CSE 527
Lecture 17, 11/24/04

RNA Secondary Structure Prediction

Outline

- What is it
- How is it Represented
- Why is it important
- Examples
- Approaches

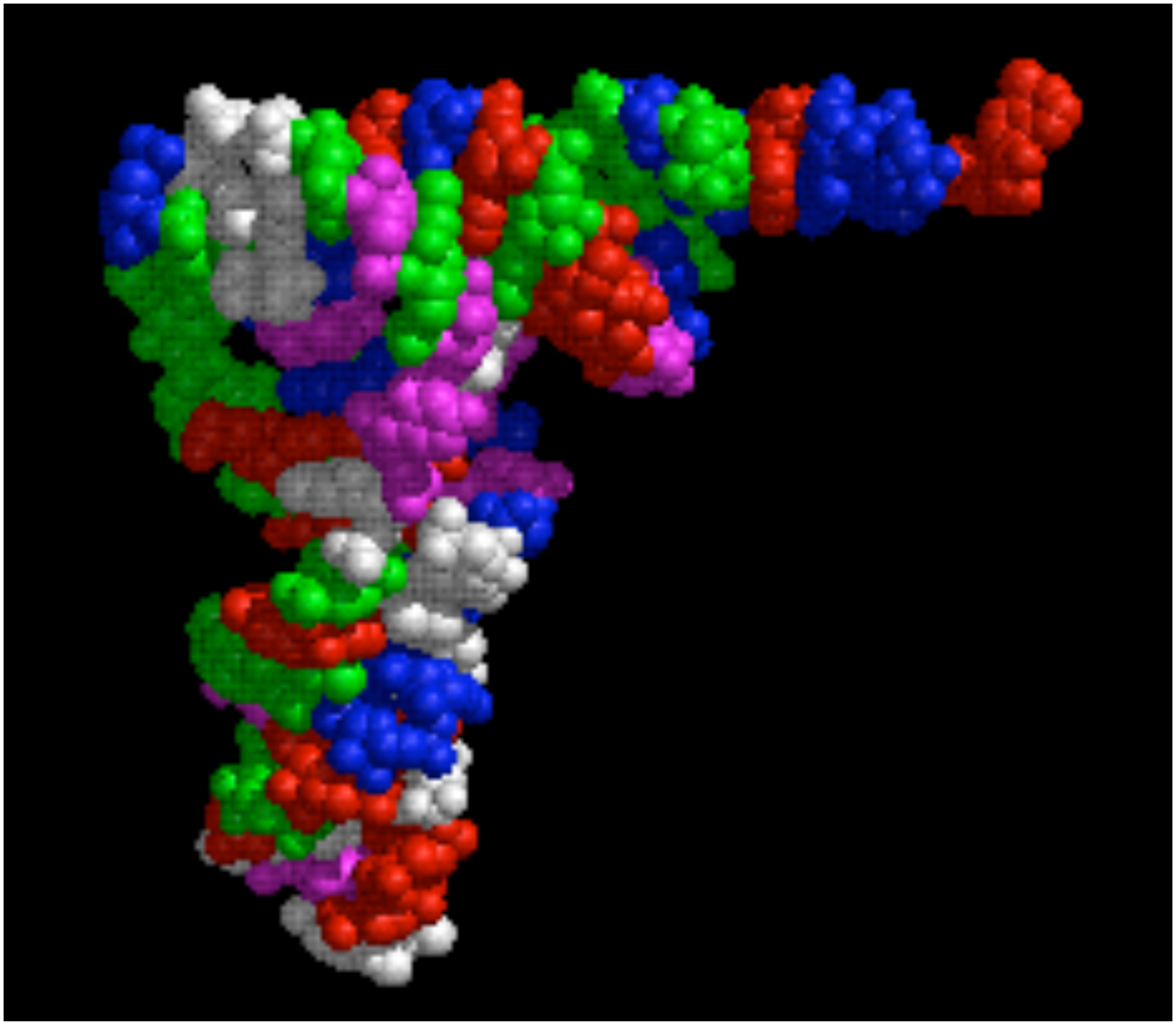
RNA Structure

- Primary Structure: Sequence
- Secondary Structure: Pairing
- Tertiary Structure: 3D shape

RNA Pairing

- Watson-Crick Pairing
  - C - G ~ 3 kcal/mole
  - A - U ~ 2 kcal/mole
- "Wobble Pair" G - U ~ 1 kcal/mole
- Non-canonical Pairs (esp. if modified)

