CS 466
Introduction to Bioinformatics
Lecture 21

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Nov 6, 2019

Outline

- Hidden Markov Models: Viterbi algorithm

Reading:

- Jones and Pevzner: Chapters 11.1-11.3
- Lecture notes

CpG Islands

Question: Given four nucleotides $\Sigma = \{A, T, C, G\}$, what is the probability of observing dinucleotide CG?

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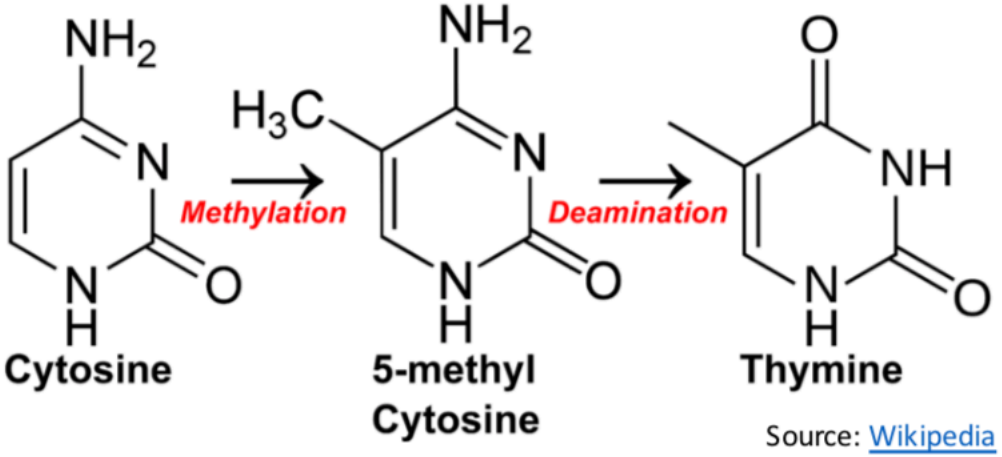

$CG \rightarrow C^*G \rightarrow TG$

CG is least observed dinucleotide as C is easily methylated and has tendency to mutate into a T afterwards

CpG Islands

- Methylation is suppressed around promoter regions of genes in a genome. So CG appears at relatively high frequency within these CpG island.
- Finding CpG islands in a genome is an important problem for annotating genes and regulatory regions.

```
CATTCCGCCTTCTCTCCCGAGGTGGCGCGTGGGA
GGTGTTTTGCTCGGGTTCTGTAAGAATAGGCCAGG
CAGCTTCCCGCCGGGATGCGCTCATCCCCTCTCGG
GGTTCCGCTCCCACCGCGCCGCGCTTCGGCCGGTT
CCGCCTGCGAGATGTTTTCCGACGGACAATGATTC
CACTCTCGGCGCCTCCCATGTTGATCCCAGCTCCT
CTGCGGGCGTCAGGACCCCTGGGCCCCGCCCCGG
CTCCACTCAGTCAATCTTTTGTCCCCGTATAAGGCG
GATTATCGGGTGGCTGGGGGCGGCTGATTCCGA
CGAATGCCCTTGGGGGTCACCCGGAGGGAACTC
CGGGCTCCGGCTTTGGCCAGCCCGCACCCCTGGT
TGAGCCGGCCCGAGGGCCACCAGGGGGCGCTCG
ATGTTCCTGCAGCCCCCCGCAGCAGCCCCACTCC
CCGGCTCACCCTACGGATTGGCTGGCCGCCCCGAG
CTCTGTGCTGTGATTGGTCACAGCCCGTGTCCGTC
GCGGGCGCCGGGCGGATACGAGGTGACGCGCA
GAGGCCCAGCTCGGGCGGTGTCCCGCGCCGGC
GACTGCGGGCGGAGTTTCGCGAGGGCCGAAGCG
GGGCAGTGTGACGGCAGCGGTCCTGGGAGGCGC
CCGCGCGCGTCGGAGCAGCTCCCCGTCCTCCGCA
GCCGTCACCGCCGGCCGTCGCCGCGCCCTGGCC
TCCCGCACTCGCGCACTCCTGTCCGCCGCCCACC
GCCCACCTCCCACCTCGATGCGGTGCCGGGCTGC
TGCGTGATGGGGCTGCGGAGCGGCGCCCTGCGG
CTCGCGGCGCCGCTGCTCGCGCTGAGGTGCGT
CGGTGCCCGGCCCCCGCGCCCCCGCGCGCCGGC
GGCTCCTGTTGACCCGGTCCGCCCGTCGGTCTGC
AGCGCGGCTGAGGTAAGGCGGCGGGGCTGGCCG
CGGTTGGCGCCGCGGTCGCGGGGTTGGGGAGGG
GGCCGCTTCGCGGGGAGGAGCGGCCGGGCCGG
GGTCCGGGCGGGGTCTGAGGGGA
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CTCTTAGTTTTGGGTGCATTTGTCTGGTCTTCCAAA
CTAGATTGAAAGCTCTGAAAAAAAAAACTATCTTGT
GTTTCTATCTGTTGAGCTCATAGTAGGTATCCAGGA
AGTAGTAGGGTTGACTGCATTGATTTGGGACTACAC
TGGGAGTTTTCTTCGCCATCTCCCTTTAGTTTTCCT
TTTTTTCTTTCTTTCTTTTCTTTTTTTTCTTTTTTTT
TTGAGATGTCGTCTTGCTCAGTCCCCCAGGCTGGA
GTGCAGTGGTGCGATCTTGGCTCACTGTAGCCTCC
ACCTCCCAGGTTCAAGCAATTCTACTGCCTTAGCCT
CCCGAGTAGCTGGGATTACAAGCACCCGCCACCAT
TCCTGGCTAATTTTTTTTTTTGTATTTTTAGTTGAGA
CAGGGTTTCACCATGTTGGTGATGCTGGTCTCAGA
CTCCTGGGGCCTAGCGATCCCCCTGCCTCAGCCT
CCCAGAGTGTTAGGATTACAGGCATGAGCCACTGT
ACCCGGCCTCTCTCCAGTTTCCAGTTGGAATCCAA
GGGAAGTAAGTTTAAGATAAAGTTACGATTTTGAAAT
CTTTGGATTCAGAAGAATTTGTCACCTTTAACACCT
AGAGTTGAACGTTCATACCTGGAGAGCCTTAACATT
AAGCCCTAGCCAGCCTCCAGCAAGTGGACATTGGT
CAGGTTTGGCAGGATTCGTCCCCTGAAGTGGACT
GAGAGCCACACCCTGGCCTGTCACCATACCCATCC
CCTATCCTTAGTGAAGCAAAACTCCTTTGTTCCCTT
CTCCTTCTCCTAGTGACAGGAAATATTGTGATCCTA
AAGAATGAAAATAGCTTGTCACCTCGTGGCCTCAG
GCCTCTTGACTTCAGGCGGTTCTGTTTAATCAAGT
GACATCTTCCCGAGGCTCCCTGAATGTGGCAGATG
AAAGAGACTAGTTCAACCCTGACCTGAGGGGAAAG
CCTTTGTGAAGGGTCAGGAG
```

