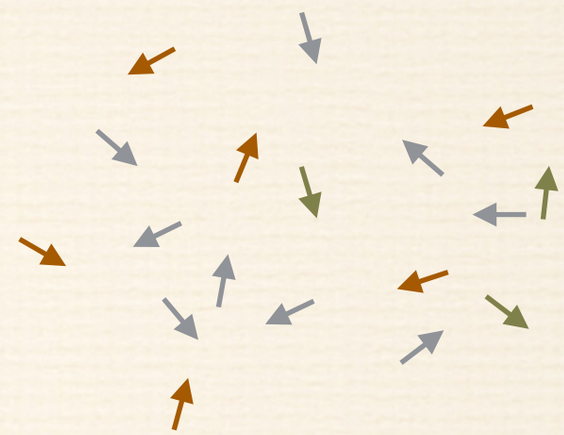
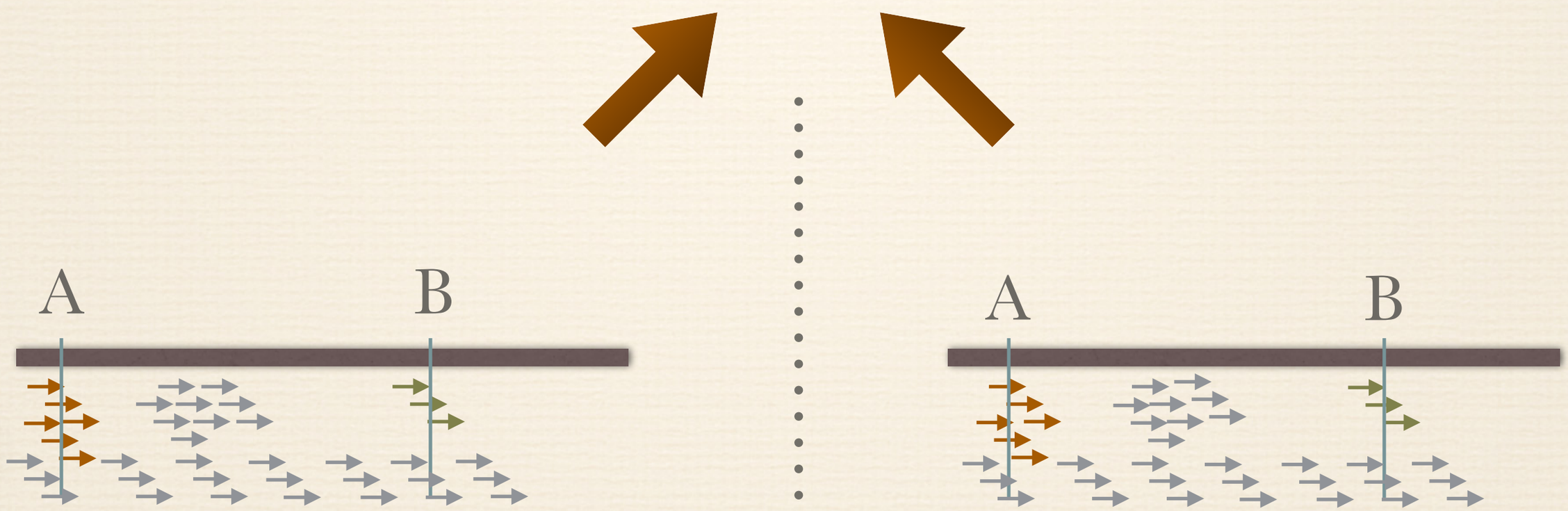




