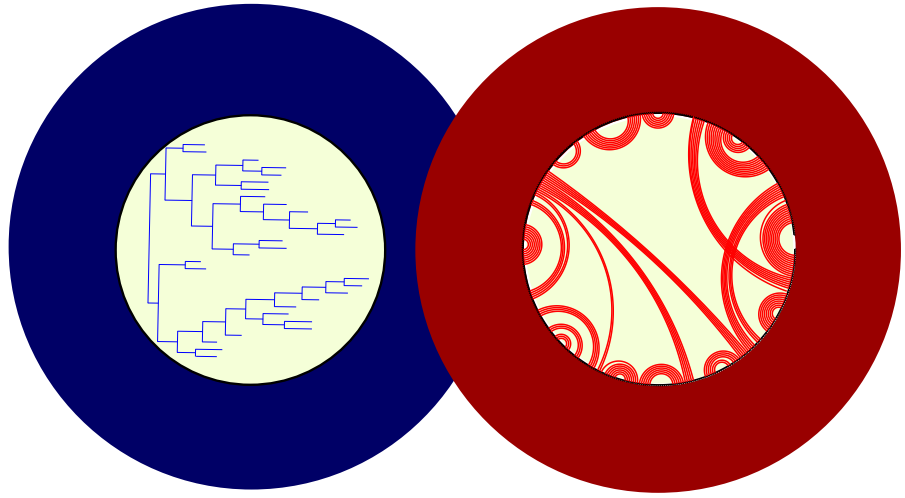
Isaac Newton Institute, 21^{st}, December 2007

A Phylogenetic Definition of Structure

Accurate Background Models for Comparative Genomic Screens



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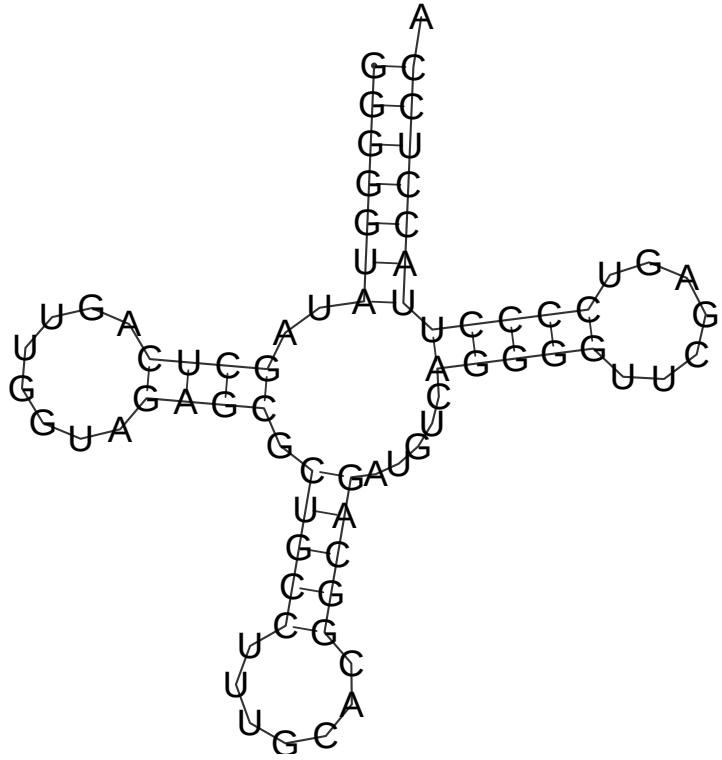
Center for Integrative Bioinformatics Vienna (CIBIV),
Max F. Perutz Laboratories (MFPL), Vienna, Austria