Left: CpG sites at 1/10 nucleotides, constituting a CpG island. The sample is of a gene-promoter, the highlighted ATG consitutes the start codon.
Right: CpG sites present at every 1/100 nucleotides, consituting a more normal example of the genome, or a region of the genome that is commonly methylated.

Source: Wikipedia

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CCGCCTGCGAGATGTTTTCCGACGGACAATGATTC
CACTCTCGGCGCCTCCCATGTTGATCCCAGCTCCT
CTGCGGGCGTCAGGACCCCTGGGCCCCGCCCCG
CTCCACTCAGTCAATCTTTTGTCCCCGTATAAGGCG
GATTATCGGGTGGCTGGGGGCGGCTGATTCCGA
CGAATGCCCTTGGGGGTCACCCGGAGGGAACTC
CGGGCTCCGGCTTTGGCCAGCCCGCACCCCTGGT
TGAGCCGGCCCGAGGGCCACCAGGGGGCGCTCG
ATGTTCCTGCAGCCCCCCGCAGCAGCCCCACTCC
CCGGCTCACCCTACGGATTGGCTGGCCGCCCCGAG
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GCGGGCGCCGGGCGGATACGAGGTGACGCGCA
GAGGCCCAGCTCGGGCGGTGTCCCGCGCCGGC
GACTGCGGGCGGAGTTTCGCGAGGGCCGAAGCG
GGGCAGTGTGACGGCAGCGGTCCTGGGAGGCGC
CCGCGCGCGTCGGAGCAGCTCCCCGTCCTCCGCA
GCCGTCACCGCCGGCCGTCGCCGCGCCCTGGCC
TCCCGCACTCGCGCACTCCTGTCCGCCGCCCACC
GCCCACCTCCCACCTCGATGCGGTGCCGGGCTGC
TGCGTGATGGGGCTGCGGAGCGGCGCCCTGCGG
CTCGCGGCGCCGCTGCTCGCGCTGAGGTGCGT
CGGTGCCCGGCCCCCGCGCCCCCGCGCGCCGGC
GGCTCCTGTTGACCCGGTCCGCCCGTCGGTCTGC
AGCGCGGCTGAGGTAAGGCGGCGGGGCTGGCCG
CGGTTGGCGCCGCGGTCGCGGGGTTGGGGAGGG
GGCCGCTTCCGCGGGGAGGAGCGGCCGGGCCGG
GGTCCGGGCGGGGTCTGAGGGGA
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CTCTTAGTTTTGGGTGCATTTGTCTGGTCTTCCAAA
CTAGATTGAAAGCTCTGAAAAAAAAAACTATCTTGT
GTTTCTATCTGTTGAGCTCATAGTAGGTATCCAGGA
AGTAGTAGGGTTGACTGCATTGATTTGGGACTACAC
TGGGAGTTTTCTTCGCCATCTCCCTTTAGTTTTCCT
TTTTTTCTTTCTTTCTTTTCTTTTTTTTCTTTTTTTT
TTGAGATGTCGTCTTGCTCAGTCCCCCAGGCTGGA
GTGCAGTGGTGCGATCTTGGCTCACTGTAGCCTCC
ACCTCCCAGGTTCAAGCAATTCTACTGCCTTAGCCT
CCCGAGTAGCTGGGATTACAAGCACCCGCCACCAT
TCCTGGCTAATTTTTTTTTTTGTATTTTTAGTTGAGA
CAGGGTTTCACCATGTTGGTGATGCTGGTCTCAGA
CTCCTGGGGCCTAGCGATCCCCCTGCCTCAGCCT
CCCAGAGTGTTAGGATTACAGGCATGAGCCACTGT
ACCCGGCCTCTCTCCAGTTTCCAGTTGGAATCCAA
GGGAAGTAAGTTTAAGATAAAGTTACGATTTTGAAAT
CTTTGGATTCAGAAGAATTTGTCACCTTTAACACCT
AGAGTTGAACGTTCATACCTGGAGAGCCTTAACATT
AAGCCCTAGCCAGCCTCCAGCAAGTGGACATTGGT
CAGGTTTGGCAGGATTCGTCCCCTGAAGTGGACT
GAGAGCCACACCCTGGCCTGTCACCATACCCATCC
CCTATCCTTAGTGAAGCAAAACTCCTTTGTTCCCTT
CTCCTTCTCCTAGTGACAGGAAATATTGTGATCCTA
AAGAATGAAAATAGCTTGTCACCTCGTGGCCTCAG
GCCTCTTGACTTCAGGCGGTTCTGTTTAATCAAGT
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Source: Wikipedia

Input: DNA sequence $\mathbf{x} = x_1 x_2 \ldots x_n$

Output: $\pi : \{1, \ldots, n\} \to \{\text{yes}, \text{no}\}$

$\mathbf{x} =$	CGAT	TG	CGA	AAAAAAT	AACGA	TTATATCG
$\pi =$	yes	no	yes	no	yes	no

Question: How do we identify CpG islands?

