Fred Hutch
Cancer Center

CANCER GENOMICS

Lecture 3: Probabilistic Methods for Profiling Copy Number Alterations

GENOME 541 Spring 2023
May 16, 2023

Gavin Ha, Ph.D.
Public Health Sciences Division
Human Biology Division

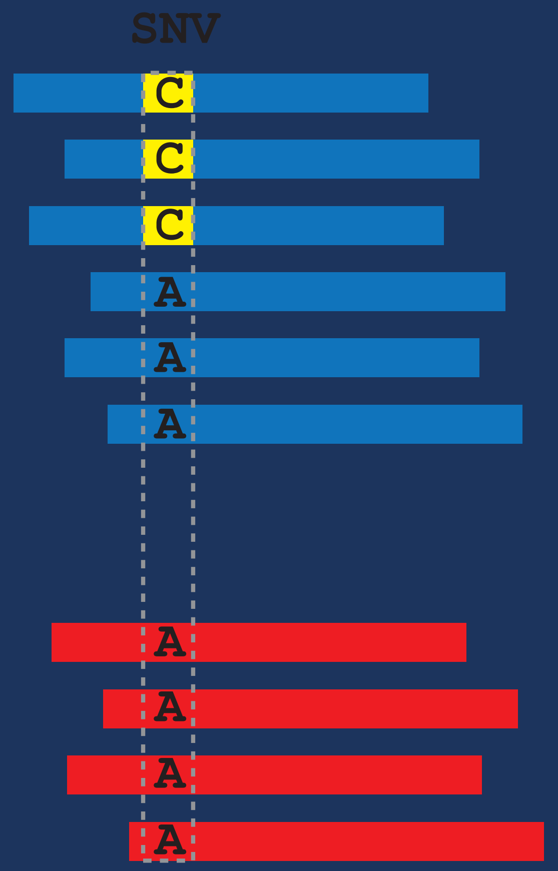
@GavinHa
gha@fredhutch.org
https://github.com/GavinHaLab
GavinHaLab.org

Outline: Probabilistic Methods for Mutation Detection

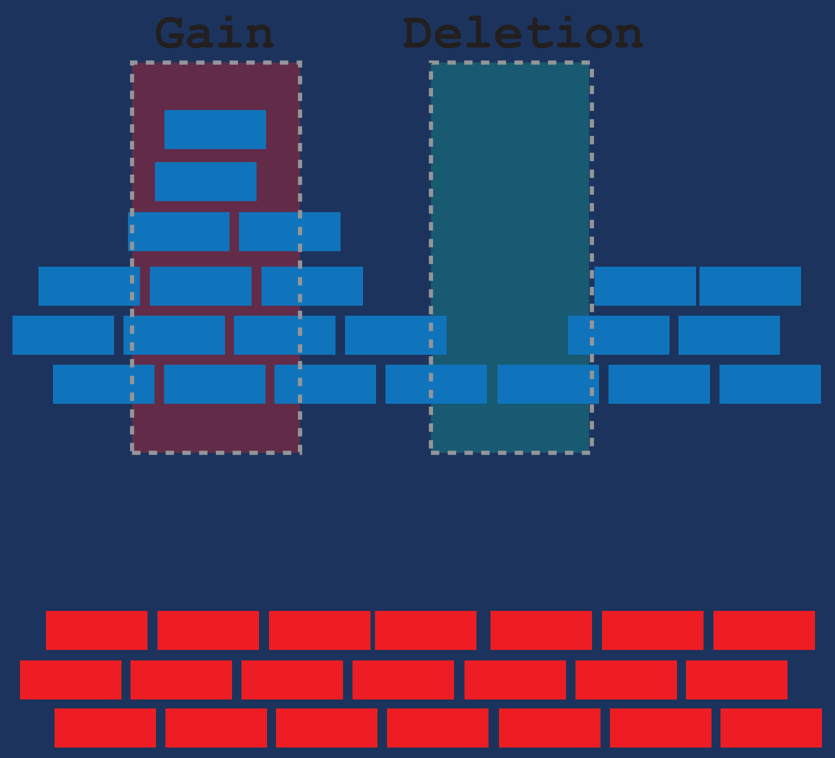
1. **Detecting Copy Number Alterations in Cancer Genomes**
   - Predicting copy number features from sequence data
   - Copy number analysis workflow
   - Data normalization
2. **Continuous Hidden Markov Model (HMM)**
   - Graphical model representation
   - Components of a continuous HMM
   - Inference & parameter estimation using expectation-maximization (EM)
3. **Copy Number Profiling using a Hidden Markov Model**
   - Probabilistic model for copy number analysis
   - Predicting copy number segments using the Viterbi algorithm

2. Detecting Mutations in Cancer Genomes

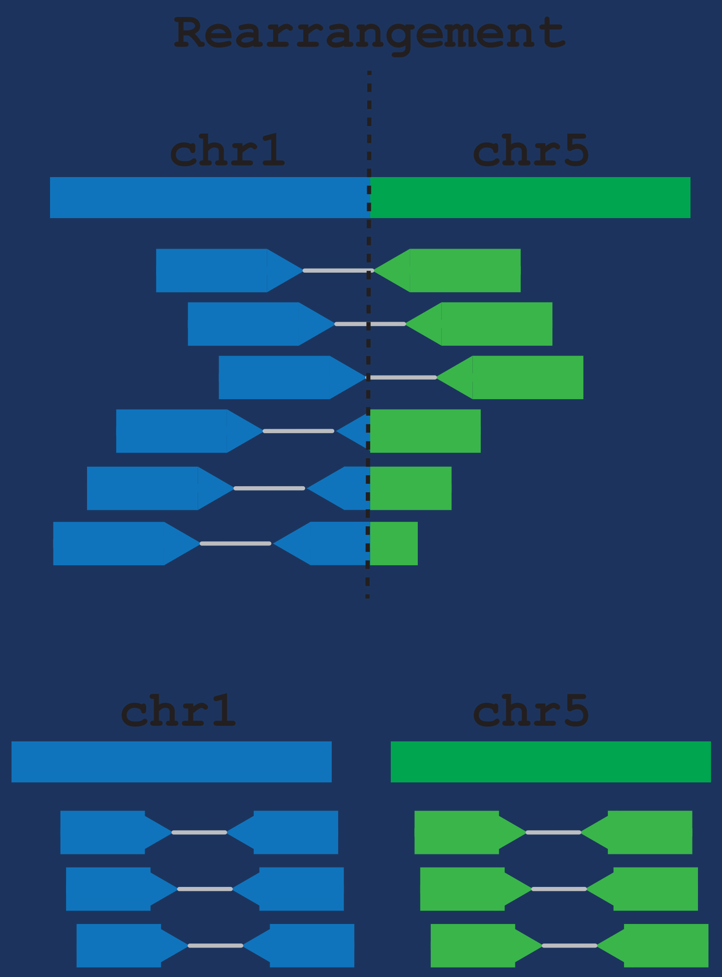
Mutations (SNV, INDEL)



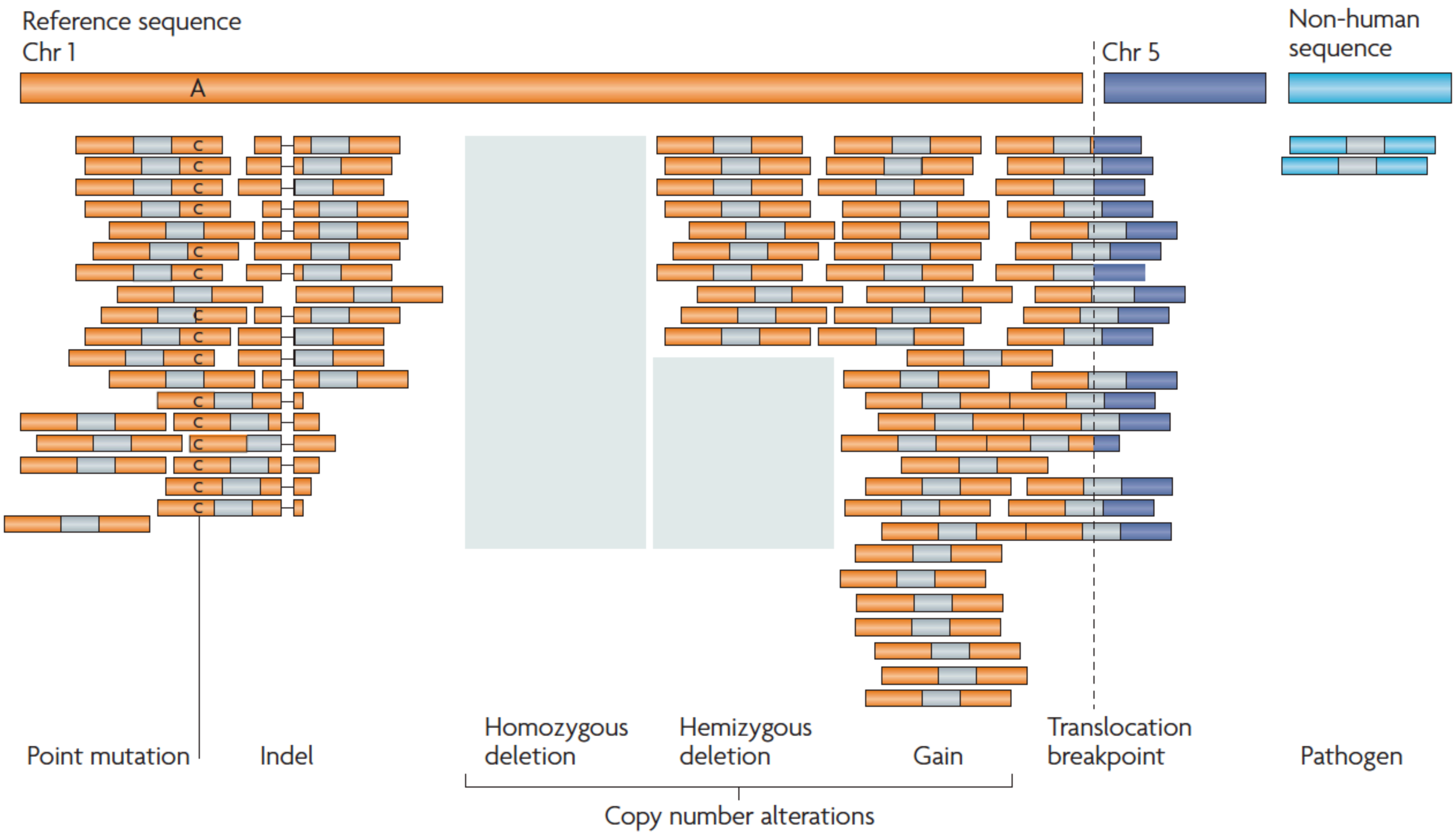
Copy Number Alterations



Structural Variants



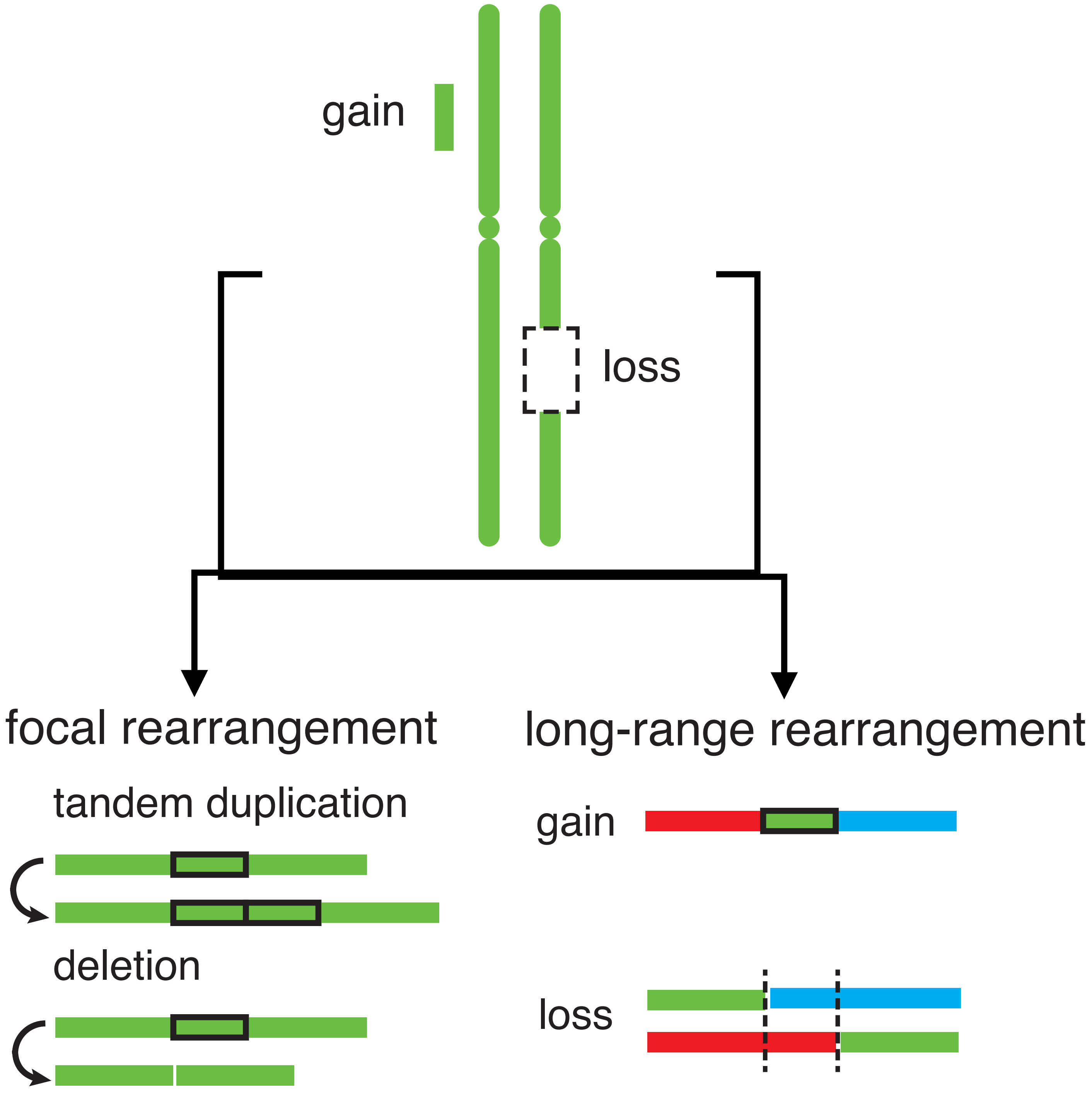
Predicting genomic alterations from sequence data



Meyerson, Gabriel & Getz. *Nature Review Genetics* **11**:685-96 (2010)

Predicting genomic alterations from sequence data

Copy number alterations
(amplitude/dosage)



gain

loss

focal rearrangement

long-range rearrangement

tandem duplication

gain

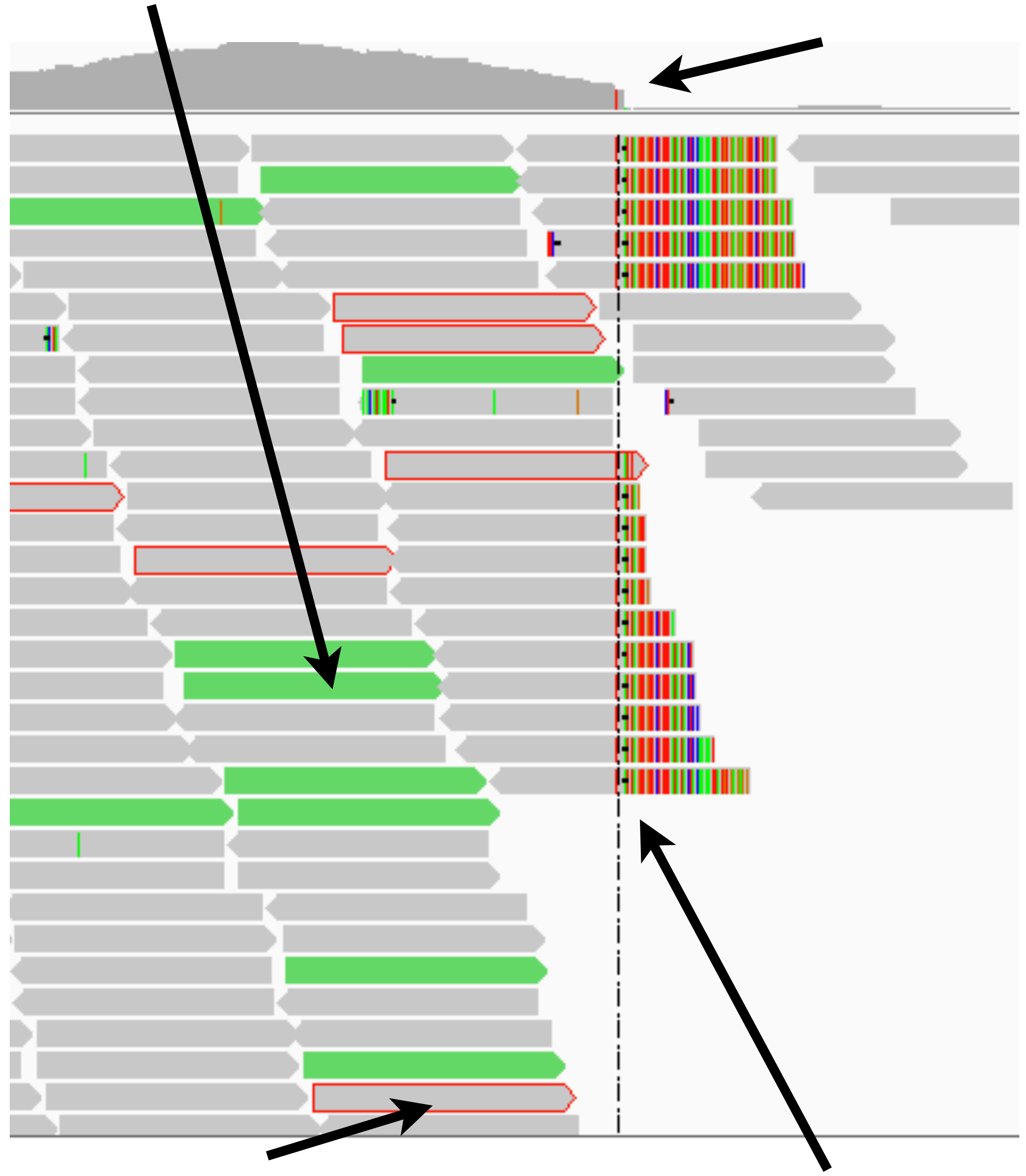
deletion

loss

Structural rearrangements
(location/configuration)

