Learning Sequence Motif Models Using Expectation Maximization (EM)

BMI/CS 776

www.biostat.wisc.edu/bmi776/

Spring 2016

Anthony Gitter

gitter@biostat.wisc.edu

Goals for Lecture

Key concepts

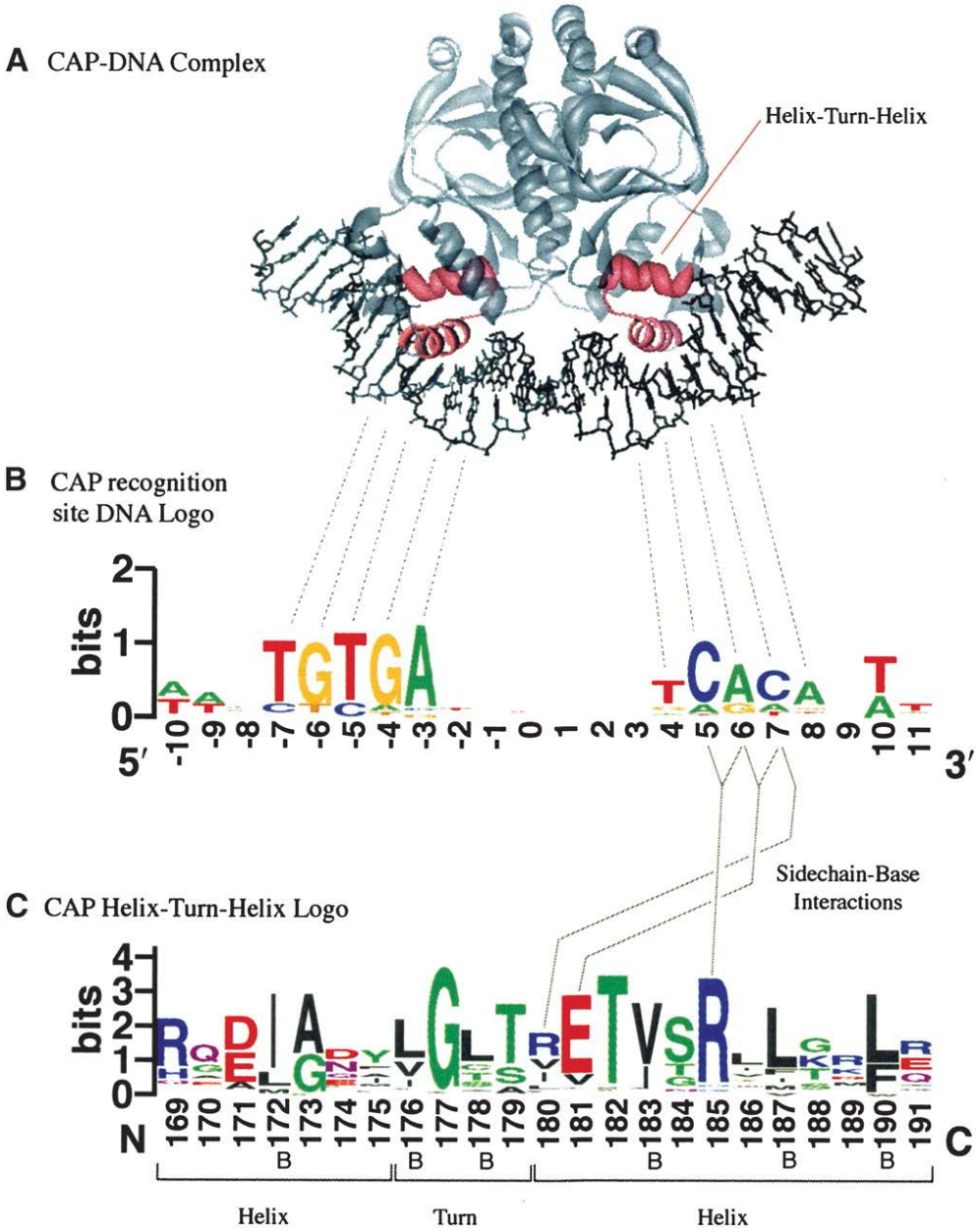
- the motif finding problem
- using EM to address the motif-finding problem
- the OOPS and ZOOPS models

Sequence Motifs

- What is a sequence *motif* ?
  - a sequence pattern of biological significance

- Examples
  - DNA sequences corresponding to protein binding sites
  - protein sequences corresponding to common functions or conserved pieces of structure

Sequence Motifs Example



CAP-binding motif model based on 59 binding sites in E.coli

helix-turn-helix motif model based on 100 aligned protein sequences

Figure from Crooks et al., *Genome Research* 14:1188-90, 2004.

