Constraint Programming and Biology: RNA secondary structure

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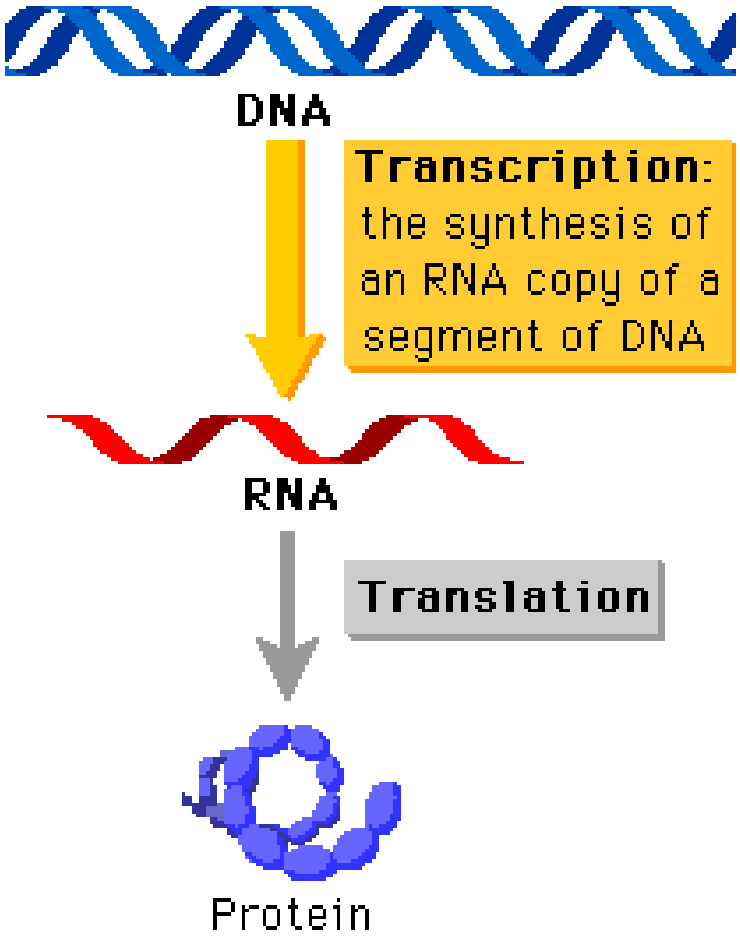
The central dogma



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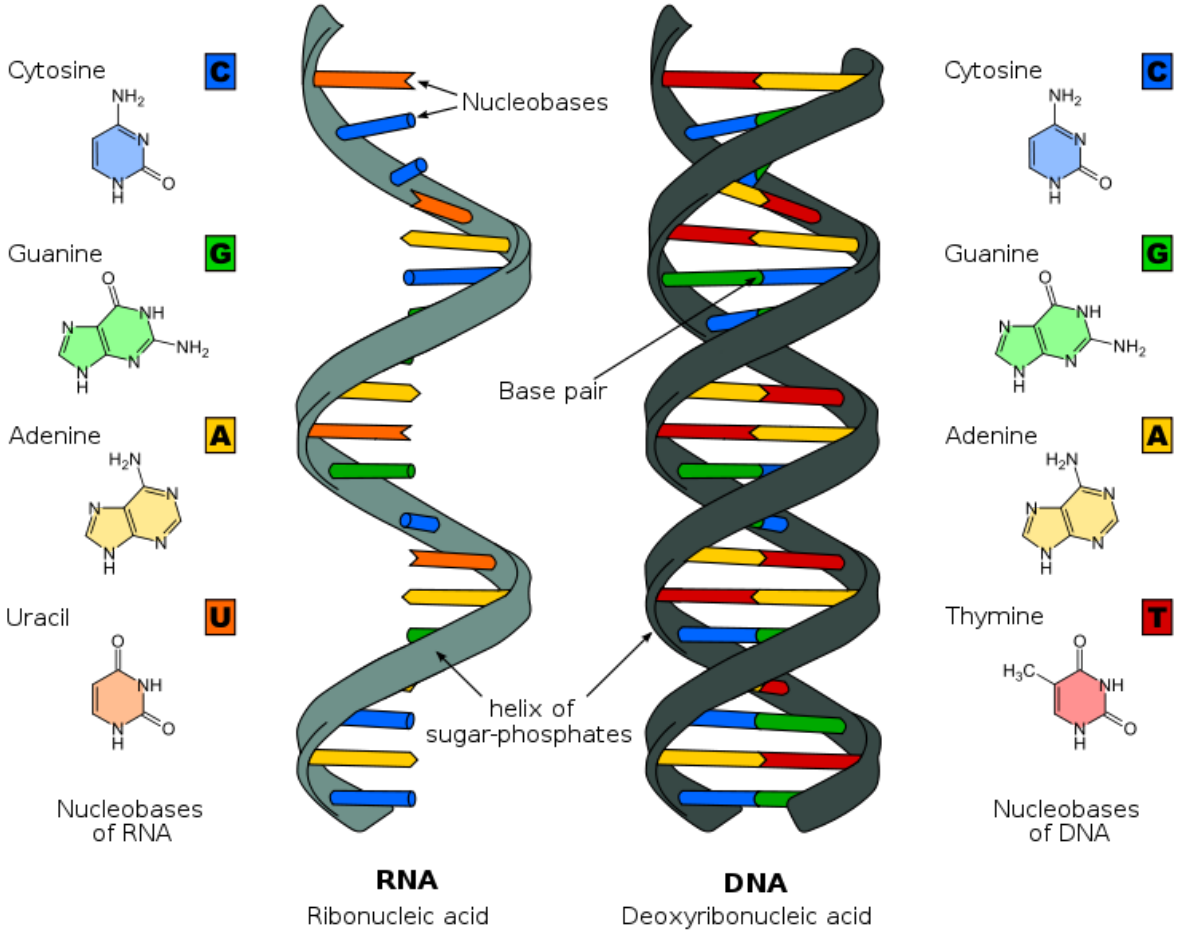