“discordant read pair”
read pairs with aberrant inferred fragment length

“copy number change”
abrupt change in read coverage



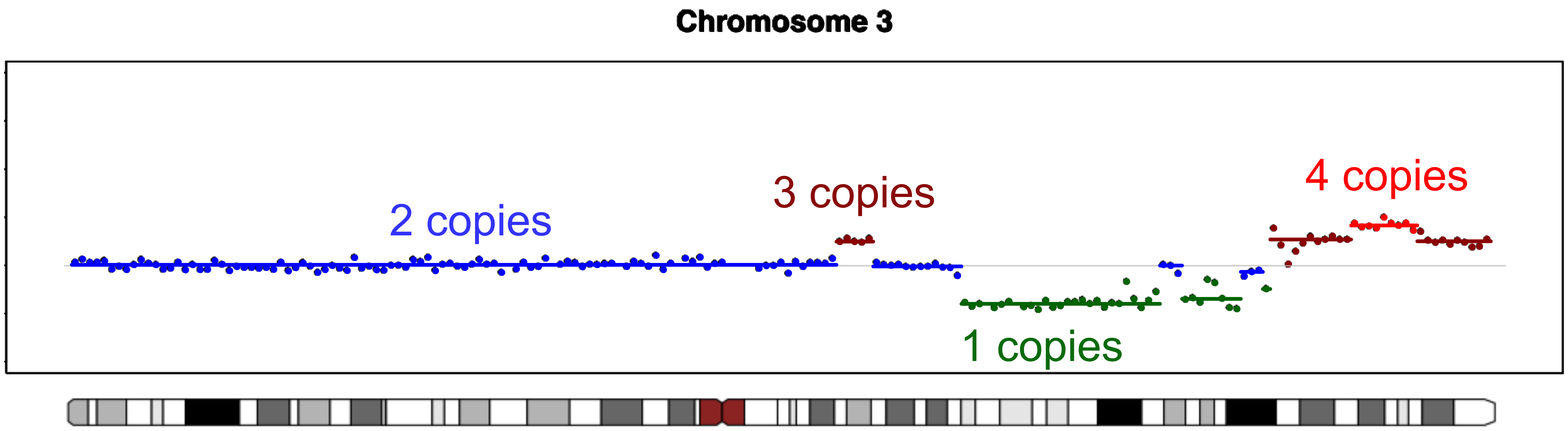
pair-mates unmapped

“split read”
split alignments

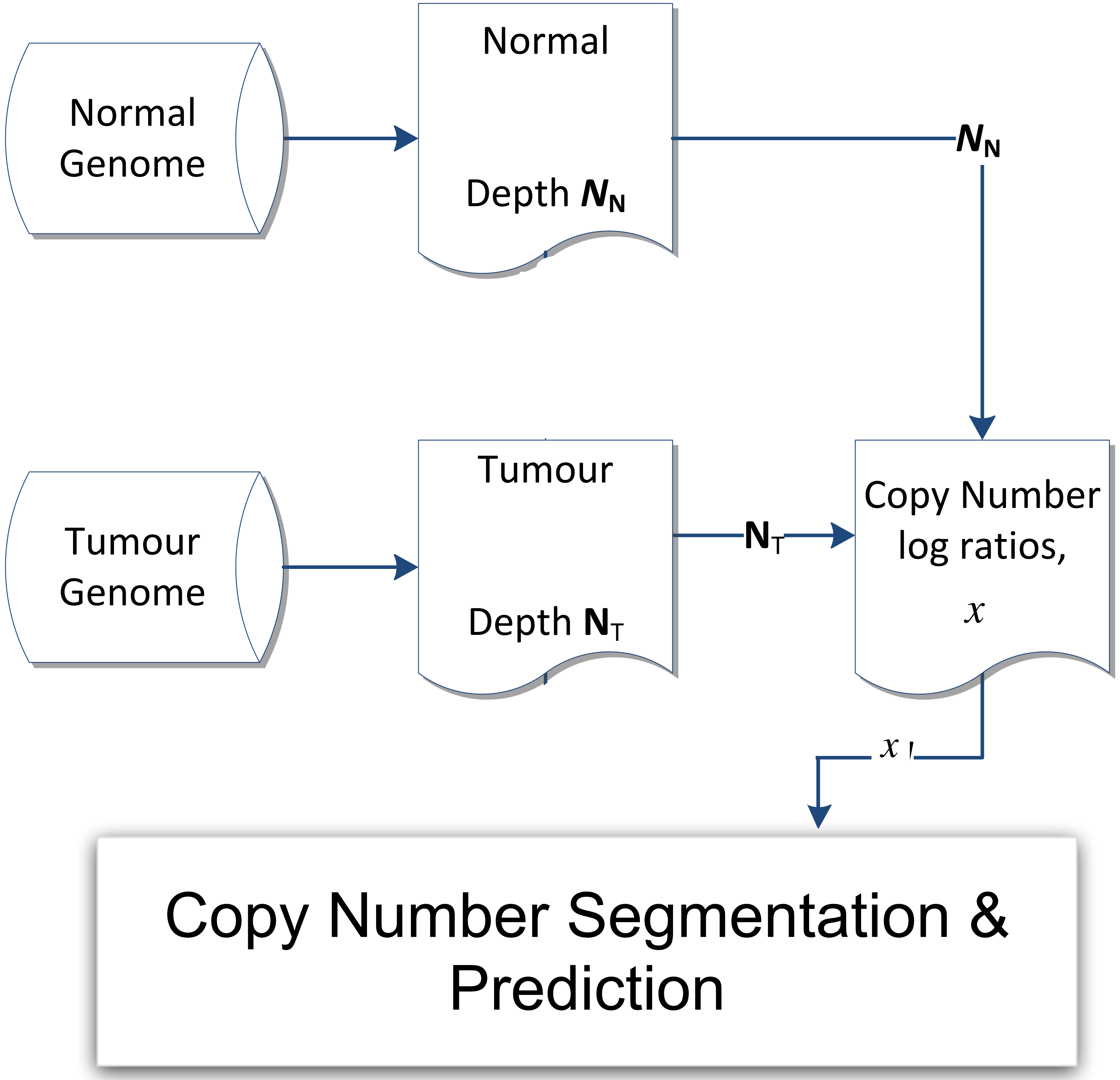
Zhang and Pellman. *CSH Symp Quant Biol.* **80**:117-37 (2016)

Tumor DNA Copy Number Analysis Strategy

1. Using sequencing read coverage as a measure for DNA copy number
2. Identifying segments of coverage changes
3. Predicting the number of copies for each segment

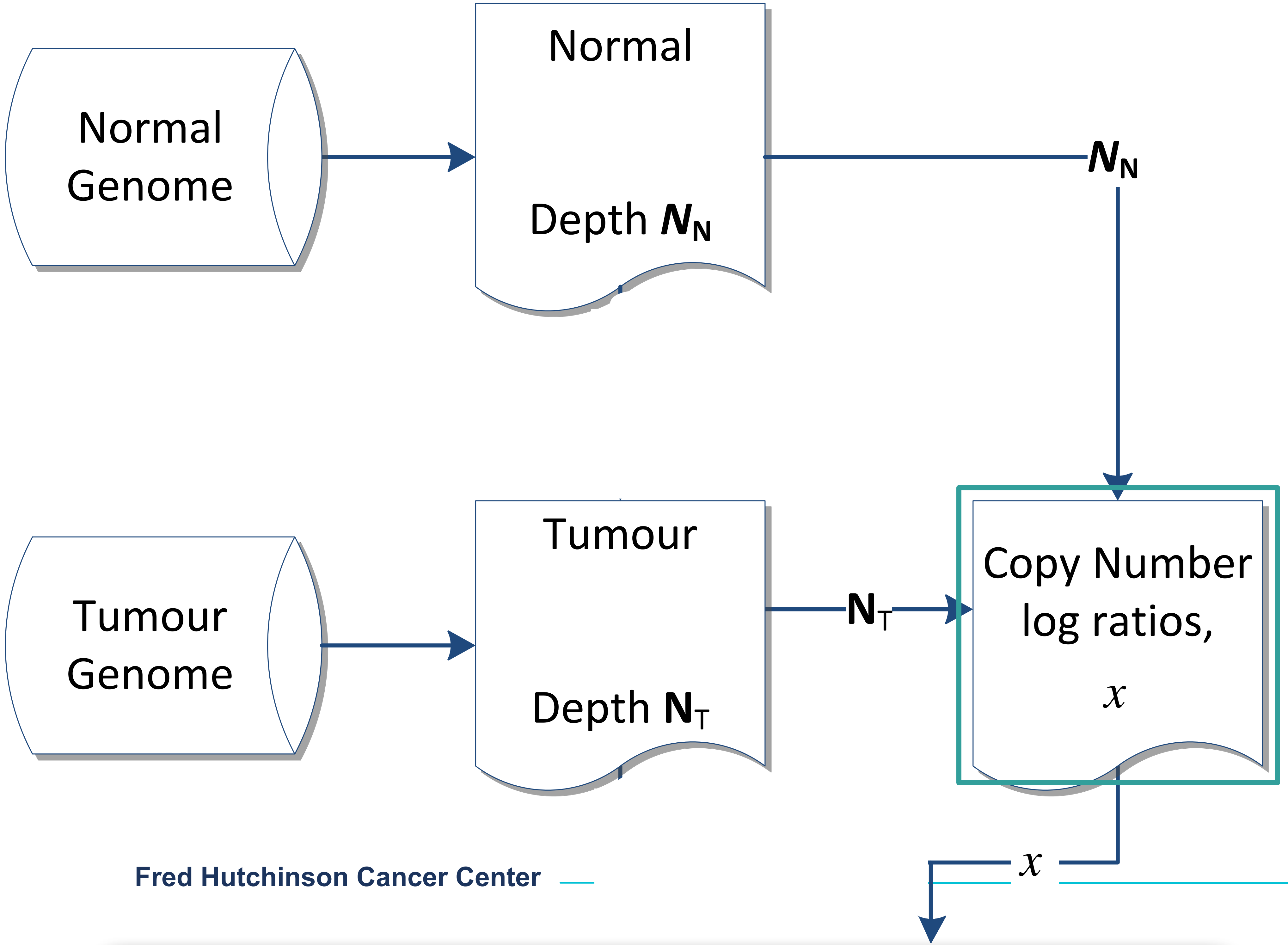


Cancer Genome Copy Number Analysis Workflow



Copy Number Analysis Workflow: Normalization

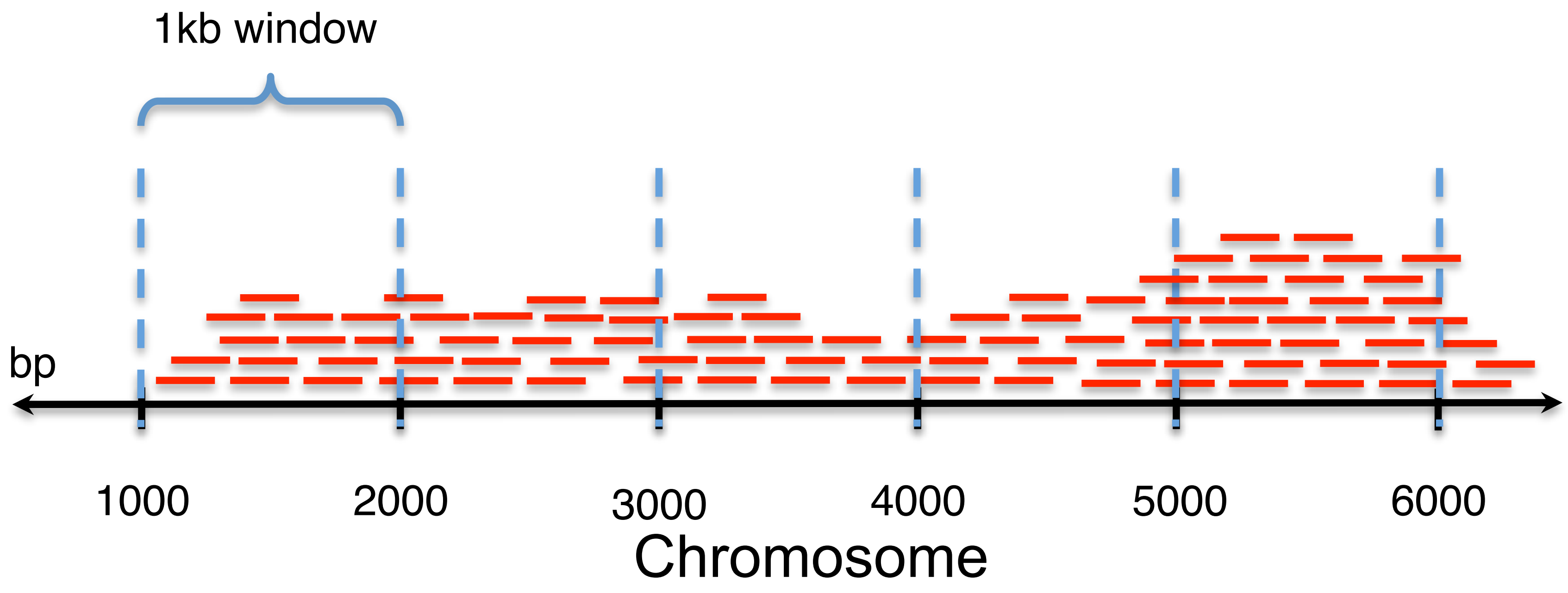
1. Correct GC/mappability biases for tumor read depth