A Related Problem: Fair Bet Casino

- Game is to flip coins, two outcomes:

Head or **T**ail

- Two coins: **F**air and **B**iased

$$\Pr(H \mid F) = \Pr(T \mid F) = 1/2$$
$$\Pr(H \mid B) = 3/4, \Pr(T \mid B) = 1/4$$

- The crooked dealer changes between Fair and Biased coins with probability 10%

CpG Islands and Fair Bet Casino

CG Islands

Input: DNA sequence $\mathbf{x} = x_1 x_2 \ldots x_n$ where $x_i \in \{A, T, C, G\}$

Output: $\pi : \{1, \ldots, n\} \to \{\text{yes}, \text{no}\}$

$\mathbf{x} =$	CGAT	TG	CGA	AAAAAAT	AACGA	TTATATCG
$\pi =$	yes	no	yes	no	yes	no

Fair Bet Casino

Input: Coin flips $\mathbf{x} = x_1 x_2 \ldots x_n$ where $x_i \in \{H, T\}$

Output: $\pi : \{1, \ldots, n\} \to \{F, B\}$

$\mathbf{x} =$ 

$\pi =$	F	F	F	B	B	B	B	B	F	F	F
$\pi' =$	F	F	F	B	B	B	F	F	F	F	F

Question: Given $\mathbf{x}$, what is more likely: $\boldsymbol{\pi}$ or $\boldsymbol{\pi}'$?

Markov Model $\mathcal{M} = (Q, A)$

- Set of states Q
  - Markov property:

$$\Pr(Q_i = q_i \mid Q_1 = q_1, \ldots, Q_{i-1} = q_{i-1}) = \Pr(Q_i = q_i \mid Q_{i-1} = q_{i-1})$$

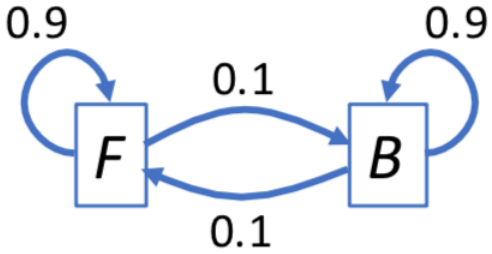
- Transition probabilities $A = [a_{ij}]$ on pairs of states
  - Rows sum to 1

Andrey Markov (source: Wikipedia)

Fair Bet Casino

$Q = \{F, B\}$

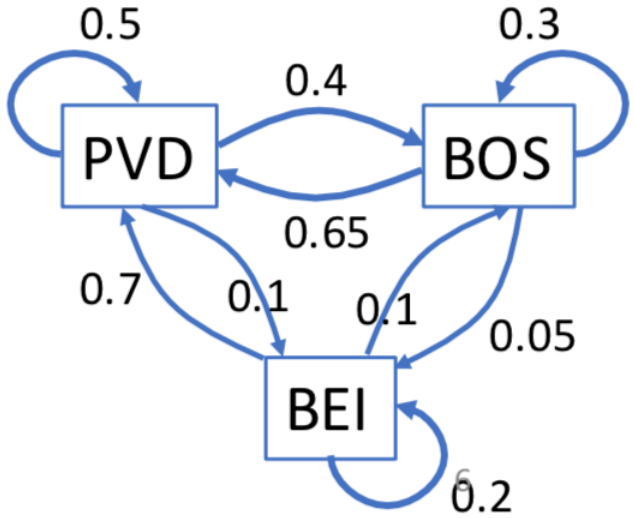
$$A = \begin{pmatrix} 0.9 & 0.1 \\ 0.1 & 0.9 \end{pmatrix}$$



Where is the professor?

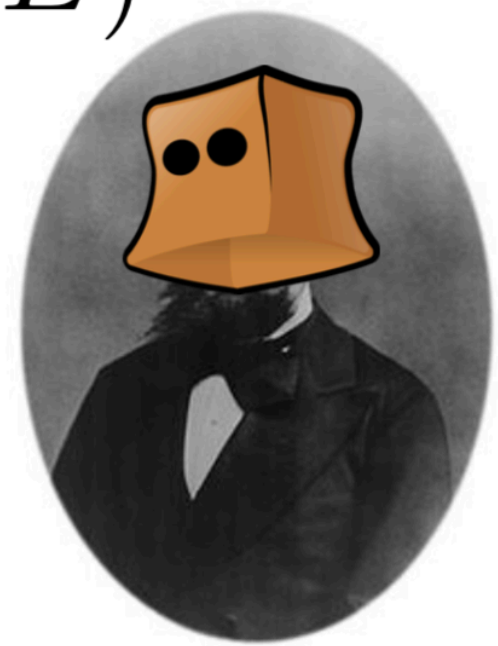
$Q = \{\text{Providence}, \text{Boston}, \text{Beijing}\}$

$$A = \begin{pmatrix} 0.5 & 0.4 & 0.1 \\ 0.65 & 0.3 & 0.05 \\ 0.7 & 0.1 & 0.2 \end{pmatrix}$$



Hidden Markov Model $\mathcal{M} = (Q, A, \Sigma, E)$

- Set of *hidden* states Q
  - Markov property
- Transition probabilities $A = [a_{ij}]$ on pairs of states
- Set of *emitted* symbols Σ
- Emission probabilities $E = [e_{ik}]$ on state-symbol pairs



Andrey Markov

<u>Two decisions:</u>

1. What symbol should I emit?
   [emission probabilities E]
2. What state should I move to next?
   [transition probabilities A]