A tRNA 3d Structure



tRNA - Alt. Representations

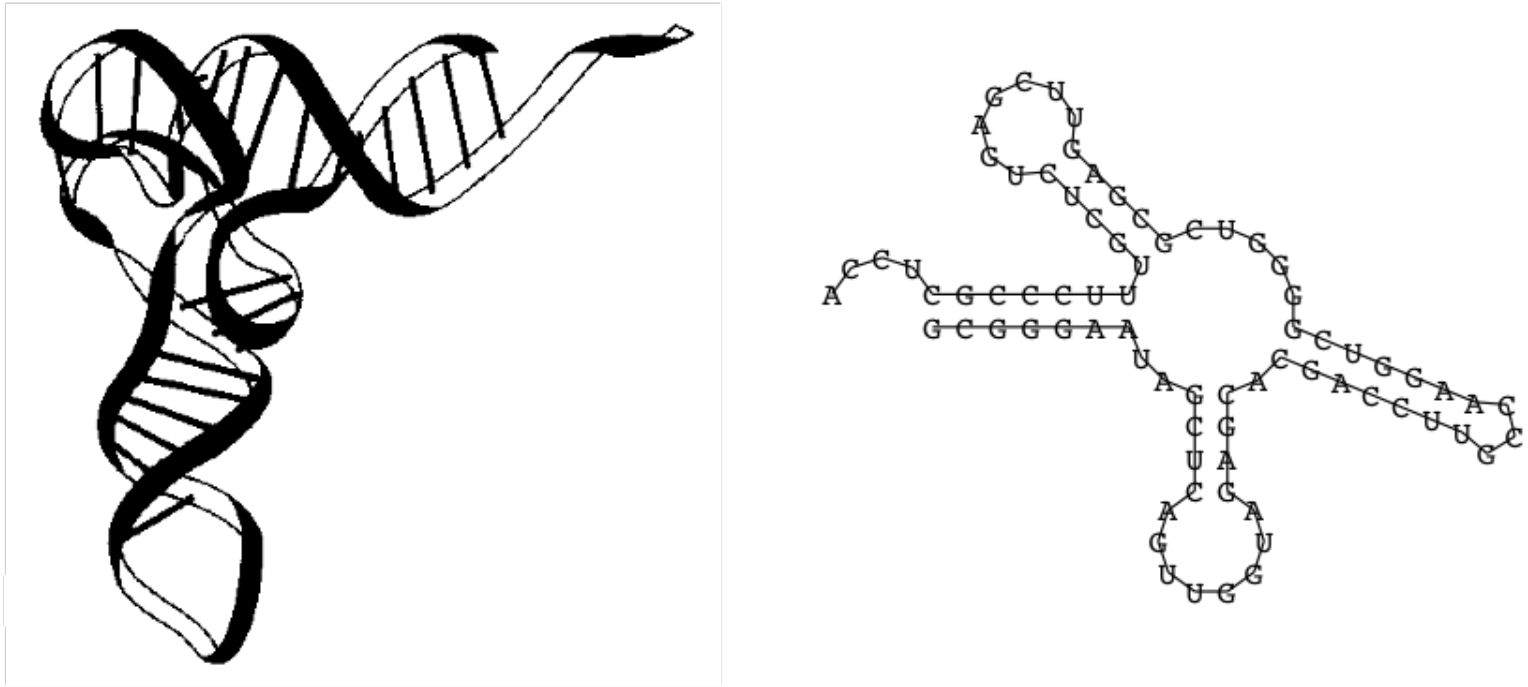


Figure 1: a) The spatial structure of the phenylalanine tRNA form yeast

b) The secondary structure extracts the most important information about the structure, namely the pattern of base pairings.