$$N^{normal} = normal\ read\ depth$$

$$N^{tumor} = tumor\ read\ depth$$

$$\frac{N^{tumor}}{N^{normal}} = copyratio$$



Copy Number Analysis Workflow: GC content bias

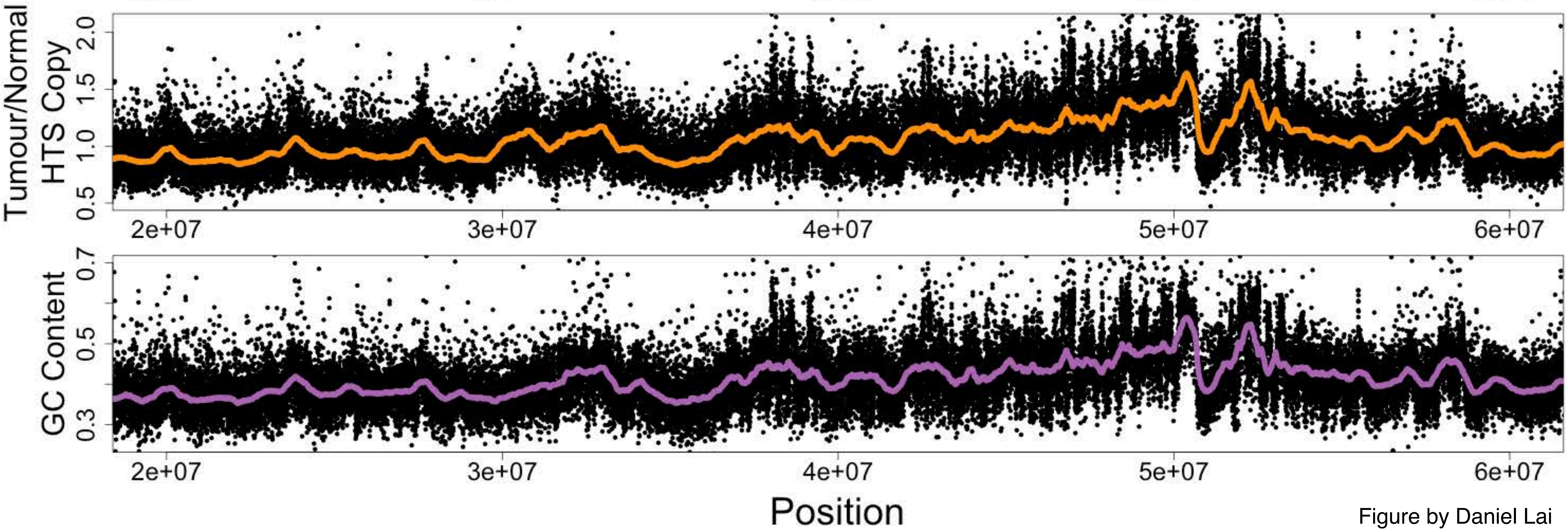
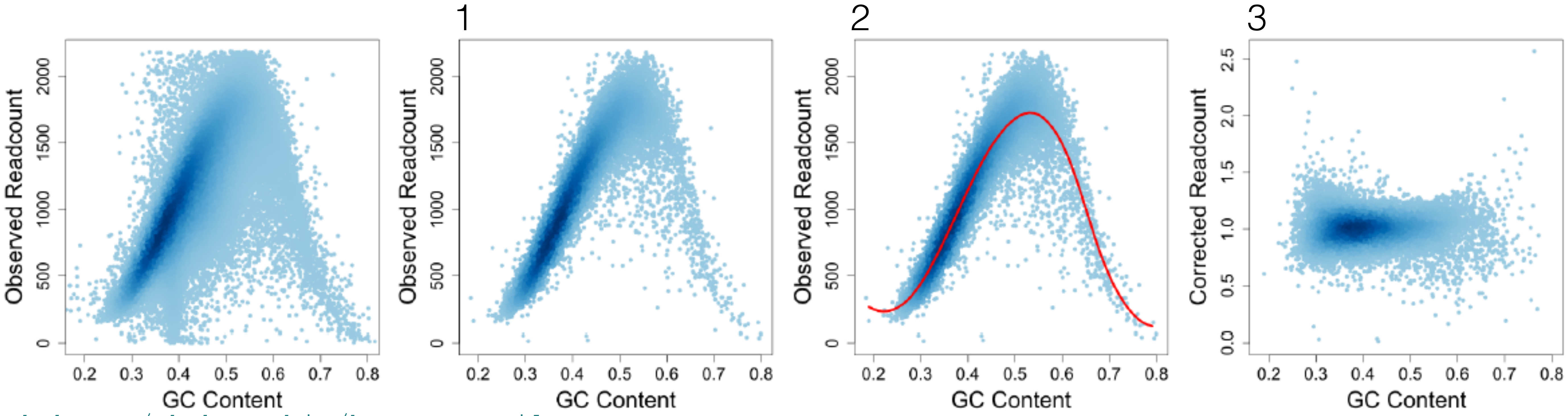


Figure by Daniel Lai

Benjamini and Speed. *Nucleic Acids Research* **40**:e72-86 (2012)
Boeva et al. *Bioinformatics* **29**(3):423-5 (2012)
Ha et al. *Genome Research* **22**:1995-2007 (2012).
Adalsteinsson*, Ha* Freeman* et al. *Nature Communications* **8**:1324 (2017)

Copy Number Analysis Workflow: GC correction (1)

1. Randomly select 50k bins and filter outliers (bottom & top 1%)
2. Fit `loess()` curve
   - local nonlinear regression
   - smoothing parameter (bandwidth): amount of local data to fit
3. $corrected\ read\ count = \frac{observed\ read\ count\ (blue\ dot)}{expected\ read\ count\ (red\ line)}$
   - relative differences between observed and predicted read counts

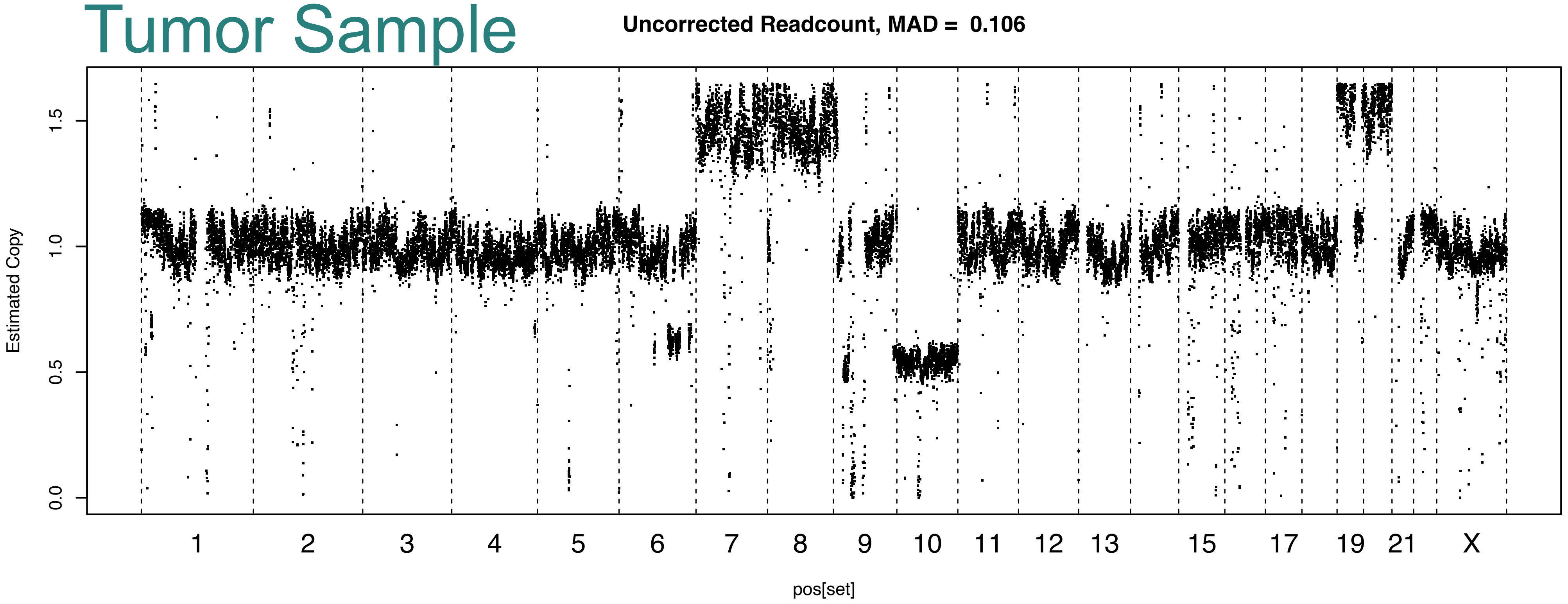


https://github.com/shahcompbio/hmmcopy_utils
https://github.com/GavinHaLab/ichorCNA

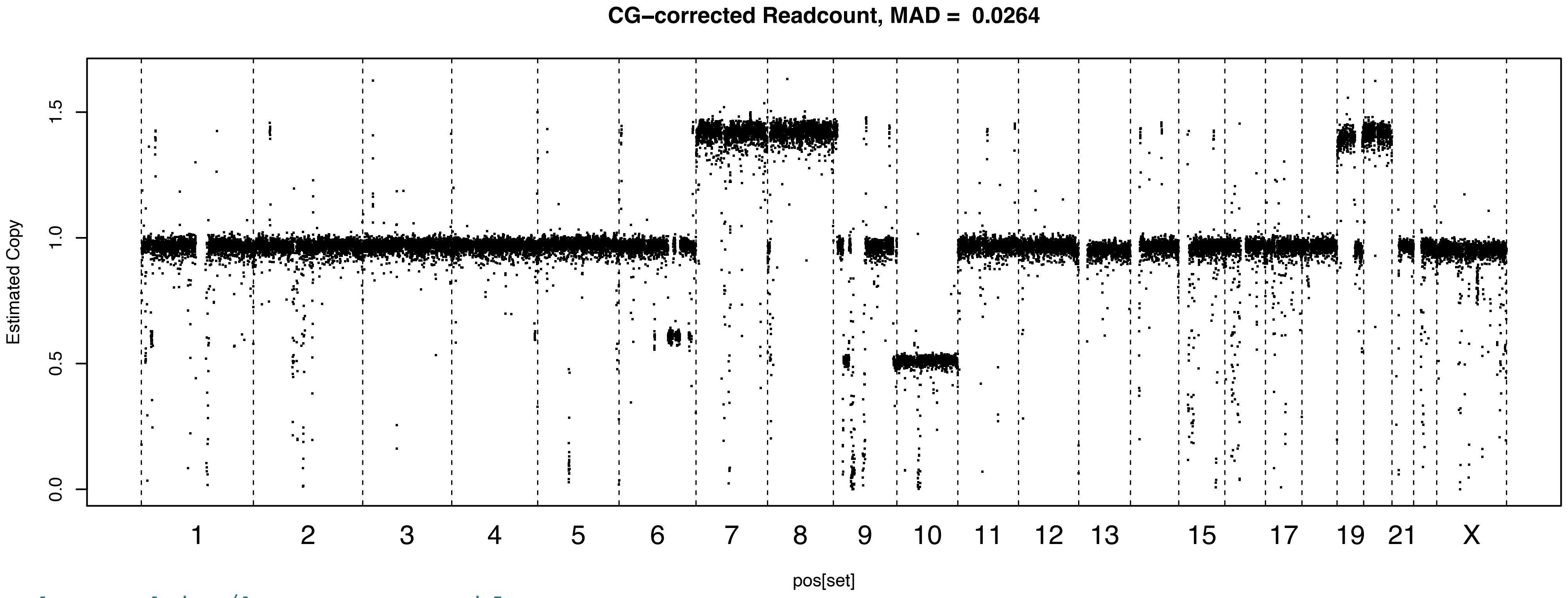
Benjamini and Speed. *Nucleic Acids Research* **40**:e72-86 (2012)
Boeva et al. *Bioinformatics* **29**(3):423-5 (2012)
Ha et al. *Genome Research* **22**:1995-2007 (2012).
Adalsteinsson*, Ha* Freeman* et al. *Nature Communications* **8**:1324 (2017)

Copy Number Analysis Workflow: GC correction (2)

Tumor Sample



Un-corrected read counts

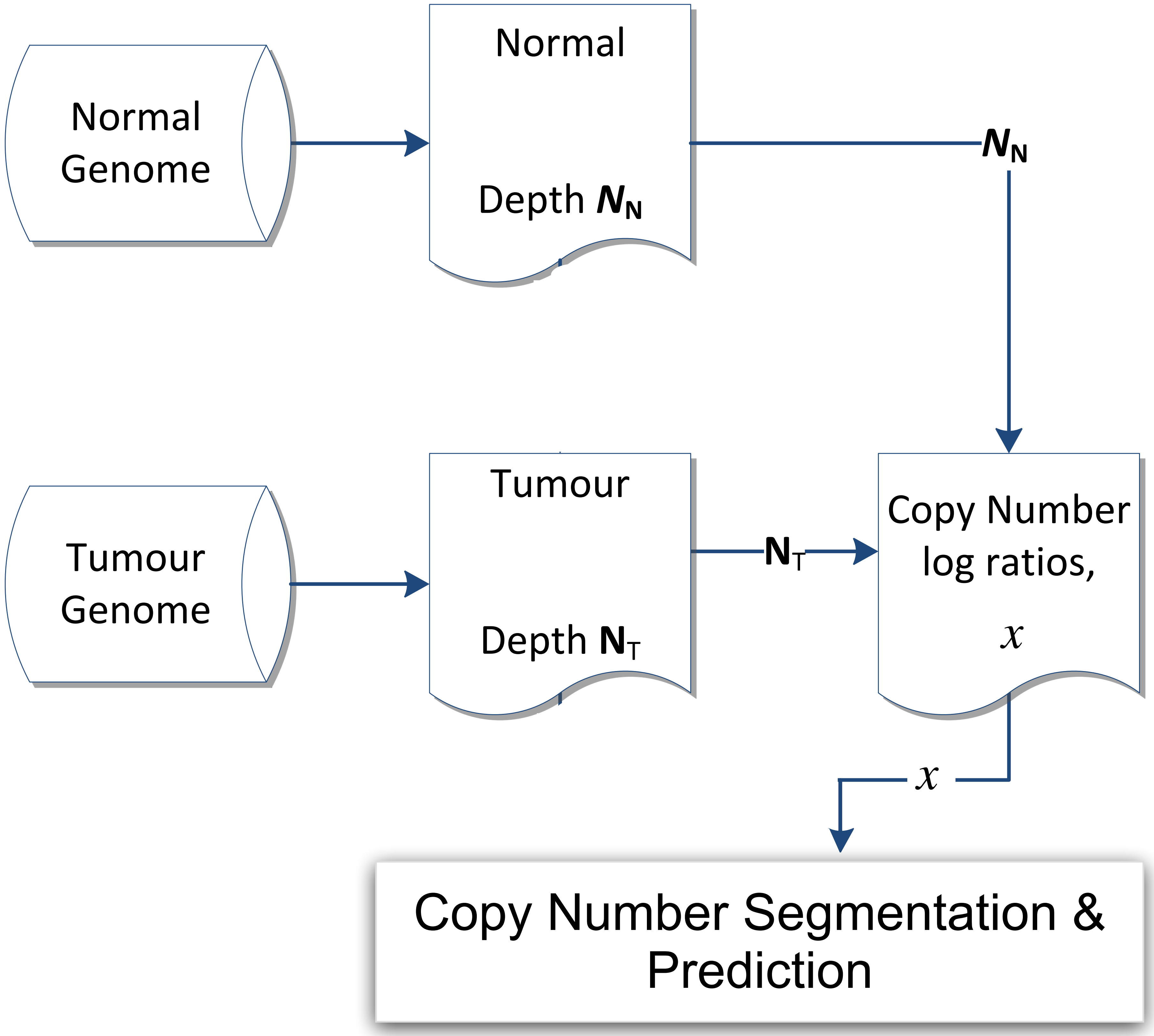


GC-corrected read counts

https://github.com/shahcompbio/hmmcopy_utils
https://github.com/GavinHaLab/ichorCNA

Benjamini and Speed. *Nucleic Acids Research* **40**:e72-86 (2012)
Boeva et al. *Bioinformatics* **29**(3):423-5 (2012)
Ha et al. *Genome Research* **22**:1995-2007 (2012).
Adalsteinsson*, Ha* Freeman* et al. *Nature Communications* **8**:1324 (2017)

Copy Number Analysis Workflow: Normalization



1. **Correct GC/mappability biases for tumor read depth**

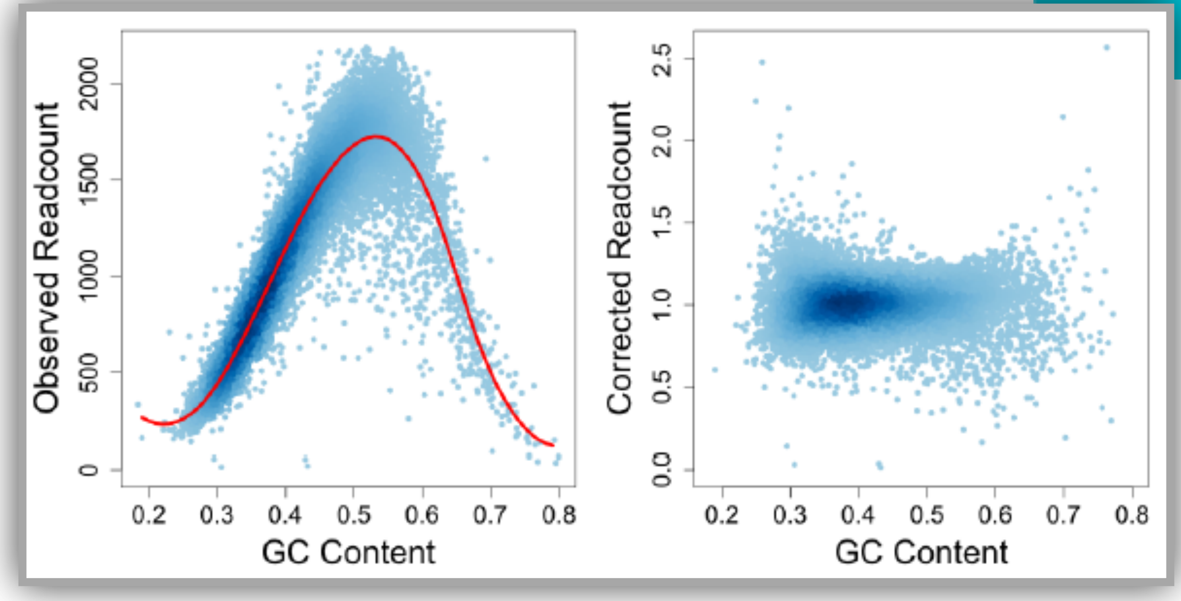
$$N^{normal} = normal\ read\ depth$$
$$N^{tumor} = tumor\ read\ depth$$
$$\hat{N}^{normal} = corrected\ normal\ read\ depth$$
$$\hat{N}^{tumor} = corrected\ tumor\ read\ depth$$
$$\log_2\left(\frac{\hat{N}^{tumor}}{\hat{N}^{normal}}\right) = corrected\ log\ ratio$$