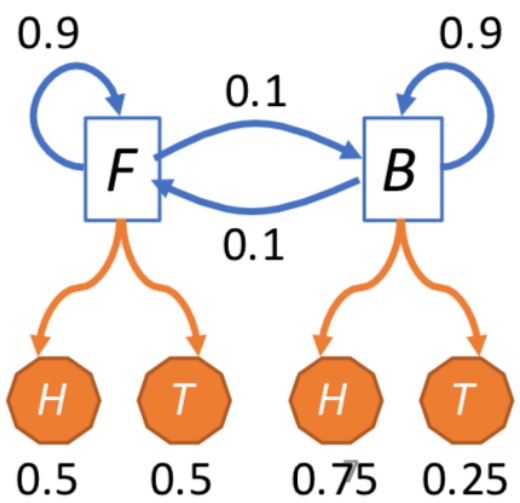
Fair Bet Casino

$Q = \{F, B\}$

$$A = \begin{array}{cc} & \begin{array}{cc} F & B \end{array} \\ & \begin{pmatrix} 0.9 & 0.1 \\ 0.1 & 0.9 \end{pmatrix} \begin{array}{c} F \\ B \end{array} \end{array}$$

$\Sigma = \{H, T\}$

$$E = \begin{array}{cc} & \begin{array}{cc} H & T \end{array} \\ & \begin{pmatrix} 0.5 & 0.5 \\ 0.75 & 0.25 \end{pmatrix} \begin{array}{c} F \\ B \end{array} \end{array}$$



Three Questions

Question 1:
What is the most probable path $\boldsymbol{\pi}^*$ that generated observations $\mathbf{x}$?

Question 2:
What is probability of observations $\mathbf{x}$ generated by any path $\boldsymbol{\pi}$?

Question 3:
What is the probability of observation x_i generated by state s?

Three Questions

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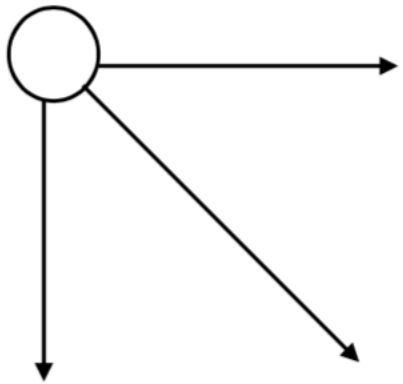
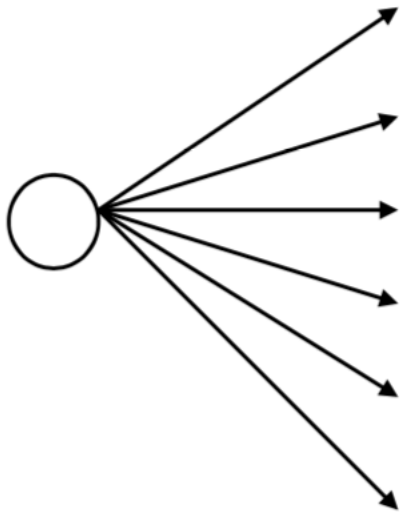
Joint Probability

Recurrence

Alignment vs. Decoding Problem



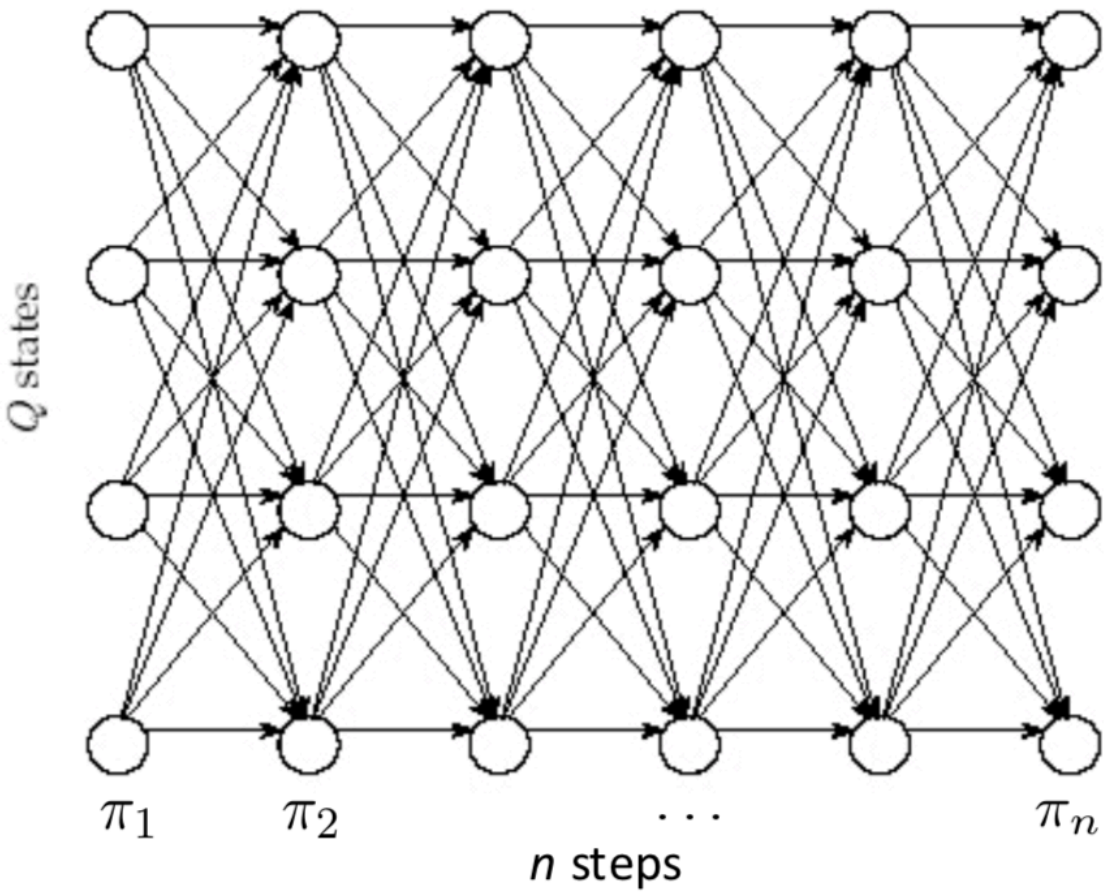
Valid directions in the *alignment problem.*



Valid directions in the *decoding problem.*

Viterbi Algorithm

- Finds path π^* with maximum $\Pr(\mathbf{x}, \pi^*)$
- Dynamic Programming algorithm
- Runs in $O(\#\text{edges}) = O(n|Q|^2)$