The Motif Model Learning Task

given: a set of sequences that are thought to contain occurrences of an unknown motif of interest

do:

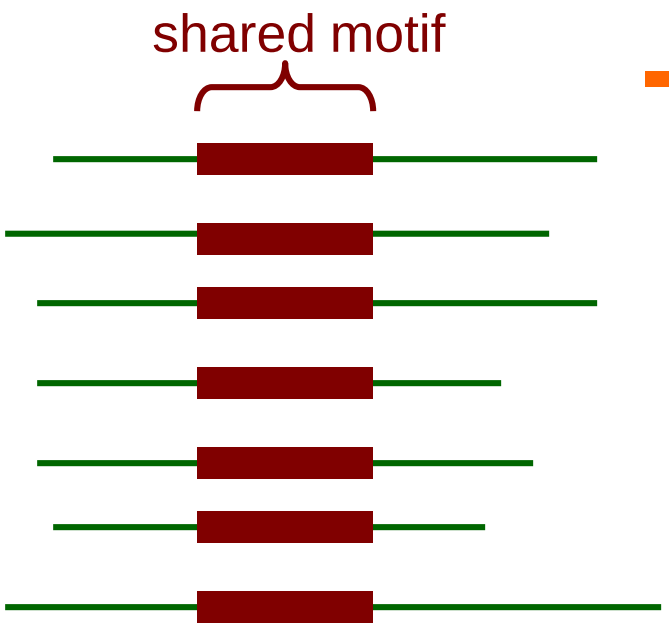
- infer a model of the motif
- predict the locations of the motif occurrences in the given sequences

Why is this important?

- To further our understanding of which regions of sequences are “functional”
- DNA: biochemical mechanisms by which the expression of genes are regulated
- Proteins: which regions of proteins interface with other molecules (e.g., DNA binding sites)
- Mutations in these regions may be significant

Motifs and *Profile Matrices* (a.k.a. *Position Weight Matrices*)

- Given a set of aligned sequences, it is straightforward to construct a profile matrix characterizing a motif of interest



sequence positions

	1	2	3	4	5	6	7	8
A	0.1	0.3	0.1	0.2	0.2	0.4	0.3	0.1
C	0.5	0.2	0.1	0.1	0.6	0.1	0.2	0.7
G	0.2	0.2	0.6	0.5	0.1	0.2	0.2	0.1
T	0.2	0.3	0.2	0.2	0.1	0.3	0.3	0.1

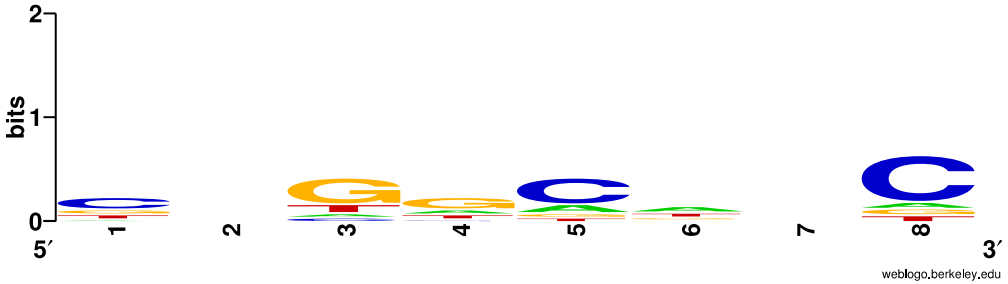
- Each element represents the probability of given character at a specified position