2. **Perform segmentation and copy number prediction**

Input Sequencing Data for Copy Number Analysis

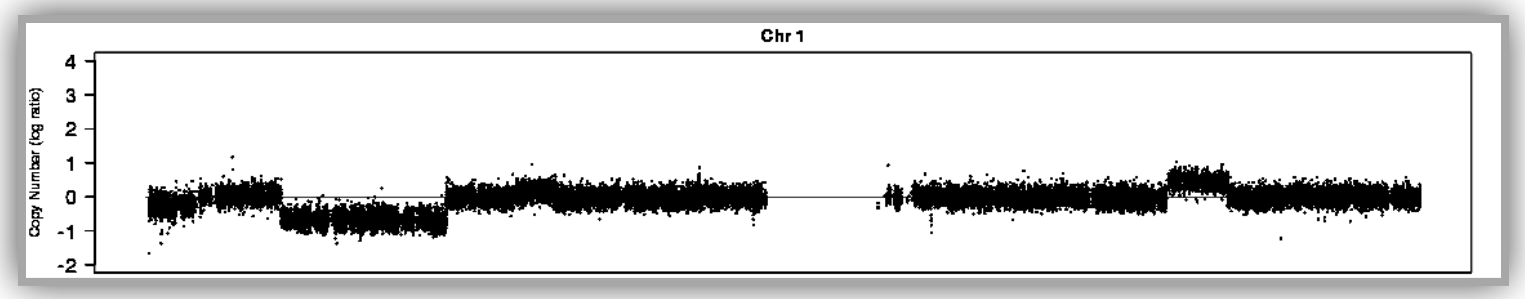
Input Data After Normalization

- GC-content bias correction applied to separately for
  - tumor sample reads $N_{1:T}^{Tumor}$
  - normal sample reads $N_{1:T}^{Normal}$



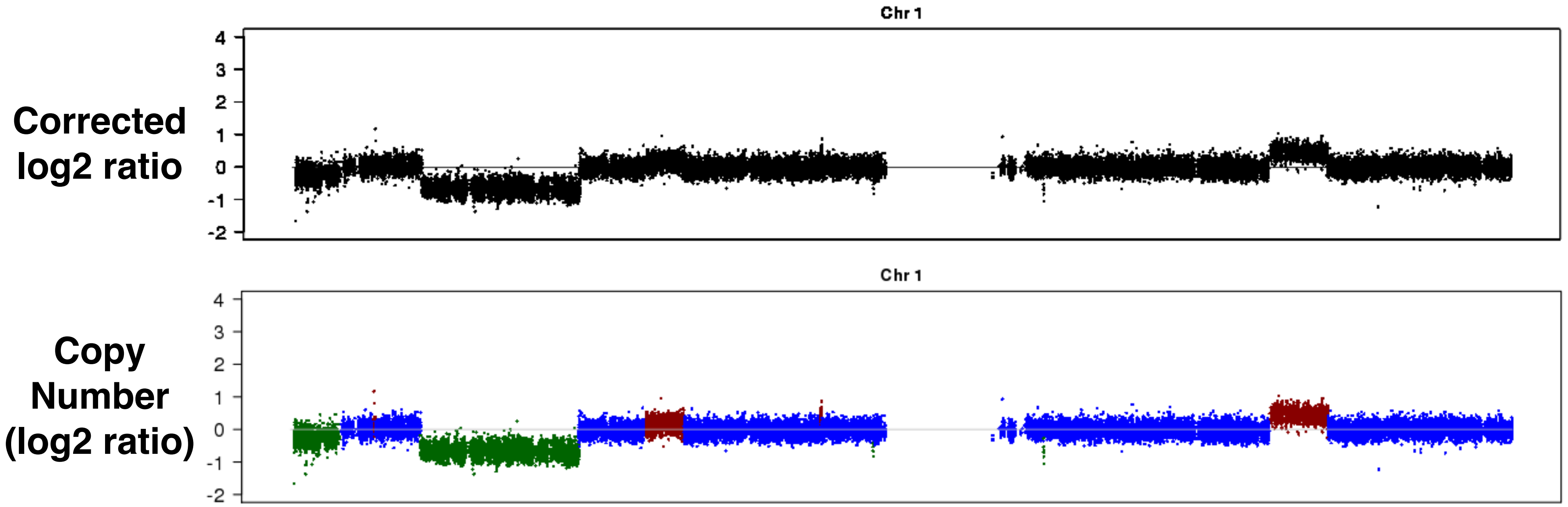
- Normalize tumor corrected read counts $\hat{N}_i^{Tumor}$ with normal corrected read counts $\hat{N}_i^{Normal}$ to obtain the log ratio for bin $t \in \{1, \ldots, T\}$

$$x_t = \log_2\left(\frac{\hat{N}_t^{Tumor}}{\hat{N}_t^{Normal}}\right)$$



Benjamini and Speed. *Nucleic Acids Research* **40**:e72-86 (2012)
Boeva et al. *Bioinformatics* **29**(3):423-5 (2012)
Ha et al. *Genome Research* **22**:1995-2007 (2012).
Adalsteinsson*, Ha* Freeman* et al. *Nature Communications* **8**:1324 (2017)

Copy Number Segmentation and Prediction



Data normalization

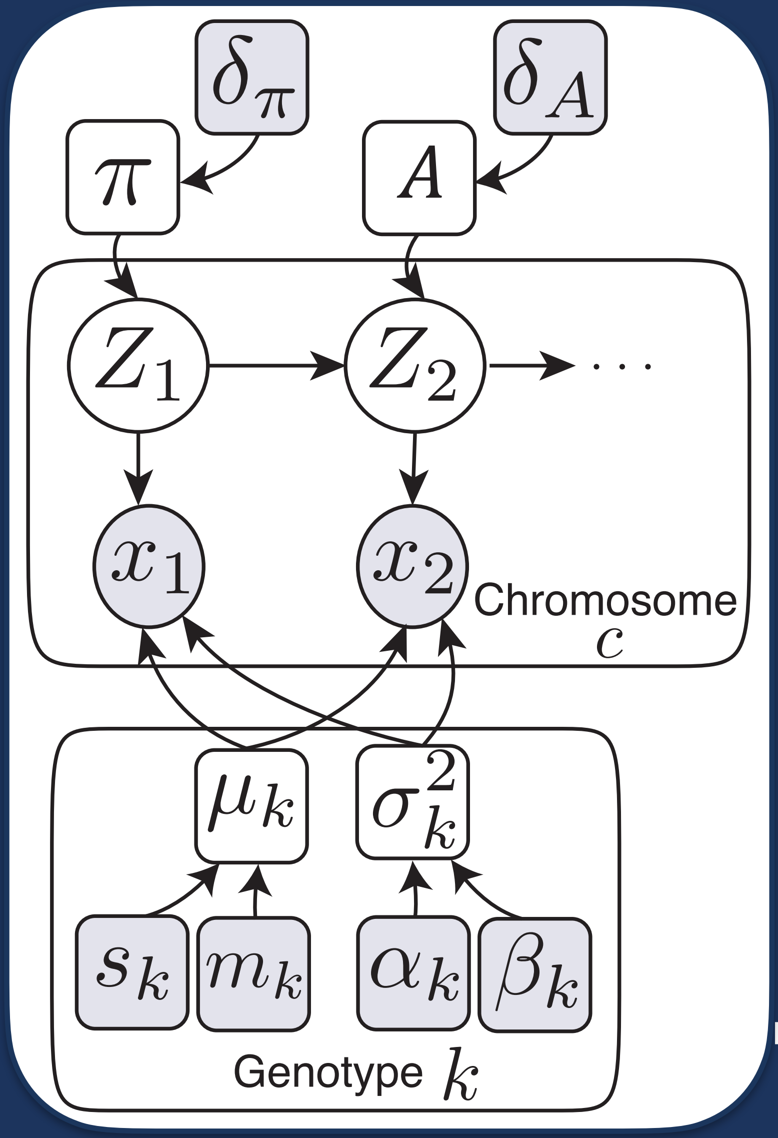
Copy Number Segmentation

- What are the genomic segments of copy number alterations?
- What is the copy number value for each segment?
- How do we account for variability/noise in the data?

Continuous hidden Markov model (HMM)

2. Continuous hidden Markov model

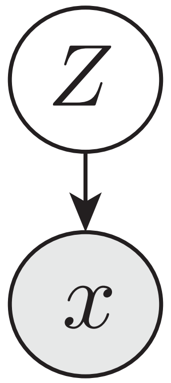
- Hidden Markov Models vs Mixture Models
- Components of a Continuous HMM
- Inference and Parameter Learning using EM
- References:
  - **HMMcopy** - Ha et al. *Genome Research* **22**:1995-2007 (2012).
  - **ichorCNA** - Adalsteinsson*, Ha* Freeman* et al. *Nature Communications* **8**:1324 (2017).
  - **TitanCNA** - Ha et al. TITAN: inference of copy number architectures in clonal cell populations from tumor whole-genome sequencing data. *Genome Research* **24**:1881-1893 (2014).
  - Murphy, K. (2012). Machine Learning: A Probabilistic Perspective. MIT Press. ISBN: 9780262018029
  - Bishop, C. M. (2006). Pattern Recognition and Machine Learning (Information Science and Statistics). Springer. ISBN: 0387310738



Probabilistic Graphical Model for HMMs

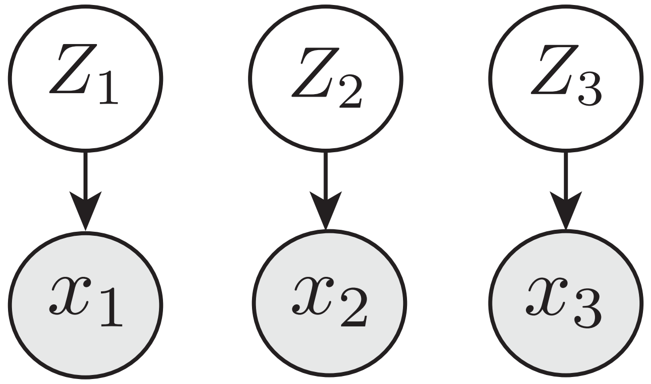
$x_{1:T}$ observed data

$Z_{1:T}$ latent variables

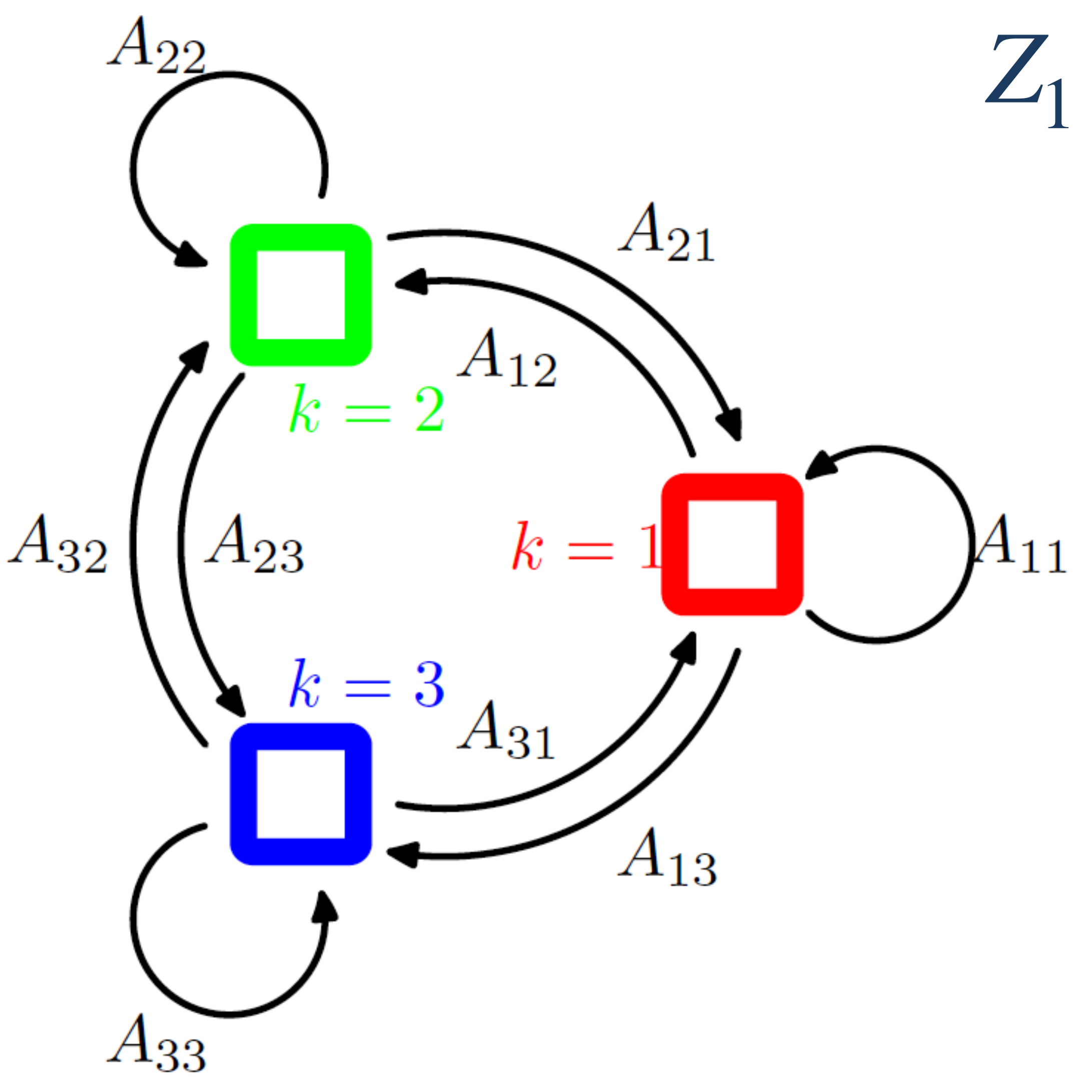


$$p(x, Z) = p(Z)p(x|Z)$$

Mixture Model

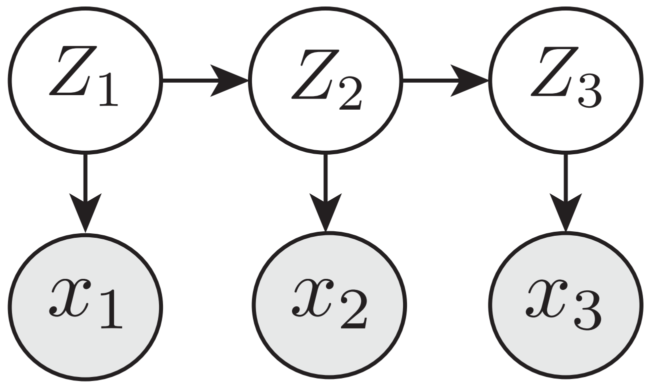


$$p(x_{1:3}, Z_{1:3}) = p(Z_{1:3})p(x_{1:3}|Z_{1:3})$$
$$= \left[\prod_{t=1}^{3} p(Z_t)\right]\left[\prod_{t=1}^{3} p(x_t|Z_t)\right]$$



Transition Diagram

Hidden Markov Model



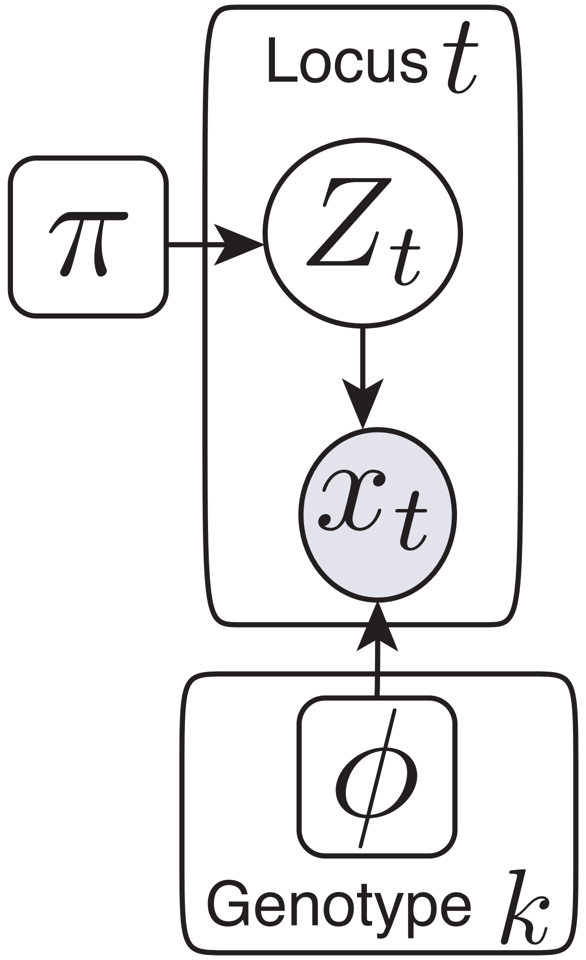
1. Markov Property $Z_3 \perp\!\!\!\perp Z_1 | Z_2$
2. Conditional independence of observations $x_3 \perp\!\!\!\perp x_{1:2} | Z_3$

Chapter 17 in Murphy (2012). Machine Learning: A Probabilistic Perspective. MIT Press
Chapter 13 in Bishop (2006). Pattern Recognition and Machine Learning. Springer

