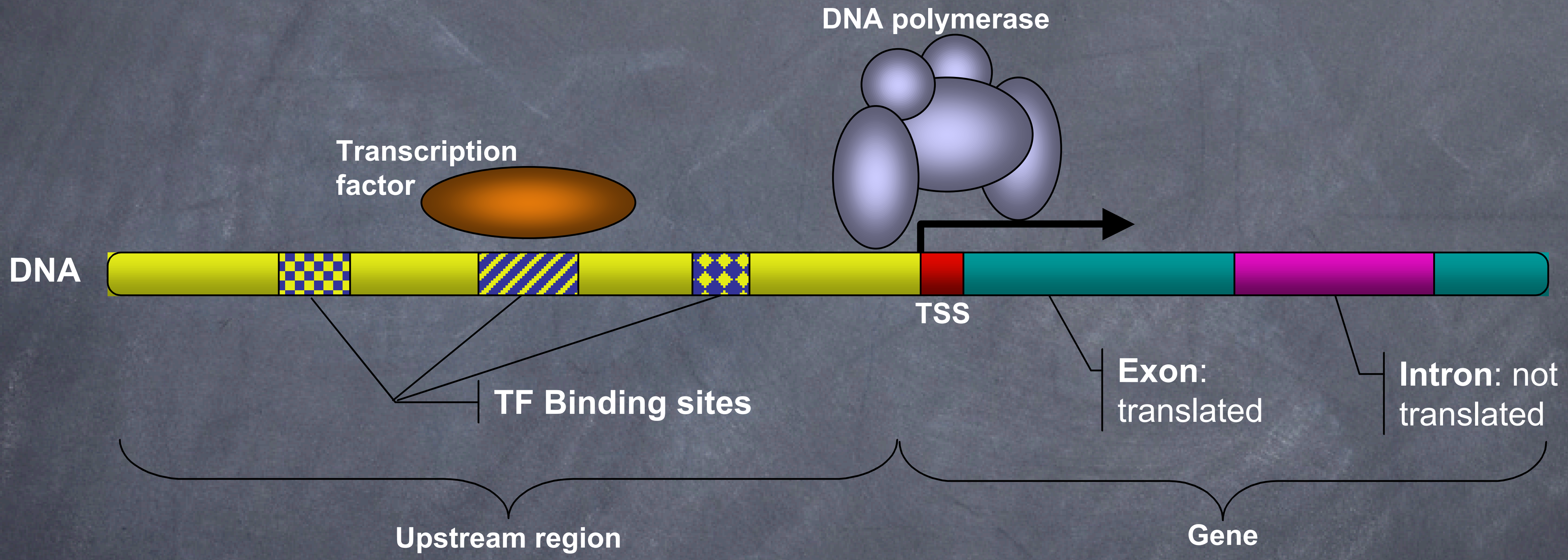
Motif Finding

CMSC 423

DNA -> mRNA -> Protein



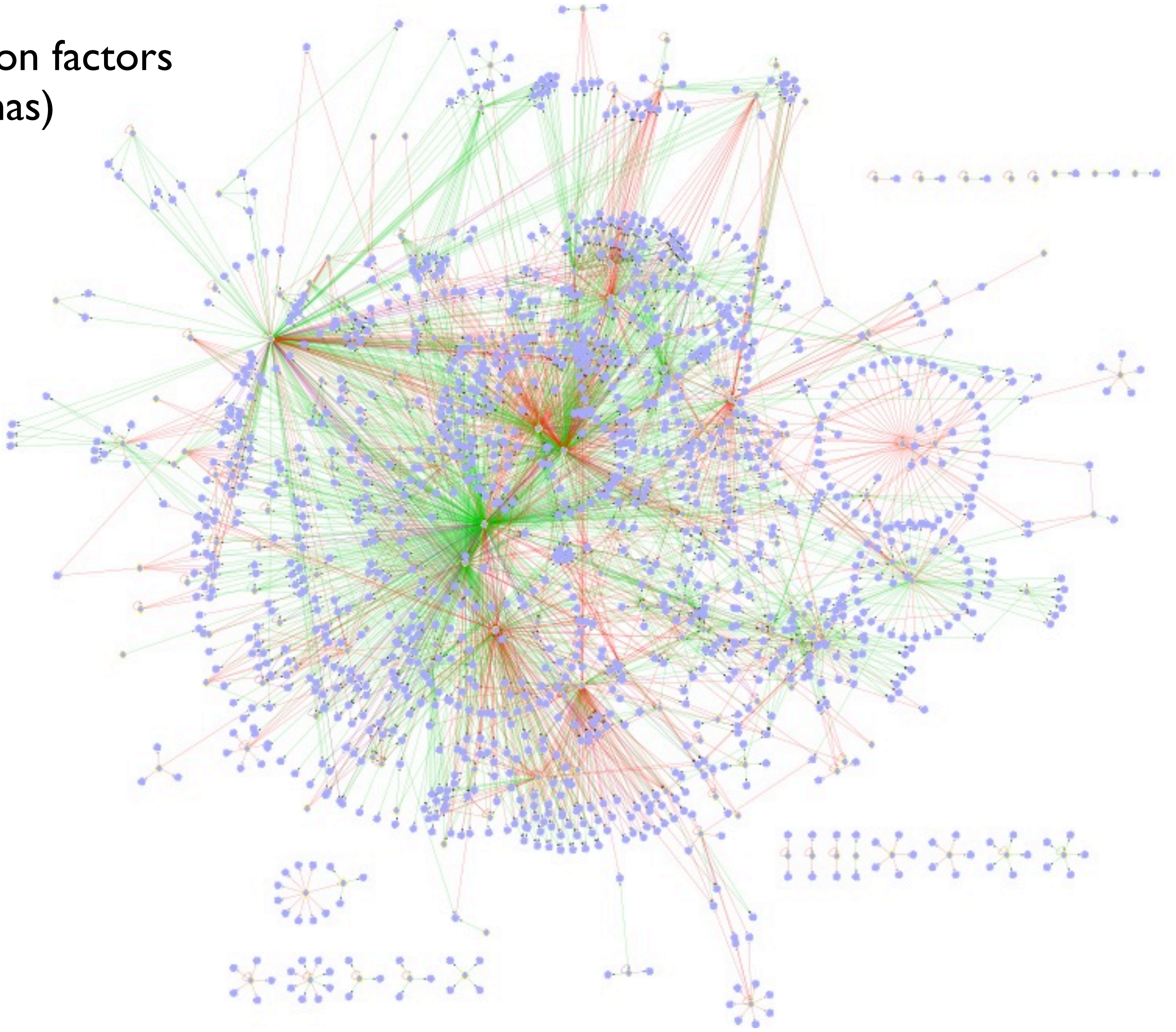
- Finding transcription factor binding sites can tell us about the cell's regulatory network.