Viterbi Algorithm – Numerical Issues

Value of products can become extremely small, leading to underflow

$$v[s,i] = \begin{cases} a_{0,s} \cdot e_{s,x_1}, & \text{if } i = 1, \\ e_{s,x_i} \max_{t \in Q}\{v[t,i-1]a_{t,s}\}, & \text{if } i > 1. \end{cases}$$

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Use logarithms!

$$\log(v[s,i]) = \begin{cases} \log(a_{0,s}) + \log(e_{s,x_1}), & \text{if } i = 1, \\ \log(e_{s,x_i}) + \max_{t \in Q}\{\log(v[t,i-1]) + \log(a_{t,s})\}, & \text{if } i > 1. \end{cases}$$

Fair Bet Casino: Example

$\mathbf{x} =$ 

	0	1 (T)	2 (H)	3 (T)	4 (H)	5 (H)	6 (H)	7 (T)	8 (H)	9 (H)	10 (H)	11 (T)
F												
B												

$Q = \{F, B\}$

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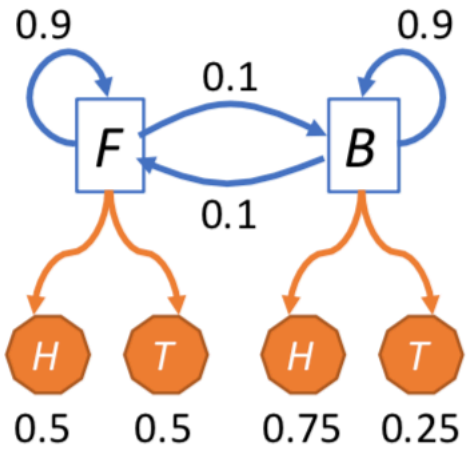

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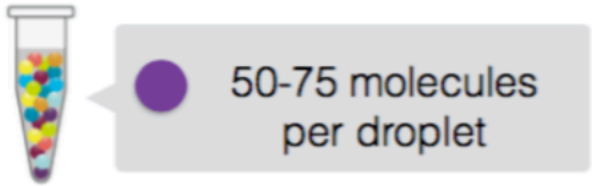
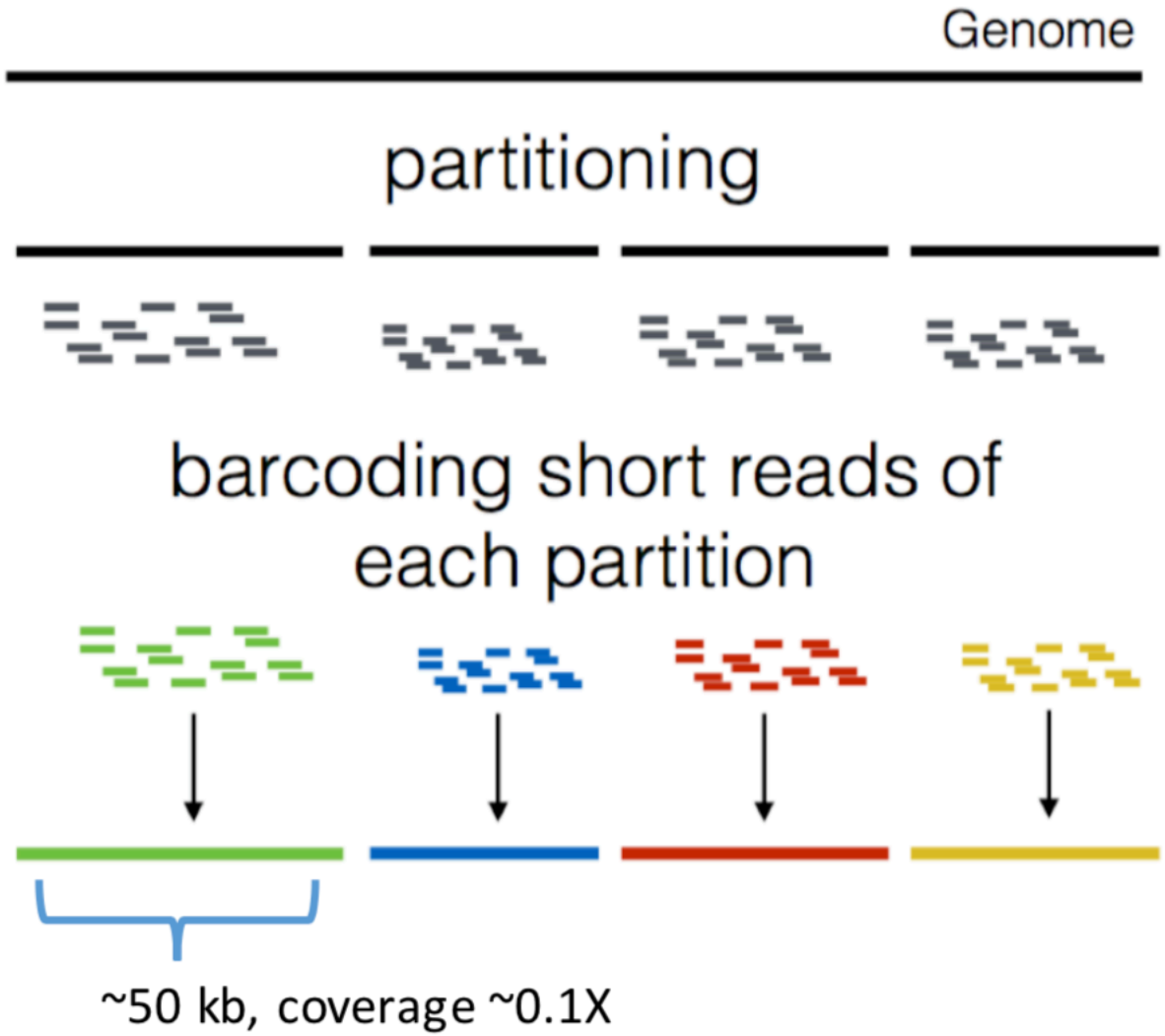
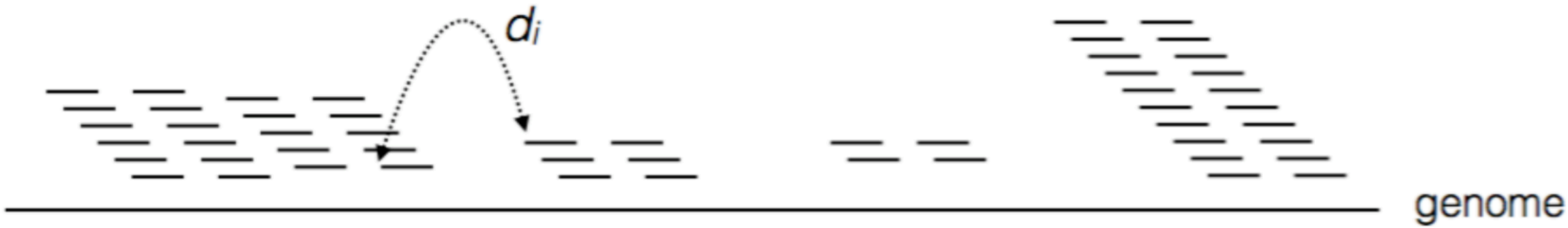
10X Genomics: Synthetic Long Reads