RNA Structure

Tertiary Structure

Secondary Structure

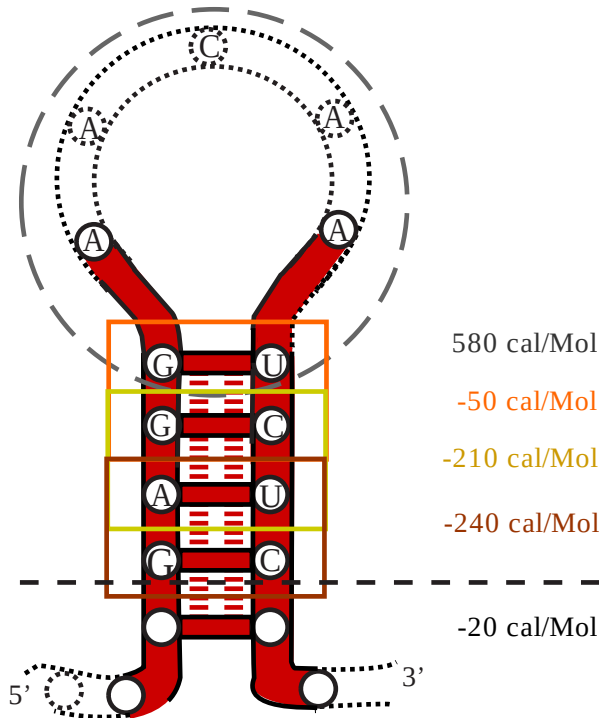


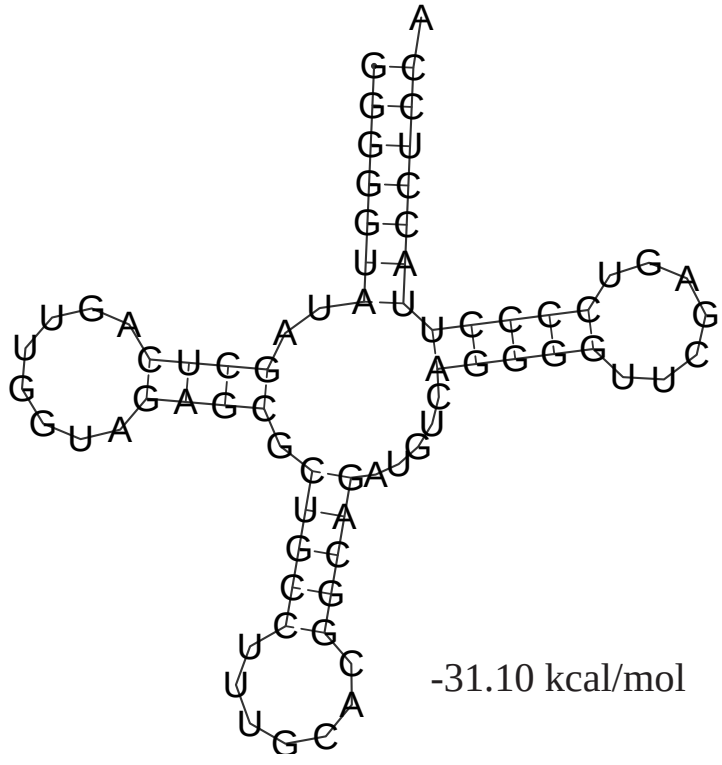
Primary Structure

GGGGGUAUAGCUCAGUUGGUAGAGCGCUGCCUUUGCACGGCAGAUGUCAGGGGUUCGAGUCCCCUUACCUCCA

RNA Structure

Minimum Free Energy Secondary Structure





The standard energy model expresses the free energy of a secondary structure $\mathcal{S}$ as the sum of the energies of its components L:

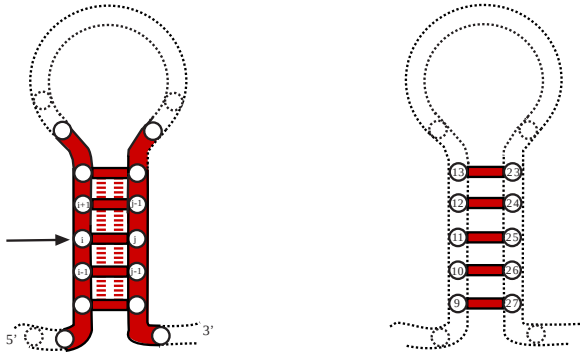
$$E(\mathcal{S}) = \sum_{L \in \mathcal{S}} E(L)$$