A "Mountain" diagram

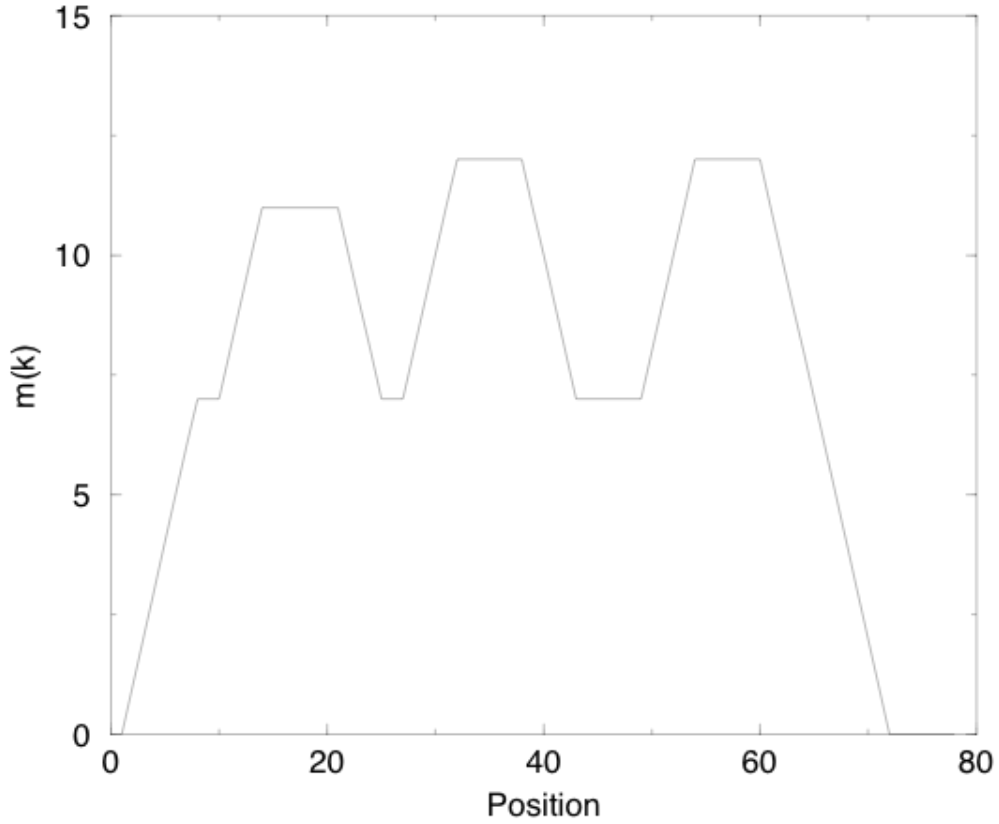
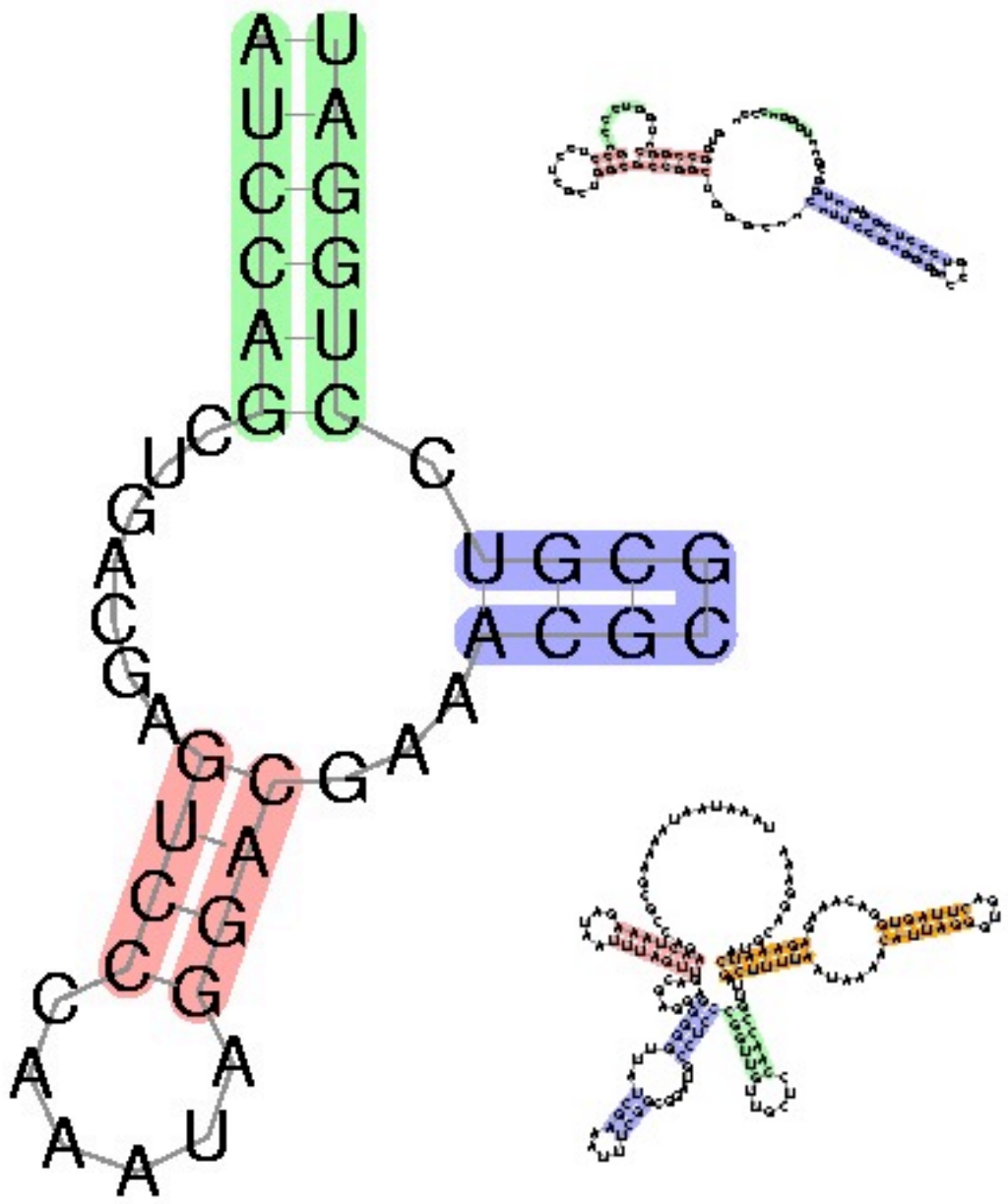


Figure 3: Mountain representation of the tRNA secondary structure shown in Figure 1. The three plateaus correspond to the three hairpin loops of the clover leave structure.