Genome indexing by partitioning and molecular barcoding





$R_j = \{r_i \mid \forall\ i,\ r_i$ contains barcode $j\}$: Paired-reads possessing barcode ***j***



Sort linked-reads and calculate distances between them

$D_j = [d_i \mid \forall\ i,\ d_i$: distance between r_i and $r_{i+1},\ r_i < r_{i+1}]$

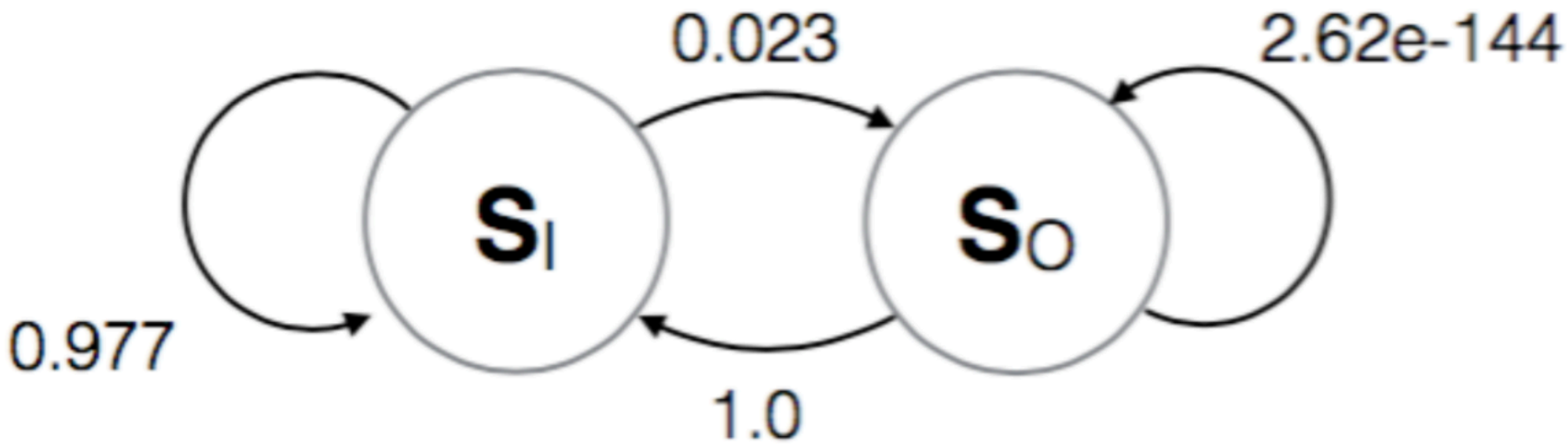
Define distances d_i as intra- or inter- long molecules

$\Sigma_i = [d_1, d_2, d_3, \ldots d_{500}, d_{501}, d_{502}, \ldots d_{1001}, d_{1002}, d_{1003}, \ldots]$

$Q_j = [\ I,\ \ I,\ \ I,\ \ldots,\ \ I,\ \ O,\ \ I, \ldots,\ \ I,\ \ O,\ \ I, \ldots.\ ]$

I: intra long molecule, O: inter long molecules

HMM



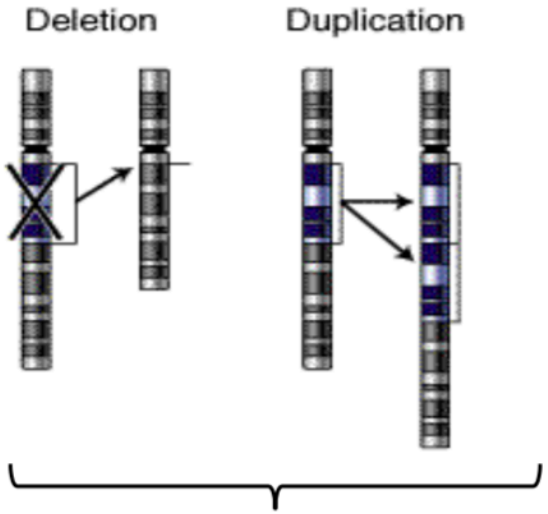
Σ_i =[d_1, d_2, d_3, … d_{500}, d_{501}, d_{502}, … d_{1001}, d_{1002}, d_{1003}, …]

Q_j =[I, I, I, …, I, O, I, …, I, O, I, ….]