Sequence logos

	1	2	3	4	5	6	7	8
A	0.1	0.3	0.1	0.2	0.2	0.4	0.3	0.1
C	0.5	0.2	0.1	0.1	0.6	0.1	0.2	0.7
G	0.2	0.2	0.6	0.5	0.1	0.2	0.2	0.1
T	0.2	0.3	0.2	0.2	0.1	0.3	0.3	0.1

or

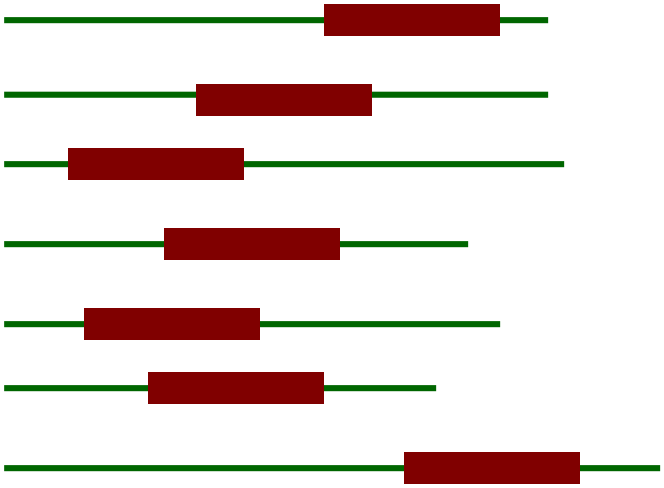
frequency logo



information content logo

Motifs and Profile Matrices

- How can we construct the profile if the sequences aren't aligned?
- In the typical case we don't know what the motif looks like.



Unaligned Sequence Example

- ChIP-chip experiment tells which probes are bound (though this protocol has been replaced by ChIP-seq)

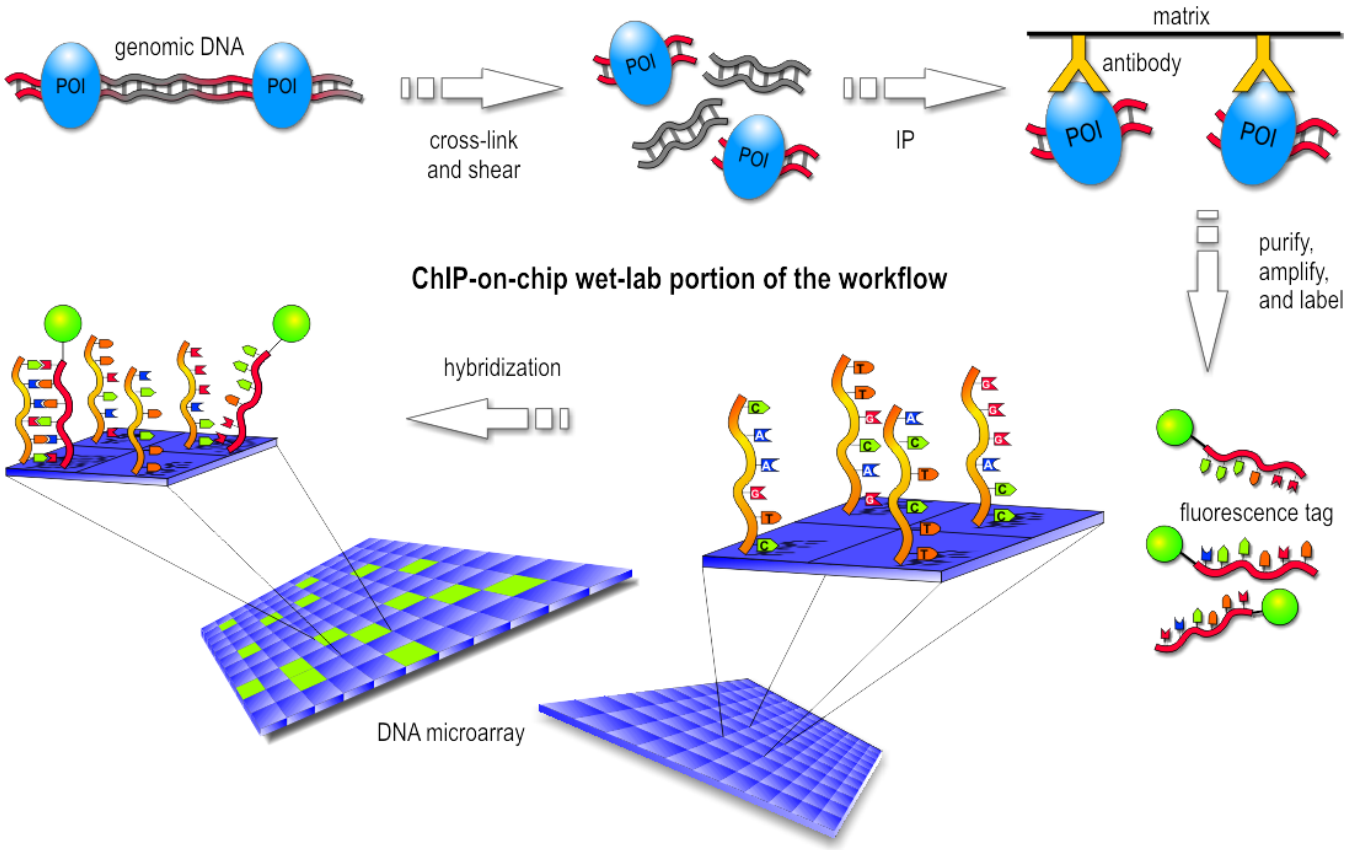
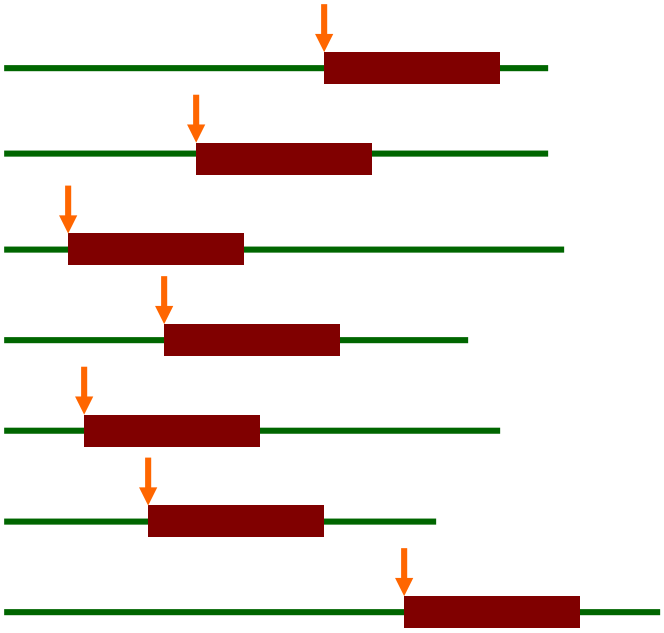