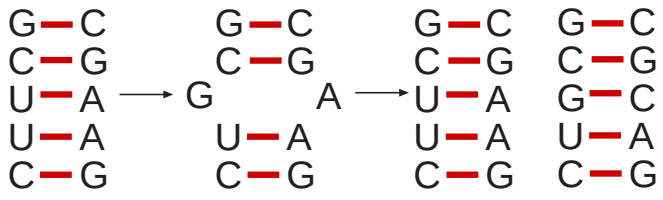
RNA Structure

Consensus Structure

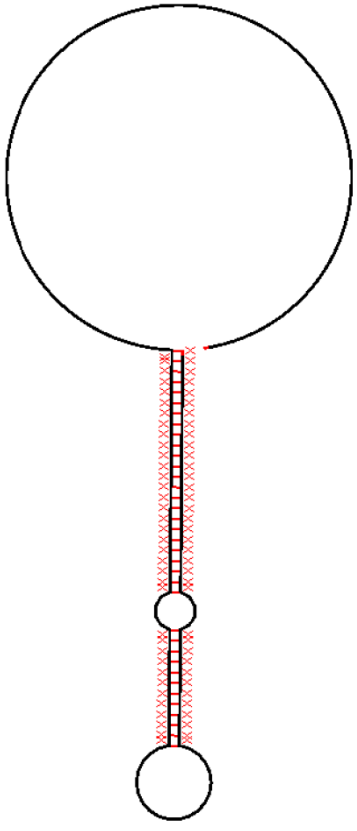
```
(((((((..(((..........))).(((((.......)))))).....(((((.......)))))))))).
GTTTCCGTAGTGTAGCGGTTATCACATTCGCCTCACACGCGAAAGGTCCCCGGTTCGATCCCGGGCGGAAACA
GTTTCCGTAGTGTAGTGGTTATCACGTTCGCCTAACACGCGAAAGGTCCCCGGTTCGAAACCGGGCGGAAACA
GTTTTCGTAGTGTAGTGGTTATCACGTGTGCTTCACACGCACAAGGTCCCCGGTTCGAACCCGGGCGAAAACA
```

1) Energy contributions of the single sequences are averaged
2) Covariance information (e.g. compensatory mutations)