ℓ_{j1} ℓ_{j2} ℓ_{j3}

I: intra long molecule, O: inter long molecules

Copy Number Variation

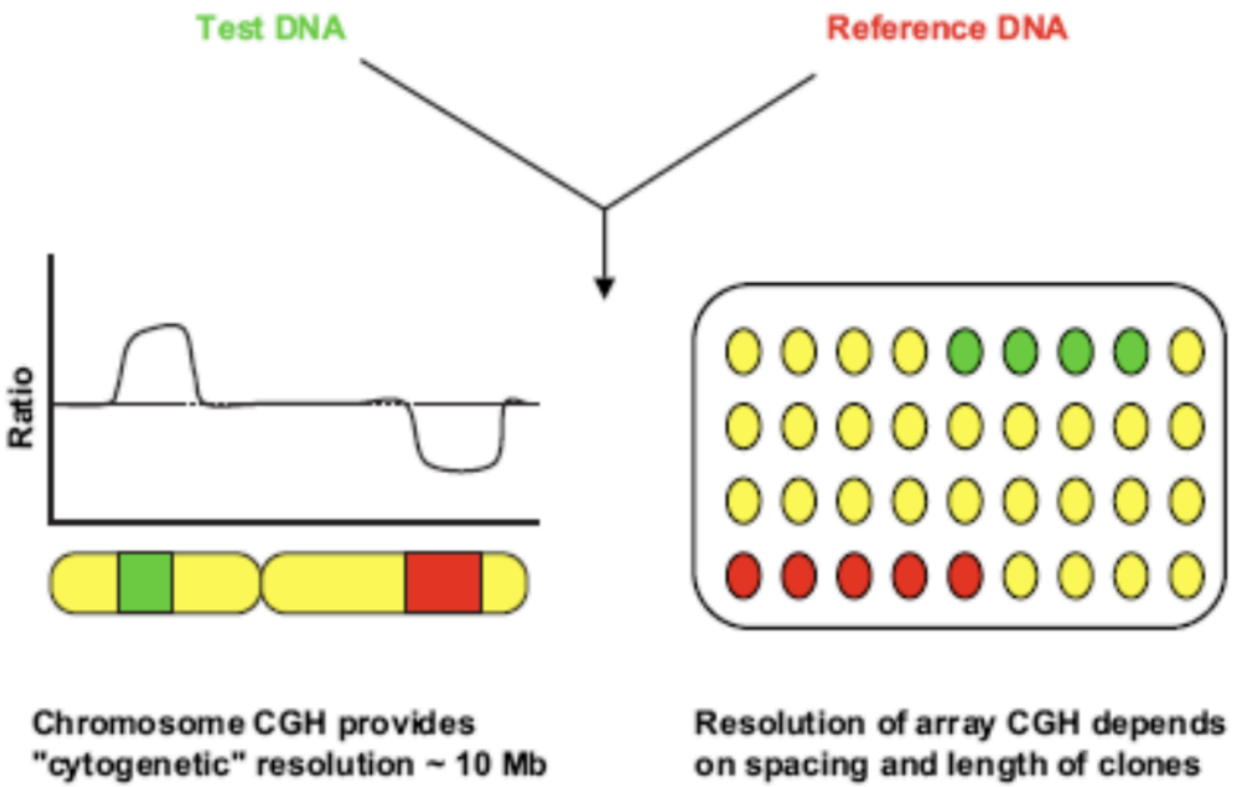


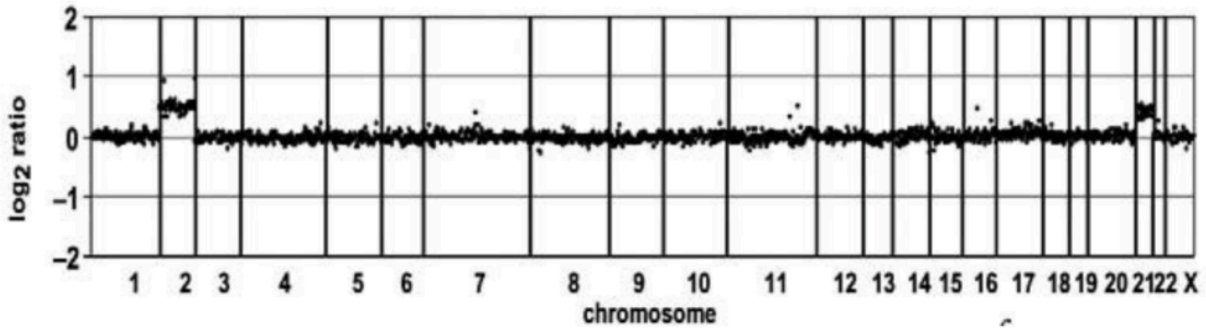
Copy Number Variants

- Different individuals may have different number of copies of segments of genome.
- These variants are associated with various diseases: autism, schizophrenia, cancer