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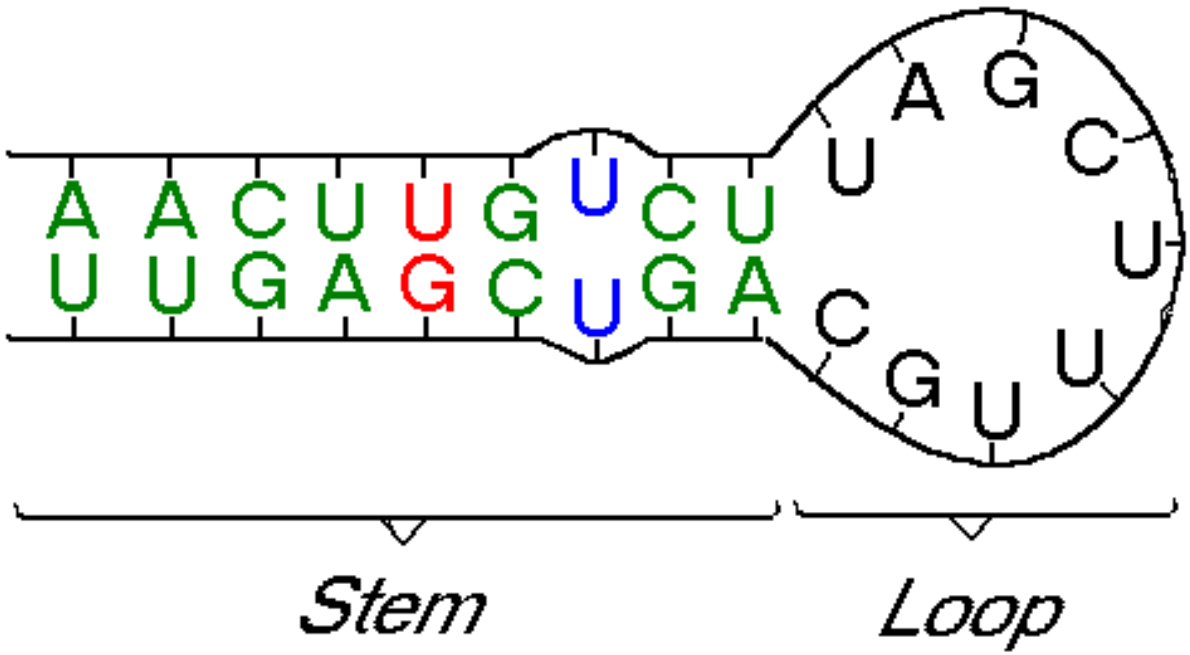

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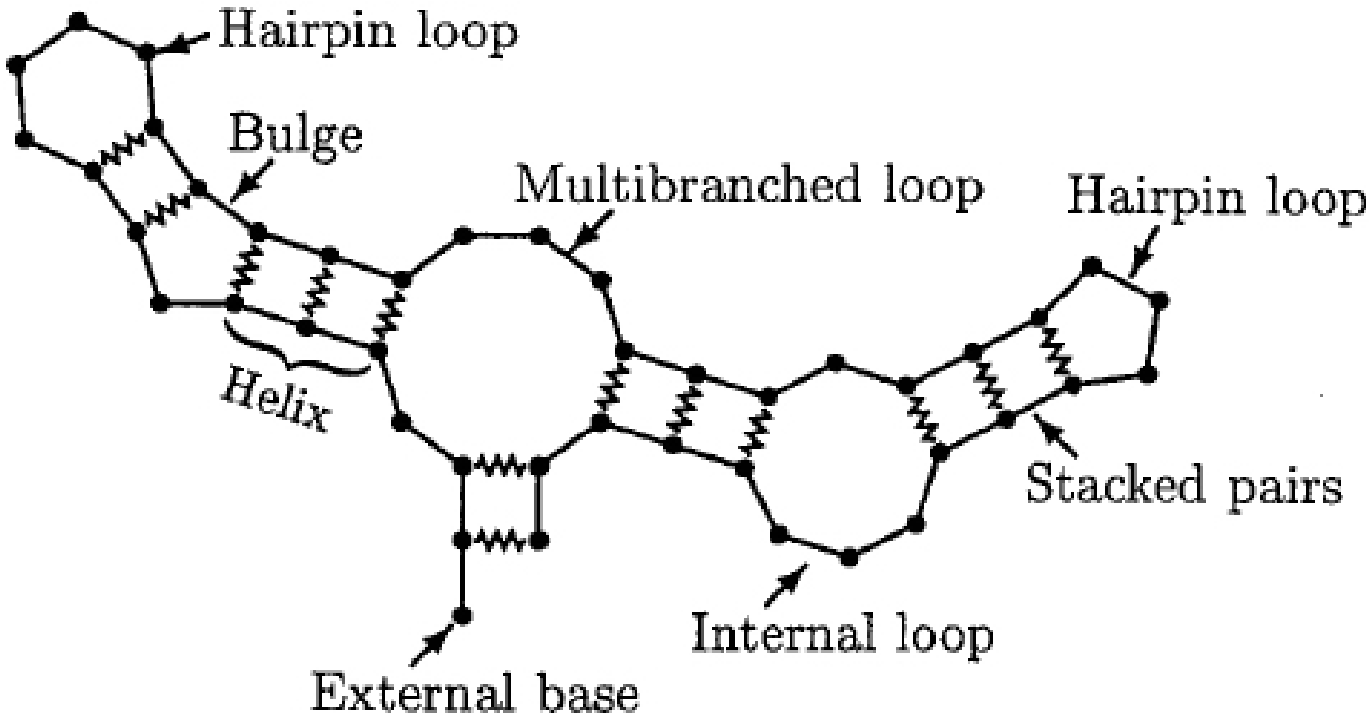

The central dogma

- RNA is a sequence of nucleotides (A,C,G,U) that (often) is just an intermediary between DNA and proteins
- The 3D structure of RNA depends largely on interactions between pairs of nucleotides (base pairing)
- The secondary structure is the set of its base pairings

Mathematically

- A RNA sequence $\vec{s} = s_1 s_2 \cdots s_n$ is a string in $\{A, C, G, U\}^*$
- A RNA secondary structure is a (partial) injective function $P \subseteq \{1, \ldots, n\}^2$ such that $(i, j) \in P \rightarrow i < j$