Transcription Network

169 transcription factors
(excluding sigmas)

3322 edges

1753 activation,
1369 repression,
185 both,
3 unknown



RNA Polymerase

b/c it makes RNA

into a polymer

is an enzyme

Discovered in 1960; Nobel prize for its discovery in 1959... oops

1959 Nobel awarded to Severo Ochoa and Arthur Kornberg for discovering what was mistakenly believed to be RNA pol.

1960 Sam Weiss and Jared Hurwitz discover the *real* RNA pol.

2006 Nobel awarded to **Roger** Kornberg (son of Arthur) for detailed structure of RNA pol.

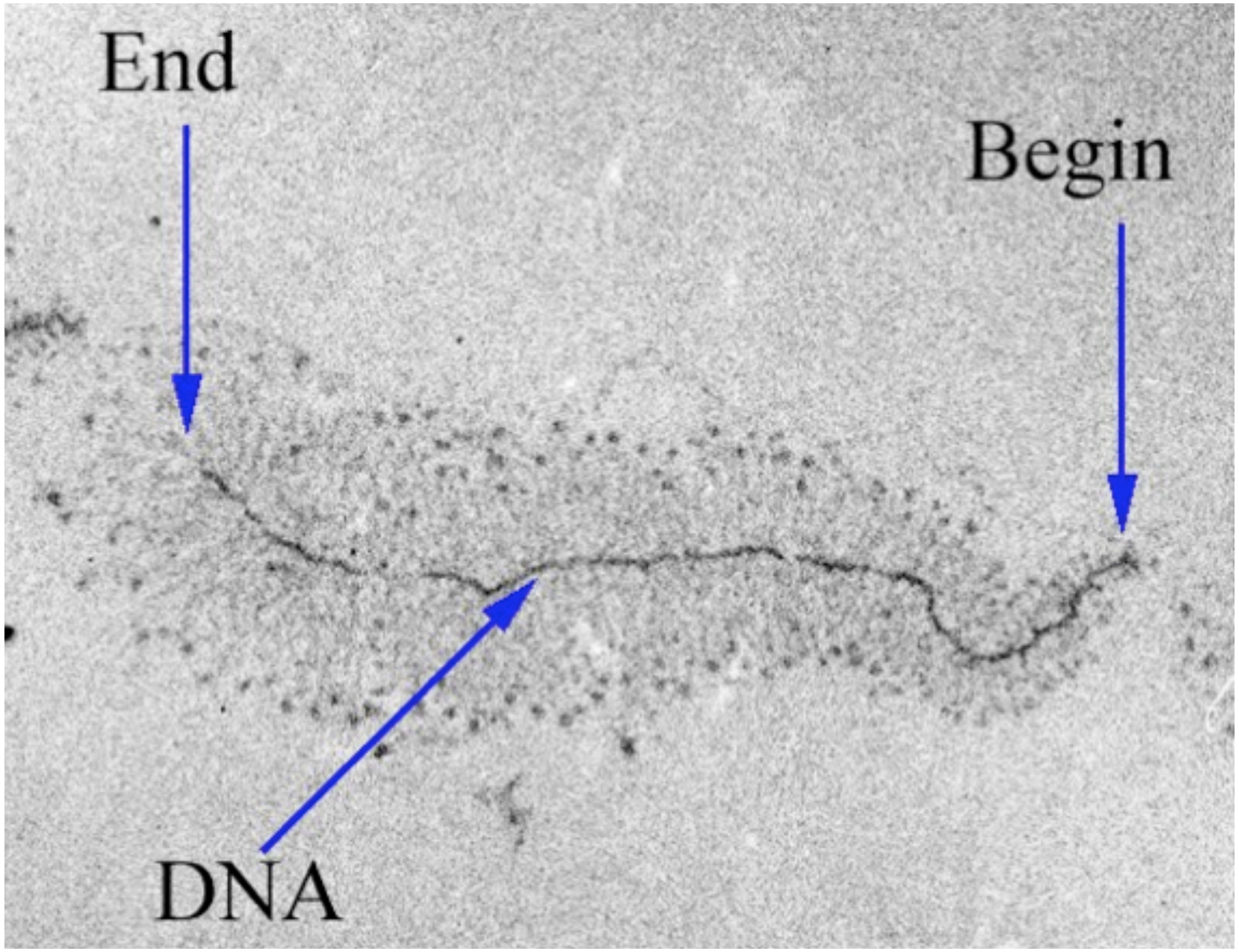
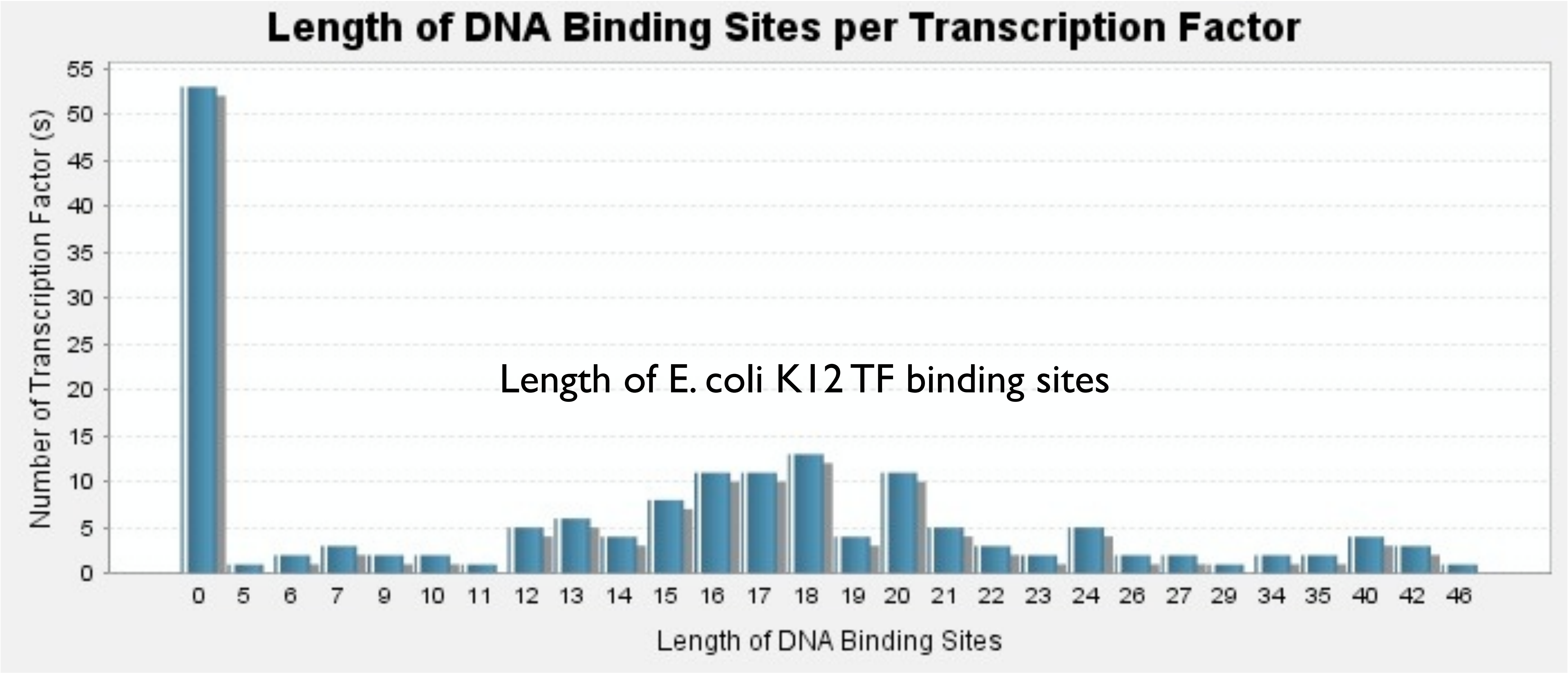


Image of transcription occurring.
Each "hair" is a piece of RNA that RNA pol is growing off of the DNA.