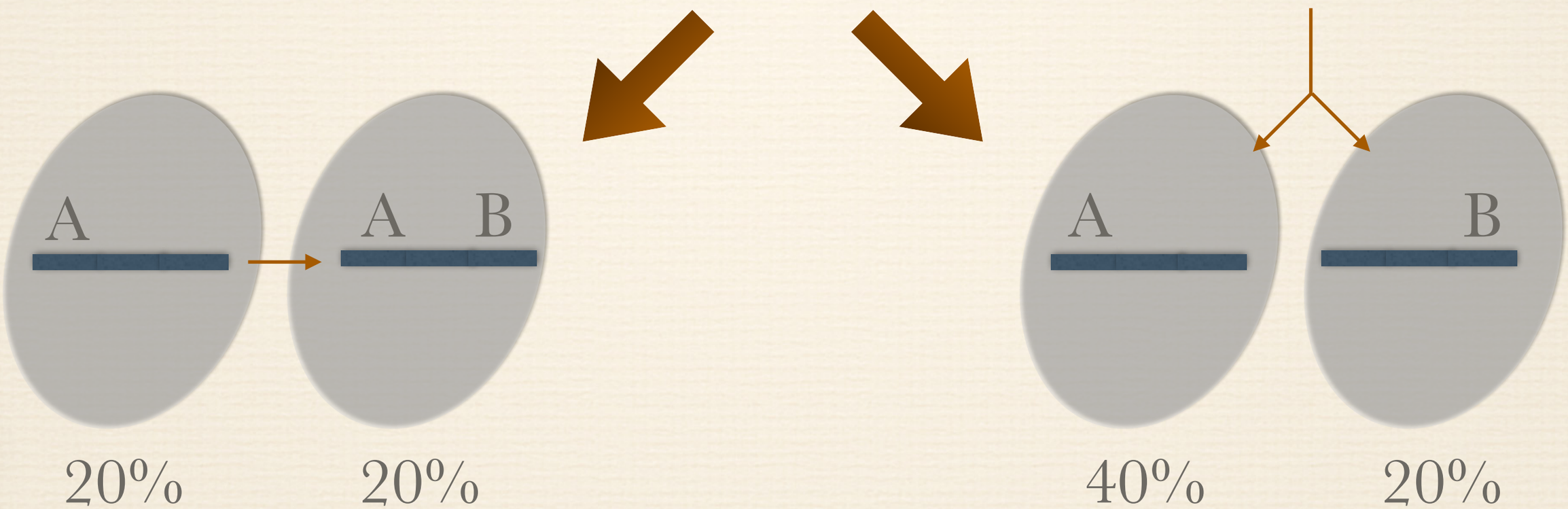
Ambiguity

Freq.	A	B
	40%	20%

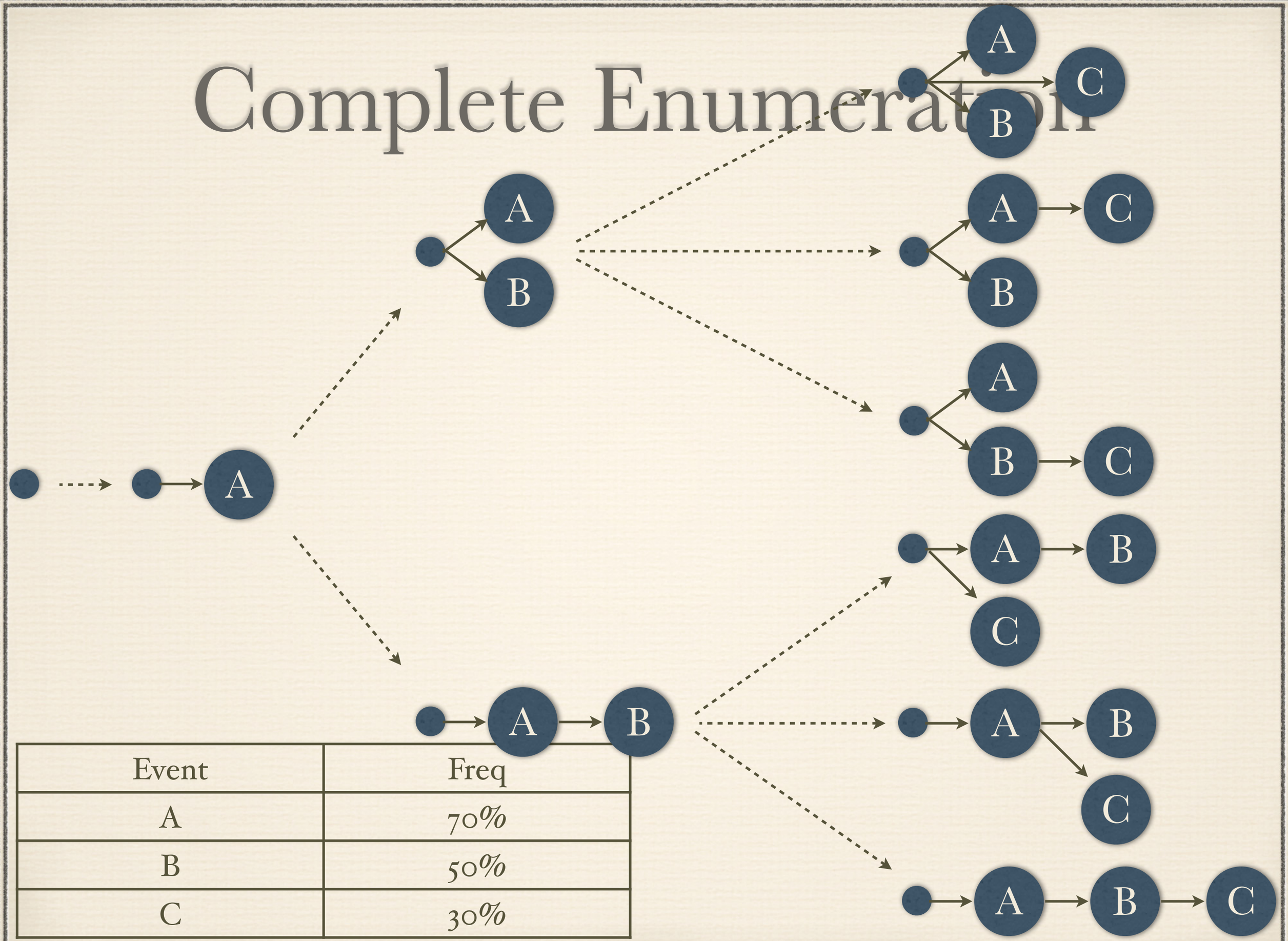


Ambiguity

Freq.	A	B
	40%	20%

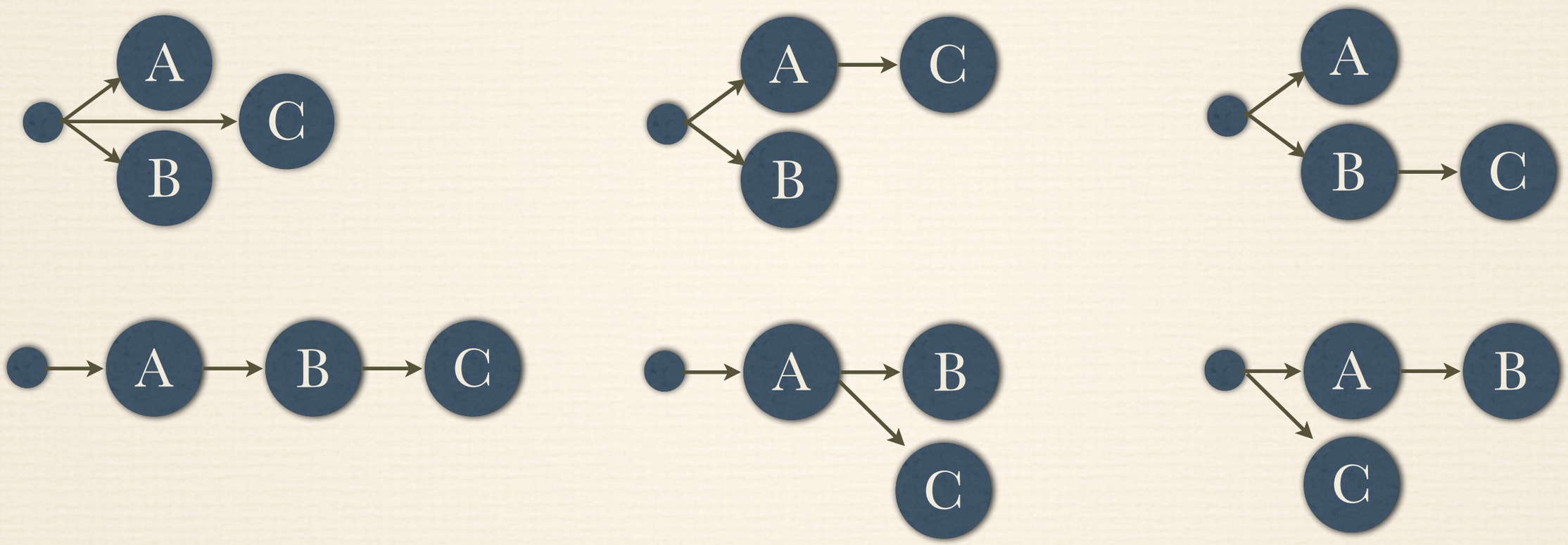


Complete Enumeration

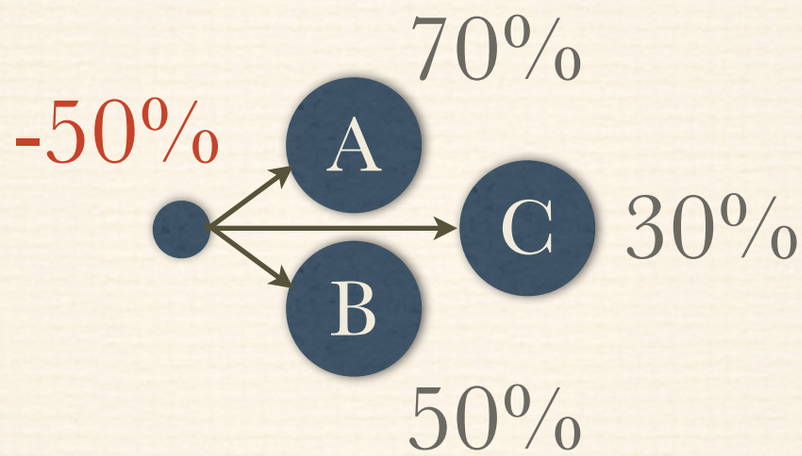


Event	Freq
A	70%
B	50%
C	30%

Complete Enumeration



Complete Enumeration



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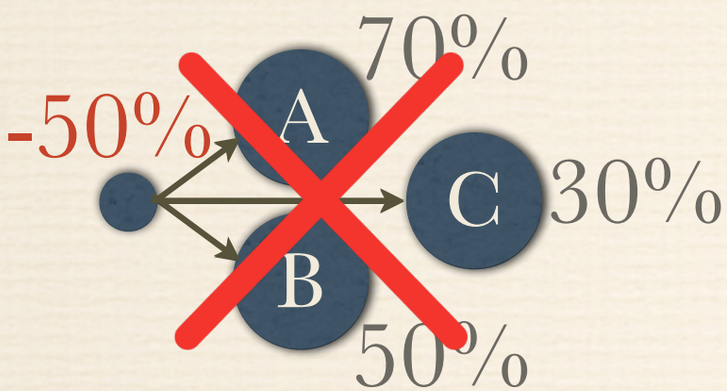
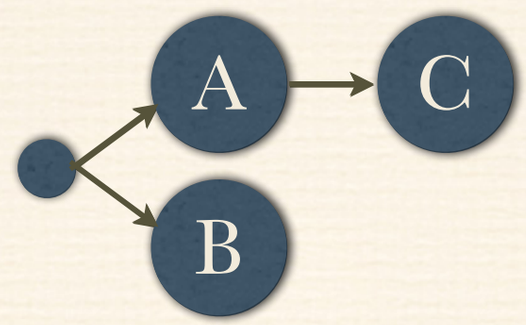
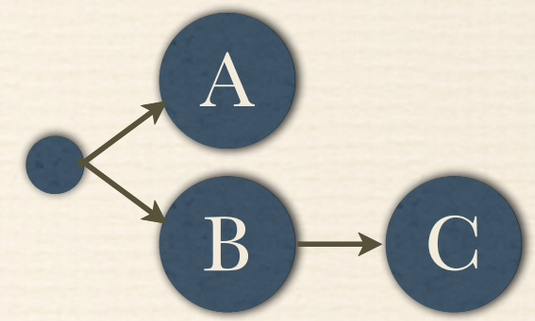
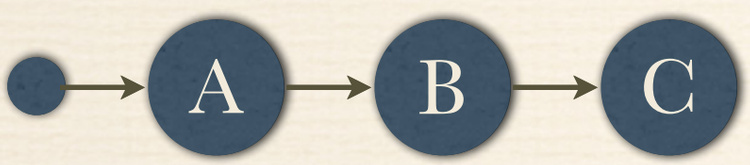
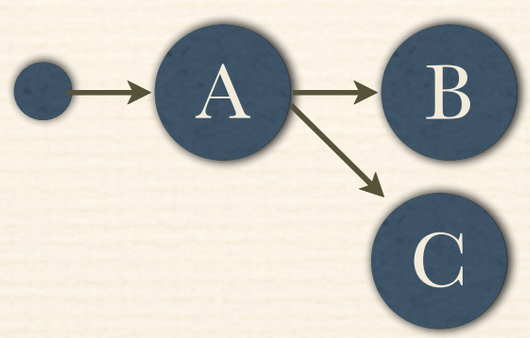
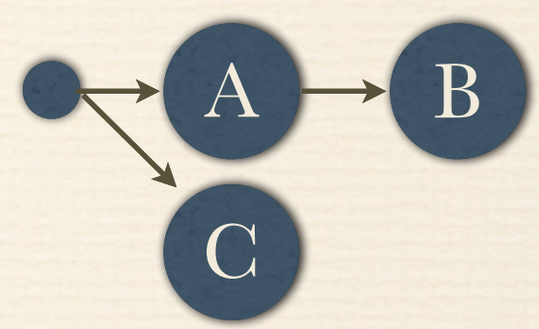