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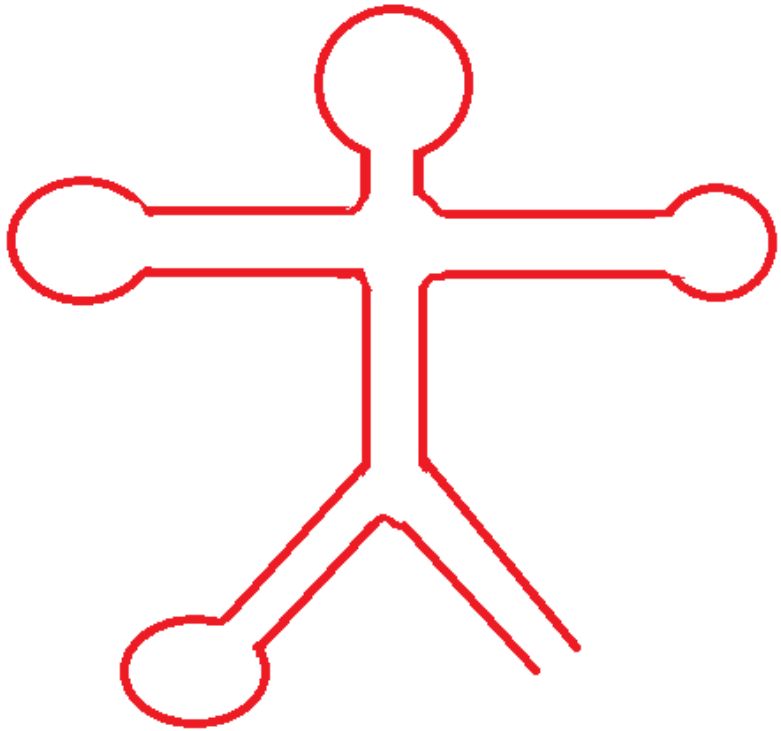
- We are interested in a pairing maximizing the pairings (and/or minimizing a more difficult energy function)

Spatial constraints (pseudo knot)



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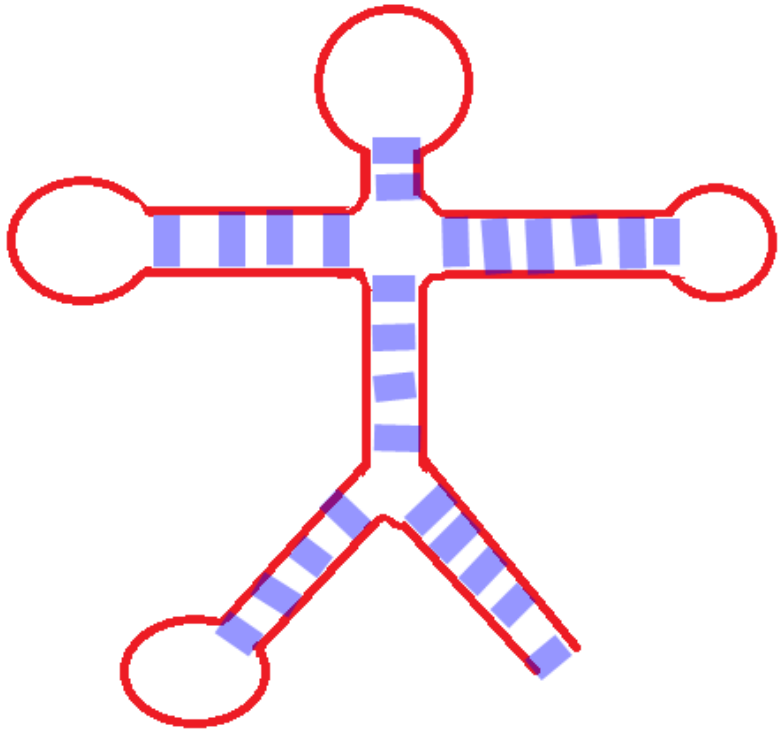

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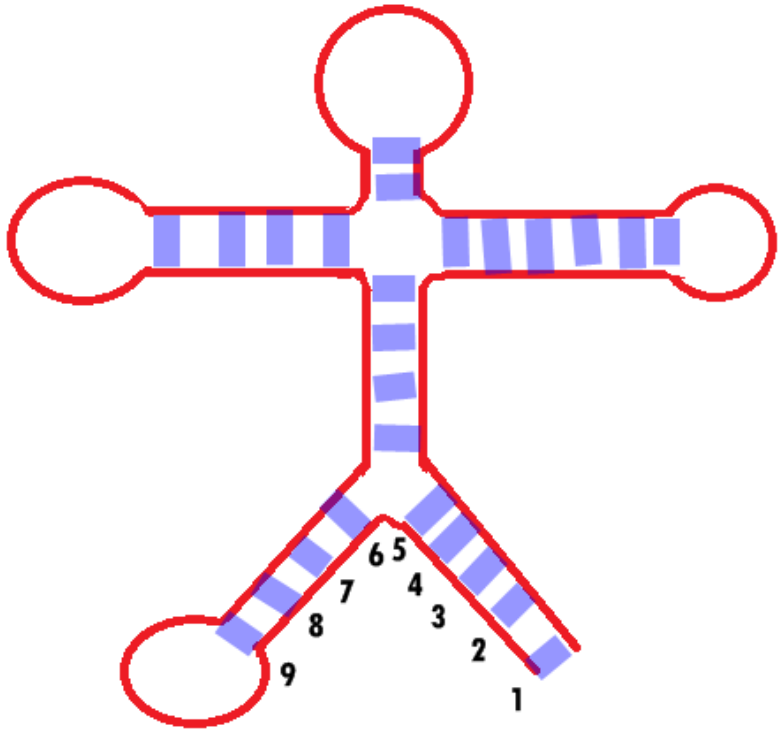

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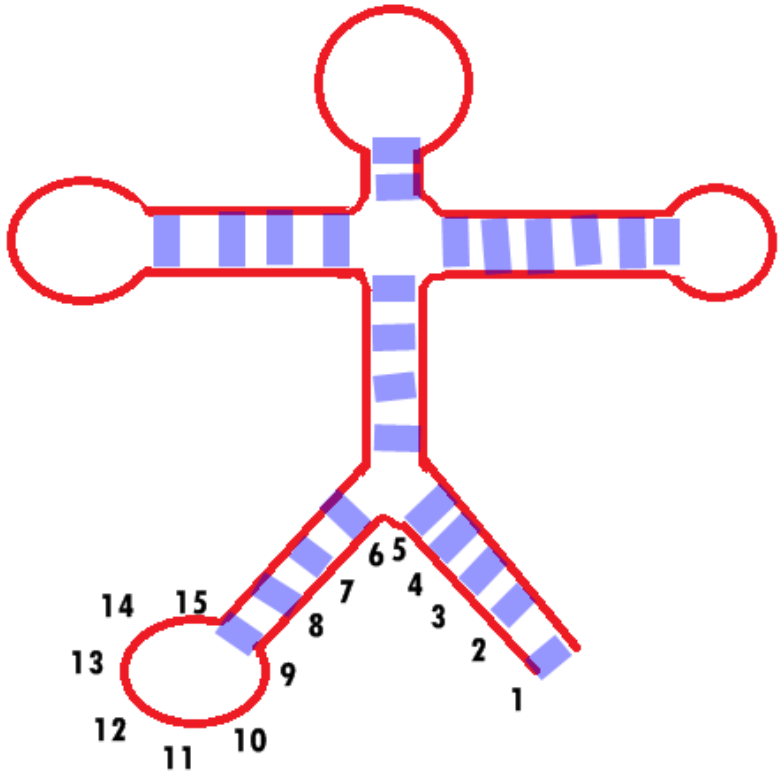