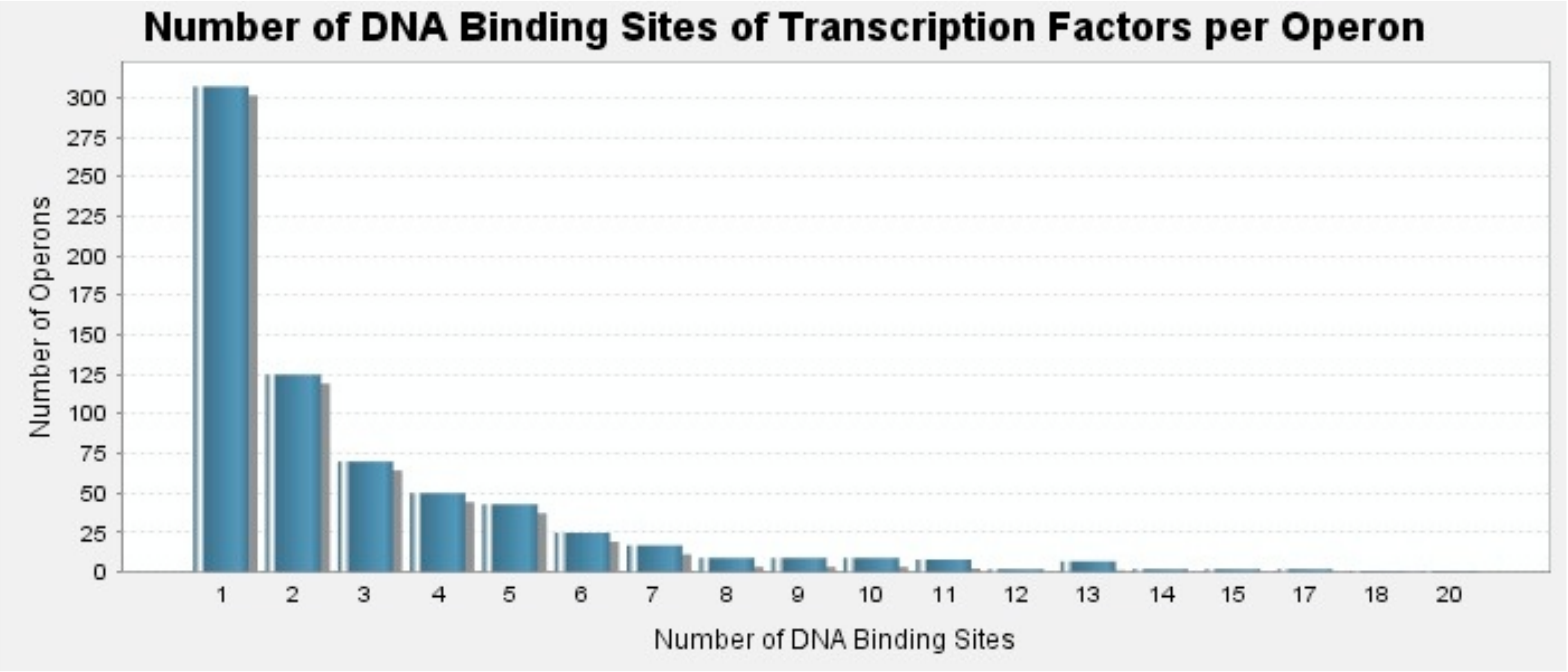
Transcription Factor Binding Sites



Length of E. coli K12 TF binding sites

RegulonDB (Feb 27, 2010)

Transcription Factor Binding Sites



RegulonDB (Feb 27, 2010)

Motif Finding

Transcription factor

```
1. ttgccacaaaataatccgccttcgcaaattgaccTACCTCAATAGCGGTAgaaaaacgcaccactgcctgacag
2. gtaagtacctgaaagttacggtctgcgaacgctattccacTGCTCCTTTATAGGTAcaacagtatagtctgatgga
3. ccacacggcaaataaggagTAACTCTTTCCGGGTAtgggtatacttcagccaatagccgagaatactgccattccag
4. ccatacccggaaagagttactccttatttgccgtgtggttagtcgcttTACATCGGTAAGGGTAgggattttacagca
5. aaactattaagattttatgcagatgggtattaaggaGTATTCCCCATGGGTAacatattaatggctctta
6. ttacagtctgttatgtggtggctgttaaTTATCCTAAAGGGGTAtcttaggaatttactt
```

Given p sequences, find the most mutually similar length-k subsequences, one from each sequence:

$$\operatorname*{argmin}_{s_1,\ldots,s_p} \sum_{i<j} \mathrm{dist}(s_i, s_j)$$

$\mathrm{dist}(s_i,s_j)$ = Hamming distance between s_i and s_j.

Hundreds of papers, many formulations (Tompa05)

Motif-finding by Gibbs Sampling

Problem. Given *p* strings and a length *k*, find the most "mutually similar" length-k substring from each string.

"Gibbs sampling" is the basis behind a general class of algorithms that is a type of local search.