Figure from https://en.wikipedia.org/wiki/ChIP-on-chip

The Expectation-Maximization (EM) Approach

[Lawrence & Reilly, 1990; Bailey & Elkan, 1993, 1994, 1995]

- EM is a family of algorithms for learning probabilistic models in problems that involve *hidden state*
- In our problem, the hidden state is where the motif starts in each training sequence



Overview of EM

- Method for finding the maximum likelihood (ML) parameters (Θ) for a model (M) and data (D)

$$\theta_{ML} = \arg\max_{\theta} P(D \mid \theta, M)$$

- Useful when
  - it is difficult to optimize $P(D \mid \theta)$ directly
  - likelihood can be decomposed by the introduction of hidden information (Z)

$$P(D \mid \theta) = \sum_{Z} P(D, Z \mid \theta)$$

  - and it is easy to optimize the function (with respect to Θ):

$$Q(\theta \mid \theta^t) = \sum_{Z} P(Z \mid D, \theta^t) \log P(D, Z \mid \theta)$$

(see text section 11.6 for details)

Applying EM to the Motif Finding Problem

- First define the probabilistic model and likelihood function $P(D \mid \theta)$
- Identify the hidden variables (Z)
  - In this application, they are the locations of the motifs
- Write out the Expectation (E) step
  - Compute the expected values of the hidden variables given current parameter values
- Write out the Maximization (M) step
  - Determine the parameters that maximize the Q function, given the expected values of the hidden variables

Representing Motifs in MEME

- MEME: **M**ultiple **E**M for **M**otif **E**licitation
- A motif is
  - assumed to have a fixed width, *W*
  - represented by a matrix of probabilities: $p_{c,\,k}$ represents the probability of character c in column k