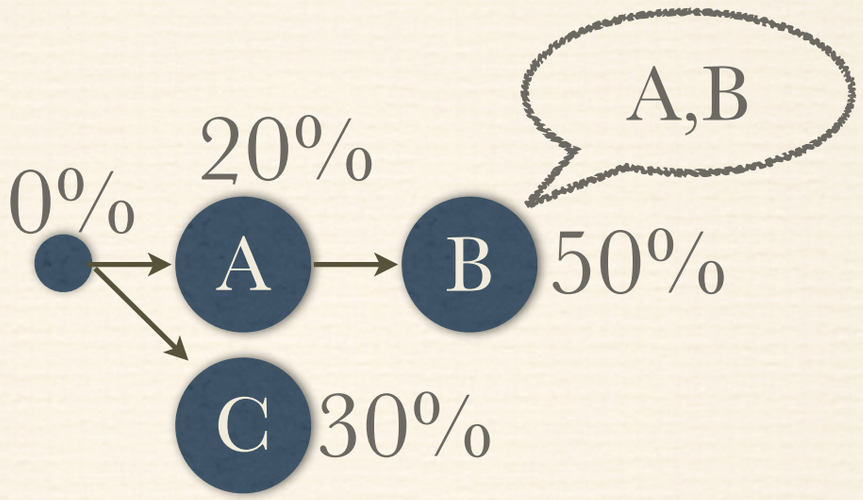






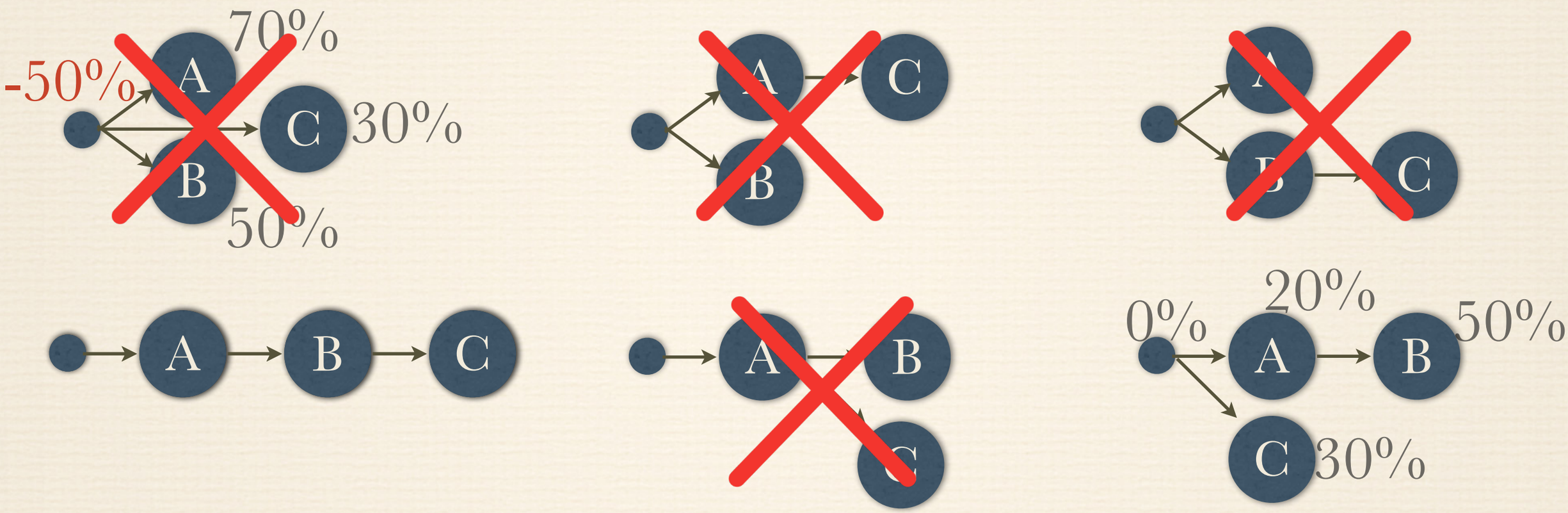


Complete Enumeration

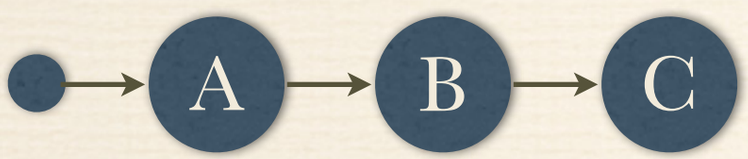


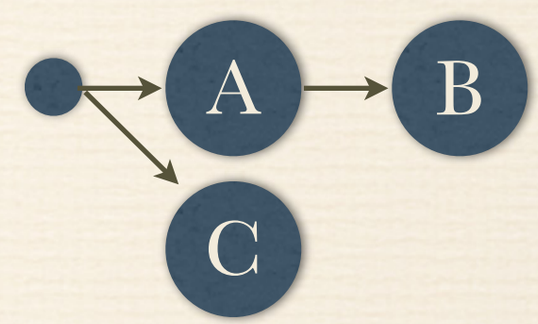
Event	Freq
A	70%
B	50%
C	30%

Complete Enumeration



Complete Enumeration





Demo 1

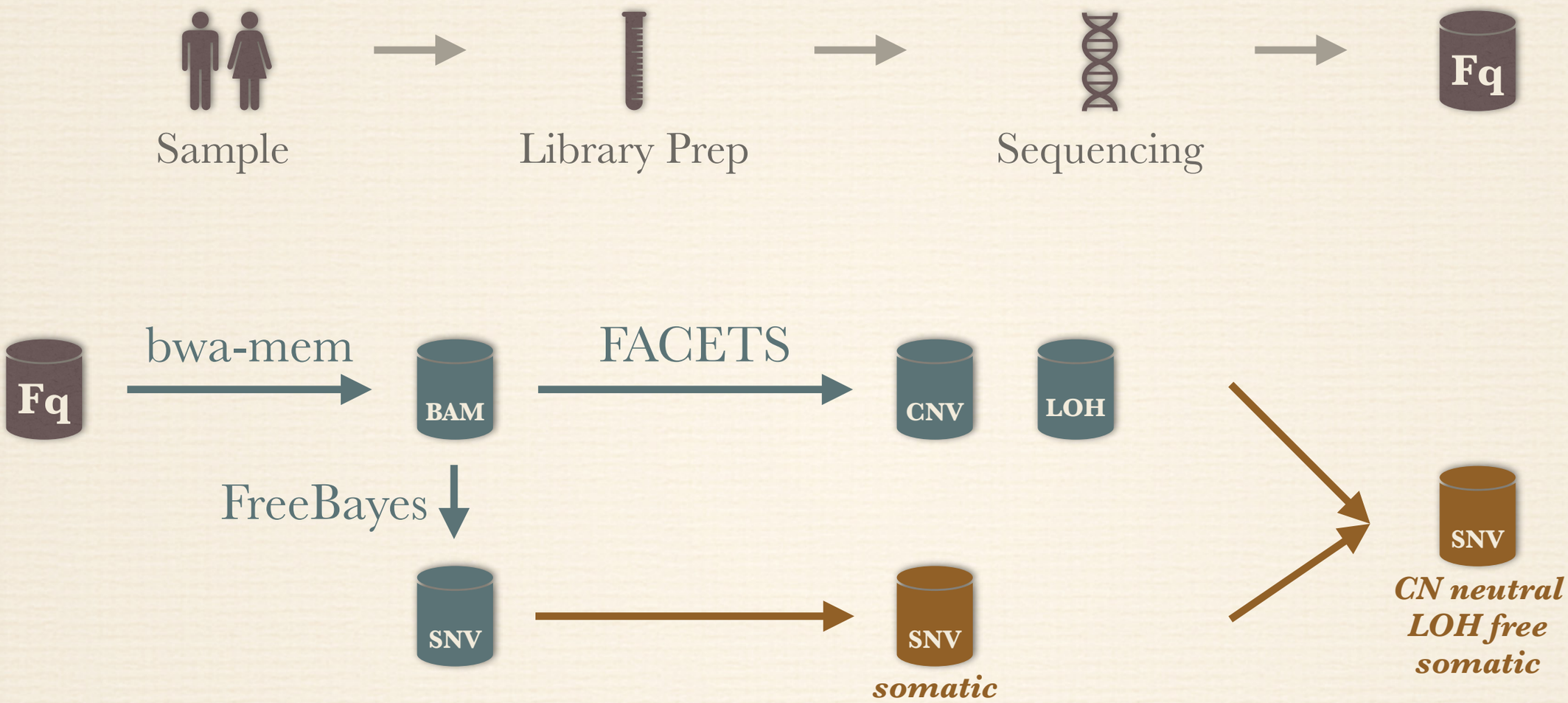
Tumor Heterogeneity Workshop

Part 2 - Genomic Pipeline

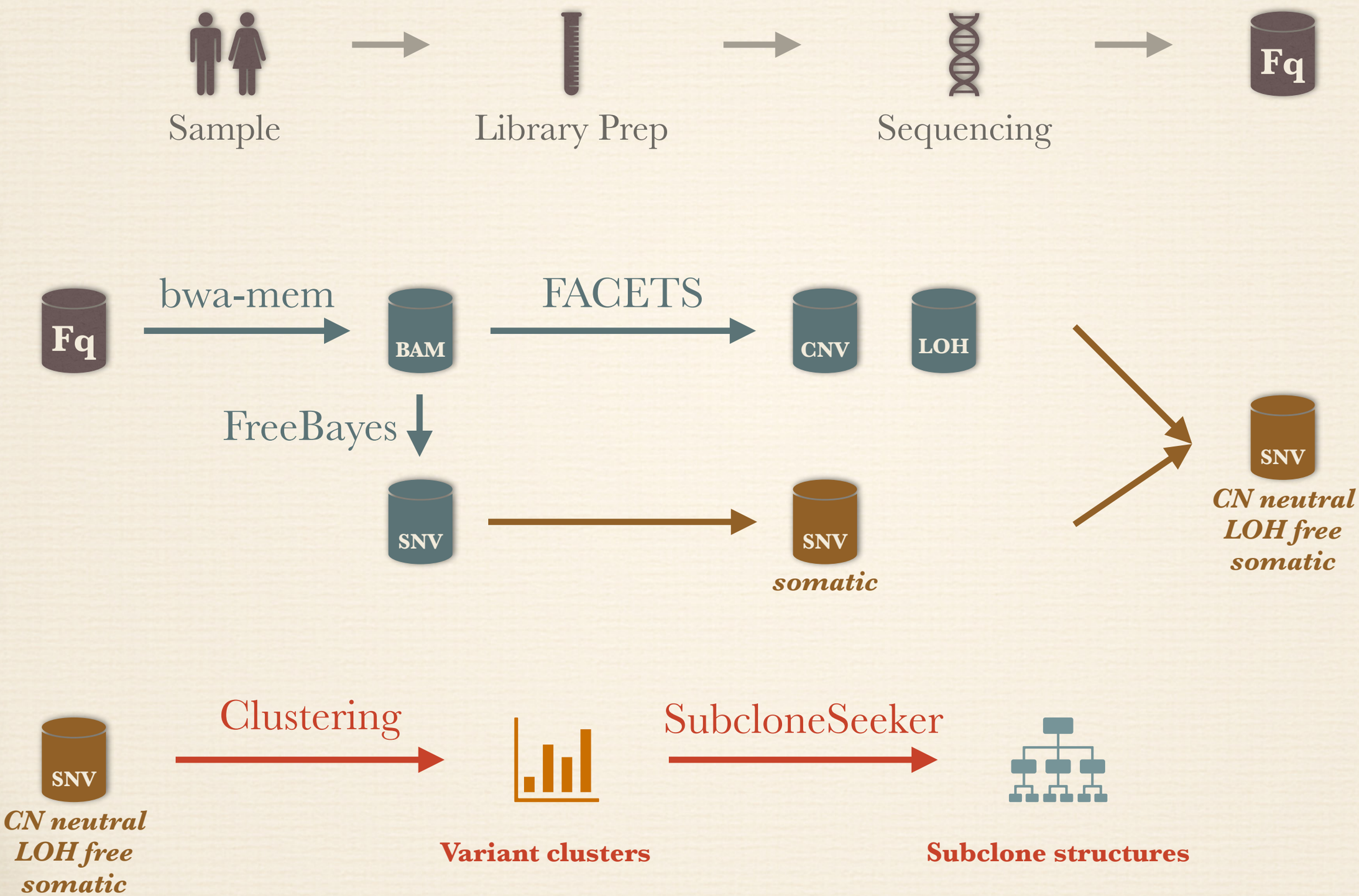
Yi Qiao, 2018

Workflow

Workflow


Sample
Library Prep
Sequencing
Fq
Fq
bwa-mem
BAM
FACETS
CNV
LOH
FreeBayes
SNV
SNV
somatic
SNV
CN neutral
LOH free
somatic