From Mixture Models to Hidden Markov Models

- Mixture model for iid data is a special case of the HMM

Mixture Model



$$p(x_{1:T}, Z_{1:T}) = p(Z_{1:T})p(x_{1:T} | Z_{1:T})$$

Joint Probability Distribution (Data likelihood)

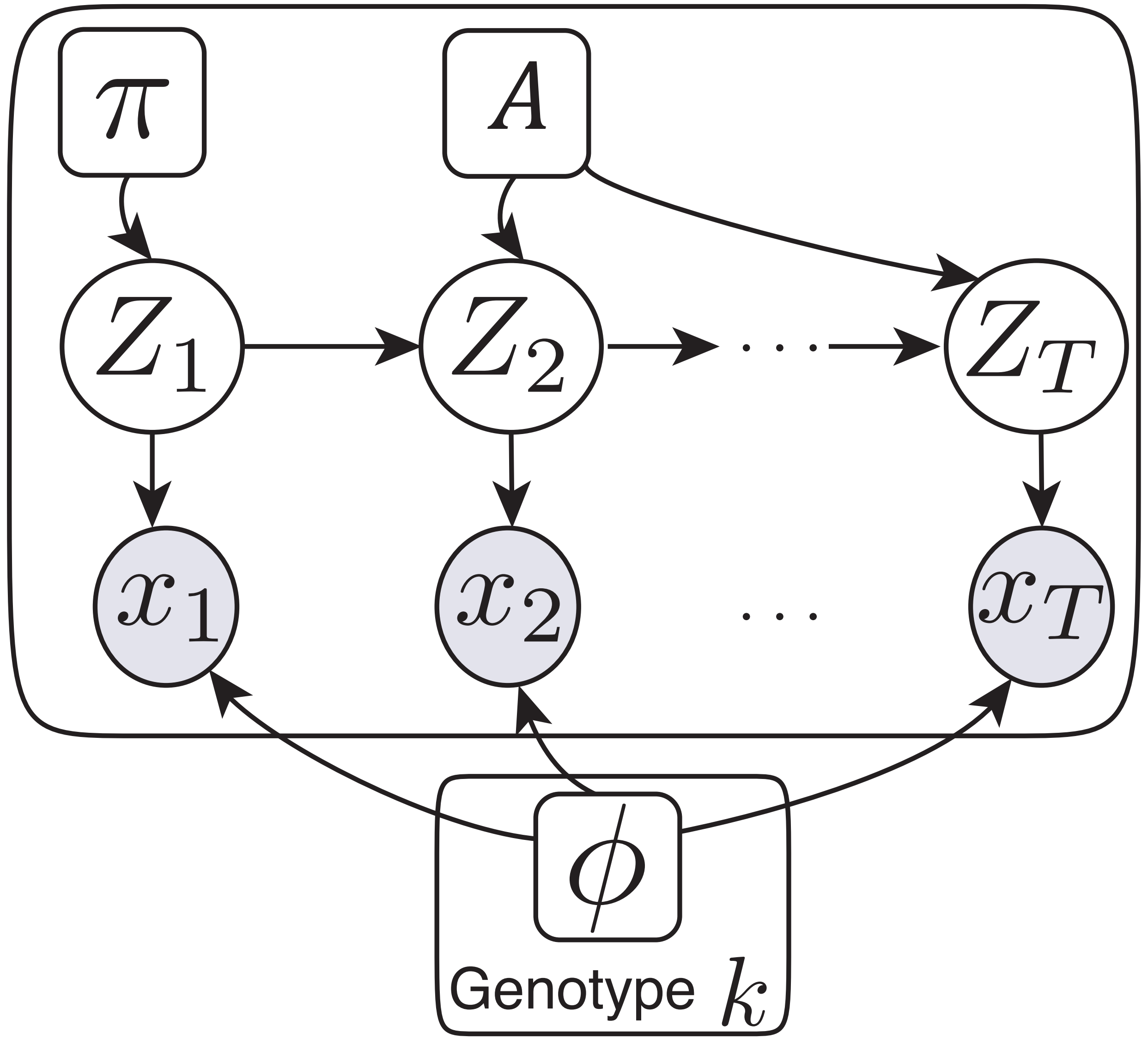
$x_{1:T}$ observed data

$Z_{1:T}$ latent variables

$\boldsymbol{\pi}$ mixture weights

$\boldsymbol{\phi}$ observation parameters

Hidden Markov Model

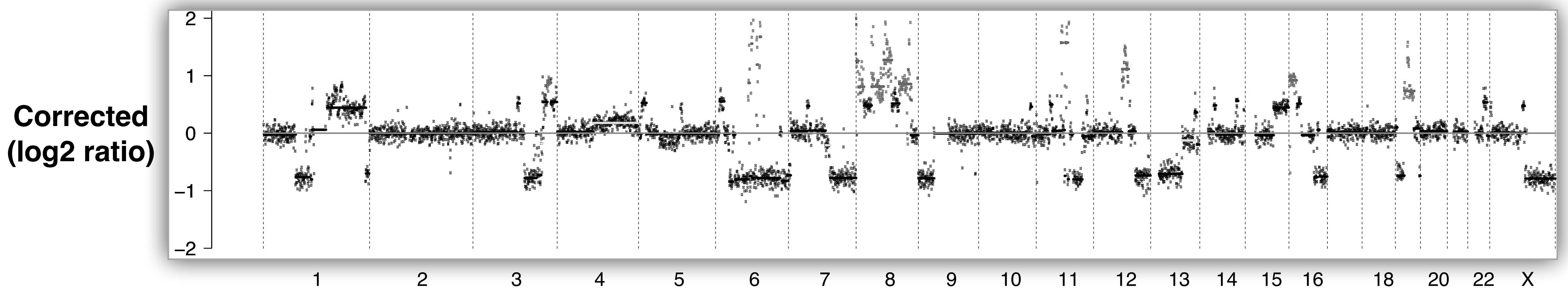


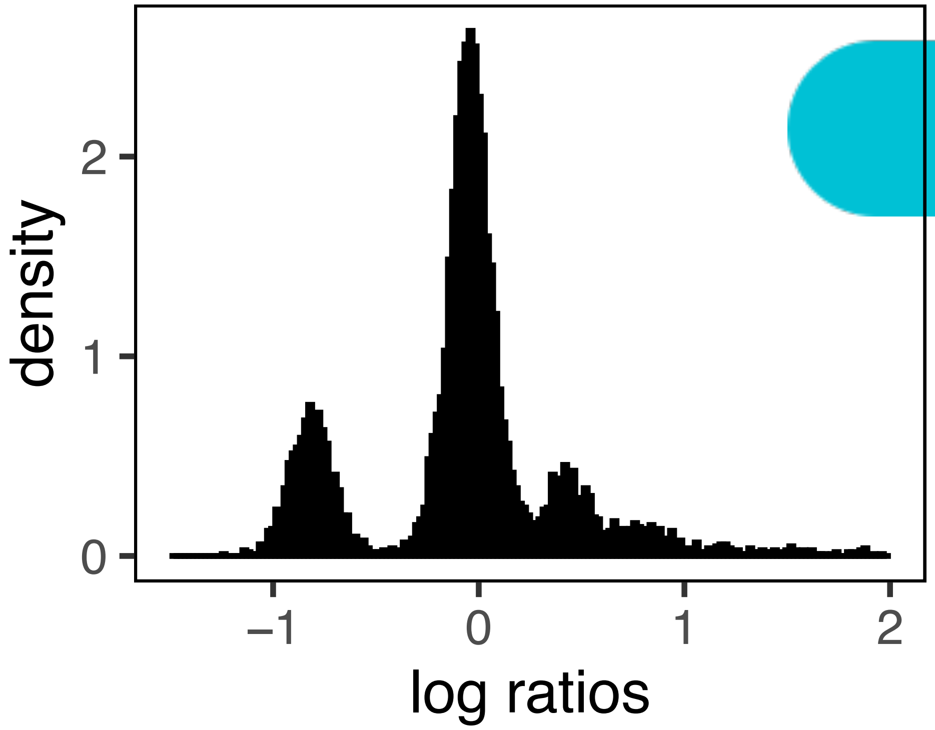
$$p(x_{1:T}, Z_{1:T} | \boldsymbol{\theta}) = \left[\prod_{t=1}^{T} p(Z_t | \boldsymbol{\pi})\right] \prod_{t=1}^{T} p(x_t | Z_t, \boldsymbol{\phi})$$

$$\boldsymbol{\theta} = \{\boldsymbol{\pi}, \boldsymbol{\phi}\}$$

$$\boldsymbol{\theta} = \{\boldsymbol{\pi}, \boldsymbol{\phi}, \boldsymbol{A}\}$$

Gaussian Mixture Model for Log Ratio Data





The ratios $\frac{\hat{r}_t^{Tumor}}{\hat{r}_t^{Normal}}$, for all t loci are log-normal distributed, so the log ratios $x_{1:T}$ follow a normal distribution.

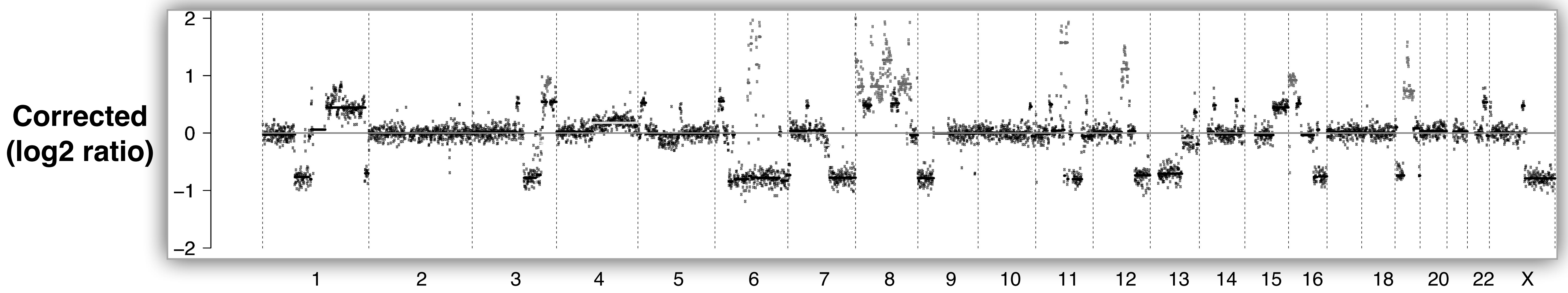
The Gaussian Distribution

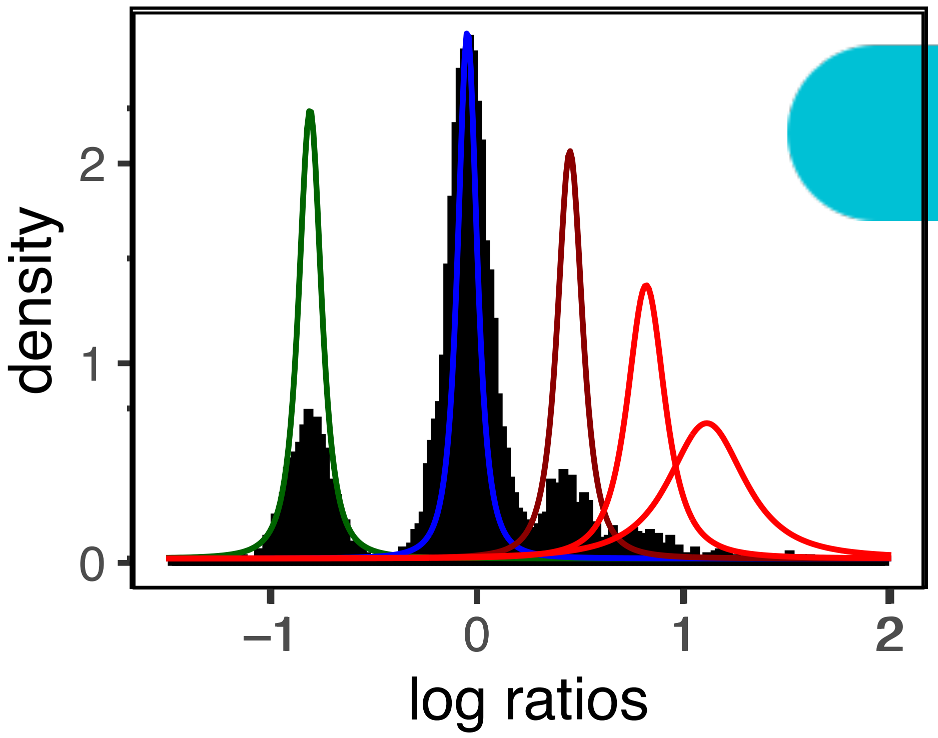
Let X be a continuous measurement with mean μ and variance σ^2, then X has a Gaussian distribution,

$X \sim \mathcal{N}(\mu, \sigma^2)$ or $p(X = x) = \mathcal{N}(x \,|\, \mu, \sigma^2)$ where

$$\mathcal{N}(x \,|\, \mu, \sigma^2) = \frac{1}{\sqrt{2\pi\sigma^2}} e^{-\frac{1}{2\sigma^2}(x-\mu)^2}$$

Gaussian Mixture Model for Log Ratio Data





The ratios $\frac{\hat{r}_t^{Tumor}}{\hat{r}_t^{Normal}}$, for all t loci are log-normal distributed, so the log ratios $x_{1:T}$ follow a normal distribution.

The Gaussian Distribution

Define a likelihood for a ***K-component mixture of Gaussians*** with means $\boldsymbol{\mu} = \{\mu_1, \ldots, \mu_K\}$ and variance $\boldsymbol{\sigma}^2 = \{\sigma_1^2, \ldots, \sigma_K^2\}$, where the observation model is a conditional Gaussian

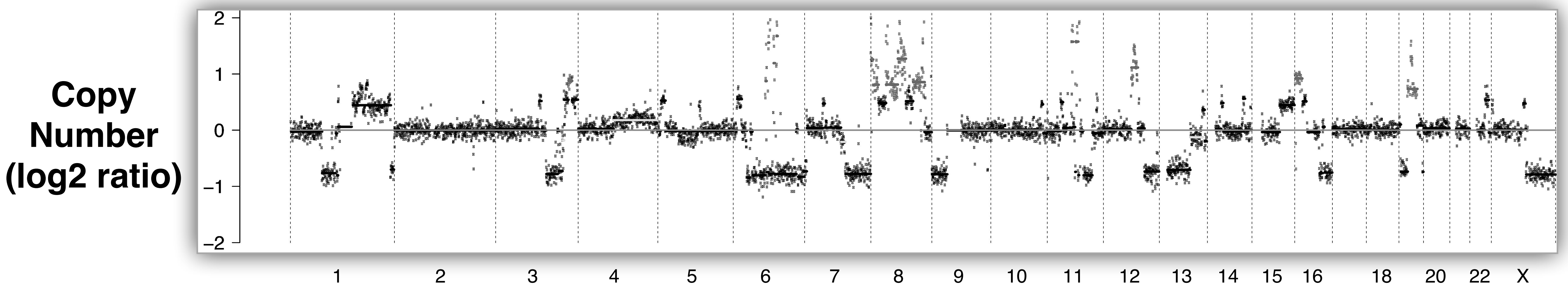
$$p(x_t \,|\, Z_t = k, \boldsymbol{\mu}, \boldsymbol{\sigma}^2) = \mathcal{N}(x_t \,|\, \mu_k, \sigma_k^2)$$

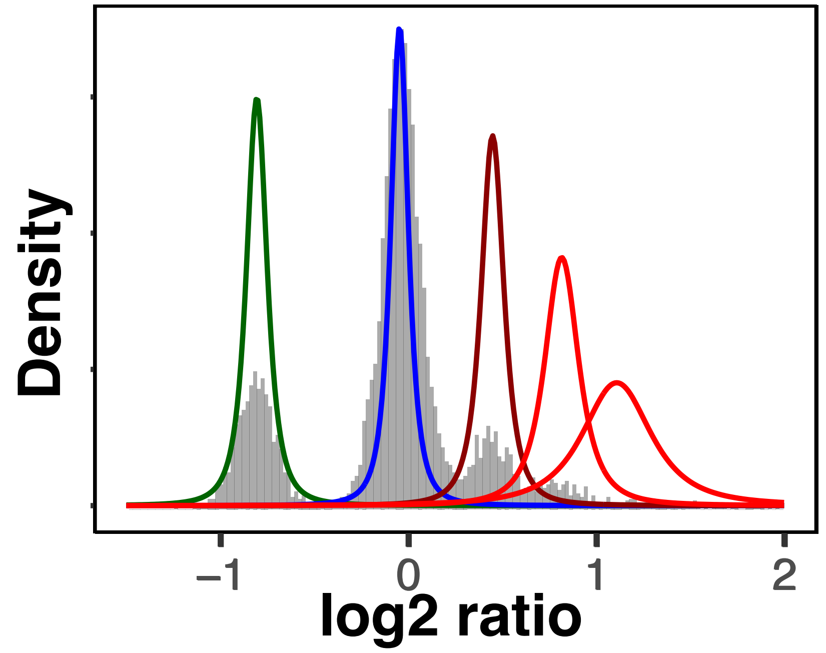
Rationale for Estimating Likelihood Parameters

Why are the data multi-modal?