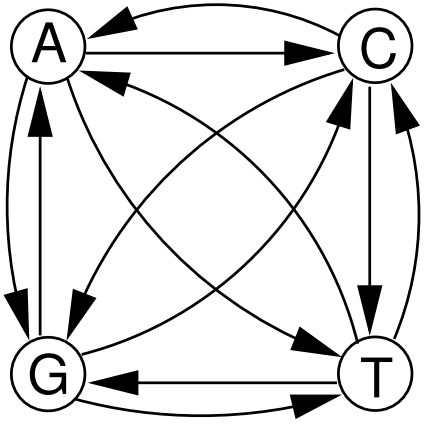




A Phylogenetic Definition of Structure

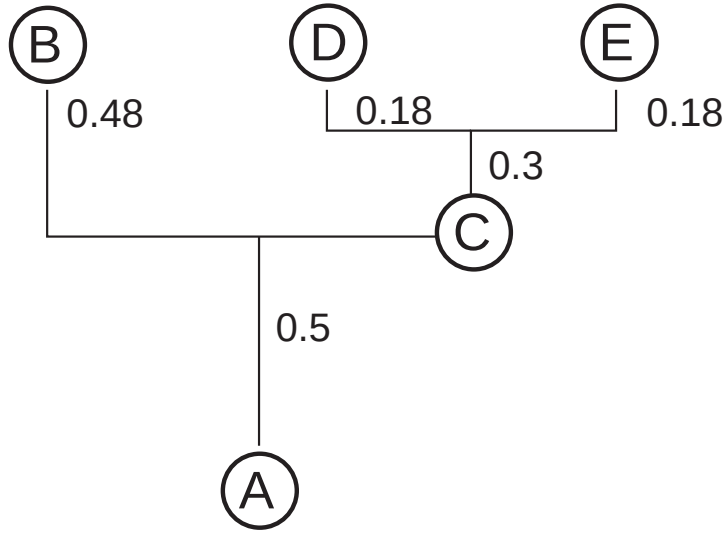


$\mathcal{N}$eighbourhood System



$\mathbf{P}(t) = \exp(\mathbf{Q}t)$

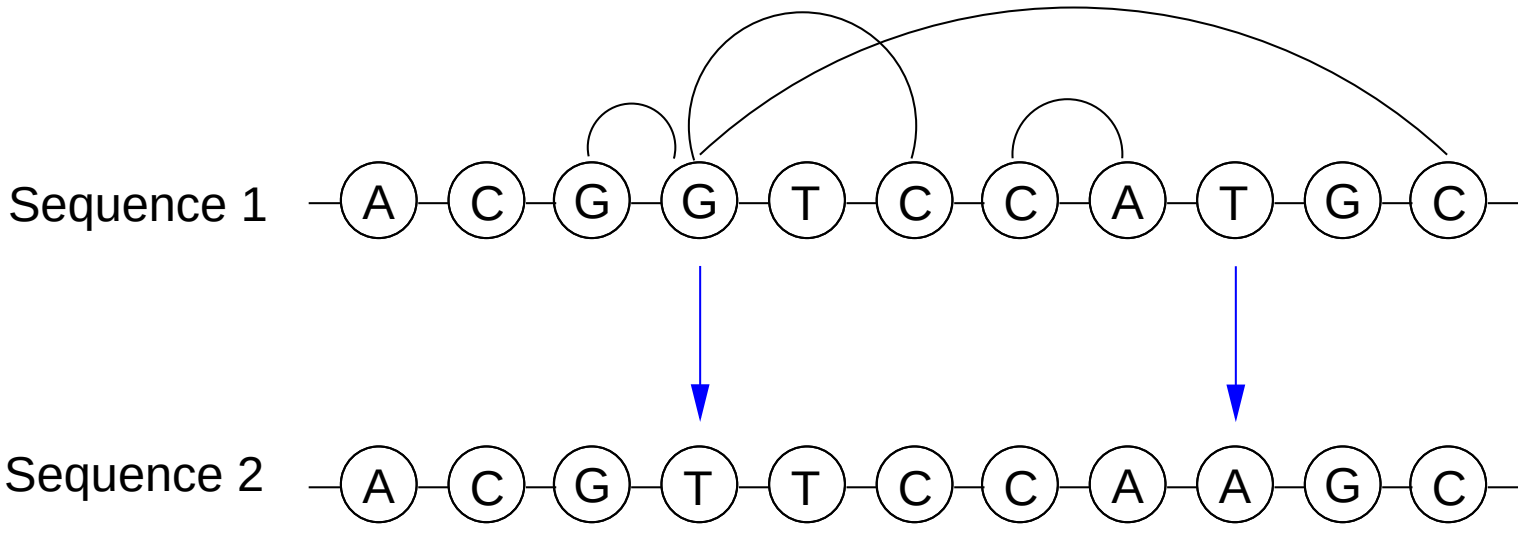
Model



Phylogenetic $\mathcal{T}$ree

Neighbourhood system

$k = 1, \cdots, l$ sites in a (nucleotide) sequence $\mathbf{x} = (x_1, \ldots, x_l)$

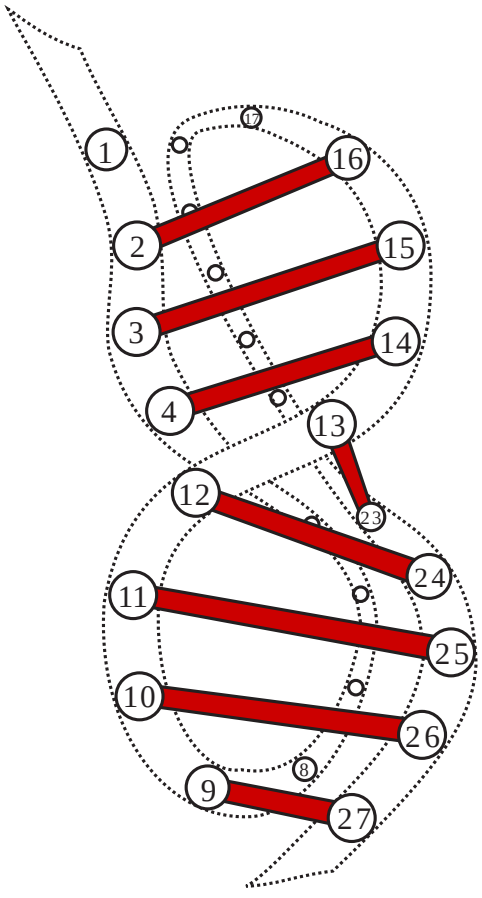


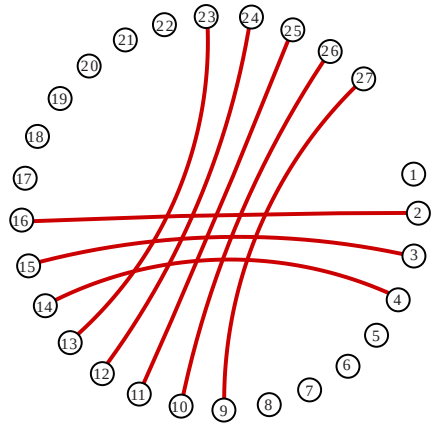
Neighbourhood system $\mathcal{N} = (N_k)_{k=1,2,\cdots,l}$:

1. $N_k \subset \{1, \ldots, l\}, k \notin N_k$ for each k
2. If $i \in N_k$ then $k \in N_i$ for each i, k.

n_k denotes the cardinality of N_k.