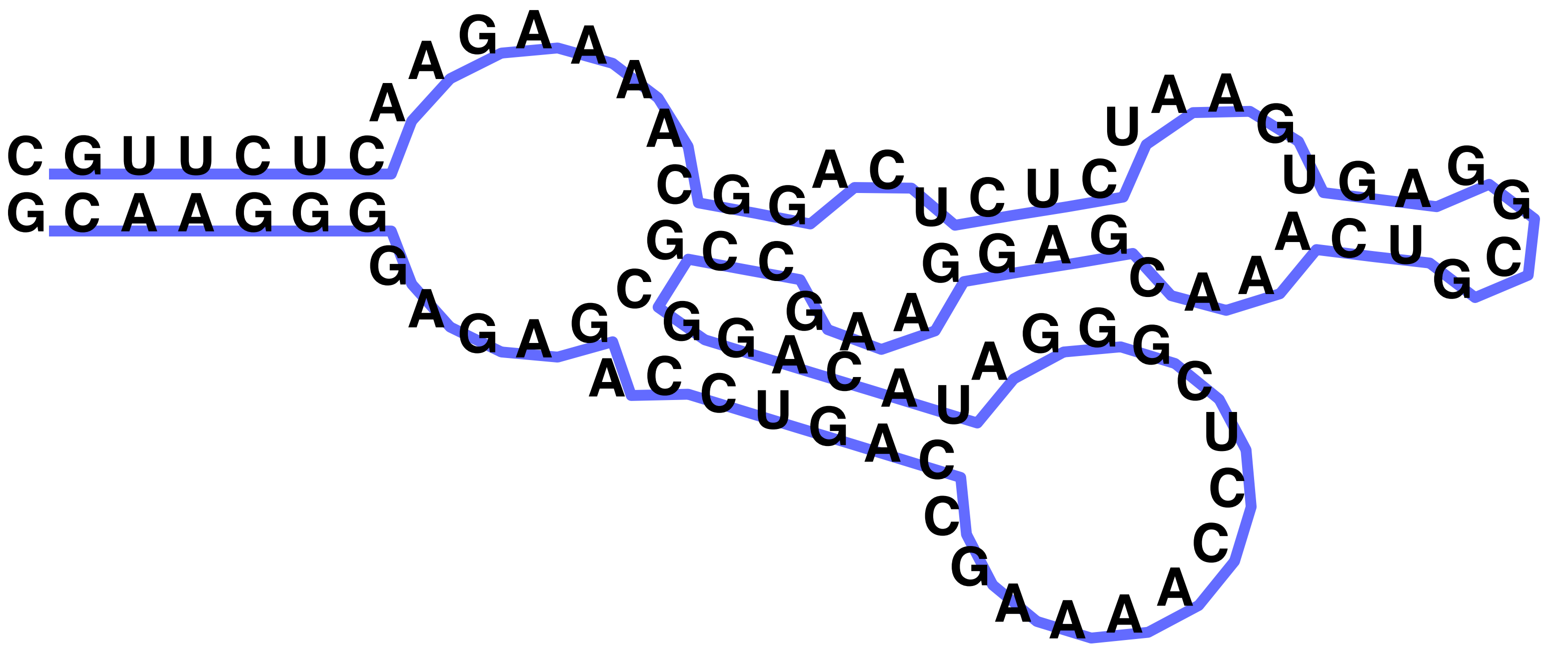
Why?

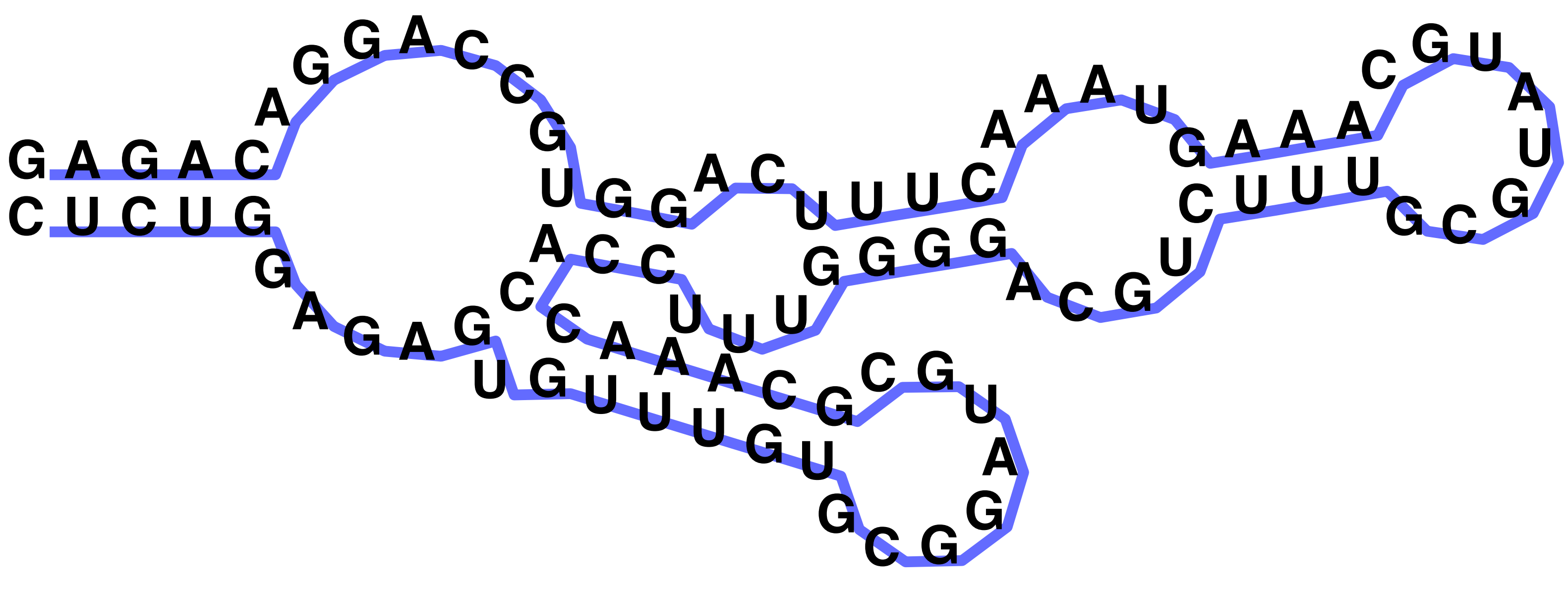
- RNA's fold, and function
- Nature uses what works