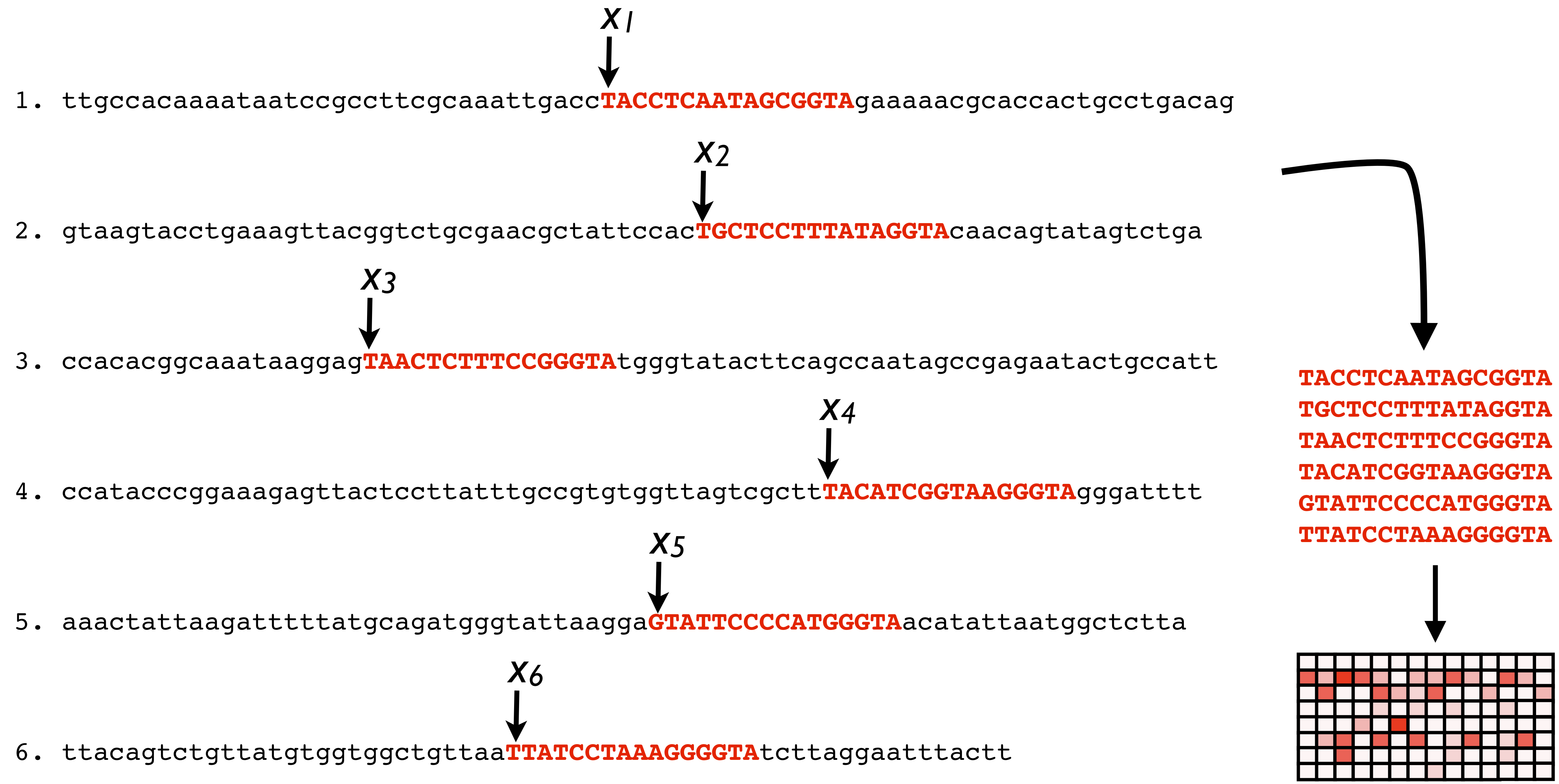
It doesn't guarantee good performance, but often works well in practice.

Assumes:

1. we know the length *k* of the motif we are looking for.
2. each input sequence contains exactly 1 real instance of the motif.

Gibbs Sampling: Profiles

If we knew the starting point of the motif in each sequence, we could construct a Sequence Profile (PSSM) for the motif:



1. ttgccacaaaataatccgccttcgcaaattgacc**TACCTCAATAGCGGTA**gaaaaacgcaccactgcctgacag