Workflow


Sample
Library Prep
Sequencing
Fq
Fq
bwa-mem
BAM
FACETS
CNV
LOH
FreeBayes
SNV
SNV
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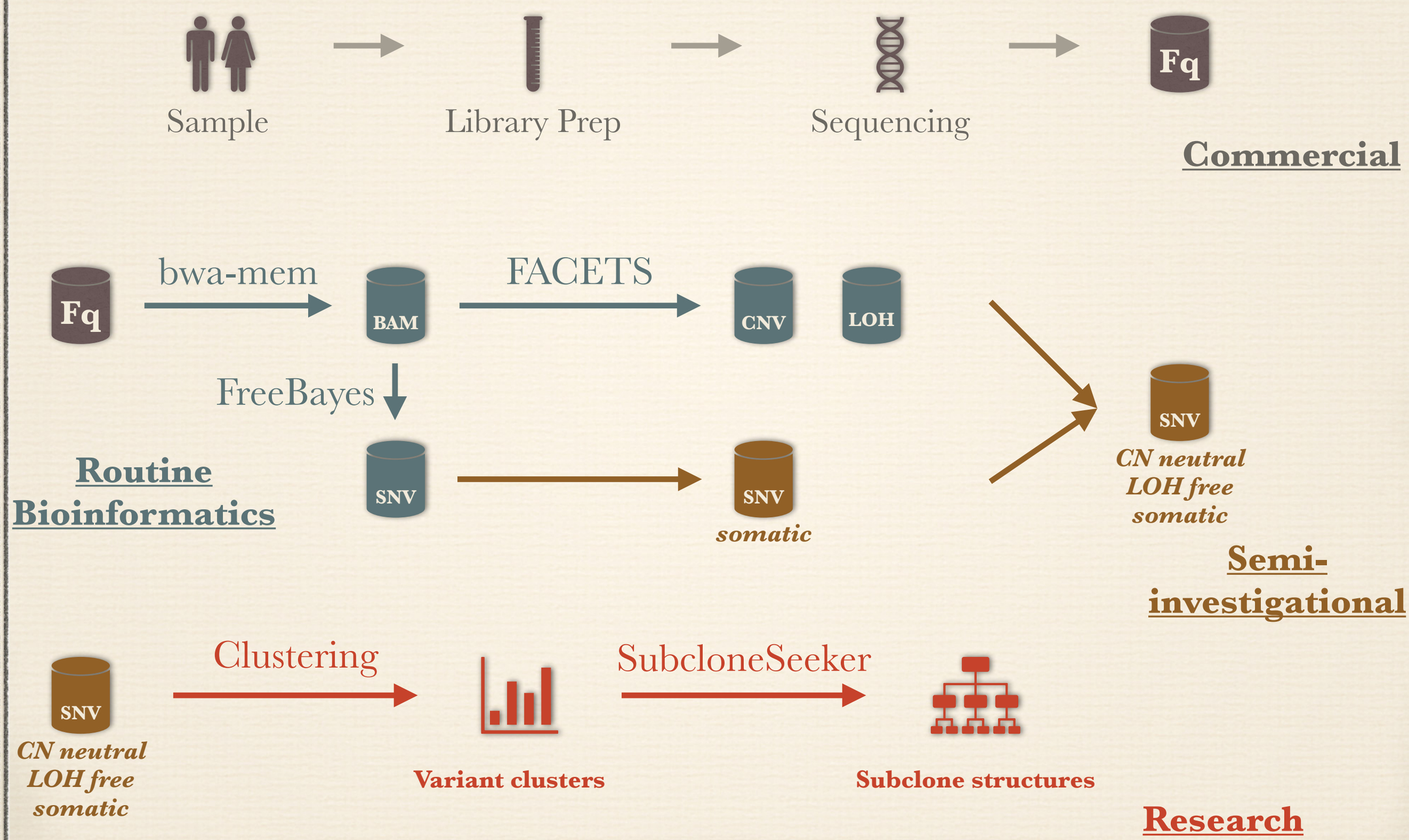
SNV
CN neutral
LOH free
somatic
SNV
CN neutral
LOH free
somatic
Clustering
Variant clusters
SubcloneSeeker
Subclone structures

Workflow


Sample
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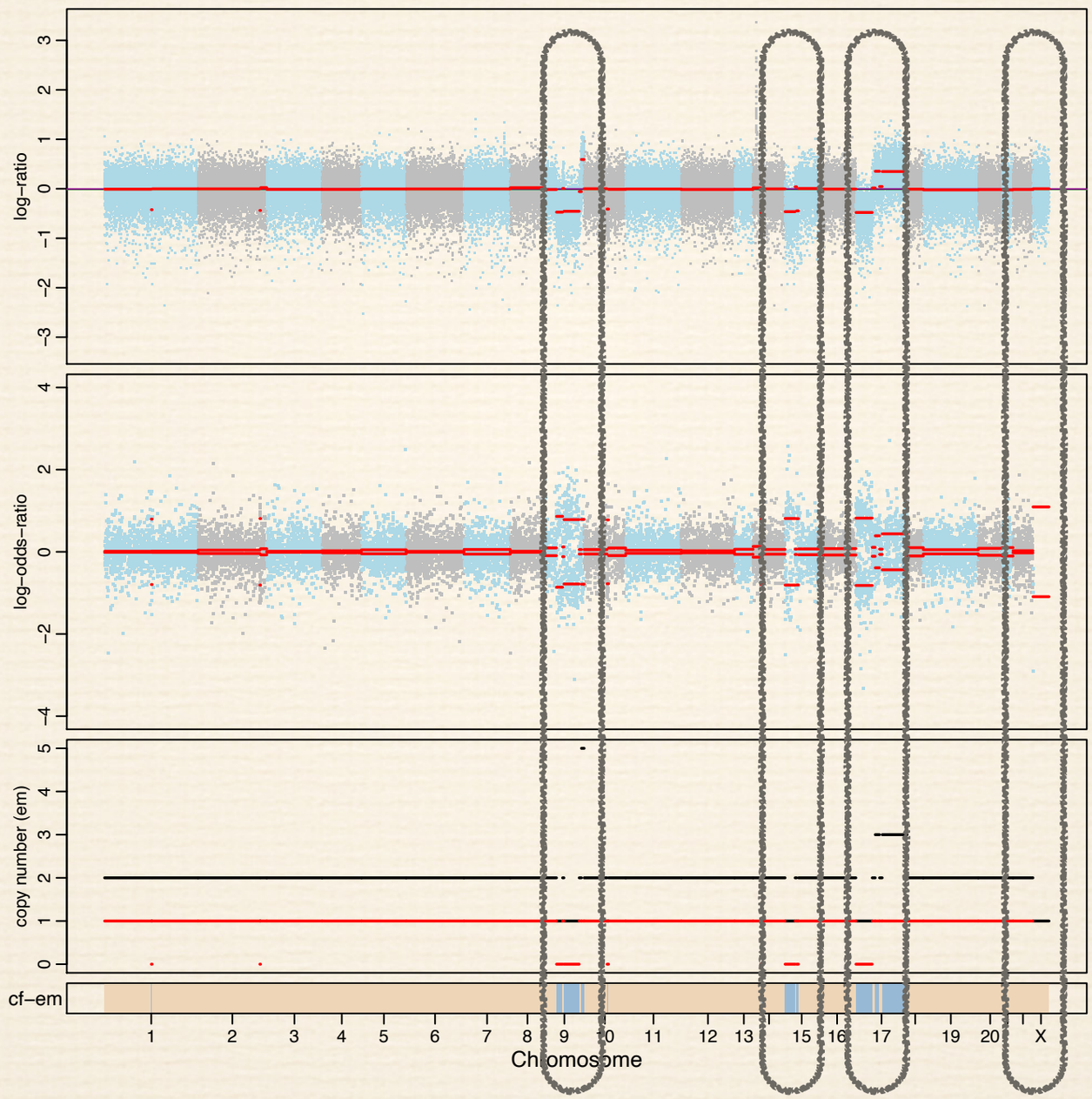
SNV
CN neutral
LOH free
somatic
Routine
Bioinformatics
Semi-
investigational
SNV
CN neutral
LOH free
somatic
Clustering
Variant clusters
SubcloneSeeker
Subclone structures
Research