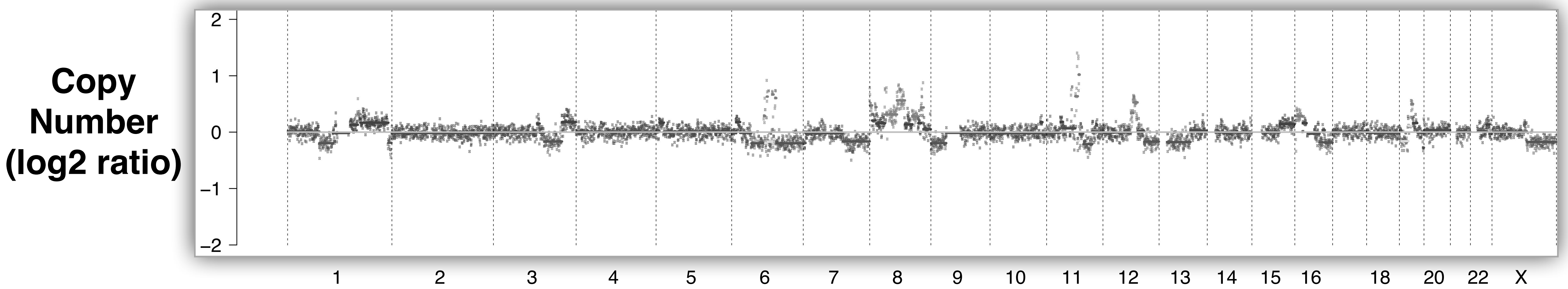
Why should we estimate the mixture distribution parameters?

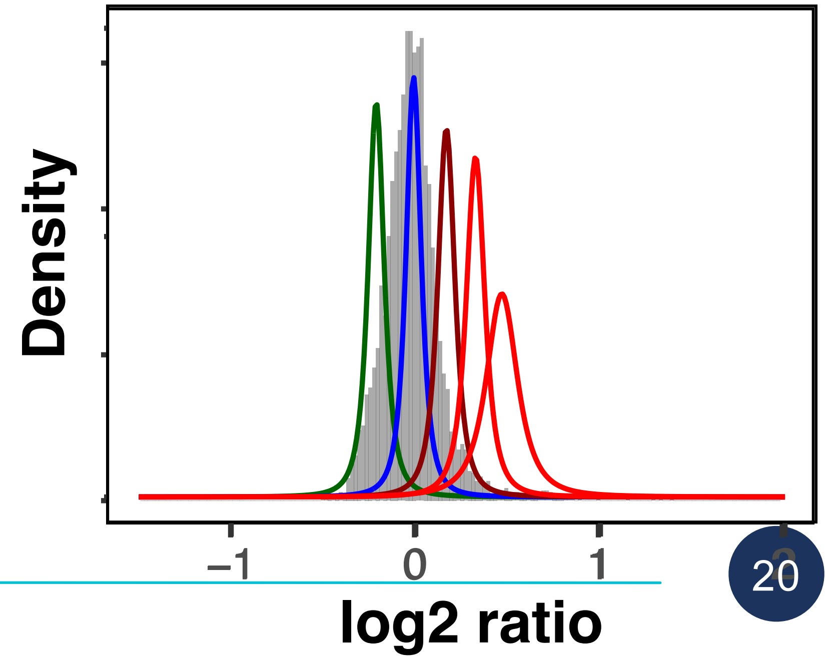
Patient 288 - Time 1





Patient 288 - Time 2





Components of a continuous HMM

Input Data: log ratios

There are T different data points with continuous values $\boldsymbol{x} = \{x_1, \ldots, x_T\}$.

Latent State Model

- The latent variables $\boldsymbol{Z} = \{Z_1, \ldots, Z_T\}$ can be assigned values from a set of K discrete states with probability

Initial state distribution

- The probabilities of the states for the first latent variable Z_1 is the parameter $\boldsymbol{\pi} = \{\pi_1, \ldots, \pi_K\}$
- $\boldsymbol{\pi}$ follows a prior distribution $p(\pi_k \mid \delta_k) = Dir(\pi_k \mid \delta_k)$

Transition Model (homogenous HMM)

- The conditional distribution between adjacent data i and j corresponds to a table A of transition probabilities

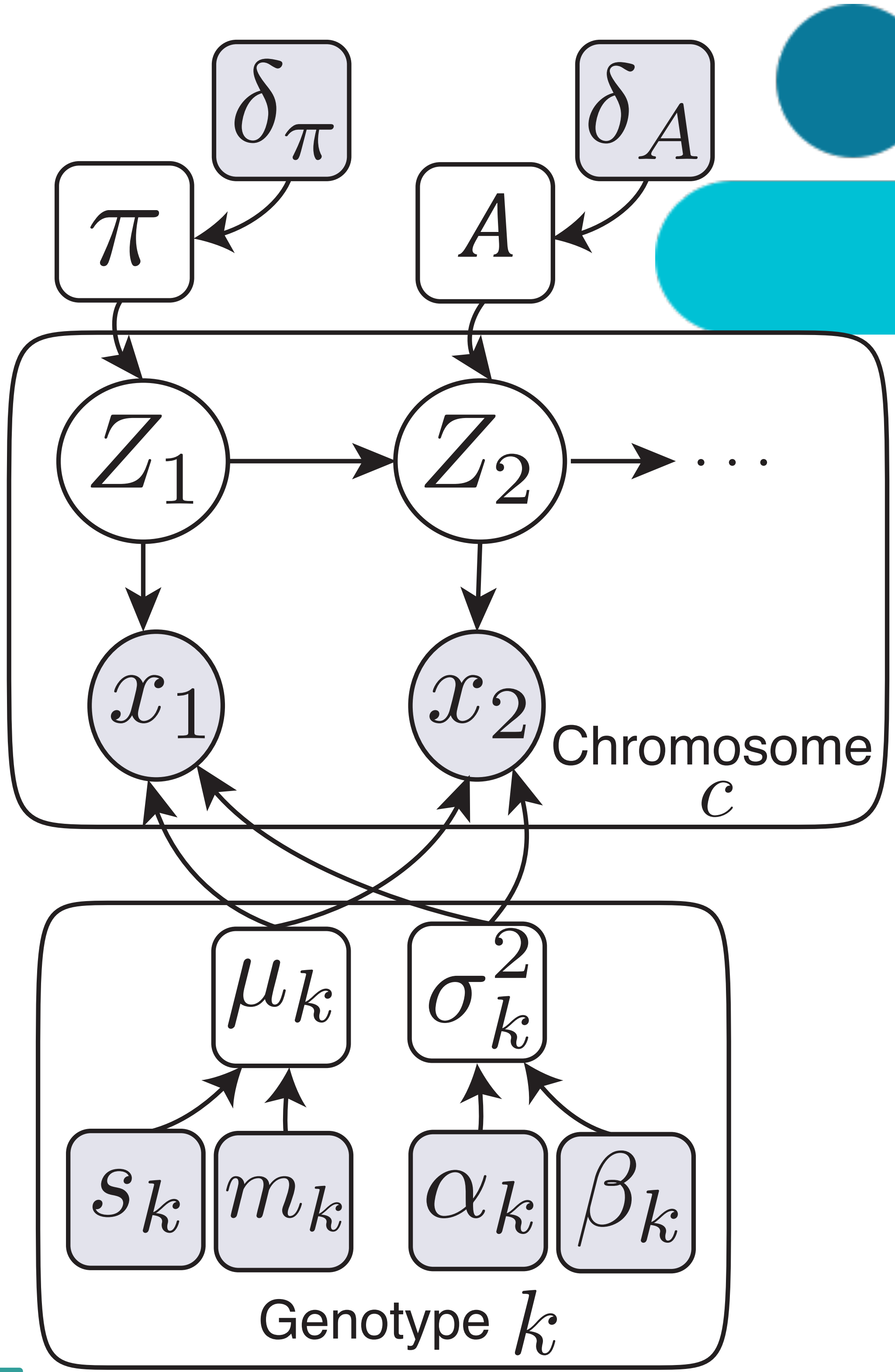
$$p(Z_t = j \mid Z_{t-1} = i) = A_{ij}$$

Emission Model (Continuous HMM)

- The emission is modeled using a mixture of Gaussians with the likelihood model

$$p(x_t \mid Z_t = k, \boldsymbol{\mu}, \boldsymbol{\sigma^2}) = \mathcal{N}(x_t \mid \mu_k, \sigma_k^2)$$

- $\boldsymbol{\mu}$ is modeled with a prior $p(\mu_k \mid m_k, s_k) = \mathcal{N}(\mu_k \mid m_k, s_k)$
- $\boldsymbol{\sigma^2}$ is modeled with prior $p(\sigma_k^2 \mid \alpha_k, \beta_k) = InvGamma(\sigma_k^2 \mid \alpha_k, \beta_k)$



Chapter 17 in Murphy (2012). Machine Learning: A Probabilistic Perspective. MIT Press
Chapter 13 in Bishop (2006). Pattern Recognition and Machine Learning. Springer

Inference & parameter estimation using EM

Expectation-Maximization: Inference and parameter training

Initialize parameters: □

E-Step: Inference using Forwards-Backwards Algorithm (Baum-Welch)

1. Compute “responsibilities” (Posterior of the latent states $\gamma(Z_{1:T})$)
   - State $Z_t = k$ is “responsible for generating observation x_t”
2. Compute “2-slice marginals” (Posterior of state transitions $\xi(Z_{t-1}, Z_t)$)
   - Expected number of transitions from state k to j

M-Step: Update parameters (learning)

1. Initial state distribution, $\boldsymbol{\pi}$
2. Transition probabilities, $\boldsymbol{A}$
3. Emission likelihood parameters, $\boldsymbol{\mu}$

Iterate between E-Step and M-Step, check when log posterior likelihood, $\log \mathbb{P}$, stops increasing.

Inference & parameter estimation using EM (E-Step)

E-Step: Forwards-backwards Algorithm (Baum-Welch; Sum-Product)

- Forward, $\alpha(\boldsymbol{Z}_t)$: joint prob. of observing all *past* data up to time t when given Z_t
- Backward, $\beta(\boldsymbol{Z}_t)$: conditional prob. of all *future* data from time $t+1$ to T when given Z_t

Forward Probabilities ($T \times K$) - Past

$$\alpha(Z_t = k) = \mathcal{N}(x_t \mid \mu_k, \sigma_k^2) \sum_{j=1}^{K} \left\{ A_{jk}\alpha(Z_{t-1} = j) \right\}$$

Backward Probabilities ($T \times K$) - Future

$$\beta(Z_t = k) = \sum_{j=1}^{K} \left\{ \mathcal{N}(x_{t+1} \mid \mu_j, \sigma_j^2) A_{kj}\beta(Z_{t+1} = j) \right\}$$

See extra slides for more details

Chapter 13 in Bishop (2006). Pattern Recognition and Machine Learning. Springer

Chapter 17 in Murphy (2012). Machine Learning: A Probabilistic Perspective. MIT Press

Inference & parameter estimation using EM (E-Step)

E-Step: Compute Responsibilities & 2-Slice Marginals

- Responsibilities, $\gamma(Z_t = k)$: is the posterior on the latent states

$$\gamma(Z_t = k) = \frac{\alpha(Z_t = k)\beta(Z_t = k)}{p(\boldsymbol{x})}$$

Responsibilities

Matrix $K \times T$

- 2-Slice Marginals, $\xi(Z_{t-1} = k, Z_t = j)$: is the expected number of transitions between k to j

$$\xi(Z_{t-1} = k, Z_t = j) = \frac{\alpha(Z_{t-1} = k)A_{kj}\mathcal{N}(x_t \,|\, \mu_j, \sigma_j^2)\beta(Z_t = j)}{p(\boldsymbol{x})}$$

2 Slice Marginals

Matrix $K \times K \times (T-1)$

- The likelihood $p(\boldsymbol{x}) = p(\boldsymbol{x} \,|\, \boldsymbol{\mu}, \boldsymbol{\sigma}^2, \boldsymbol{\pi})$ is computed in the forwards recursion

$$\ell = \log p(\boldsymbol{x}) = \sum_{t=1}^{T} \log \left(\sum_{k=1}^{K} \alpha(Z_t = k) \right)$$

Log likelihood

See extra slides for more details

Chapter 13 in Bishop (2006). Pattern Recognition and Machine Learning. Springer

Chapter 17 in Murphy (2012). Machine Learning: A Probabilistic Perspective. MIT Press

Inference & parameter estimation using EM (M-Step)

Expected complete data log likelihood

$$Q = \overset{\text{Initial State Dist}}{\boxed{\sum_{k=1}^{K} \gamma(Z_1 = k) \log \pi_k}} + \overset{\text{Transition}}{\boxed{\sum_{t=2}^{T} \sum_{j=1}^{K} \sum_{k=1}^{K} \xi(Z_{t-1} = k, Z_t = j) \log A_{kj}}} + \overset{\text{Emission}}{\boxed{\sum_{t=1}^{T} \sum_{k=1}^{K} \gamma(Z_t = k) \log \mathcal{N}(x_t \,|\, \mu_k, \sigma_k^2)}} + \overset{\text{Priors}}{priors}$$

M-Step: update parameters, $\boldsymbol{\pi}$, $\boldsymbol{\mu}, \boldsymbol{\sigma}^2$

$$\hat{\pi}_k = \frac{\gamma(Z_1 = k) + \delta^{\pi}(k) - 1}{\sum_{j=1}^{K} \left\{ \gamma(Z_1 = j) + \delta^{\pi}(j) - 1 \right\}}$$

MAP for initial state distribution

$$\hat{\mu}_k = \frac{s_k \sum_{t=1}^{T} \gamma(Z_t = k) x_t + m\sigma_k^2}{s_k \sum_{t=1}^{T} \gamma(Z_t = k) + \sigma_k^2}$$

MAP for Gaussian means

$$\hat{\sigma}_k^2 = \frac{\sum_{t=1}^{T} \gamma(Z_t = k) \left(x_t - \bar{x}_k \right)^2 + 2\beta_k}{\sum_{t=1}^{T} \gamma(Z_t = k) + 2(\alpha_k + 1)}$$

MAP for Gaussian variance terms

Where $\bar{x} = \dfrac{\sum_{t=1}^{T} \gamma(Z_t = k) x_t}{\sum_{t=1}^{T} \gamma(Z_t = k)}$

See extra slides for more details

https://www.cs.ubc.ca/~murphyk/Papers/bayesGauss.pdf

Inference & parameter estimation using EM (M-Step)

M-Step: Update transition matrix, A

Expected number of transitions from k to j

Prior counts

$$\hat{A}_{kj} = \frac{\sum_{t=2}^{T} \xi(Z_{t-1} = k, Z_t = j) + \delta_j^A(k)}{\sum_{l=1}^{K} \left\{ \sum_{t=2}^{T} \xi(Z_{t-1} = k, Z_t = l) + \delta_j^A(l) \right\}}$$

“Pseudo-counts”

Expected number of transitions from k to any other state

Evaluate the log posterior

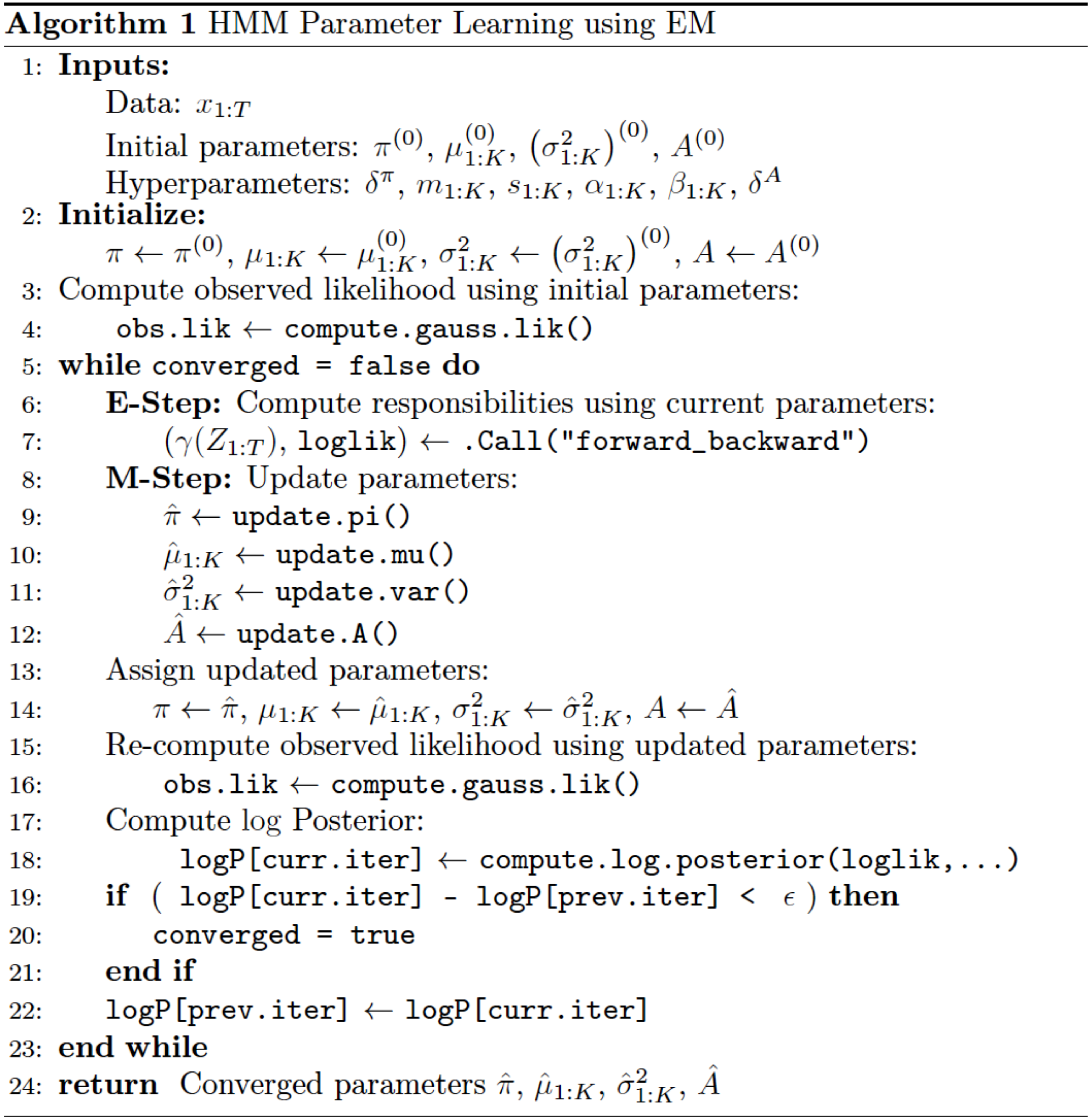
$$\log \mathbb{P} = \ell + \boxed{\log Dir(\hat{\boldsymbol{\pi}} \,|\, \boldsymbol{\delta}) + \sum_{k=1}^{K} \left\{ \log \mathcal{N}(\hat{\mu}_k \,|\, m_k, s_k) + \log InvGamma(\hat{\sigma}_k^2 \,|\, \alpha_k, \beta_k) + \log Dir(A_{k,1:K}^{(0)} \,|\, \hat{A}_{k,1:K}) \right\}}$$

Log likelihood

Log priors

Iterate between E-Step and M-Step: stop when $\log \mathbb{P}$ changes less than ϵ compared to previous EM iteration.