2. gtaagtacctgaaagttacggtctgcgaacgctattccac**TGCTCCTTTATAGGTA**caacagtatagtctga
3. ccacacggcaaataaggag**TAACTCTTTCCGGGTA**tgggtatacttcagccaatagccgagaatactgccatt
4. ccatacccggaaagagttactccttatttgccgtgtggttagtcgctt**TACATCGGTAAGGGTA**gggatttt
5. aaactattaagatttttatgcagatgggtattaagga**GTATTCCCCATGGGTA**acatattaatggctctta
6. ttacagtctgttatgtggtggctgttaa**TTATCCTAAAGGGGTA**tcttaggaatttactt

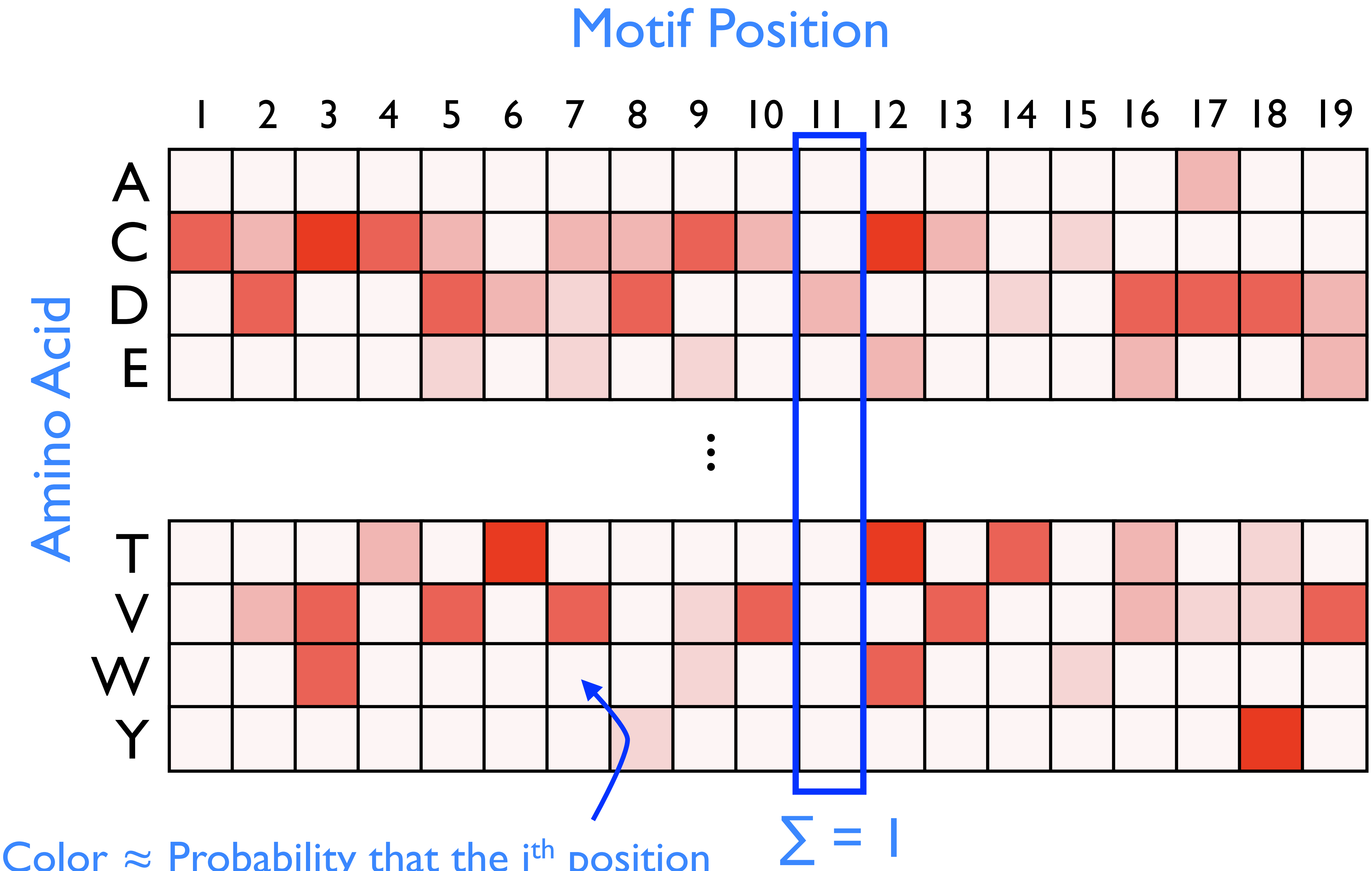
TACCTCAATAGCGGTA
TGCTCCTTTATAGGTA
TAACTCTTTCCGGGTA
TACATCGGTAAGGGTA
GTATTCCCCATGGGTA
TTATCCTAAAGGGGTA