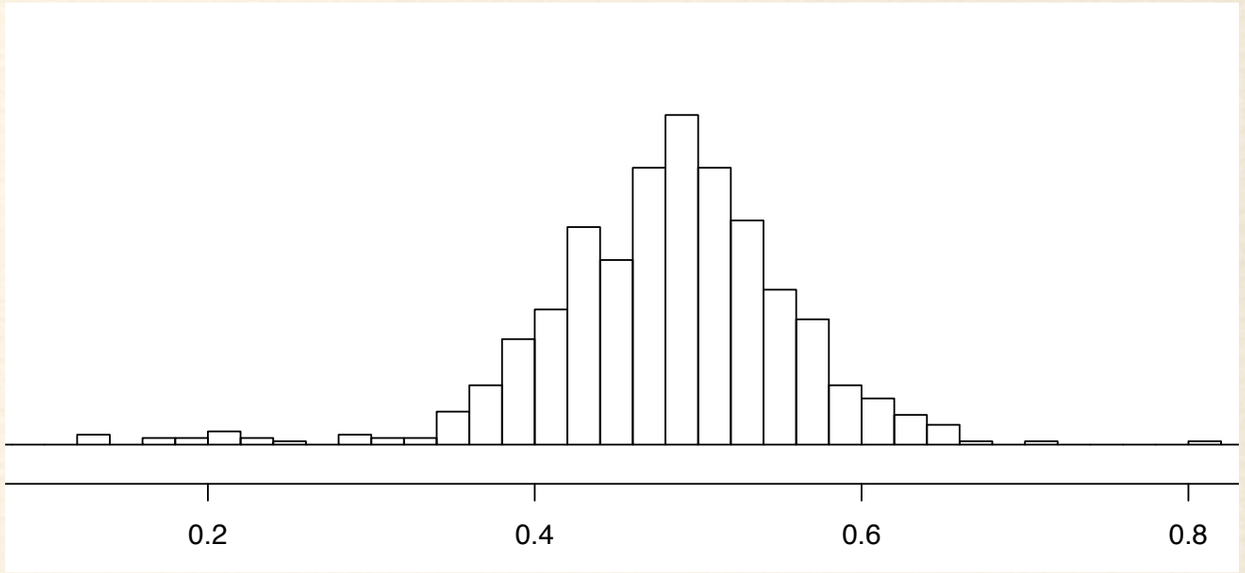
FACETS example

Exclude chr 9, 15, 17, X



SNV Allele Frequencies

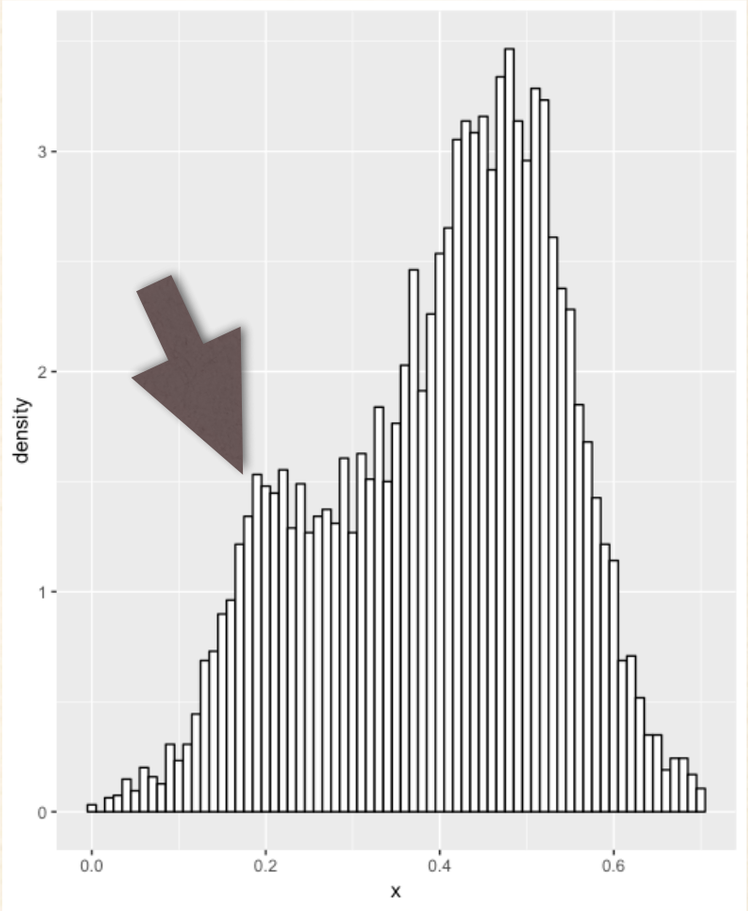
- Individual SNV allele frequency measurement is noisy



SNV Allele Frequencies

- Individual SNV allele frequency measurement is noisy

histogram

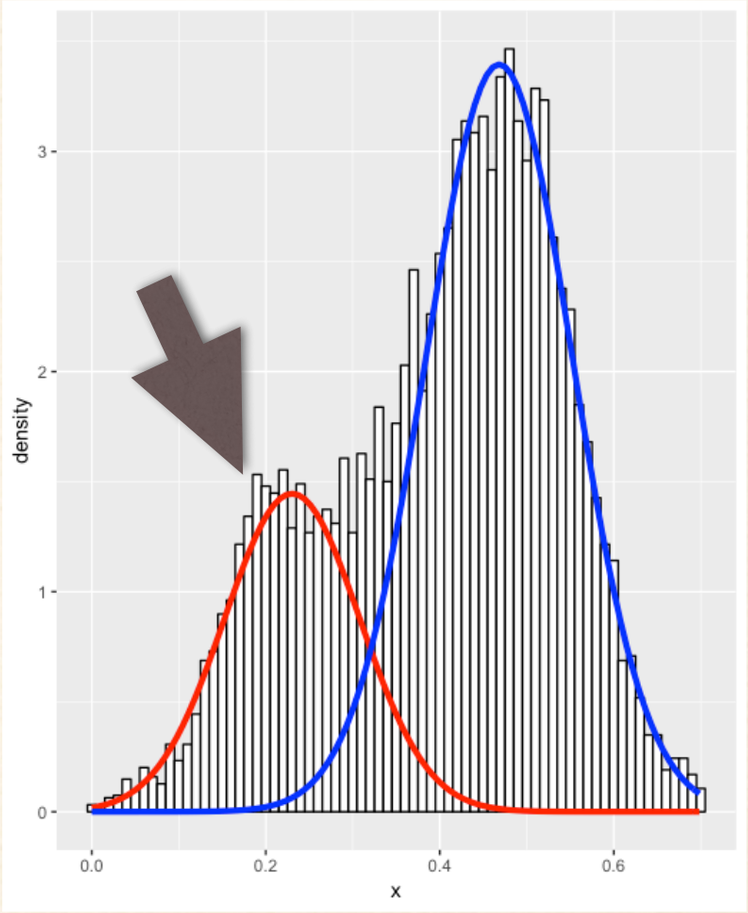


Variant Allele Frequency (subclonal)

SNV Allele Frequencies

- Individual SNV allele frequency measurement is noisy
- Group SNVs with similar AF, and use each cluster as a unit of inherence.

histogram



Variant Allele Frequency (subclonal)