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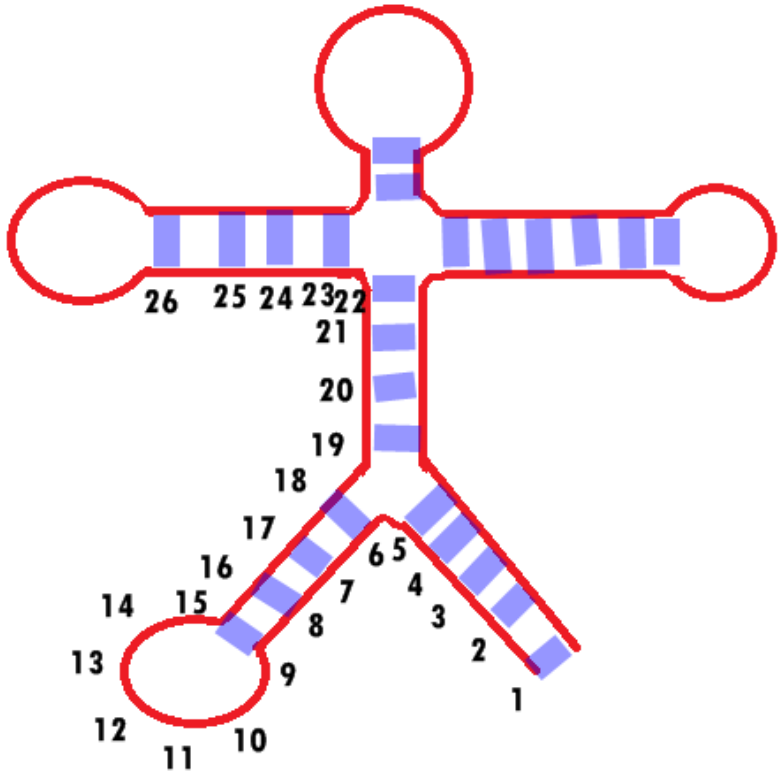

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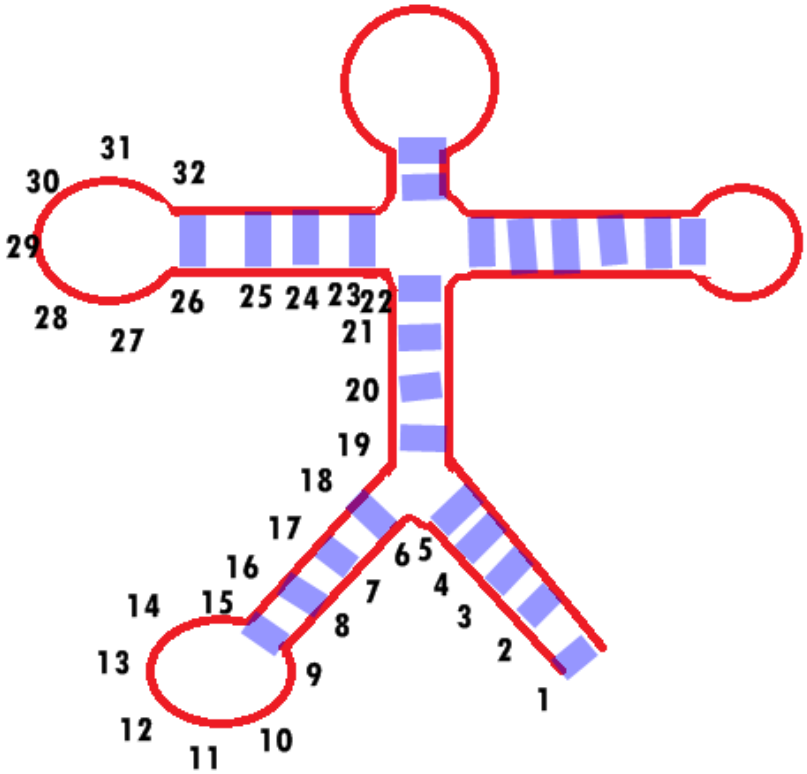

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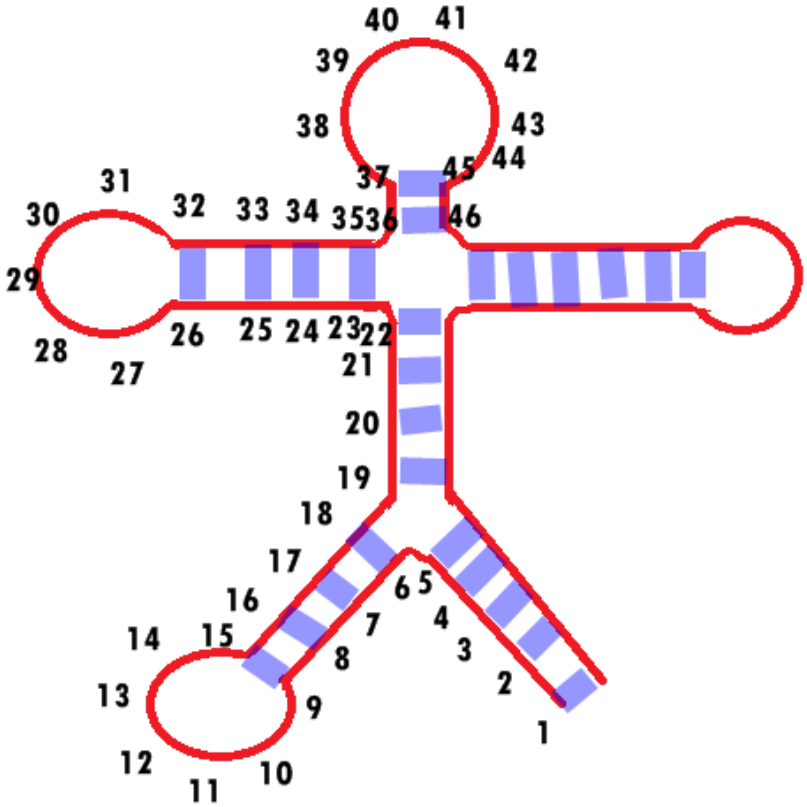