Importance

- Ribozymes (RNA Enzymes)
- Retroviruses
- Effects on transcription, translation, splicing...
- Functional RNAs: rRNA, tRNA, snRNA, snoRNA, micro RNA, RNAi, riboswitches, regulatory elements in 3' & 5' UTRs, ...





Definitions

- Sequence $^{5'}\ r_1\ r_2\ r_3 \ldots r_n\ ^{3'}$ in {A, C, G, T}
- A Secondary Structure is a set of pairs i•j s.t.

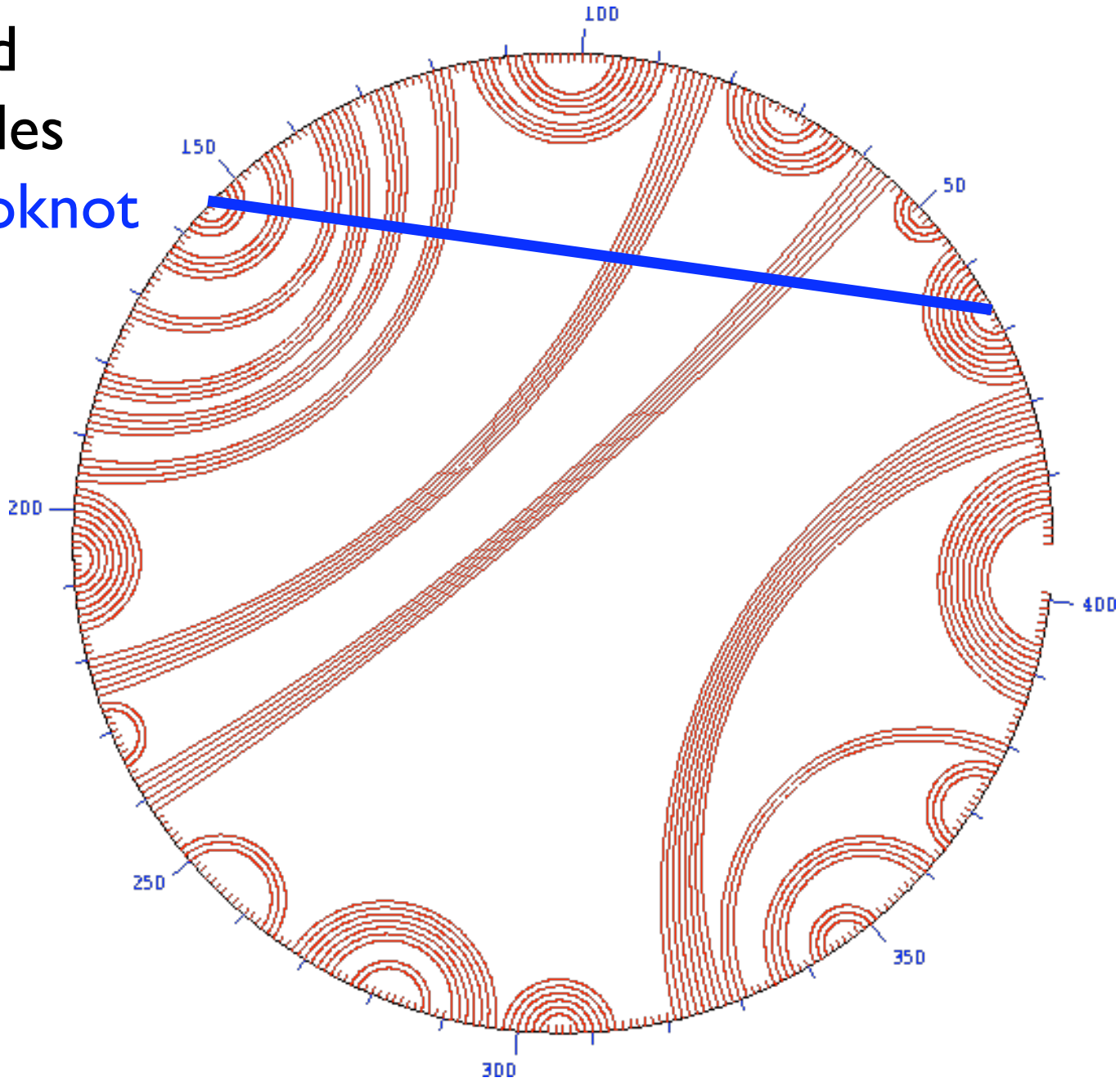
1. $i < j-4$
2. if i•j & i'•j' are two pairs with $i \leq i'$, then
   - A. $i = i'$ & $j = j'$, or
   - B. $j < i'$, or
   - C. $i < i' < j' < j$