Example: $\mathcal{N}$(Pseudoknot)





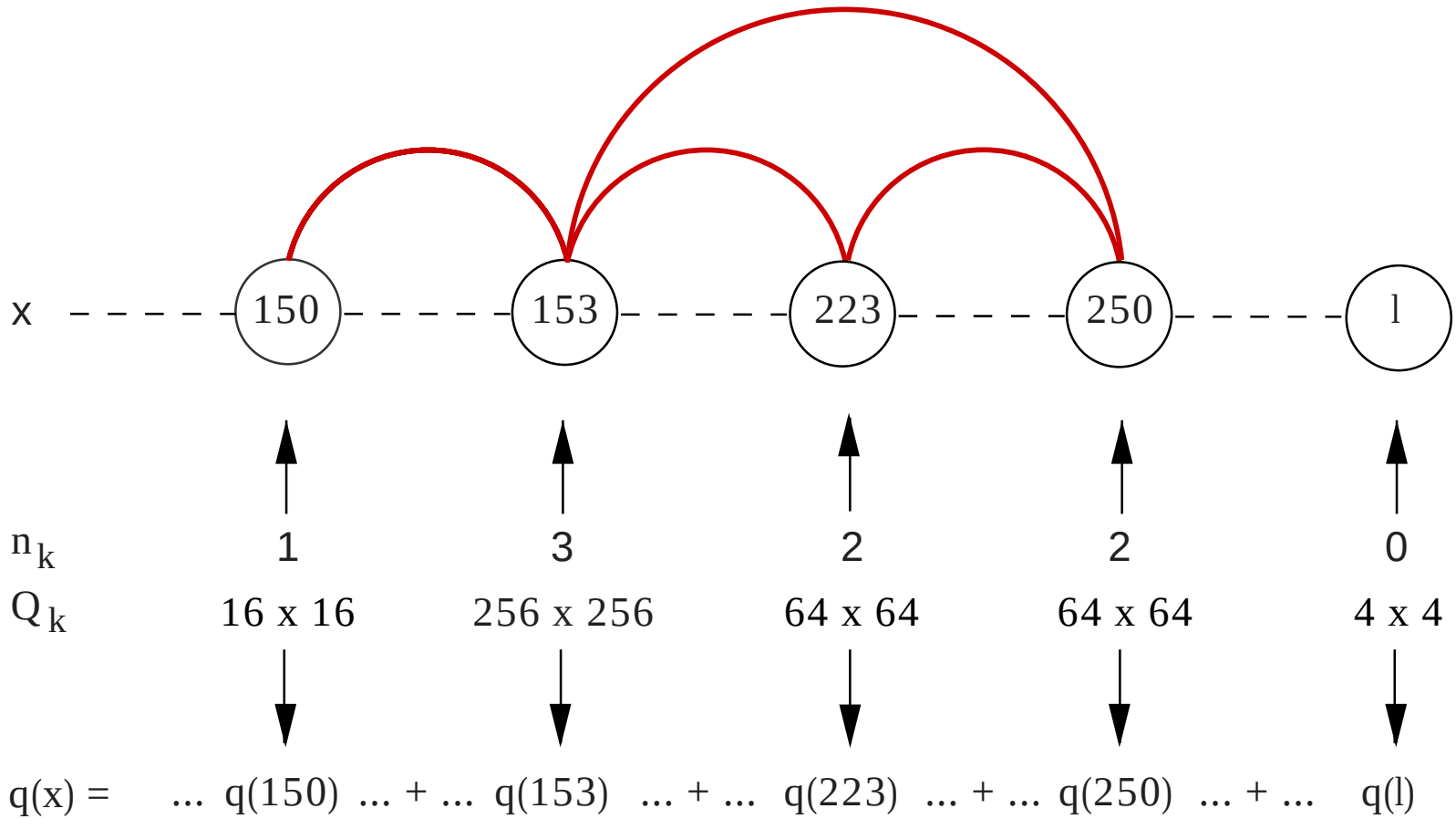
$N_1 = \{\}$
$N_2 = \{16\}$
$N_3 = \{15\}$
$N_4 = \{14\}$
$N_5 = \{\}$
$N_6 = \{\}$
$N_7 = \{\}$
$N_8 = \{\}$
$N_9 = \{27\}$

$N_{10} = \{26\}$
$N_{11} = \{25\}$
$N_{12} = \{24\}$
$N_{13} = \{23\}$
$N_{14} = \{4\}$
$N_{15} = \{3\}$
$N_{16} = \{2\}$
$N_{17} = \{\}$
$N_{18} = \{\}$

$N_{19} = \{\}$
$N_{20} = \{\}$
$N_{21} = \{\}$
$N_{22} = \{\}$
$N_{23} = \{13\}$
$N_{24} = \{12\}$
$N_{25} = \{11\}$
$N_{26} = \{10\}$
$N_{27} = \{9\}$

A model, that represents a universal description of arbitrary complex dependencies among sites.