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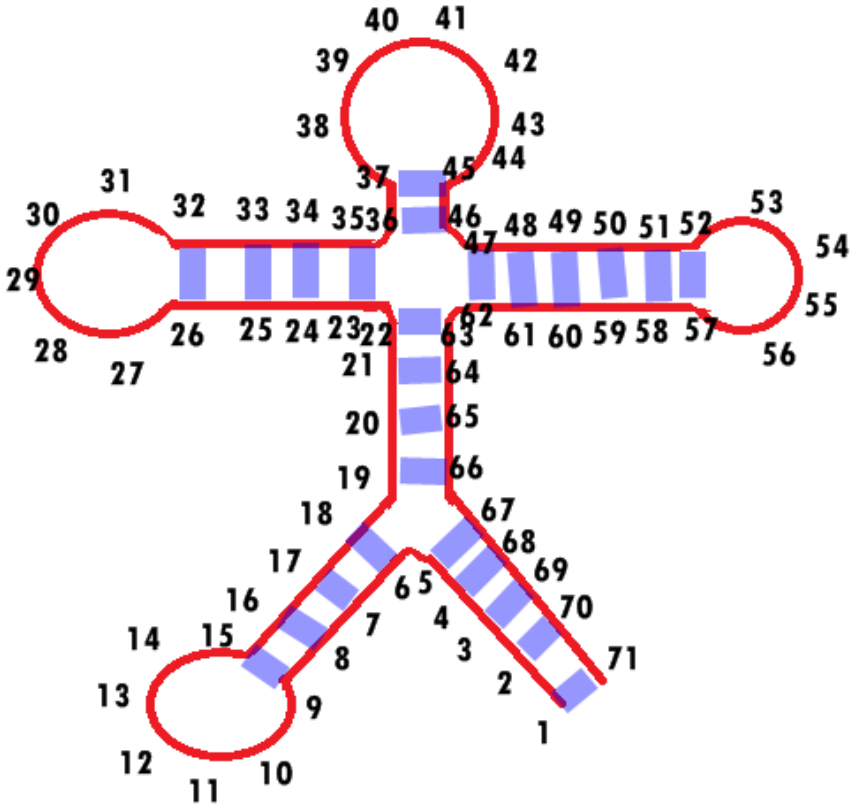

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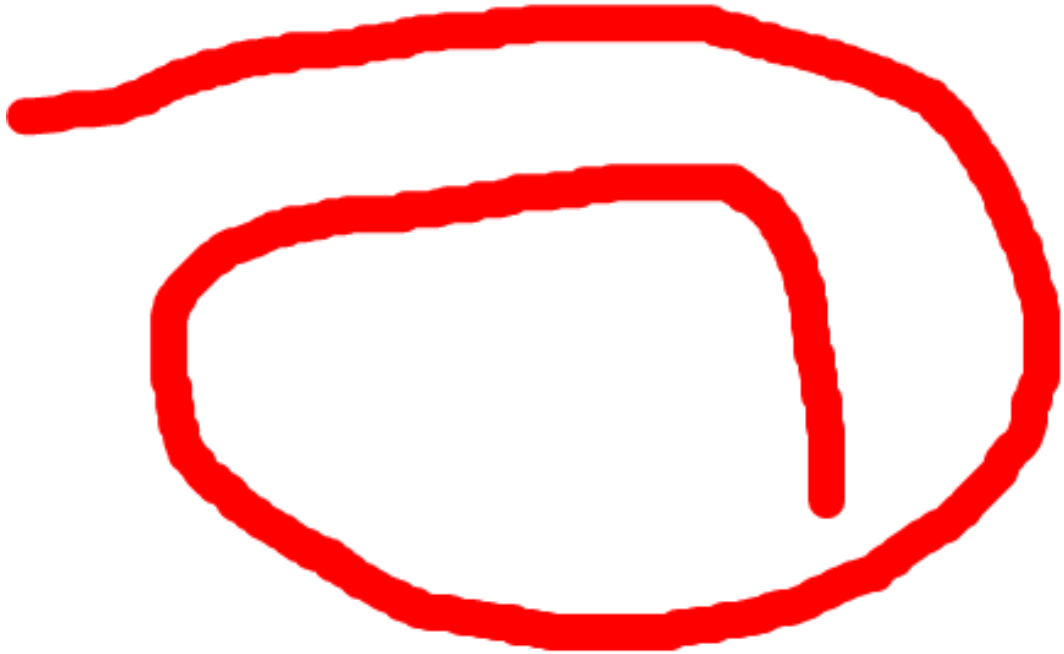

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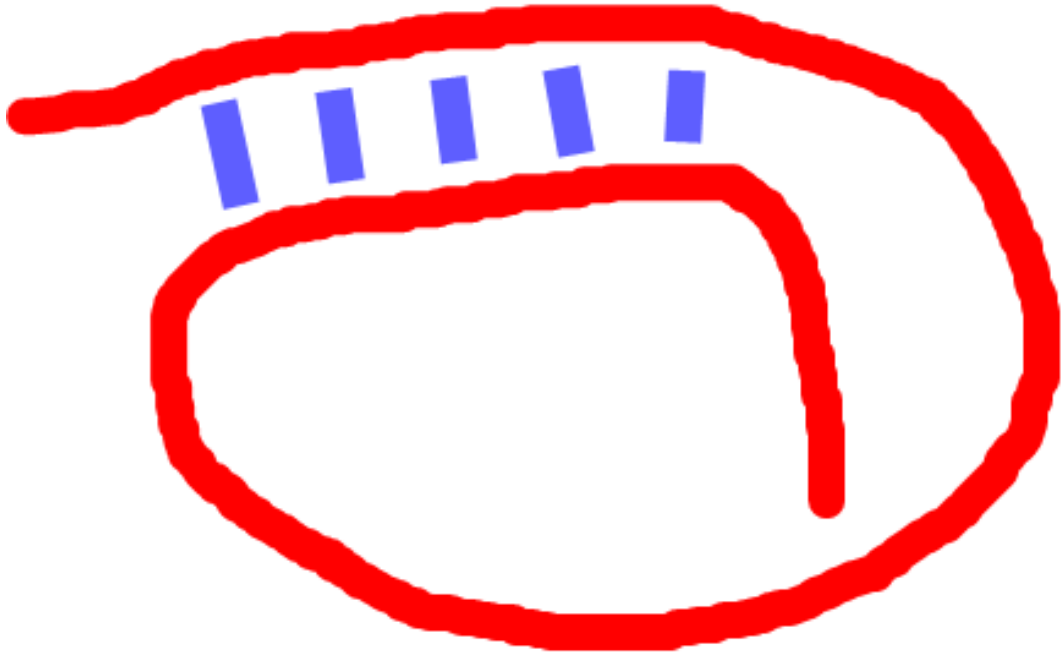