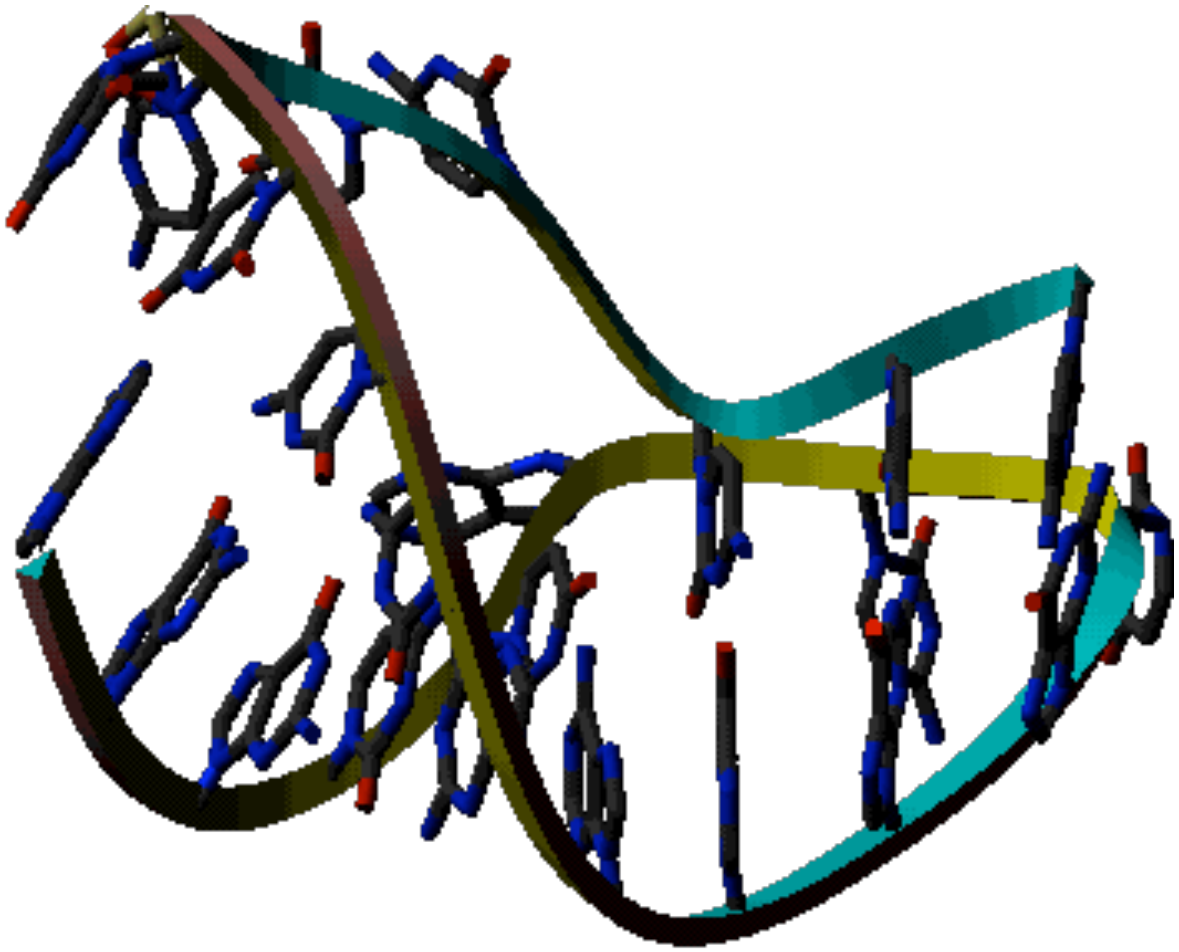
} First pair precedes 2nd, or is nested within it. No "pseudoknots."

Nested

Precedes

Pseudoknot





The corresponding secondary structure is:

```
                  A–C
3' – A–G–G–C–U/    U
     U–C–C–G–A–G–G–G
      U          C–C–C – 5'
       C––U––C/
```

A Pseudoknot

Approaches to Structure Prediction

- Maximum Pairing
  + simple
  - too inaccurate
- Minimum Energy
  + Works on single sequences
  - Ignores pseudoknots
  - Only finds “optimal” fold
- Partition Function
  + Finds all folds
  - Ignores pseudoknots

Approaches, II

- Comparative sequence analysis
  + handles all pairings (incl. pseudoknots)
  - requires several (many?) aligned, appropriately diverged
- Stochastic Context-free Grammars
  Roughly combines min energy & comparative, but no pseudoknots
- Physical experiments (x-ray crystalography, NMR)

Nussinov: Max Pairing

- B(i,j) = # pairs in optimal pairing of ri ... rj
- B(i,j) = 0 for all i, j with i ≥ j-4; otherwise
- B(i,j) = max of:

  1. B(i+1,j)
  2. B(i,j-1)
  3. B(i+1,j-1) +(if ri pairs with rj then 1 else 0)
  4. max { B(i,k)+B(k+1,j) | i < k < j }

Time: $O(n^3)$

Pair-based Energy Minimization

- E(i,j) = energy of pairs in optimal pairing of ri ... rj
- E(i,j) = ∞ for all i, j with i ≥ j-4; otherwise
- E(i,j) = min of:

  1. E(i+1,j)
  2. E(i,j-1)
  3. E(i+1,j-1) + e(ri,rj)
  4. min { E(i,k)+E(k+1,j) | i < k < j }

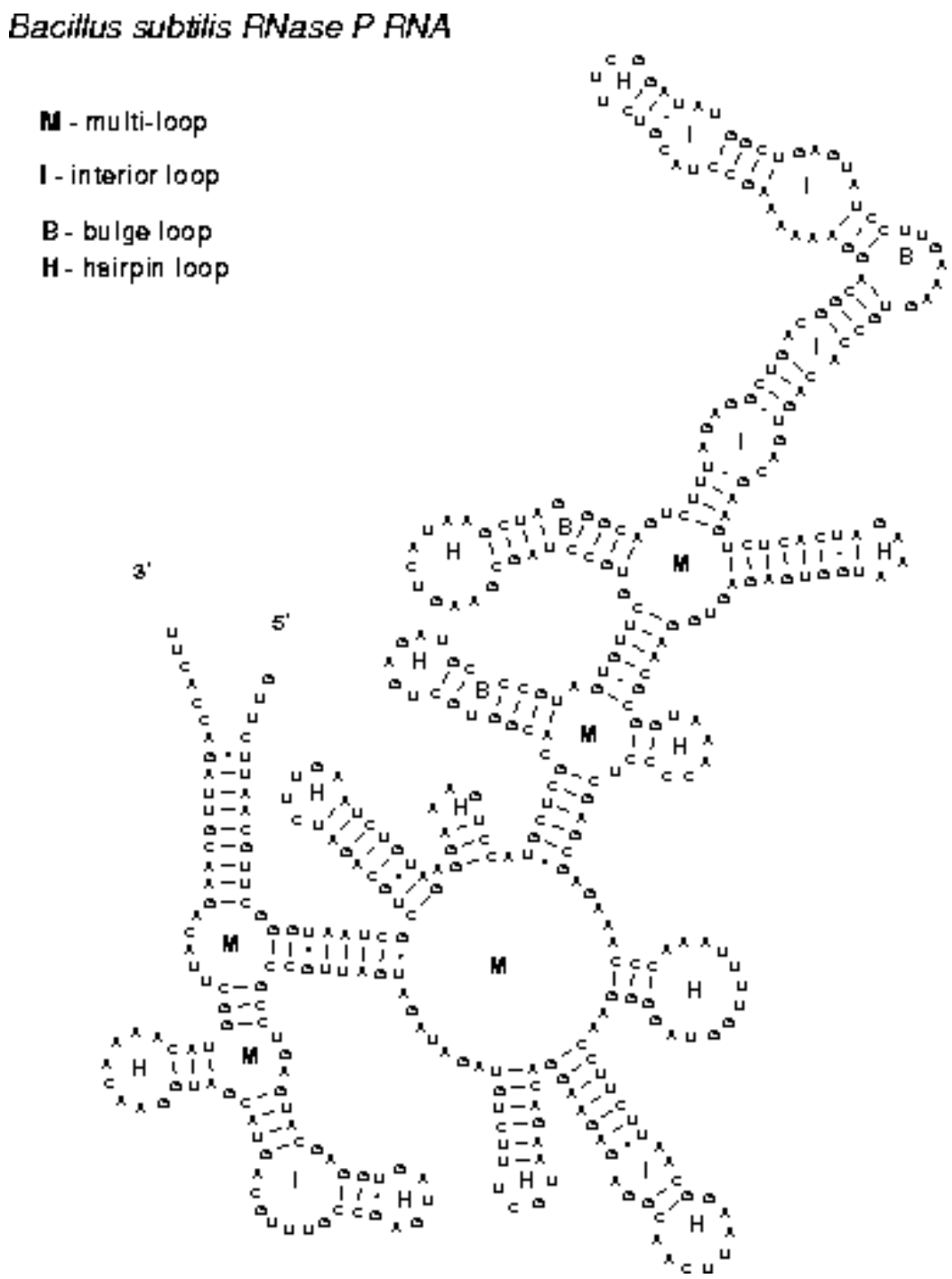
energy of one pair

Time: $O(n^3)$

Loop-based Energy Minimization

- Detailed experiments show that it's more accurate to model based on loops, rather than just pairs
- Loop types
  - Stack
  - Hairpin loop
  - Bulge
  - Interior loop

Loop Examples



Zuker: Loop-based Energy, I

- W(i,j) = energy of optimal pairing of ri ... rj
- V(i,j) = as above, but forcing pair i•j
- W(i,j) = V(i,j) = ∞ for all i, j with i ≥ j-4
- W(i,j) = min(W(i+1,j), W(i,j-1), V(i+1,j-1),
  min { E(i,k)+E(k+1,j) | i < k < j })