See extra slides for more details

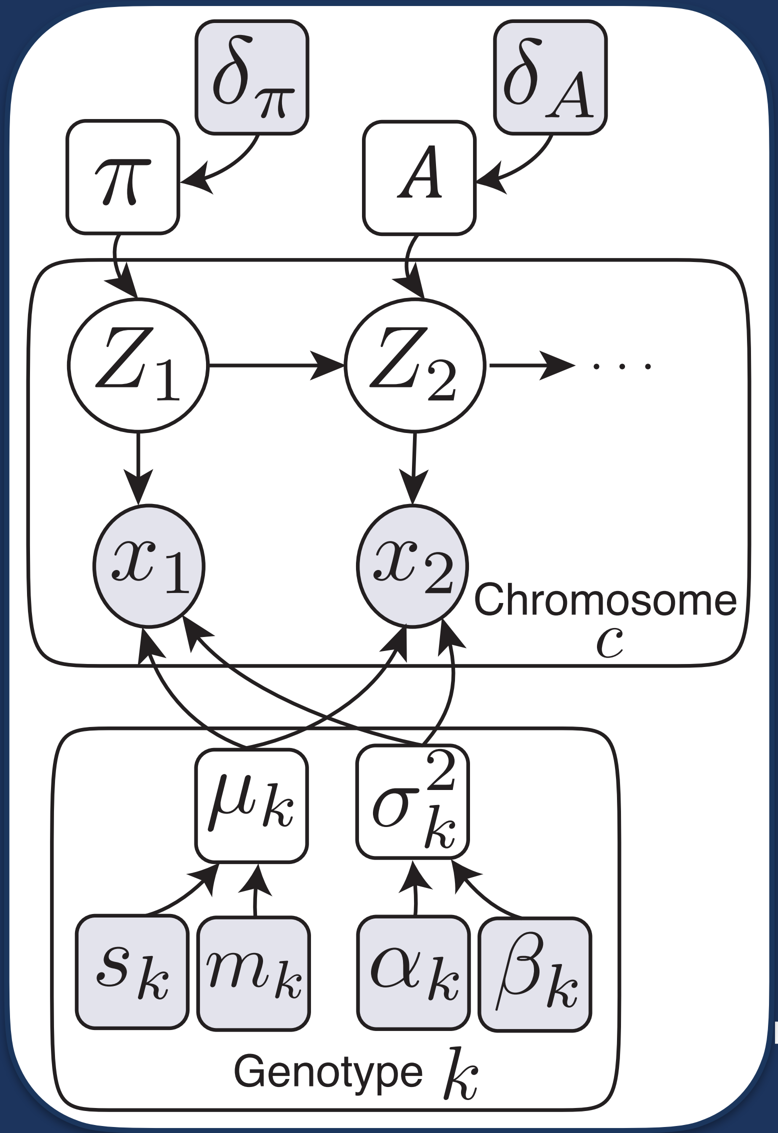


Algorithm 1 HMM Parameter Learning using EM

```
Inputs:
    Data: $x_{1:T}$
    Initial parameters: $\pi^{(0)}$, $\mu_{1:K}^{(0)}$, $(\sigma^2_{1:K})^{(0)}$, $A^{(0)}$
    Hyperparameters: $\delta^{\pi}$, $m_{1:K}$, $s_{1:K}$, $\alpha_{1:K}$, $\beta_{1:K}$, $\delta^{A}$
Initialize:
    $\pi \leftarrow \pi^{(0)}$, $\mu_{1:K} \leftarrow \mu_{1:K}^{(0)}$, $\sigma^2_{1:K} \leftarrow (\sigma^2_{1:K})^{(0)}$, $A \leftarrow A^{(0)}$
Compute observed likelihood using initial parameters:
     obs.lik ← compute.gauss.lik()
while converged = false do
    E-Step: Compute responsibilities using current parameters:
        $(\gamma(Z_{1:T})$, loglik$)$ ← .Call("forward_backward")
    M-Step: Update parameters:
        $\hat{\pi}$ ← update.pi()
        $\hat{\mu}_{1:K}$ ← update.mu()
        $\hat{\sigma}^2_{1:K}$ ← update.var()
        $\hat{A}$ ← update.A()
    Assign updated parameters:
        $\pi \leftarrow \hat{\pi}$, $\mu_{1:K} \leftarrow \hat{\mu}_{1:K}$, $\sigma^2_{1:K} \leftarrow \hat{\sigma}^2_{1:K}$, $A \leftarrow \hat{A}$
    Re-compute observed likelihood using updated parameters:
        obs.lik ← compute.gauss.lik()
    Compute log Posterior:
         logP[curr.iter] ← compute.log.posterior(loglik,...)
    if ( logP[curr.iter] - logP[prev.iter] < $\epsilon$ ) then
        converged = true
    end if
    logP[prev.iter] ← logP[curr.iter]
end while
return  Converged parameters $\hat{\pi}$, $\hat{\mu}_{1:K}$, $\hat{\sigma}^2_{1:K}$, $\hat{A}$
```

3. Copy Number Profiling using a HMM

- Defining the HMM for copy number analysis
- Copy number segmentation using Viterbi
- References:
  - **HMMcopy** - Ha et al. *Genome Research* **22**:1995-2007 (2012).
  - **ichorCNA** - Adalsteinsson*, Ha* Freeman* et al. *Nature Communications* **8**:1324 (2017).
  - **TitanCNA** - Ha et al. TITAN: inference of copy number architectures in clonal cell populations from tumor whole-genome sequencing data. *Genome Research* **24**:1881-1893 (2014).
  - Murphy, K. (2012). Machine Learning: A Probabilistic Perspective. MIT Press. ISBN: 9780262018029
  - Bishop, C. M. (2006). Pattern Recognition and Machine Learning (Information Science and Statistics). Springer. ISBN: 0387310738



Probabilistic Model for Copy Number Analysis

Input Data: log ratios

There are T different genomic bins with log ratio data $\boldsymbol{x} = \{x_1, \dots, x_T\}$.

Latent State Model: copy number states

There are 5 different possible copy number states (genotypes), $K = \{1, 2, 3, 4, 5\}$

1. A specific genotype $k \in K$ can be assigned to the each of the **latent states** $\boldsymbol{Z} = \left\{Z_1, \dots, Z_T\right\}$
2. The **initial state distribution** $\boldsymbol{\pi} = \{\pi_1, \dots, \pi_5\}$ is used for the first latent state Z_1

Transition Model

3. The probabilities for transitioning to copy number state j in bin t from state i in bin $t-1$ are contained in matrix $\boldsymbol{A} \in \mathbb{R}^{K \times K}$

$$p(Z_t = j \,|\, Z_{t-1} = i) = A_{ij}$$

Emission Model: likelihood for log ratio data

For each copy number state, the log ratio means are $\boldsymbol{\mu} = \{\mu_1, \dots, \mu_5\}$ and variance $\boldsymbol{\sigma^2} = \{\sigma_1^2, \dots, \sigma_5^2\}$

4. The **emission model** is a mixture of Gaussians with *unknown* parameters, $\boldsymbol{\mu}$ and $\boldsymbol{\sigma^2}$,

$$p(x_t \,|\, Z_t = k, \boldsymbol{\mu}, \boldsymbol{\sigma^2}) = \mathcal{N}(x_t \,|\, \mu_k, \sigma_k^2)$$

Prior Model

5. The **priors** in the model have hyper-parameters $\boldsymbol{\delta^\pi}$, $m_{1:K}$, $s_{1:K}$, $\alpha_{1:K}$, $\beta_{1:K}$, $\boldsymbol{\delta^A_{1:K}}$

$$p(\boldsymbol{\pi} \,|\, \boldsymbol{\delta^\pi}) = Dirichlet(\boldsymbol{\pi} \,|\, \boldsymbol{\delta^\pi})$$

$$p(\mu_k \,|\, m_k, s_k) = \mathcal{N}(\mu_k \,|\, m_k, s_k)$$

$$p(\sigma_k^2 \,|\, \alpha_k, \beta_k) = InvGamma(\sigma_k^2 \,|\, \alpha_k, \beta_k)$$

$$p(\boldsymbol{A_{k,1:K}} \,|\, \boldsymbol{\delta^A}) = Dirichlet(\boldsymbol{A_{k,1:K}} \,|\, \boldsymbol{\delta^A_k})$$

$$\begin{array}{cc} & j \\ i & \begin{array}{|c|ccc|} \hline \mathbf{A} & \mathbf{0} & \mathbf{\dots} & \mathbf{5} \\ \hline \mathbf{0} & & & \\ \mathbf{\dots} & & & \\ \mathbf{5} & & & \\ \hline \end{array} \end{array} \quad \sum_{j=1}^{K} A_{ij} = 1$$

Ha et al. *Genome Research* **22**:1995-2007 (2012).
Adalsteinsson*, Ha* Freeman* et al. *Nat Commun* **8**:1324 (2017)

Probabilistic Model for Copy Number Analysis

Input Data: log ratios

There are T different genomic bins with log ratio data $\boldsymbol{x} = \{x_1, \ldots, x_T\}$.

Latent State Model: copy number states

There are 5 different possible copy number states (genotypes), $K = \{1, 2, 3, 4, 5\}$

1. A specific genotype $k \in K$ can be assigned to the each of the $\boxed{\textbf{latent states } \boldsymbol{Z} = \{Z_1, \ldots, Z_T\}}$
2. The **initial state distribution** $\boxed{\boldsymbol{\pi} = \{\pi_1, \ldots, \pi_5\}}$ is used for the first latent state Z_1

Transition Model

3. The probabilities for transitioning to copy number state j in bin t from state i in bin $t-1$ are contained in $\boxed{\text{matrix } \boldsymbol{A} \in \mathbb{R}^{K \times K}}$

$$p(Z_t = j \mid Z_{t-1} = i) = A_{ij}$$

Emission Model: likelihood for log ratio data

For each copy number state, the log ratio means are $\boxed{\boldsymbol{\mu} = \{\mu_1, \ldots, \mu_5\} \text{ and variance } \boldsymbol{\sigma^2} = \{\sigma_1^2, \ldots, \sigma_5^2\}}$

4. The **emission model** is a mixture of Gaussians with *unknown* parameters, $\boldsymbol{\mu}$ and $\boldsymbol{\sigma^2}$,

$$p(x_t \mid Z_t = k, \boldsymbol{\mu}, \boldsymbol{\sigma^2}) = \mathcal{N}(x_t \mid \mu_k, \sigma_k^2)$$

Prior Model

5. The **priors** in the model have hyper-parameters $\boldsymbol{\delta^\pi}$, $m_{1:K}$, $s_{1:K}$, $\alpha_{1:K}$, $\beta_{1:K}$, $\boldsymbol{\delta^A_{1:K}}$

$$p(\boldsymbol{\pi} \mid \boldsymbol{\delta^\pi}) = Dirichlet(\boldsymbol{\pi} \mid \boldsymbol{\delta^\pi})$$

$$p(\mu_k \mid m_k, s_k) = \mathcal{N}(\mu_k \mid m_k, s_k)$$

$$p(\sigma_k^2 \mid \alpha_k, \beta_k) = InvGamma(\sigma_k^2 \mid \alpha_k, \beta_k)$$

$$p(\boldsymbol{A_{k,1:K}} \mid \boldsymbol{\delta^A}) = Dirichlet(\boldsymbol{A_{k,1:K}} \mid \boldsymbol{\delta^A_k})$$

E-Step:
Compute
Responsibilities

M-Step:
Update parameters

j

i

A	0	…	5
0			
…			
5			

$$\sum_{j=1}^{K} A_{ij} = 1$$

Ha et al. *Genome Research* **22**:1995-2007 (2012).
Adalsteinsson*, Ha* Freeman* et al. *Nat Commun* **8**:1324 (2017)

Copy number segmentation using Viterbi

Viterbi algorithm (Max-Sum)

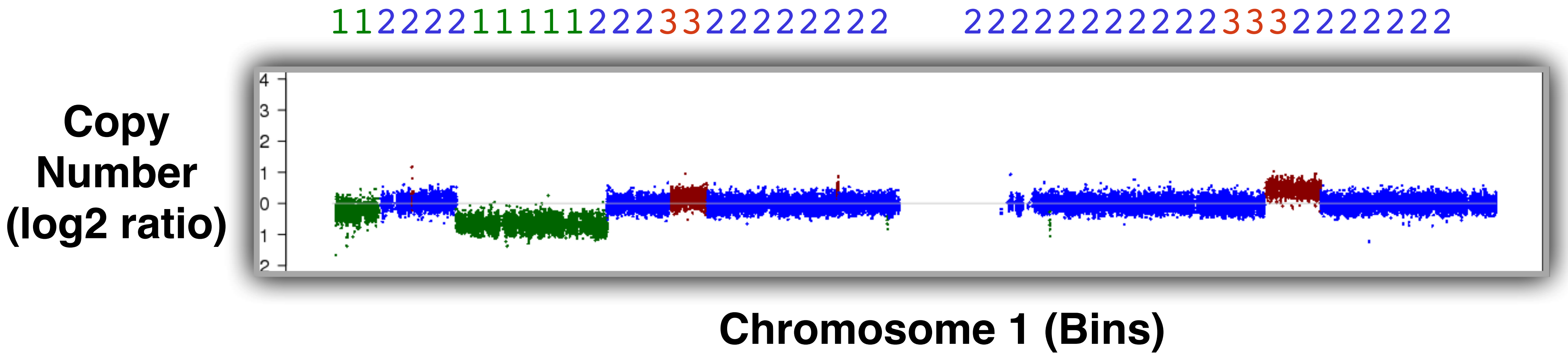
- Find the most probable sequence of copy number states

$$\hat{Z}_{1:T} = \max_{Z_{1:T}} \log p(Z_{1:T} \mid x_{1:T})$$

- Perform max-sum of probabilities in trellis

$$\omega(Z_{t+1} = k) = \log \mathcal{N}(x_{t+1} \mid \mu_k, \sigma_k^2) + \max_{Z_t} \left\{ \log A_{Z_t,k} + \log \omega(Z_t) \right\}$$

- Back trace from $\omega(Z_T)$ to find overall most probable path

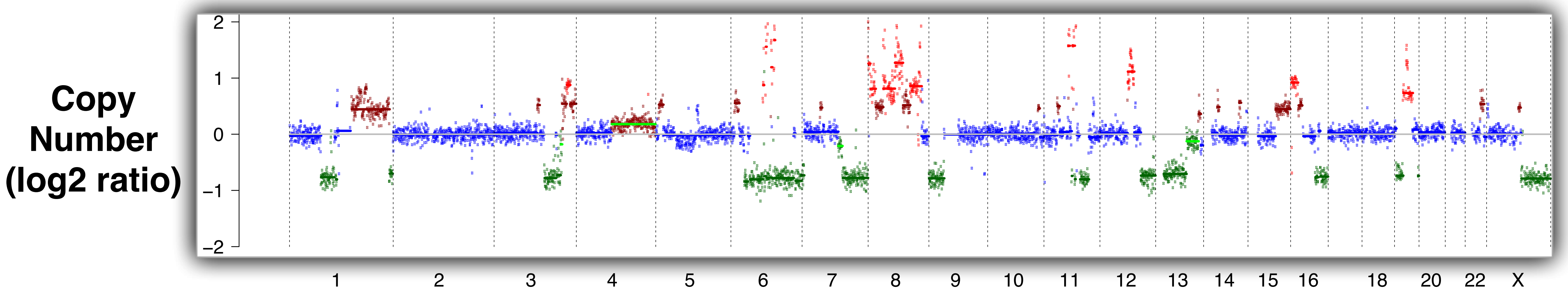


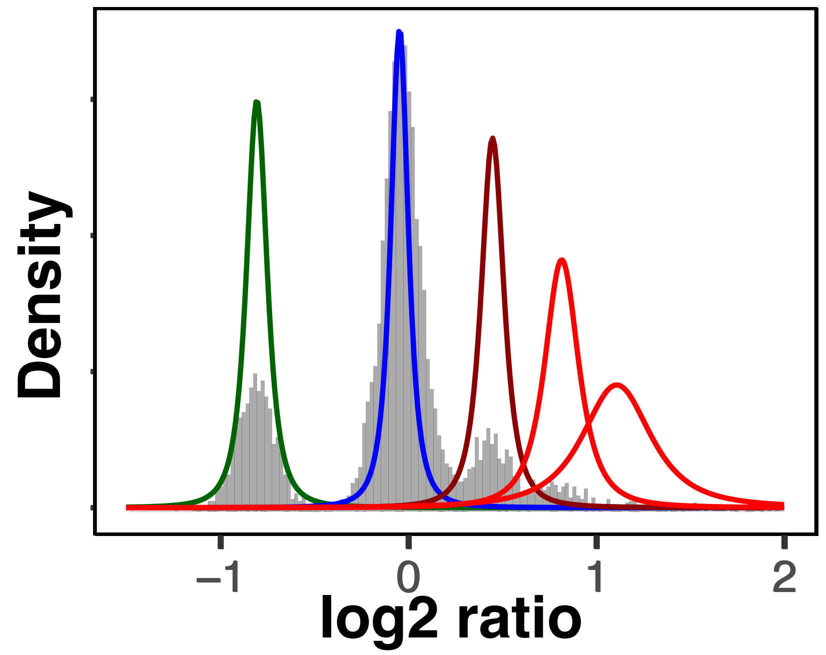
Rationale for Estimating Likelihood Parameters

Why should we estimate the mixture distribution parameters?

