Incorporating RNA-Protein Interactions into RNA Secondary Structure Prediction

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Overview

1. Introduction
2. Physical extension
3. Interaction with proteins
4. Putting it all together
5. Summary

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RNA secondary structure prediction

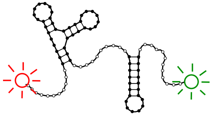
Secondary structure prediction

Prediction of RNA secondary structures in solution is mature field: Nussinov, Waterman, McCaskill, MFOLD, Dynafold, Vienna package, Sfold, . . .

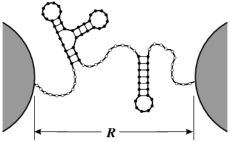
But . . .

Solution structure may not always be what we want ⇒

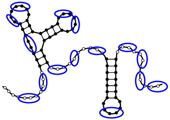
FRET



Force-extension



Protein-RNA interactions



Partition function approach

Partition function approach (McCaskill, 1990)

- Each secondary structure S is a state
- Each state S has a (free) energy

$$E[S] = \sum_{(i,j)\in S} \varepsilon_{i,j}$$

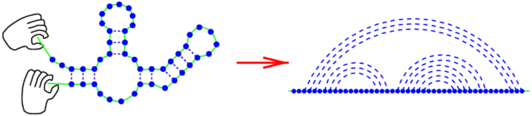
(in practice use much more detailed energy model)

- All thermodynamic quantities can be derived from partition function

$$Z = \sum_{S} \exp\left(-E[S]/k_B T\right)$$

Arc diagrams

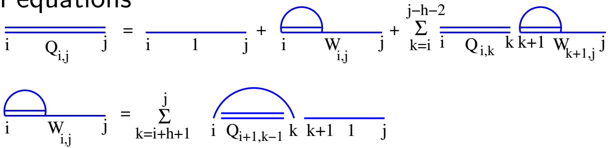
Secondary structures correspond one to one to arc diagrams



Partition functions as recursion equations

- Partition function $Q_{i,j}$  for substrand from base *i* to base *j*
- Partition function $W_{i,j}$  for substrand from base *i* to base *j* where base *i* is paired to the last paired base

⇒ Recursion equations



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    - Restricted partition function
    - Polymer Physics
    - FRET
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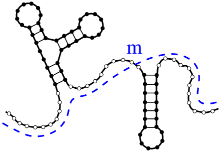
External bases

Physical extension of a given secondary structure depends only on the external bases m.



Restricted partition functions

Define $Q(m)$ to be the partition function over all structures with fixed number of external bases m.

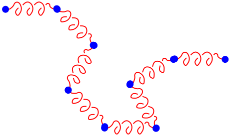
- Can be calulated by similar recursion equation for $Q_{1,j}(m)$
- $Z = \sum_{m=\Delta}^{N} Q(m)$
- $P(m) = Q(m)/Z$ is the probability to observe m external bases

U. Gerland, RB, and T. Hwa, Biophys J., 2001

Polymer Physics

Backbone model

- For given m backbone is a single-stranded RNA molecule
- Well described by extensible freely jointed chain
- Distribution $W(R|m)$ of end-to-end-distance R well known



Including secondary structure

End-to-end distance distribution

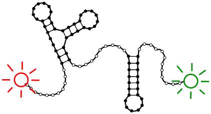
$$P(R) = \sum_{m=\Delta}^{N} P(m) W(R|m)$$

Force-distance relationship

$$F(R) = -k_B T \frac{\partial}{\partial R} \log \sum_{m=\Delta}^{N} Q(m) W(R|m)$$

FRET

Problem



- Energy transfer proportional to $(R_0/R)^6$
- Backbone moves faster than FRET time scale

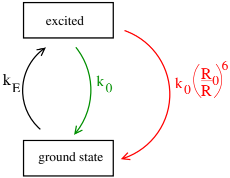
$\Rightarrow$ Energy transfer is not given by $(R_0/\langle R\rangle)^6$