Zuker: Loop-based Energy, II

- V(i,j) =min(eh(i,j), es(i,j)+V(i+1,j-1), VBI(i,j), VM(i,j))

  hairpin, stack, bulge/interior, multi-loop

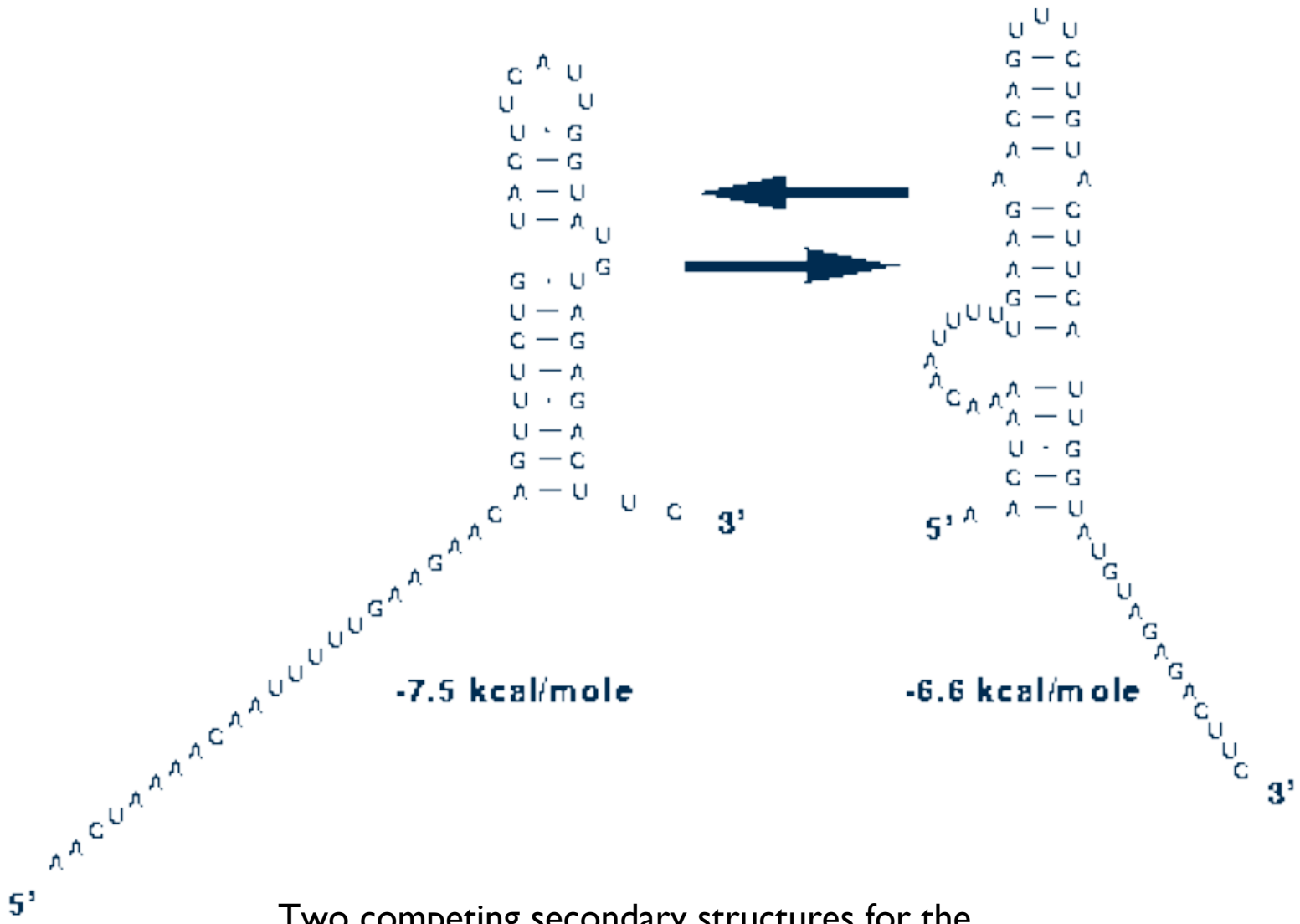
- VM(i,j) = min { W(i,k)+W(k+1,j) | i < k < j })
- VBI(i,j) = min { ebi(i,j,i',j') + V(i', j') |
  i < i' < j' < j & i'-i+j-j' > 2 }

Time: $O(n^4)$

$O(n^3)$ possible if ebi(.) is "nice"

Suboptimal Energy

- There are always alternate folds with near-optimal energies. Thermodynamics predicts that populations of identical molecules will exist in different folds; individual molecules even flicker among different folds
- Zuker's algorithm can be modified to find suboptimal folds
- McCaskill gives a more elaborate dynamic programming algorithm calculating the "partition function," which defines the probability distribution over all these states.



Two competing secondary structures for the Leptomonas collosoma spliced leader mRNA.

Example of suboptimal folding

Black dots:
pairs in opt fold

Colored dots:
pairs in folds
2-5% worse
than
optimal fold