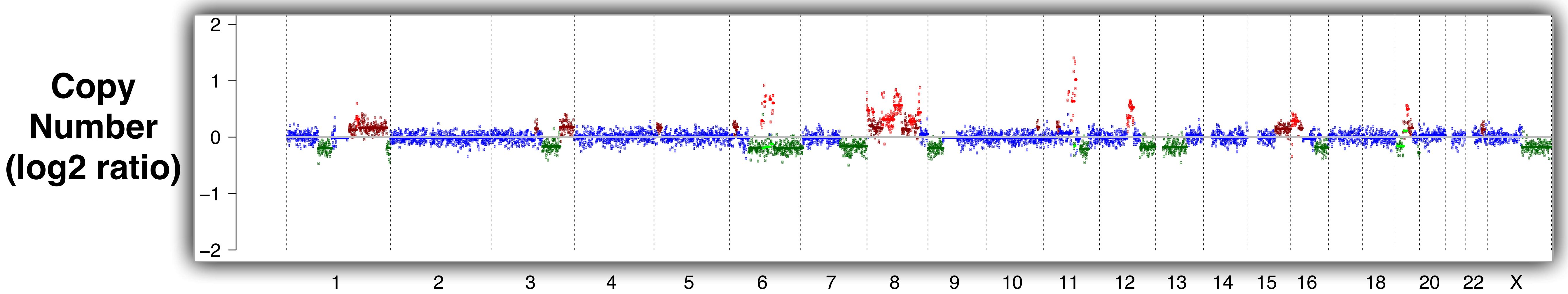
- Can account for technical and biological “noise” by estimating model parameters $\boldsymbol{\mu} = \{\mu_0, \ldots, \mu_5\}$ and $\boldsymbol{\sigma}^2 = \{\sigma_0^2, \ldots, \sigma_5^2\}$?

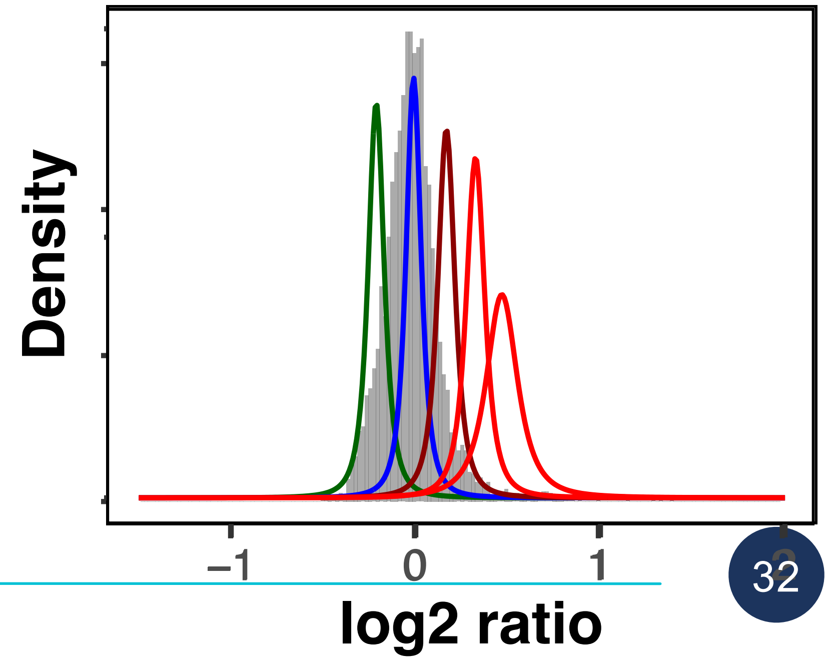
Patient 288 - Time 1





Patient 288 - Time 2





🟦 Copy Neutral 🟩 Deletion 🟥 Gain 🟥 Amplification

Homework #8: Profiling copy number alterations

A. Implement a copy number alteration (CNA) caller described in Lecture 3

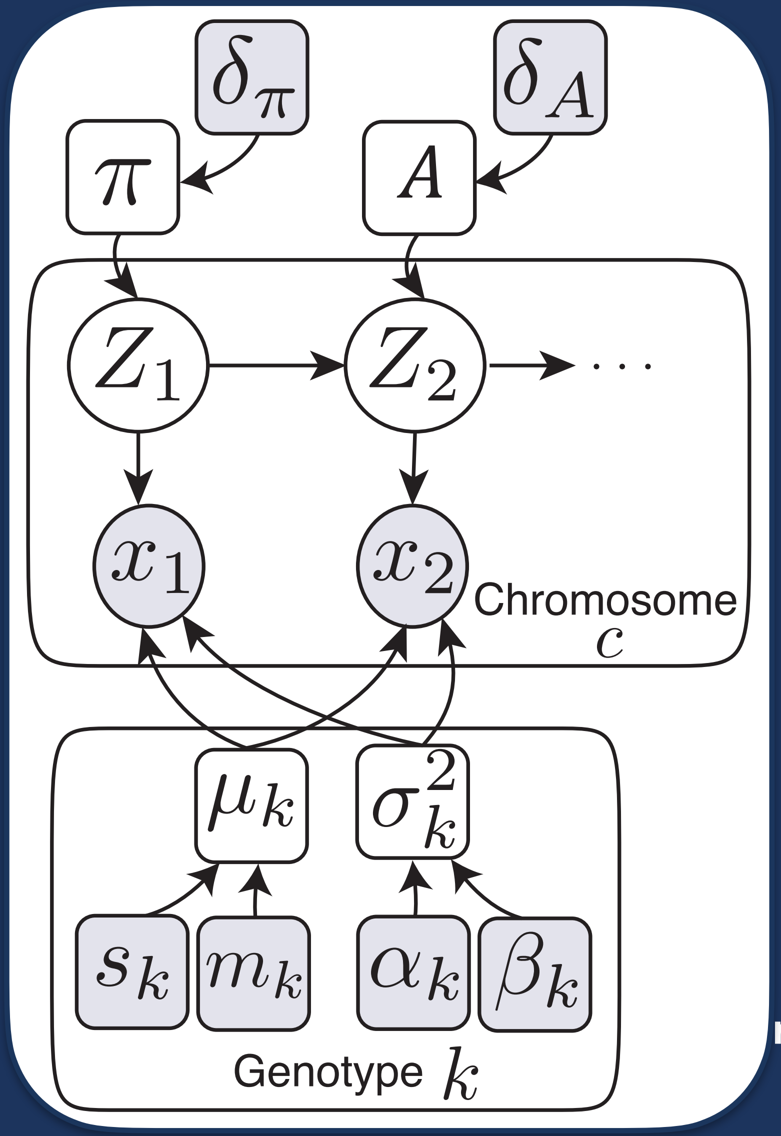
- Implement components of a continuous HMM in a Bayesian framework
- Learn the parameters and infer the genotypes using EM
- Predict the copy number alteration segments for a chromosome.
- Expected outputs for each question will be provided so that you can check your code.

B. Power calculations for mutation detection described in Lecture 4

Due: May 26th, 2023

Extra Slides

- Continuous hidden Markov models (HMMs)
- Parameter inference using EM and copy number segmentation
- References:
  - **ichorCNA** - Adalsteinsson*, Ha* Freeman* et al. *Nature Communications* **8**:1324 (2017).
  - **HMMcopy** - Ha et al. *Genome Research* **22**:1995-2007 (2012).
  - **TitanCNA** - Ha et al. TITAN: inference of copy number architectures in clonal cell populations from tumor whole-genome sequencing data. *Genome Research* **24**:1881-1893 (2014).
  - Murphy, K. (2012). Machine Learning: A Probabilistic Perspective. MIT Press. ISBN: 9780262018029
  - Bishop, C. M. (2006). Pattern Recognition and Machine Learning (Information Science and Statistics). Springer. ISBN: 0387310738



ichorCNA: Model inference using EM (extra slide 1)

Complete data likelihood: joint distribution of latent and observed variables

$$p(x_{1:T}, Z_{1:T} \,|\, \boldsymbol{\theta}) = p(Z_1 \,|\, \pi_{1:K}) \left[\prod_{t=2}^{T} p(Z_t \,|\, Z_{t-1}, \boldsymbol{A}) \right] \prod_{t=1}^{T} p(x_t \,|\, Z_t, \mu, \sigma^2)$$

$$= \prod_{k=1}^{K} \pi_k^{\mathbb{I}(Z_i=k)} \left[\prod_{t=2}^{T} \prod_{k=1}^{K} \prod_{j=1}^{K} A_{jk}^{\mathbb{I}(Z_{t-1}=j)\mathbb{I}(Z_t=k)} \right] \prod_{t=1}^{T} \prod_{k=1}^{K} \mathcal{N}(x_t \,|\, \mu_k, \sigma_k^2)^{\mathbb{I}(Z_t=k)}$$

where $\boldsymbol{\theta} = \left\{ \pi_{1:K},\ \mu_{1:K},\ \sigma^2_{1:K},\ \boldsymbol{A} \right\}$

Complete data log likelihood

$$\log p(x_{1:T}, Z_{1:T} \,|\, \boldsymbol{\theta}) = \sum_{k=1}^{K} \mathbb{I}(Z_i = k) \log \pi_k + \sum_{t=2}^{T} \sum_{j=1}^{K} \sum_{k=1}^{K} \mathbb{I}(Z_{t-1} = j, Z_t = k) \log A_{jk} + \sum_{t=1}^{T} \sum_{k=1}^{K} \mathbb{I}(Z_i = k) \log \mathcal{N}(x_t \,|\, \mu_k, \sigma_k^2)$$

Expected complete data log likelihood

$$Q = \sum_{k=1}^{K} \gamma(Z_1 = k) \log \pi_k + \sum_{t=2}^{T} \sum_{j=1}^{K} \sum_{k=1}^{K} \xi(Z_{t-1} = j, Z_t = k) \log A_{jk} + \sum_{t=1}^{T} \sum_{k=1}^{K} \gamma(Z_t = k) \log \mathcal{N}(x_t \,|\, \mu_k, \sigma_k^2)$$

Chapter 13 in Bishop (2006). Pattern Recognition and Machine Learning. Springer

Additional definitions for your reference

ichorCNA: Model inference using EM (extra slide 2)

E-Step: compute responsibilities using the forwards-backwards algorithm (Baum-Welch)

$$\gamma(\mathbf{Z}_t) = p(\mathbf{Z}_t | \boldsymbol{x}, \theta^{old}) = \frac{p(\boldsymbol{x} | \mathbf{Z}_t | \boldsymbol{\theta}^{\boldsymbol{old}}) p(\mathbf{Z}_t | \theta^{old})}{p(\boldsymbol{x} | \theta^{old})}$$

$$\gamma(\mathbf{Z}_t) = \frac{p(x_1, \ldots, x_t, \mathbf{Z}_t) p(x_{t+1}, \ldots, x_T | \mathbf{Z}_t)}{p(\boldsymbol{x})}$$

$$\gamma(\mathbf{Z}_t) = \frac{\alpha(\mathbf{Z}_t)\beta(\mathbf{Z}_t)}{p(\boldsymbol{x})}$$

Responsibilities

Matrix $K \times T$

Where $\alpha(Z_t = k) = \mathcal{N}(x_t | Z_t = k) \sum_{j=1}^{K} \left\{ A_{jk} \alpha(Z_t = j) \right\}$ is the forward recursion probability

Forward Probabilities

Matrix $K \times T$

Where $\beta(Z_t = k) = \sum_{j=1}^{K} \left\{ \mathcal{N}(x_{t+1} | Z_{t+1} = j) A_{kj} \alpha(Z_{t+1} = j) \right\}$ is the backward recursion probability

Backward Probabilities

Matrix $K \times T$

$$\xi(\mathbf{Z}_{t-1}, \mathbf{Z}_t) = p(\boldsymbol{x} | \mathbf{Z}_{t-1}, \mathbf{Z}_t) P(\mathbf{Z}_{t-1}, \mathbf{Z}_t)$$

$$\xi(\mathbf{Z}_{t-1}, \mathbf{Z}_t) = \frac{\alpha(\mathbf{Z}_{t-1}) p(x_t | \mathbf{Z}_t) p(\mathbf{Z}_t | \mathbf{Z}_{t-1}) \beta(\mathbf{Z}_t)}{p(\boldsymbol{x})}$$

2 Slice Marginals

Matrix $K \times K \times (T-1)$

Likelihood function

$$\ell = \log p(\boldsymbol{x}) = \sum_{t=1}^{T} \log \left(\sum_{k=1}^{K} \alpha(Z_t = k) \right)$$

Chapter 13 in Bishop (2006). Pattern Recognition and Machine Learning. Springer

Additional definitions for your reference

ichorCNA: Model inference using EM (extra slide 3)

M-Step: Update the parameters given the responsibilities

$$\mathbb{P}rior(\pi_{1:K}, \mu_{1:K}, \sigma^2_{1:K}, \boldsymbol{A}) = \prod_{k=1}^{K} Dir(\pi_k \,|\, \delta_k) Dir(A_k \,|\, \delta_A) \mathcal{N}(\mu_k \,|\, \alpha, \beta) InvGamma(\sigma_k^2 \,|\, \alpha_k, \beta_k)$$

Priors

$$\mathcal{O} = Q + \log \mathbb{P}(\pi_{1:K}, \mu_{1:K}, \sigma^2_{1:K}, \boldsymbol{A})$$

Complete data log likelihood + log priors

- The object function $\mathcal{O}$ is used to obtain the update equations for $\pi_{1:K}$ and $\mu_{1:K}$

$$\frac{\partial \mathcal{O}}{\partial \pi_k} = 0, \text{ find } \hat{\pi}_k$$

MAP for initial state distribution

$$\frac{\partial \mathcal{O}}{\partial \mu_k} = 0, \text{ find } \hat{\mu}_k$$

MAP for for Gaussian means

$$\frac{\partial \mathcal{O}}{\partial \sigma_k^2} = 0, \text{ find } \hat{\sigma}_k^2$$

MAP for for Gaussian variance

$$\frac{\partial \mathcal{O}}{\partial A_{jk}} = 0, \text{ find } \hat{A}_{jk}$$

MAP for transition probabilities

EM Convergence: after each iteration, monitor the log posterior

$$\ell = \log p(\boldsymbol{x}) = \sum_{t=1}^{T} \log \left(\sum_{k=1}^{K} \alpha(Z_t = k) \right)$$

Incomplete Data Log likelihood

$$\log \mathbb{P} = \ell + \log \mathbb{P}rior(\pi_{1:K}, \mu_{1:K}, \sigma^2_{1:K}, \boldsymbol{A})$$

Log posterior

Chapter 13 in Bishop (2006). Pattern Recognition and Machi
Learning. Springer

Additional definitions for your reference