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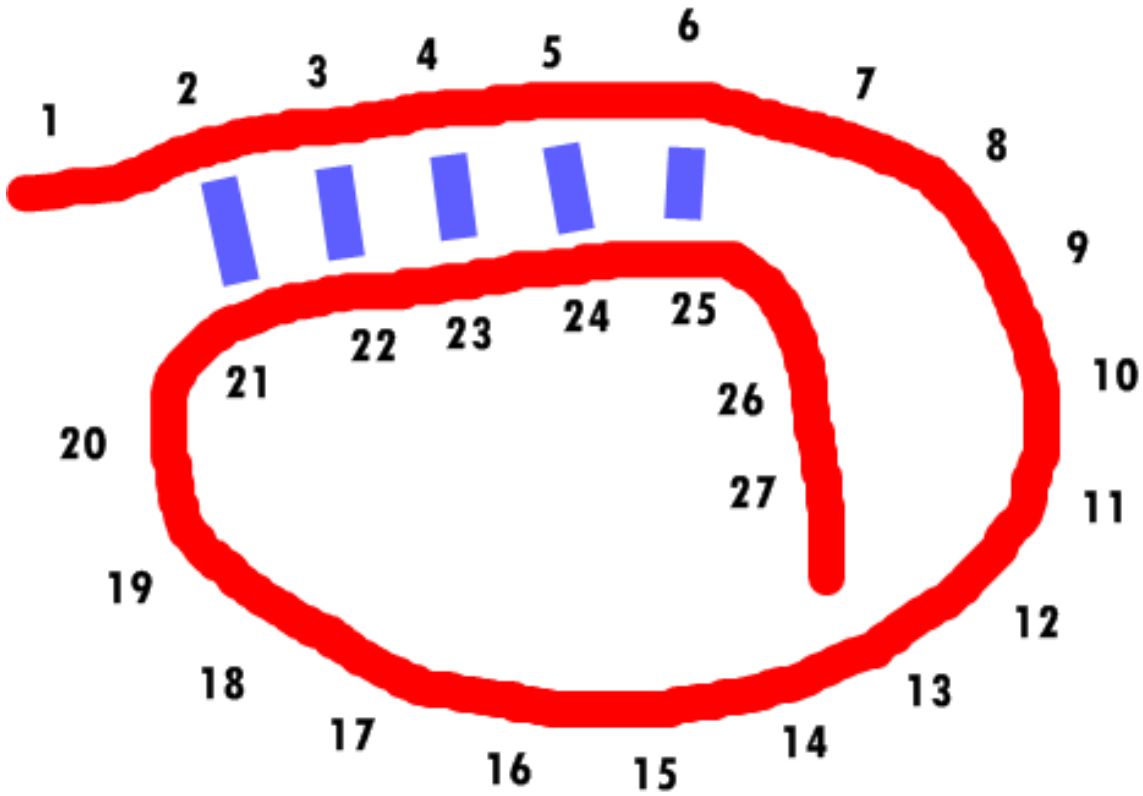

If $i < \ell < j$ and $(i, j) \in P$, and $((\ell, k) \in P$ or $(k, \ell) \in P)$, then $i < k < j$.
NO!

Results

- The pseudo-knot constraint is sensible. Adding it there are polynomial-time algorithms (`mfold`: dynamic programming. `http://mfold.rna.albany.edu`)
- Without the pseudo-knot constraint the problem is NP complete.

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- The pseudo-knot constraint is sensible. Adding it there are polynomial-time algorithms (`mfold`: dynamic programming. `http://mfold.rna.albany.edu`)
- Without the pseudo-knot constraint the problem is NP complete.
- Actually, what problem?

NP Completeness

Lyngsø and Pedersen, 2000

- Let $\vec{s} = s_1 \cdots s_n$ be a RNA sequence, and P a secondary structure. Then

$$E(\vec{s}, P) = \sum_{(i,j) \in P, i<j} E(\vec{s}, i, j, P)$$

- where $E(\vec{s}, i, j, P)$ depend on s_i and s_j and, moreover, on the s_z such that $(i+1, z) \in P$ or $(j-1, z) \in P$.

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- In the NP-completeness proof they first assume to have an infinite set of complementary bases (e.g., $(A_1, U_1), (A_2, U_2), (A_3, U_3), \ldots$) and define E as follows:

$$E(\vec{s}, i, j, P) = \begin{cases} -1 & \text{If } s_i \text{ and } s_j \text{ are complementary symbols and} \\ & (\forall z \in \{1, \ldots, i-1, j+1, \ldots, n\}) \\ & (\{(i+1, z), (z, i+1), (j-1, z), (z, j-1)\} \cap P = \emptyset) \\ 0 & \text{otherwise} \end{cases}$$

$$E(\vec{s}, i, j, P) = 0$$

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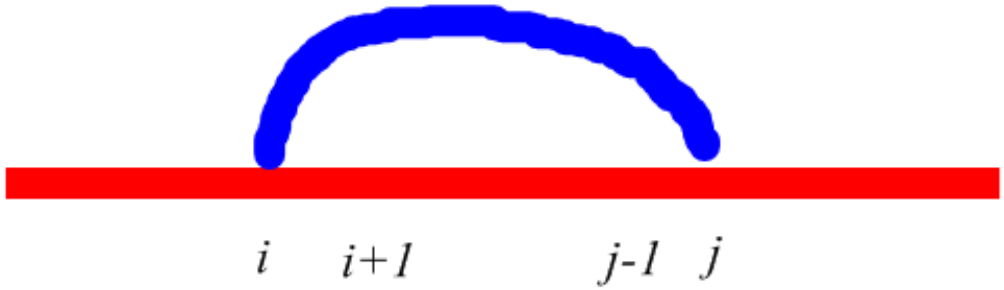