- Also represent the "background" (i.e. sequence outside the motif): $p_{c,0}$ represents the probability of character c in the background

Representing Motifs in MEME

- Example: a motif model of length 3

$p =$

	0	1	2	3
A	0.25	0.1	0.5	0.2
C	0.25	0.4	0.2	0.1
G	0.25	0.3	0.1	0.6
T	0.25	0.2	0.2	0.1

background motif positions

Representing Motif Starting Positions in MEME

- The element $Z_{i,j}$ of the matrix Z is an indicator random variable that takes value 1 if the motif starts in position j in sequence i (and takes value 0 otherwise)
- Example: given DNA sequences of length 6, where *W*=3

G T C A G G

G A G A G T

A C G G A G

C C A G T C

$Z =$

	1	2	3	4
seq1	0	0	1	0
seq2	1	0	0	0
seq3	0	0	0	1
seq4	0	1	0	0

Probability of a Sequence Given a Motif Starting Position

$$P(X_i \mid Z_{i,j} = 1, p) = \underbrace{\prod_{k=1}^{j-1} p_{c_k,0}}_{\text{before motif}} \underbrace{\prod_{k=j}^{j+W-1} p_{c_k,k-j+1}}_{\text{motif}} \underbrace{\prod_{k=j+W}^{L} p_{c_k,0}}_{\text{after motif}}$$

X_i is the i th sequence

$Z_{i,j}$ is 1 if motif starts at position j in sequence i

c_k is the character at position k in sequence i

Sequence Probability Example

$$X_i = \mathbf{G\ C\ \boxed{T\ G\ T}\ A\ G}$$

$$p = \begin{array}{cccccc} & & \mathbf{0} & \mathbf{1} & \mathbf{2} & \mathbf{3} \\ & \mathbf{A} & \mathbf{0.25} & \mathbf{0.1} & \mathbf{0.5} & \mathbf{0.2} \\ & \mathbf{C} & \mathbf{0.25} & \mathbf{0.4} & \mathbf{0.2} & \mathbf{0.1} \\ & \mathbf{G} & \mathbf{0.25} & \mathbf{0.3} & \mathbf{0.1} & \mathbf{0.6} \\ & \mathbf{T} & \mathbf{0.25} & \mathbf{0.2} & \mathbf{0.2} & \mathbf{0.1} \end{array}$$

$$P(X_i \mid Z_{i3} = 1, p) =$$

$$p_{\mathrm{G},0} \times p_{\mathrm{C},0} \times p_{\mathrm{T},1} \times p_{\mathrm{G},2} \times p_{\mathrm{T},3} \times p_{\mathrm{A},0} \times p_{\mathrm{G},0} =$$

$$0.25 \times 0.25 \times 0.2 \times 0.1 \times 0.1 \times 0.25 \times 0.25$$

Likelihood function

$$P(D \mid p) = \prod_i P(X_i \mid p)$$

$$= \prod_i \sum_j P(X_i \mid Z_{ij} = 1, p) P(Z_{ij} = 1)$$

$$= (L - W + 1)^{-n} \prod_i \sum_j P(X_i \mid Z_{ij} = 1, p)$$

- EM will (indirectly) optimize this function

Basic EM Approach

given: length parameter *W*, training set of sequences

t=0

set initial values for $p^{(0)}$

do

++t

re-estimate $Z^{(t)}$ from $p^{(t-1)}$ (E-step)

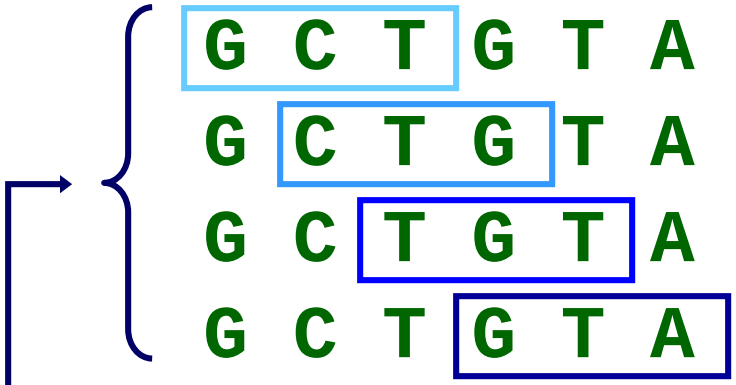
re-estimate $p^{(t)}$ from $Z^{(t)}$ (M-step)

until change in $p^{(t)} < \varepsilon$ (or change in likelihood is $< \varepsilon$)

return: $p^{(t)}$, $Z^{(t)}$

Warning: Notation Abuse!