Measuring Copy Number Variants

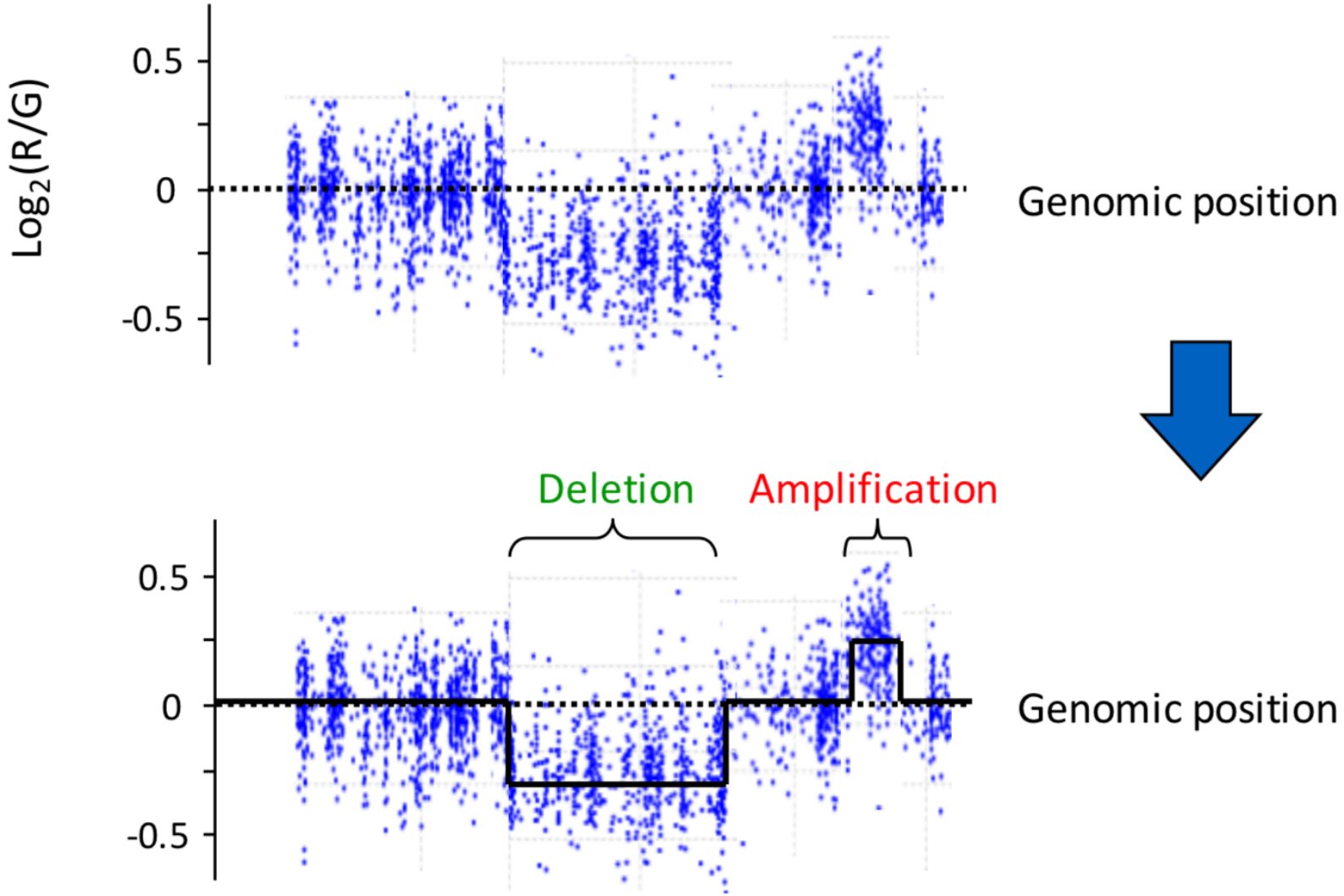
Comparative Genomic Hybridization (CGH)





Segmentation and Copy Number Calling

Divide genome into segments of equal copy number



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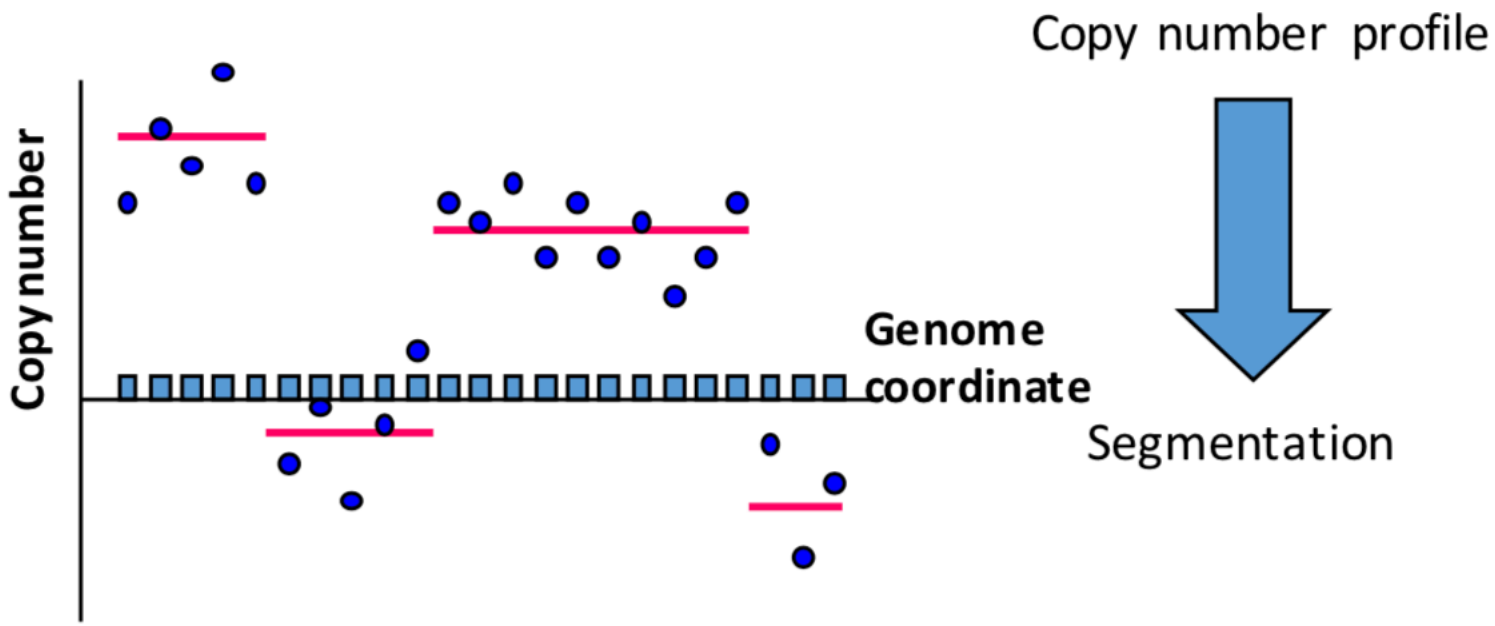

Input: $X_i = \log_2 T_i / R_i$, clone $i = 1, \ldots, N$

Output: Assignment $s(i) \in \{S_1, \ldots, S_K\}$ where S_i represent *copy number states*

Summary

- Markov property – Current state depends only on previous state
- Hidden Markov Models: states are not given only emitted symbols
- Viterbi algorithm: Find the most likely sequence of states given a set of observations

Reading:

- Jones and Pevzner: Chapters 11.1-11.3
- Lecture notes