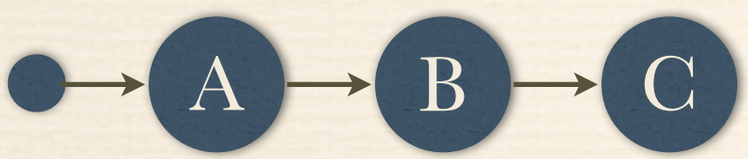
Demo 2

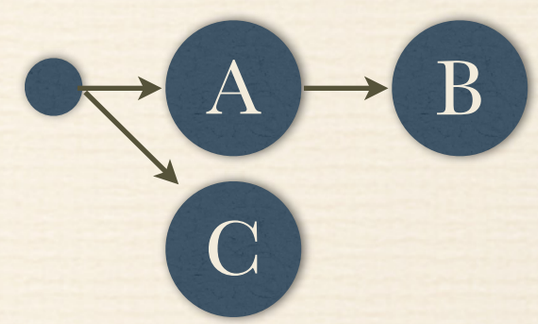
Tumor Heterogeneity Workshop

Part 3 - Multisample Analysis

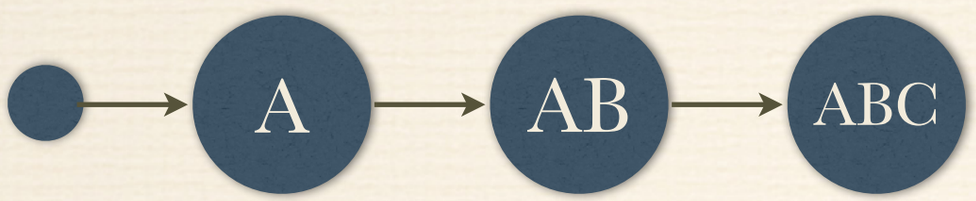
Yi Qiao, 2018

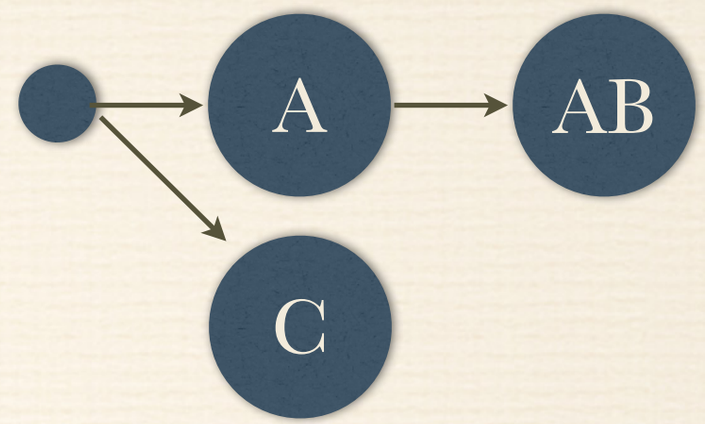
Complete Enumeration

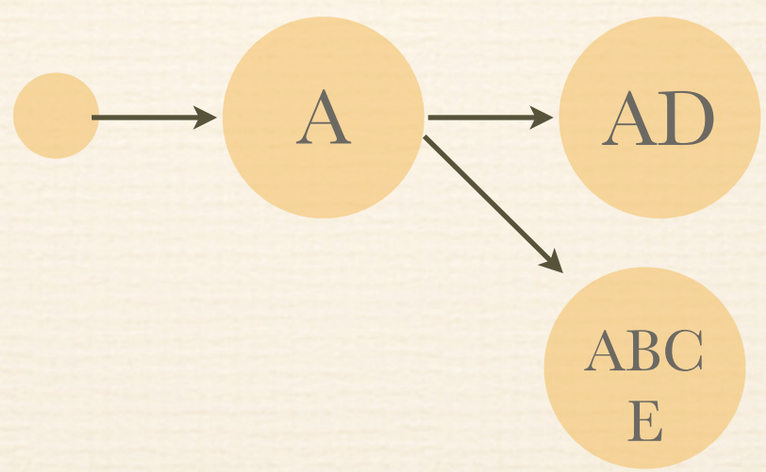