Gibbs Sampling, Version 1: Pseudocode

```
Set $(x_1, x_2, ..., x_p)$ to random positions in each input string.

**repeat until** the answer $(x_1, x_2, ..., x_p)$ doesn't change
    **for** i = 1 ... p:
        Build a profile Q using sequences at $(x_1, x_2, ..., x_p)$ except $x_i$
        Set $x_i$ to where the profile Q matches **best** in string *i*.
```

Sequence Profiles (PSSM)

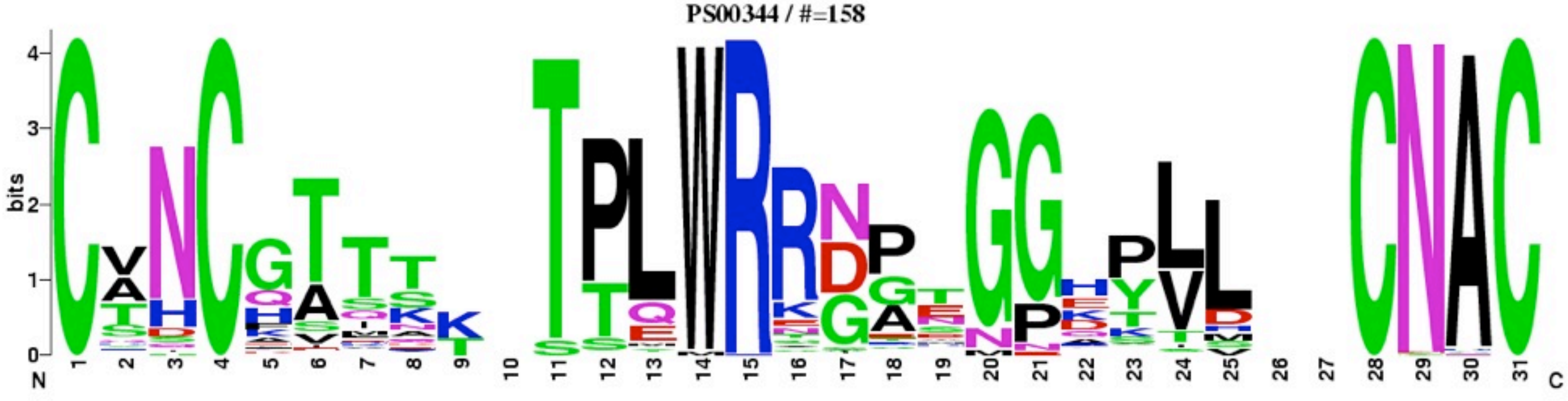


Color ≈ Probability that the i^{th} position has the given amino acid = $e_i(x)$.