- During the E-step, we compute the expected values of Z given $p^{(t-1)}$
- We denote these expected values by $Z^{(t)} = E[Z \mid p^{(t-1)}]$
- For example:



$Z^{(t)} =$

	1	2	3	4
seq1	0.1	0.1	0.2	0.6
seq2	0.4	0.2	0.1	0.3
seq3	0.3	0.1	0.5	0.1

The E-step: Computing $Z^{(t)}$

- To estimate the starting positions in Z at step t

$$Z_{i,j}^{(t)} = \frac{P(X_i \mid Z_{i,j} = 1, p^{(t-1)}) P(Z_{i,j} = 1)}{\sum_{k=1}^{L-W+1} P(X_i \mid Z_{i,k} = 1, p^{(t-1)}) P(Z_{i,k} = 1)}$$

- This comes from Bayes' rule applied to

$$P(Z_{i,j} = 1 \mid X_i, p^{(t-1)})$$

The E-step: Computing $Z^{(t)}$

- Assume that it is equally likely that the motif will start in any position

$$Z_{i,j}^{(t)} = \frac{P(X_i \mid Z_{i,j} = 1, p^{(t-1)}) \cancel{P(Z_{i,j} = 1)}}{\sum_{k=1}^{L-W+1} P(X_i \mid Z_{i,k} = 1, p^{(t-1)}) \cancel{P(Z_{i,k} = 1)}}$$

Example: Computing $Z^{(t)}$

$X_i =$ **G C T G T A G**

$p^{(t-1)} =$

	0	1	2	3
A	0.25	0.1	0.5	0.2
C	0.25	0.4	0.2	0.1
G	0.25	0.3	0.1	0.6
T	0.25	0.2	0.2	0.1

$$Z_{i,1}^{(t)} \propto P(X_i \mid Z_{i,1} = 1, p^{(t-1)}) = 0.3 \times 0.2 \times 0.1 \times 0.25 \times 0.25 \times 0.25 \times 0.25$$
$$Z_{i,2}^{(t)} \propto P(X_i \mid Z_{i,2} = 1, p^{(t-1)}) = 0.25 \times 0.4 \times 0.2 \times 0.6 \times 0.25 \times 0.25 \times 0.25$$

⋮

- Then normalize so that $\sum_{j=1}^{L-W+1} Z_{i,j}^{(t)} = 1$

The M-step: Estimating p

- Recall $p_{c,k}$ represents the probability of character c in position k ; values for $k=0$ represent the background

$$p_{c,k}^{(t)} = \frac{n_{c,k} + d_{c,k}}{\sum_b (n_{b,k} + d_{b,k})}$$

pseudo-counts

$$n_{c,k} = \begin{cases} \sum_i \sum_{\{j | X_{i,j+k-1} = c\}} Z_{i,j}^{(t)} & k > 0 \\ n_c - \sum_{j=1}^{W} n_{c,j} & k = 0 \end{cases}$$

sum over positions where c appears

total # of c's in data set

Example: Estimating p

A C A G C A

$$Z_{1,1}^{(t)} = 0.1,\quad Z_{1,2}^{(t)} = 0.7,\quad Z_{1,3}^{(t)} = 0.1,\quad Z_{1,4}^{(t)} = 0.1$$

A G G C A G

$$Z_{2,1}^{(t)} = 0.4,\quad Z_{2,2}^{(t)} = 0.1,\quad Z_{2,3}^{(t)} = 0.1,\quad Z_{2,4}^{(t)} = 0.4$$

T C A G T C

$$Z_{3,1}^{(t)} = 0.2,\quad Z_{3,2}^{(t)} = 0.6,\quad Z_{3,3}^{(t)} = 0.1,\quad Z_{3,4}^{(t)} = 0.1$$

$$p_{\mathrm{A},1}^{(t)} = \frac{Z_{1,1}^{(t)} + Z_{1,3}^{(t)} + Z_{2,1}^{(t)} + Z_{3,3}^{(t)} + 1}{Z_{1,1}^{(t)} + Z_{1,2}^{(t)} \ \ldots \ + Z_{3,3}^{(t)} + Z_{3,4}^{(t)} + 4}$$