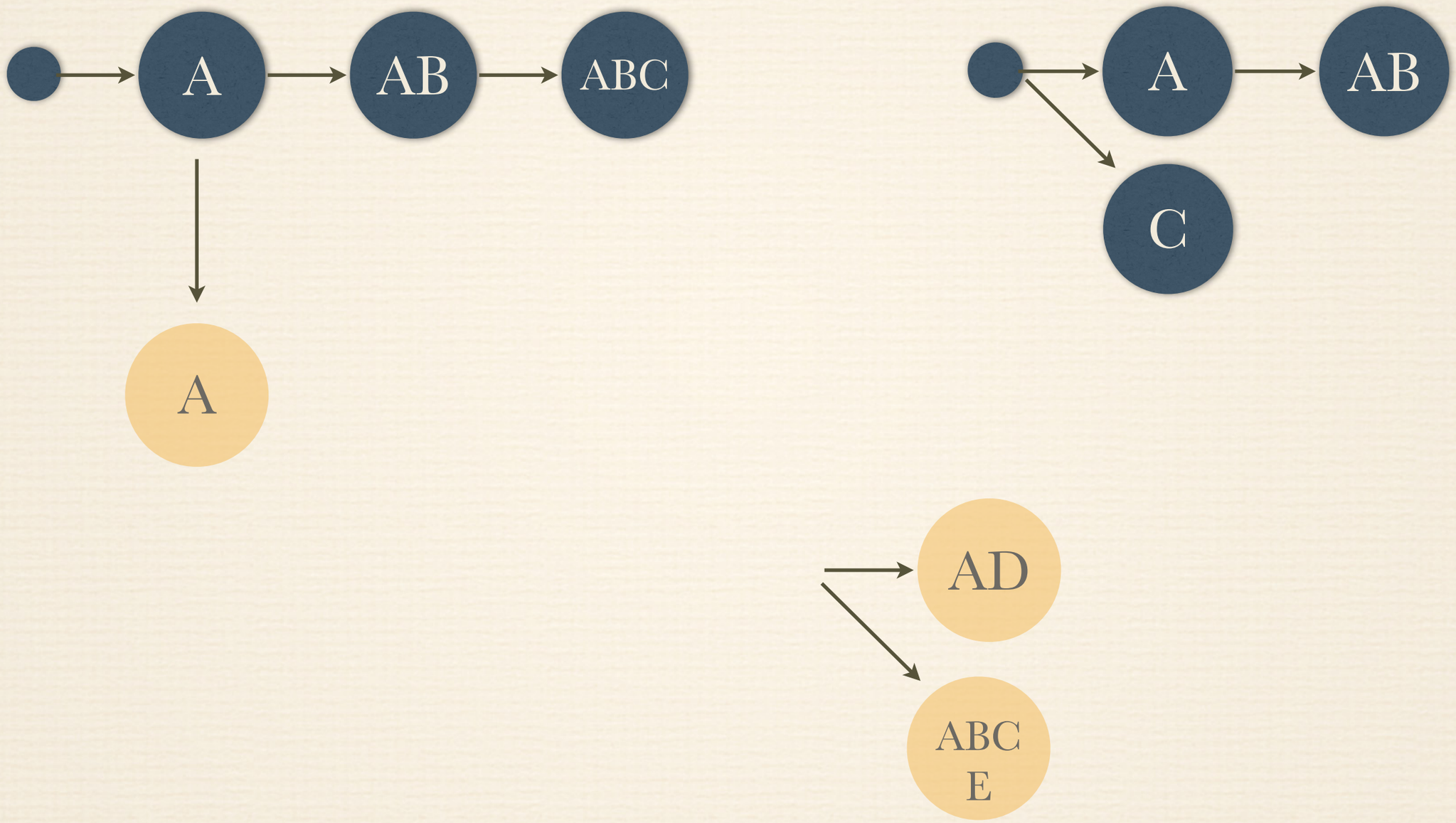
Complete Enumeration



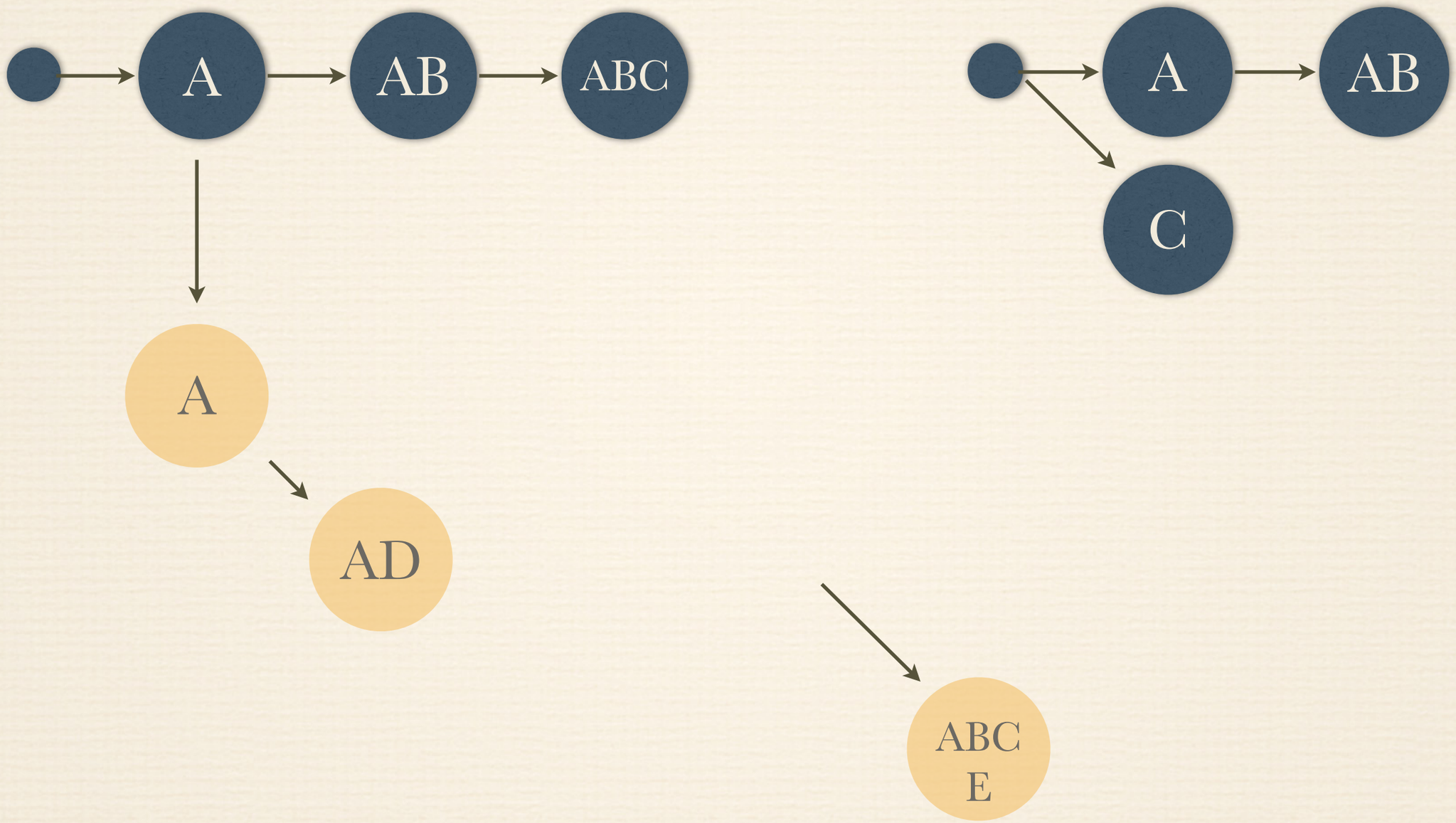




Complete Enumeration



Complete Enumeration



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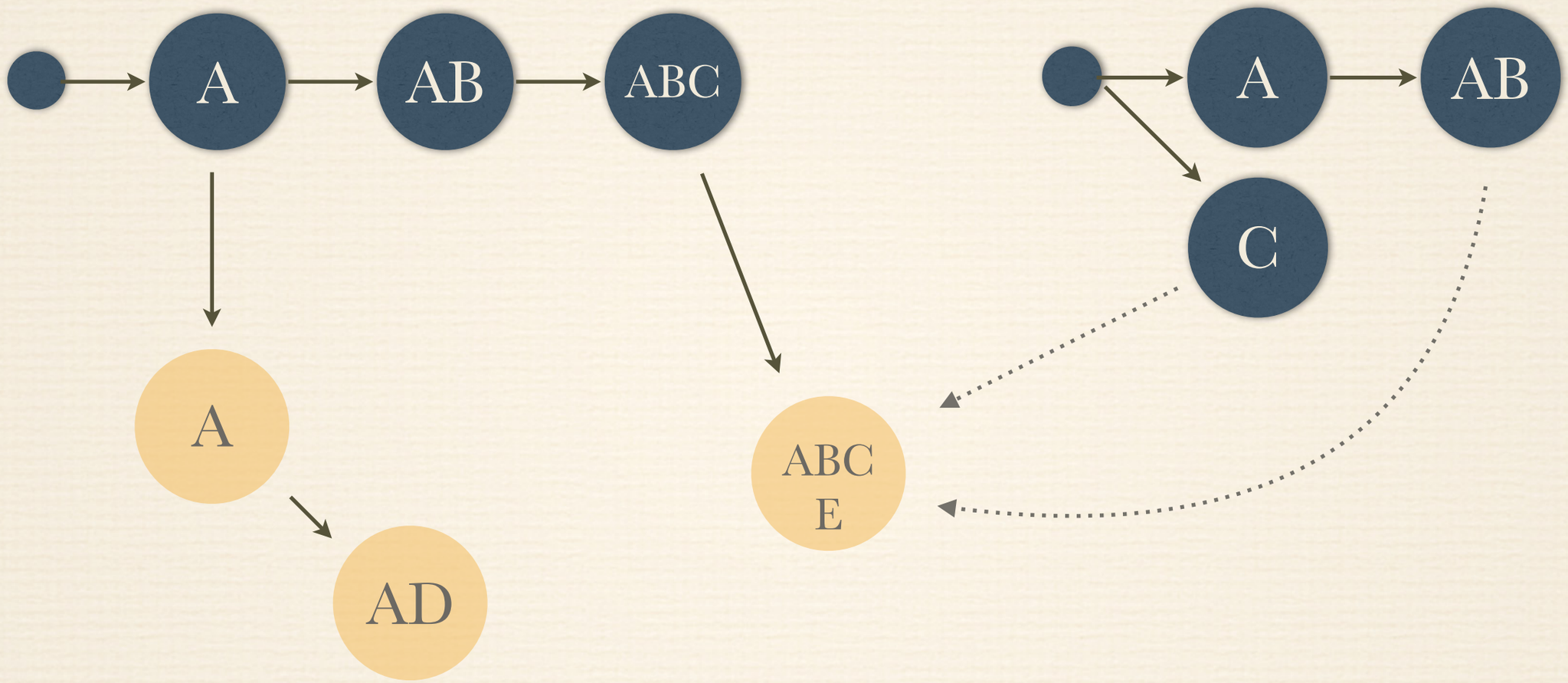
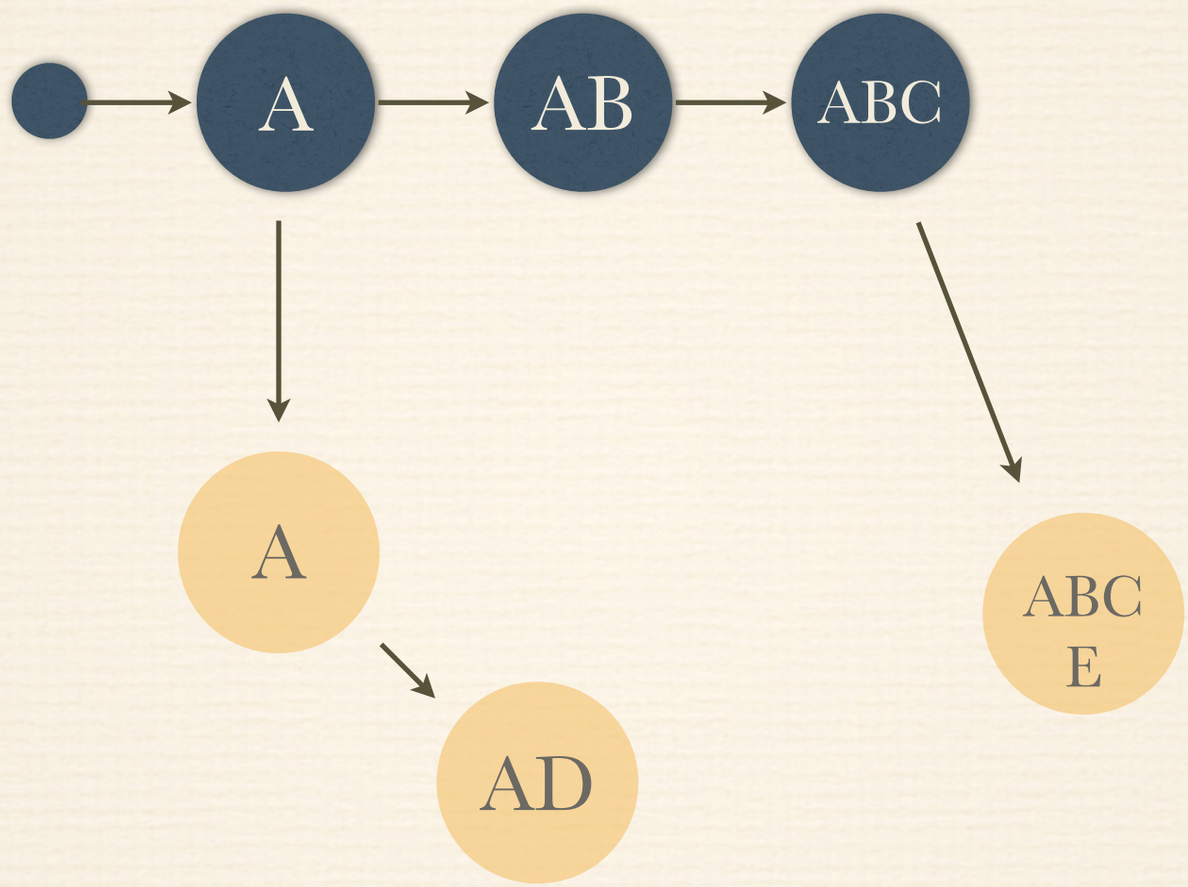
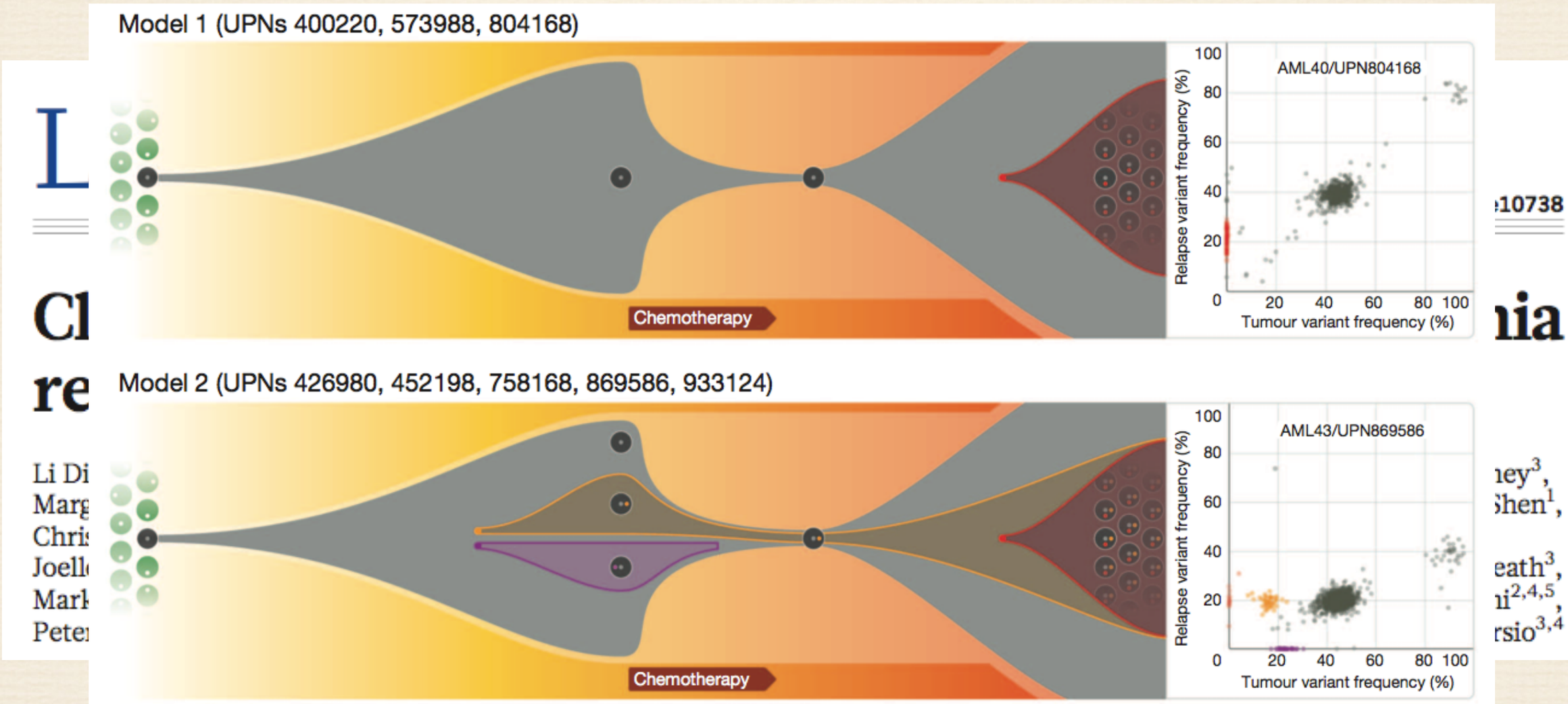