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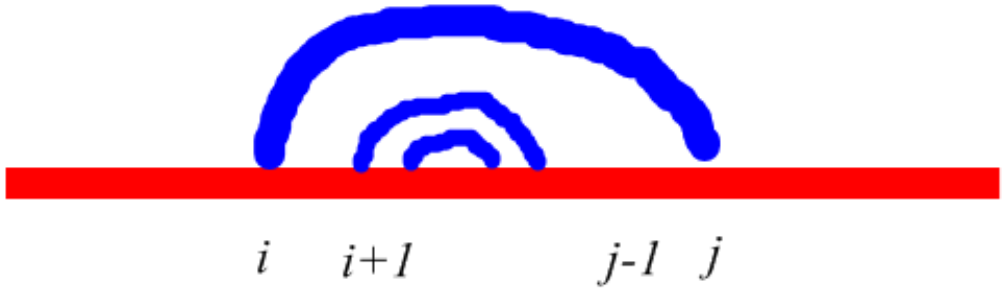

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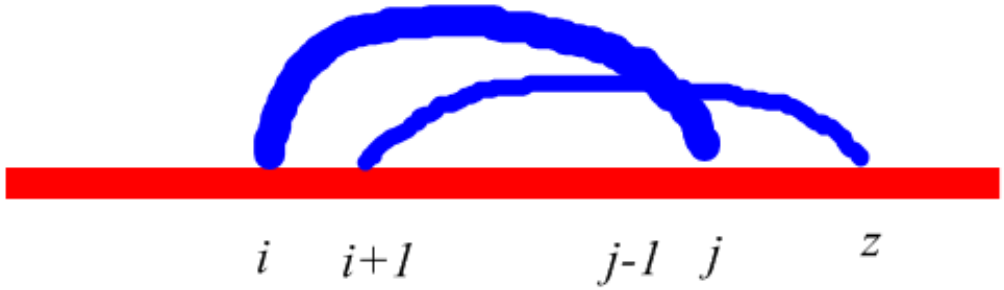

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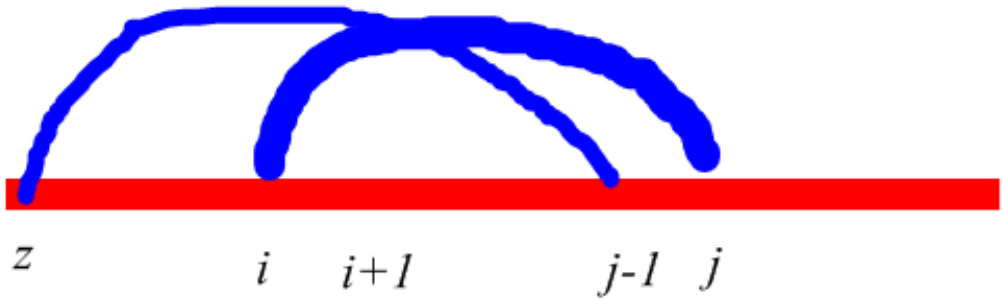

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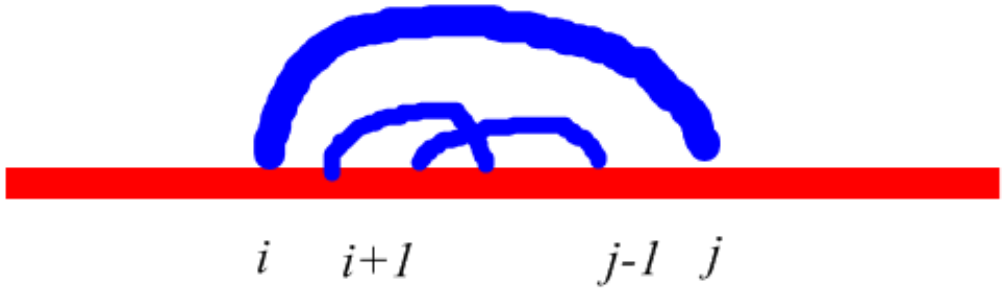

$$E(\vec{s}, i, j, P) = -1$$

NP Completeness

Lyngsø and Pedersen, 2000

- To prove NP hardness they start from 3SAT with the further requirement that each literal occurs at most twice (for a variable X, you can have X zero, one or two times and $\neg X$ zero, one, or two times). Prove it is NP complete (exercise)
- For a clause $c_i = \ell_1 \vee \ell_2 \vee \ell_3$ they introduce a gadget:

$$\mathcal{C}_i = c_{i,1}(\ell_1)_{1/2}\overline{c_{i,1}}\,c_{i,2}(\ell_2)_{1/2}c_{i,1}\overline{c_{i,2}}(\ell_3)_{1/2}c_{i,2}$$

  (1/2 according to the leftmost/rightmost occurrence of that literal)
- For a variable X_i that occurs twice positively and twice negatively, introduce a gadget (a substring in case of less occurrences)

$$\mathcal{V}_i = v_i\overline{(X_i)_2}\,\overline{(X_i)_1}\,\overline{v_i}\,v_i\overline{(\neg X_i)_2}\,\overline{(\neg X_i)_1}\,\overline{v_i}$$

- The encoding of a formula $c_1 \wedge \cdots \wedge c_m$ on variables $X_1 \ldots X_n$ is

$$\mathcal{C}_1 \cdots \mathcal{C}_m \mathcal{V}_1 \cdots \mathcal{V}_n$$

NP Completeness

Main idea

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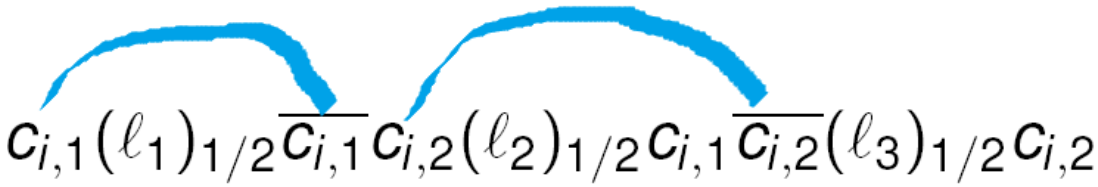