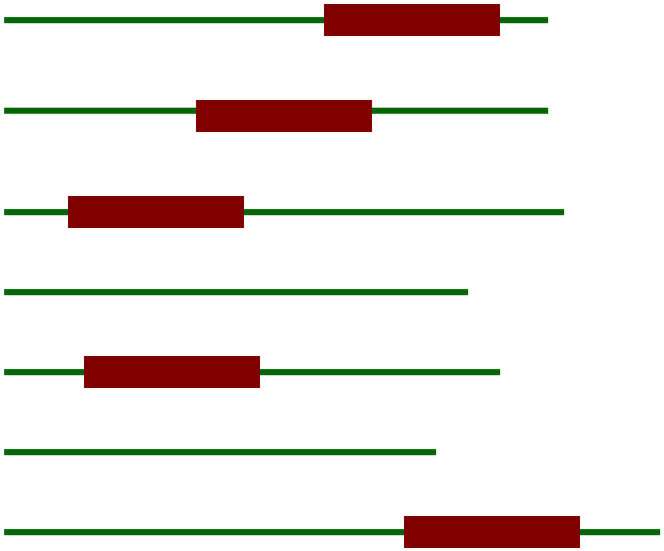
$$p_{\mathrm{C},2}^{(t)} = \frac{Z_{1,1}^{(t)} + Z_{1,4}^{(t)} + Z_{2,3}^{(t)} + Z_{3,1}^{(t)} + 1}{Z_{1,1}^{(t)} + Z_{1,2}^{(t)} \ \ldots \ + Z_{3,3}^{(t)} + Z_{3,4}^{(t)} + 4}$$

$$\vdots$$

The ZOOPS Model

- The approach as we've outlined it, assumes that each sequence has exactly one motif occurrence per sequence; this is the OOPS model

- The ZOOPS model assumes *zero or one* occurrences per sequence



E-step in the ZOOPS Model

- We need to consider another alternative: the *i*th sequence doesn't contain the motif
- We add another parameter (and its relative)

$$\gamma$$

- prior probability of a sequence containing a motif

$$\lambda = \frac{\gamma}{(L - W + 1)}$$

- prior probability that any position in a sequence is the start of a motif

E-step in the ZOOPS Model

$$Z_{i,j}^{(t)} = \frac{P(X_i \mid Z_{i,j} = 1, p^{(t-1)})\lambda^{(t-1)}}{P(X_i \mid Q_i = 0, p^{(t-1)})(1-\gamma^{(t-1)}) + \sum_{k=1}^{L-W+1} P(X_i \mid Z_{i,k} = 1, p^{(t-1)})\lambda^{(t-1)}}$$

- Q_i is a random variable for which $Q_i = 1$ if sequence X_i contains a motif, $Q_i = 0$ otherwise

$$Q_i = \sum_{j=1}^{L-W+1} Z_{i,j}$$

$$P(X_i \mid Q_i = 0, p^{(t-1)}) = \prod_{j=1}^{L} p_{c_j,0}^{(t-1)}$$

M-step in the ZOOPS Model

- Update p same as before
- Update γ as follows:

$$\gamma^{(t)} \equiv (L - W + 1)\lambda^{(t)} = \frac{1}{n}\sum_{i=1}^{n} Q_i^{(t)}$$

Extensions to the Basic EM Approach in MEME

- Varying the approach (TCM model) to assume *zero or more* motif occurrences per sequence
- Choosing the width of the motif
- Finding multiple motifs in a group of sequences