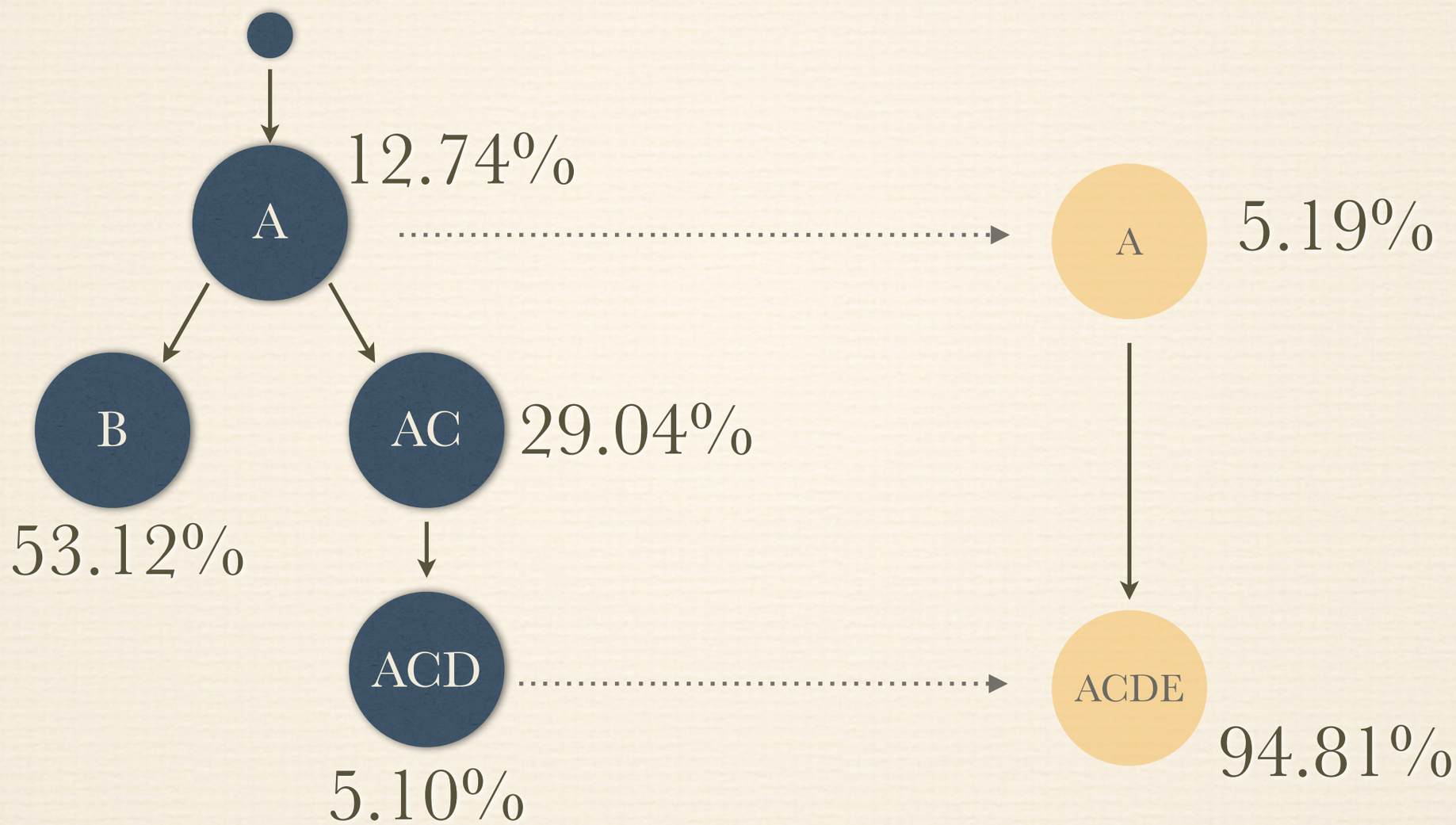
Complete Enumeration



WashU AML Dataset



UPN933124



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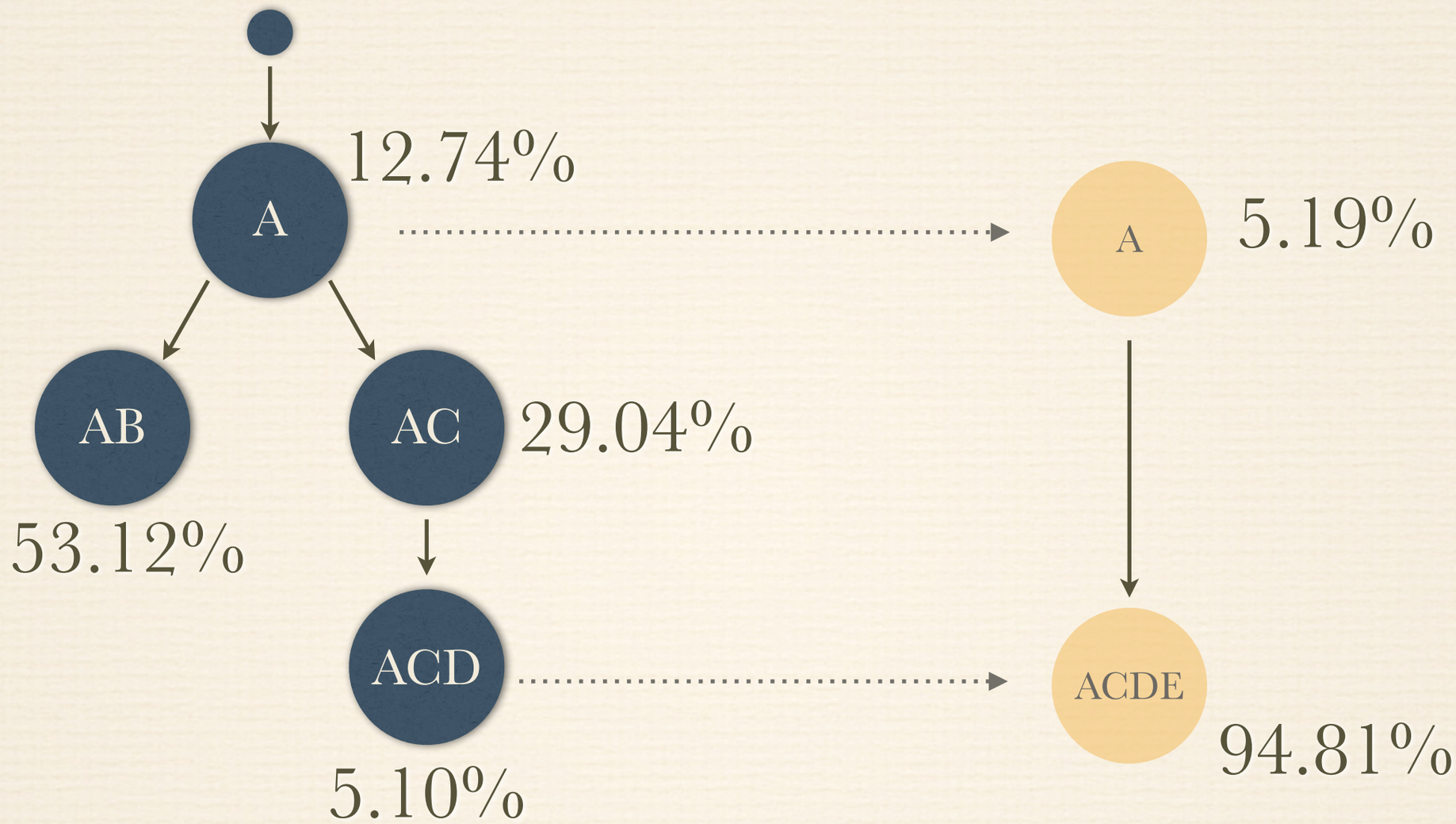
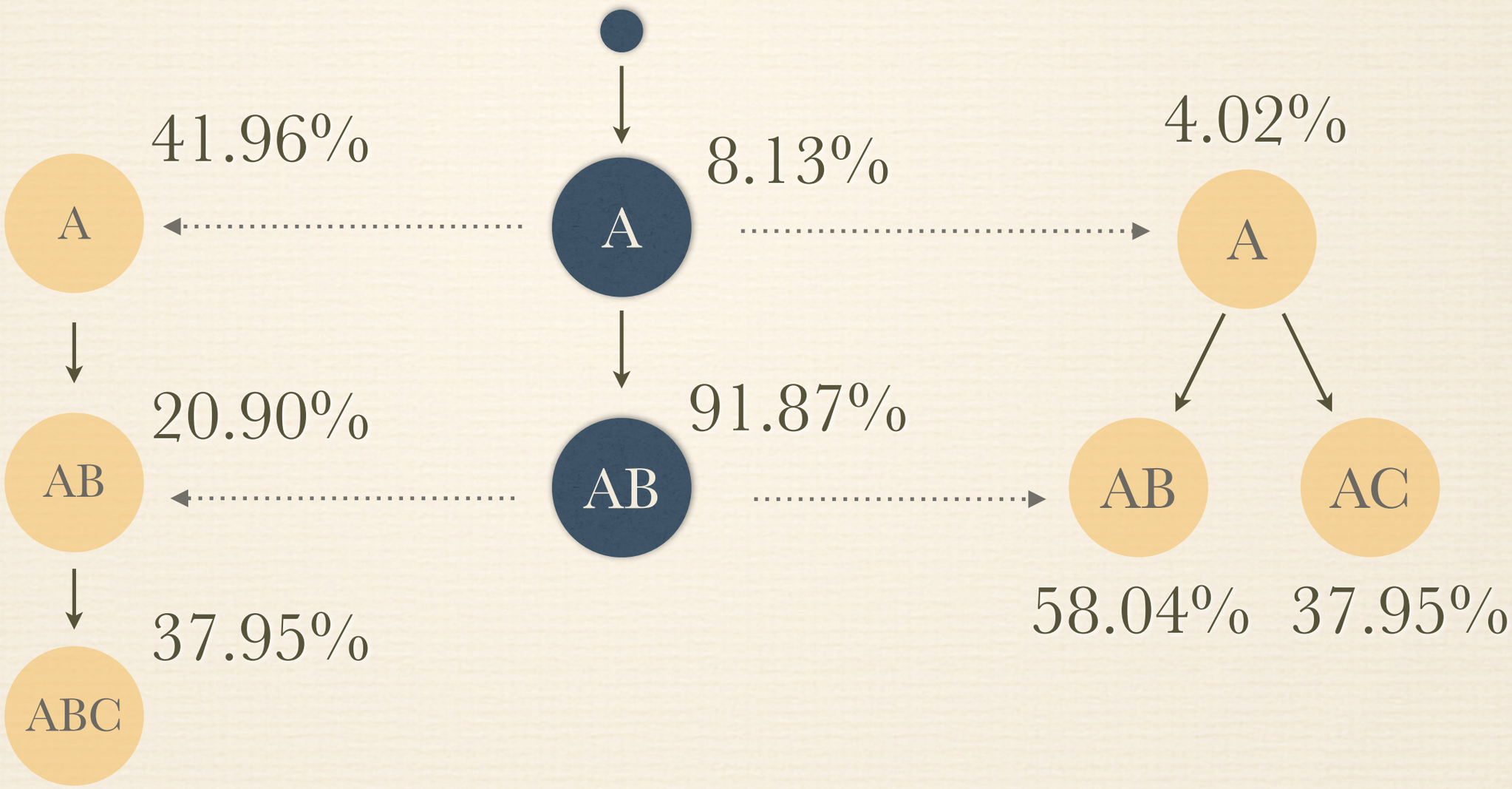


A minor clone carrying the vast majority of the primary tumor mutations survived and expanded at relapse

UPN758168



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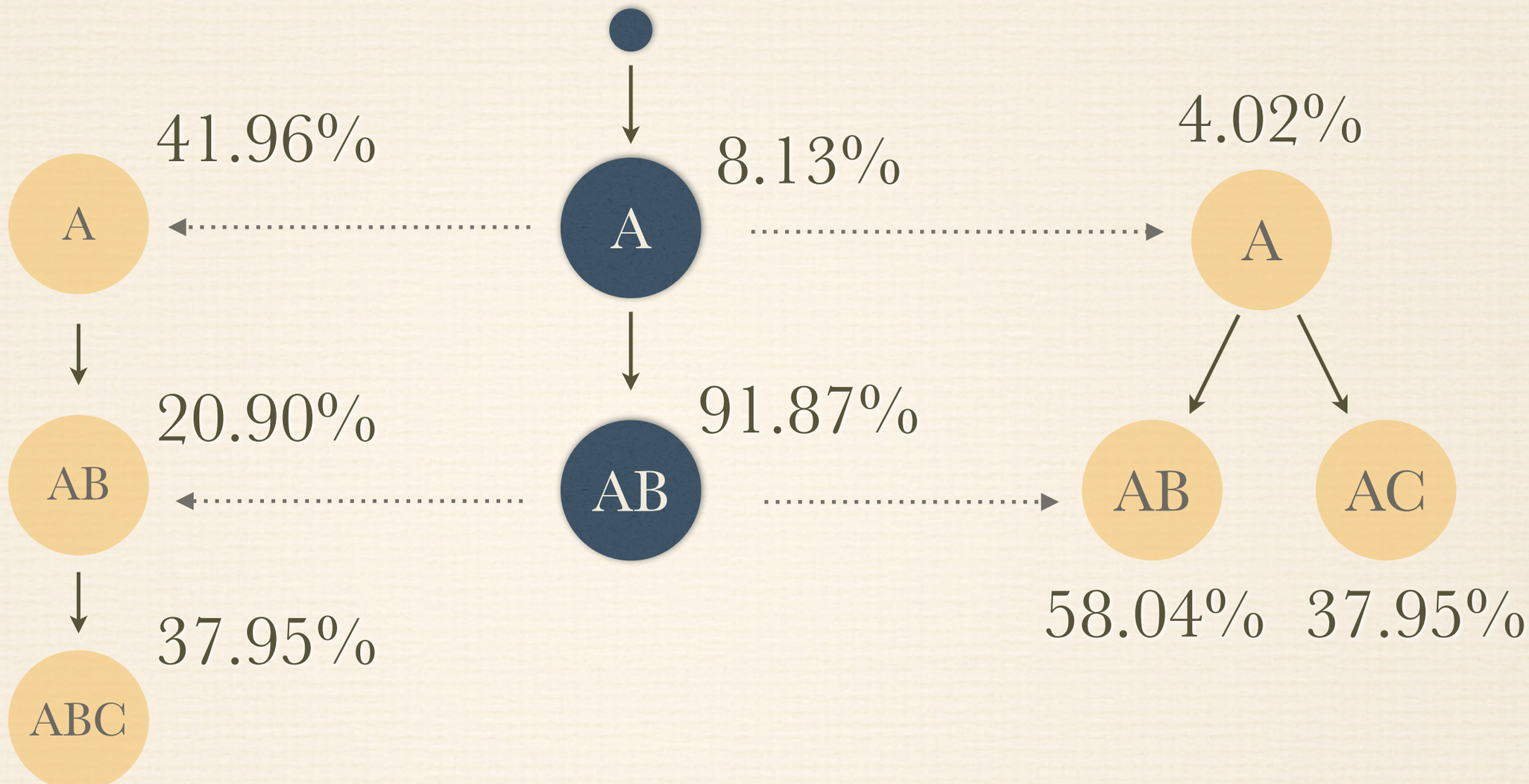

A minor clone carrying the vast majority of the primary tumor mutations survived and expanded at relapse

Demo 3