Sequence Logos

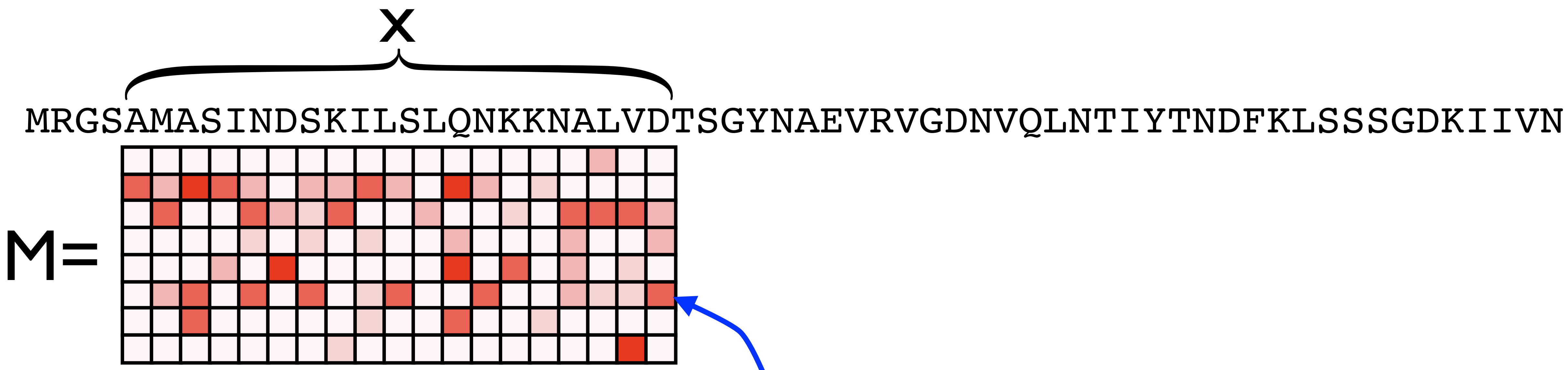
Height of letter ≈ fraction of time that letter is observed at that position.

(Height of all the letters in a column ≈ to how conserved the column is)



Motif Position

Scoring a Sequence


x
MRGSAMASINDSKILSLQNKKNALVDTSGYNAEVRVGDNVQLNTIYTNDFKLSSSGDKIIVN
M=

Color ≈ Probability that the ith position has the given amino acid = $e_i(x)$.

$$\text{Score}(x) = \Pr(x \mid M) = \prod_{i=1}^{L} e_i(x_i)$$

Score of a string according to profile M = Product of the probabilities you would observe the given letters.

Background Frequencies

Interested in how different this motif position is from we expect by chance.

Correct for "expect by chance" by dividing by the probability of observing x in a random string:

$$\text{ScoreCorrected}(x) = \frac{\Pr(x \mid M)}{\Pr(x \mid \text{background})} = \prod_{i=1}^{L} \frac{e_i(x_i)}{b(x_i)}$$

$b(x_i)$:= probability of observing character x_i at random.
Usually computed as (# x_i in entire string) / (length of string)

Often, to avoid multiplying lots of terms, we take the log and then sum:

$$\text{ScoreCorrectedLog}(x) = \log \prod_{i=1}^{L} \frac{e_i(x_i)}{b(x_i)} = \sum_{i=1}^{L} \log\left(\frac{e_i(x_i)}{b(x_i)}\right)$$

Gibbs Example

```
gibbs(["thequickdog", "browndog", "dogwood"], k=3)
1: [8, 1, 2] ['dog', 'row', 'gwo']
2: [8, 5, 0] ['dog', 'dog', 'dog']
F: [8, 5, 0] ['dog', 'dog', 'dog']
```

random starting positions

Small bias toward “o” in the middle is correct.

```
gibbs(["thequickdog", "browndog", "dogwood"], k=3)
1: [4, 3, 1] ['uic', 'wnd', 'ogw']
2: [6, 2, 4] ['ckd', 'own', 'ood']
3: [8, 5, 0] ['dog', 'dog', 'dog']
F: [8, 5, 0] ['dog', 'dog', 'dog']
```

```
gibbs(["thequickdog", "browndog", "dogwood"], k=3)
1: [2, 0, 1] ['equ', 'bro', 'ogw']
2: [7, 4, 2] ['kdo', 'ndo', 'gwo']
F: [7, 4, 2] ['kdo', 'ndo', 'gwo']
```

Might not find the optimal.

```python
def gibbs(Seqs, k):
   """Seqs is a list of strings. Find the best motif."""

   # start with random indices
   I = [random.randint(0, len(x) - k) for x in Seqs]

   LastI = None
   while I != LastI:     # repeat until nothing changes
      LastI = list(I)

      # iterate through every string
      for i in xrange(len(Seqs)):
         # compute the profile for the sequences except i
         P = profile_for([
                x[j : j + k] for q, (x, j) in enumerate(zip(Seqs, I))
                   if q != i
             ])

         # find the place the profile matches best
         best = None
         for j in xrange(len(Seqs[i]) - k + 1):
            score = profile_score(P, Seqs[i][j : j + k])
            if score > best or best is None:
               best = score
               bestpos = j
         # update the ith position with the best
         I[i] = bestpos

   return I, [x[j : j + k] for x, j in zip(Seqs, I)]
```

Another Example

```
gibbs(["aaa123", "678aaa45", "9a7aaab", "32aa19a8aaa"], 3)
1: [0, 5, 0, 2] ['aaa', 'a45', '9a7', 'aa1']
2: [1, 3, 3, 8] ['aa1', 'aaa', 'aaa', 'aaa']
3: [0, 3, 3, 8] ['aaa', 'aaa', 'aaa', 'aaa']
F: [0, 3, 3, 8] ['aaa', 'aaa', 'aaa', 'aaa']
```

Bias toward “a” in the profile quickly leads to finding the implanted “aaa”