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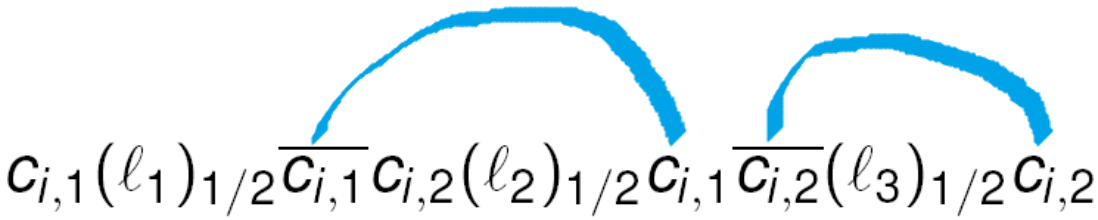

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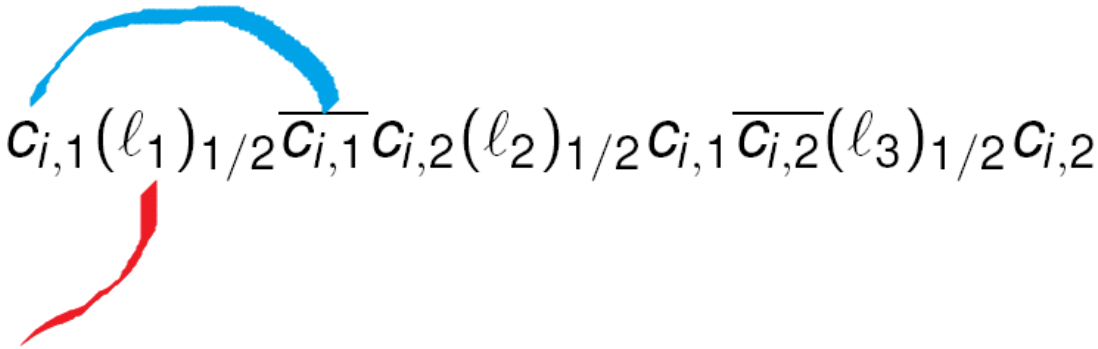

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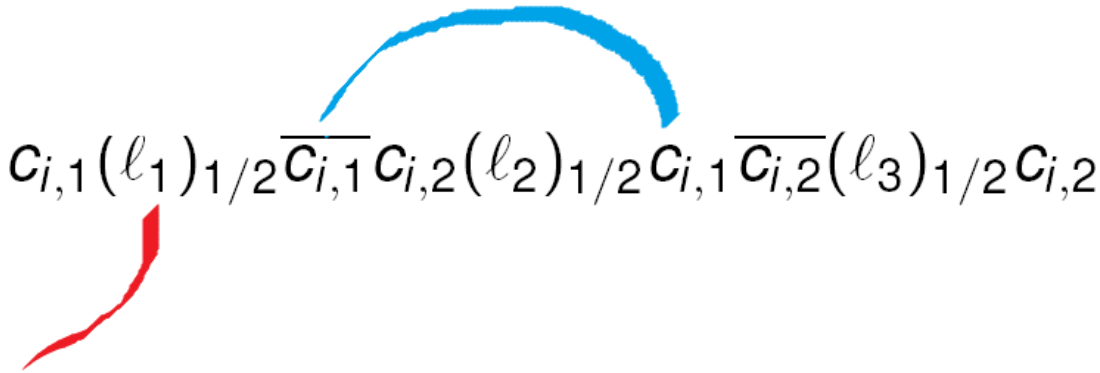

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They prove that φ is satisfiable iff there is a secondary structure with energy -(3m+n) [Nice exercise]

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Then they complete the proof without the hypothesis of infinite alphabet (this is a nice reading—too long for explaining it in this course).

A simple CLP encoding

- Input $s_1, \ldots, s_n$
- Variables $Pairs = [P_1, \ldots, P_n]$.
- Let $S_x = \{i \in \{1, \ldots, n\} \mid s_i = x\}$.
  If $s_i = A$, then $\text{dom}(P_i) = \{0\} \cup S_U$.
  If $s_i = C$, then $\text{dom}(P_i) = \{0\} \cup S_G$.
  If $s_i = G$, then $\text{dom}(P_i) = \{0\} \cup S_C \cup S_U$.
  If $s_i = U$, then $\text{dom}(P_i) = \{0\} \cup S_A \cup S_G$.
- For $i = 1, \ldots, n$, if $P_i > 0$ then $P_{P_i} = I$. If $P_i = 0$ no constraint. It can be stated compactly as:

$$element(P + 1, [I|Pairs], I)$$

- Pseudo-knots: If $P_i > 0$ then $(P_{i+1} \in [i + 3..P_{P_i} - 1]) \vee (P_{i+1} = 0)$

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- a solution close to the statistics, namely 35% for AU, 53% for CG, 12% for GU.
- Let $NC = n - \#\text{contacts}$
- We minimize therefore a weighted sum of the form

$$c_1 \frac{NC}{n} + c_2 \frac{\#(AU) - .35(n - NC)}{n} + c_3 \frac{\#(CG) - .53(n - NC)}{n}$$

(c_1, c_2, c_3 constants that can be changed. The denominator n can be omitted for minimization)

- Let us see some execution of `RNA_alignment.pl`

(Some) References

- M. Zucker and P. Stiegler. Optimal computer folding of large RNA sequences using thermodynamics and auxiliary information. Nucleid Acid Research, 9(1):133–148, 2981.
- R.B. Lyngsø and C.N.S Pedersen. RNA Pseudoknot prediction in Energy-Based Models. J. of Computational Biology 7(3/4), 2000.
- G. Blin, G. Fertin, I. Rusu, and C. Sinoquet. Extending the hardness of RNA secondary structure comparison. LNCS 4614, pp. 140–151, 2007.
- M. Bauer, G.W. Klau, and K. Reinert. Accurate multiple sequence-structure alignment of RNA sequences using combinatorial optimization. BMC Bioinformatics, 8, 2007.
- M. Bavarian and V. Dahl. Constraint Based Methods for Biological Sequence Analysis. J. Universal Computer Science 12(11):1500–1520, 2006 (also in WCB 05).
- A. Dal Palù, M. Möhl, S. Will. A Propagator for Maximum Weight String Alignment with Arbitrary Pairwise Dependencies. CP 2010: 167-175 (also in WCB 10)