Solution

- Monte-Carlo simulation of polymer dynamics

  Murphy et al., Biophys. J. 2004
- Provides $P(E_{FRET}|m)$
- Total expected FRET-distribution

$$P(E_{FRET}) = \sum_{m=\Delta}^{N} P(E_{FRET}|m)P(m)$$



excited

ground state

k_E

k_0

$k_0\left(\frac{R_0}{R}\right)^6$

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    - Protein on single-stranded RNA
    - Including secondary structure
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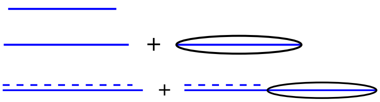
Protein on single-stranded RNA

Main quantity

Define $O_{i,j}$ to be the partition function of the single-stranded RNA from base i to base j in the presence of a single-stranded binding protein at concentration C with footprint F and dissociation constant K_D.

Calculation

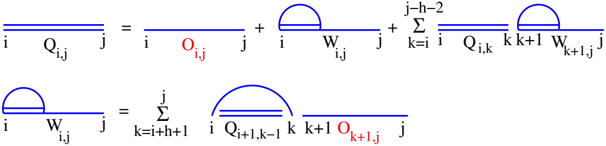


- $O_{i,j} = 1$ if $j - i + 1 < F$
- $O_{i,i+F-1} = 1 + C/K_D$
- $O_{i,j} = O_{i,j-1} + C/K_D O_{i,j-F}$
- Can include sequence dependence and cooperativity

RNA secondary structure with protein interactions

Secondary structure recursion

Protein-RNA interaction can be incorporated into secondary structure recursions by replacing 1 by $O_{i,j}$



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    - Base pairing and protein binding
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FRET prediction

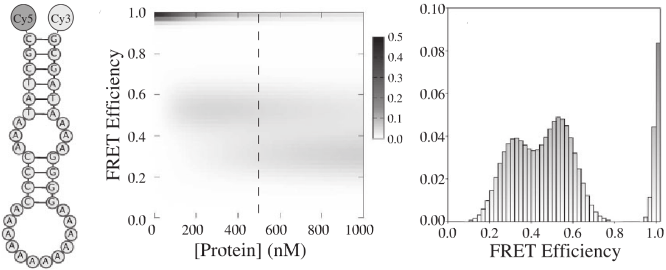
Implementation

- Incorporate everything into Vienna package

  Hofacker et al., Monatsh. Chemie, 1994
- Uses full nearest neighbor model and Turner parameters

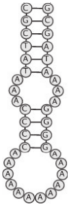
Example

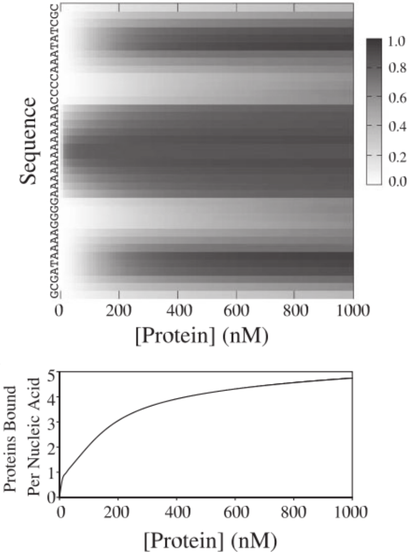
Protein with binding energy of 10kcal/mol at 1M and 6nt footprint



Other observables

- Can extend ability to calculate pair probabilities to case with proteins
- Can calculate protein occupancy of every base
- Can calculate number of bound proteins by taking numerical derivative





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Conclusions

- Physical end-to-end distance can be incorporated into RNA secondary structure prediction
- Protein-RNA interactions can be included in RNA secondary structure

Future work

- Apply approach to extract mechanistic parameters from experiments
- Double-stranded RNA-binding proteins
- RNA kinetics in the presence of proteins

Acknowledgements

Bob Forties

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