Can be multiple optimal answers

```
gibbs(["aaabbb", "bbbaaabb", 'babaaab', 'ababacaaabac', 'abbbababaaabbbaba'], 3)
1: [1, 4, 0, 4, 11] ['aab', 'aab', 'bab', 'aca', 'bbb']
2: [1, 4, 4, 7, 9]  ['aab', 'aab', 'aab', 'aab', 'aab']
F: [1, 4, 4, 7, 9]  ['aab', 'aab', 'aab', 'aab', 'aab']
gibbs(["aaabbb", "bbbaaabb", 'babaaab', 'ababacaaabac', 'abbbababaaabbbaba'], 3)
1: [0, 3, 3, 3, 8] ['aaa', 'aaa', 'aaa', 'bac', 'aaa']
2: [0, 3, 3, 6, 8] ['aaa', 'aaa', 'aaa', 'aaa', 'aaa']
F: [0, 3, 3, 6, 8] ['aaa', 'aaa', 'aaa', 'aaa', 'aaa']
```

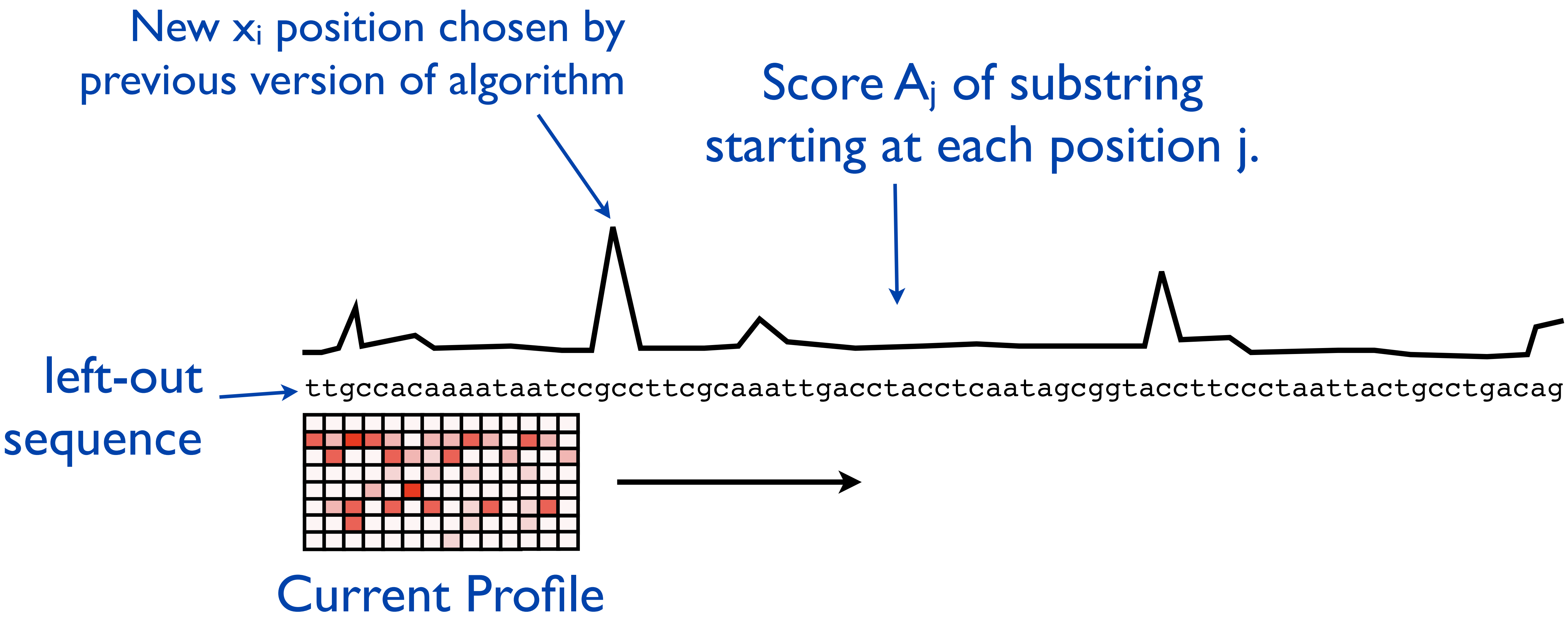
Randomness: Gibbs Sampling

- Run the Gibbs sampling multiple times to make it more likely you find the global optimal.
- Can increase the use of randomness to further avoid getting stuck in local optima by choosing new x_i randomly.

```
Set $(x_1, x_2, ..., x_p)$ to random positions in each input string.

repeat until the best $(x_1, x_2, ..., x_p)$ doesn't change too often
    for $i = 1 ... p$:
        Build a profile Q using sequences at $(x_1, x_2, ..., x_p)$ except $x_i$
        Choose $x_i$ according to the profile probability distribution of Q
        in string $i$.
```

Profile Probability Distribution



New x_i position chosen by previous version of algorithm

Score A_j of substring starting at each position j.

left-out sequence

ttgccacaaaataatccgccttcgcaaattgacctacctcaatagcggtaccttccctaattactgcctgacag

Current Profile

Instead of choosing the position with the best match, choose a position randomly such that:

$$\text{Probability of choosing position } j = \frac{A_j}{\sum_i A_i}$$

(Lawrence, et al., *Science*, 1994)

Recap

- “Motif finding” is the problem of finding a set of common substrings within a set of strings.

- Useful for finding transcription factor binding sites.

- **Gibbs sampling:** repeatedly leave one sequence out and optimize the motif location in the left-out sequence.
- Doesn’t guarantee finding a good solution, but often works.