✓ Choosing good starting points for the parameters

✓ Using background knowledge to bias the parameters

Starting Points in MEME

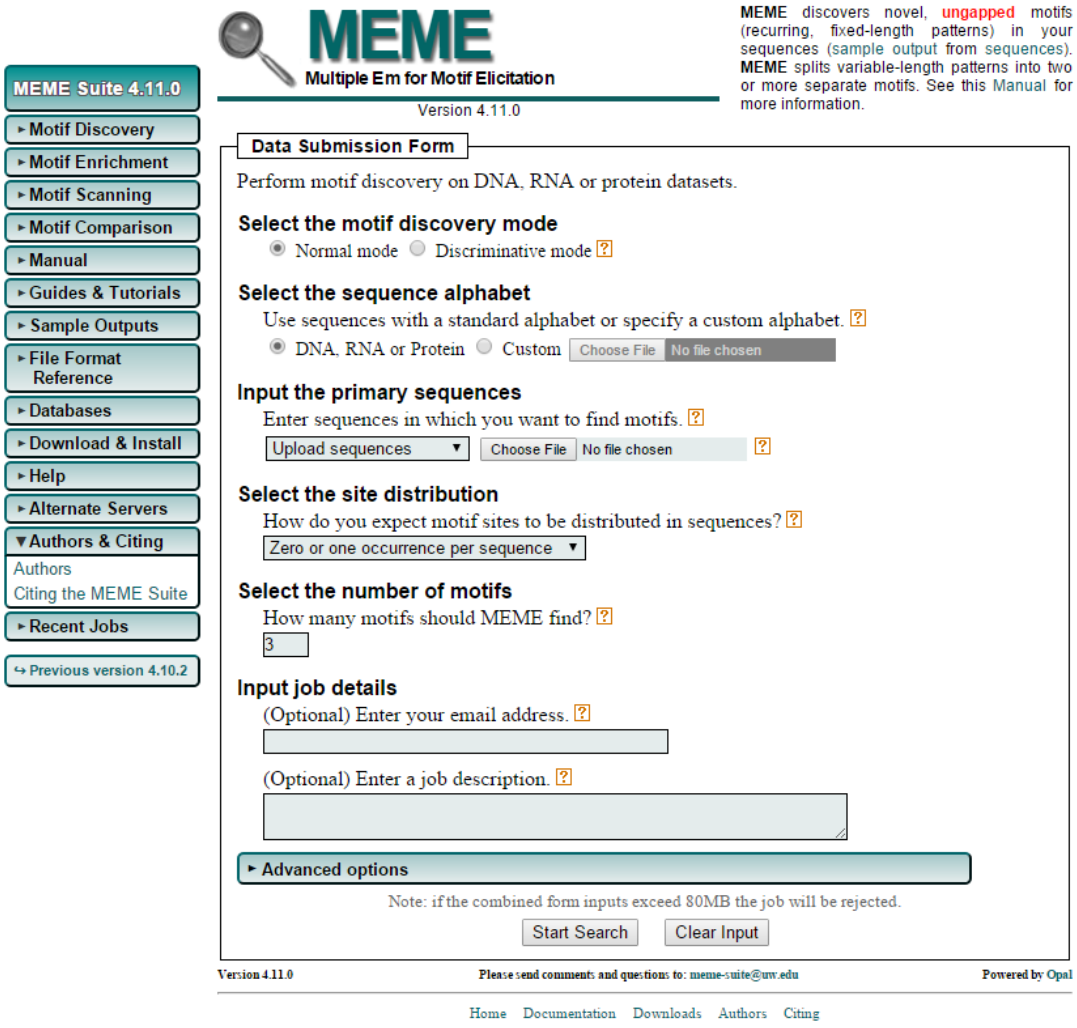
- EM is susceptible to local maxima, so it's a good idea to try multiple starting points
- Insight: motif must be similar to *some* subsequence in data set
- For every distinct subsequence of length *W* in the training set
  - derive an initial *p* matrix from this subsequence
  - run EM for 1 iteration
- Choose motif model (i.e. *p* matrix) with highest likelihood
- Run EM to convergence

Using Subsequences as Starting Points for EM

- Set values matching letters in the subsequence to some value π
- Set other values to $(1-\pi)/(M-1)$ where M is the length of the alphabet
- Example: for the subsequence TAT with $\pi = 0.7$

$$p = \begin{array}{c|ccc} & 1 & 2 & 3 \\ A & 0.1 & 0.7 & 0.1 \\ C & 0.1 & 0.1 & 0.1 \\ G & 0.1 & 0.1 & 0.1 \\ T & 0.7 & 0.1 & 0.7 \end{array}$$

MEME web server



http://meme-suite.org/