Modeling Sequence Evolution with Arbitrary Dependencies



Basic idea: Different substitution matrix for each site

$\rightarrow$ only one mutation is allowed at the current site.

$n_k = 1$

	AA	AC	AG	AU	CA	CC	CG	CU	GA	GC	GG	GU	UA	UC	UG	UU
AA	$*$	-	-	-	π_{CA}	-	-	-	π_{GA}	-	-	-	π_{UA}	-	-	-
AC	-	$*$	-	-	-	π_{CC}	-	-	-	π_{GC}	-	-	-	π_{UC}	-	-
AG	-	-	$*$	-	-	-	π_{CG}	-	-	-	π_{GG}	-	-	-	π_{UG}	-
AU	-	-	-	$*$	-	-	-	π_{CU}	-	-	-	π_{GU}	-	-	-	π_{UU}
CA	π_{AA}	-	-	-	$*$	-	-	-	π_{GA}	-	-	-	π_{UA}	-	-	-
CC	-	π_{AC}	-	-	-	$*$	-	-	-	π_{GC}	-	-	-	π_{UC}	-	-
CG	-	-	π_{AG}	-	-	-	$*$	-	-	-	π_{GG}	-	-	-	π_{UG}	-
CU	-	-	-	π_{AU}	-	-	-	$*$	-	-	-	π_{GU}	-	-	-	π_{UU}
GA	π_{AA}	-	-	-	π_{CA}	-	-	-	$*$	-	-	-	π_{UA}	-	-	-
GC	-	π_{AC}	-	-	-	π_{CC}	-	-	-	$*$	-	-	-	π_{UC}	-	-
GG	-	-	π_{AG}	-	-	-	π_{CG}	-	-	-	$*$	-	-	-	π_{UG}	-
GU	-	-	-	π_{AU}	-	-	-	π_{CU}	-	-	-	$*$	-	-	-	π_{UU}
UA	π_{AA}	-	-	-	π_{CA}	-	-	-	π_{GA}	-	-	-	$*$	-	-	-
UC	-	π_{AC}	-	-	-	π_{CC}	-	-	-	π_{GC}	-	-	-	$*$	-	-
UG	-	-	π_{AG}	-	-	-	π_{CG}	-	-	-	π_{GG}	-	-	-	$*$	-
UU	-	-	-	π_{AU}	-	-	-	π_{CU}	-	-	-	π_{GU}	-	-	-	$*$

$n_k = 1$

(k,i)	AA	CA	GA	UA	AC	CC	GC	UC	AG	CG	GG	UG	AU	CU	GU	UU
AA	$*$	π_{CA}	π_{GA}	π_{UA}	–	–	–	–	–	–	–	–	–	–	–	–
CA	π_{AA}	$*$	π_{GA}	π_{UA}	–	–	–	–	–	–	–	–	–	–	–	–
GA	π_{AA}	π_{CA}	$*$	π_{UA}	–	–	–	–	–	–	–	–	–	–	–	–
UA	π_{AA}	π_{CA}	π_{GA}	$*$	–	–	–	–	–	–	–	–	–	–	–	–
AC	–	–	–	–	$*$	π_{CC}	π_{GC}	π_{UC}	–	–	–	–	–	–	–	–
CC	–	–	–	–	π_{AC}	$*$	π_{GC}	π_{UC}	–	–	–	–	–	–	–	–
GC	–	–	–	–	π_{AC}	π_{CC}	$*$	π_{UC}	–	–	–	–	–	–	–	–
UC	–	–	–	–	π_{AC}	π_{CC}	π_{GC}	$*$	–	–	–	–	–	–	–	–
AG	–	–	–	–	–	–	–	–	$*$	π_{CG}	π_{GG}	π_{UG}	–	–	–	–
CG	–	–	–	–	–	–	–	–	π_{AG}	$*$	π_{GG}	π_{UG}	–	–	–	–
GG	–	–	–	–	–	–	–	–	π_{AG}	π_{CG}	$*$	π_{UG}	–	–	–	–
UG	–	–	–	–	–	–	–	–	π_{AG}	π_{CG}	π_{GG}	$*$	–	–	–	–
AU	–	–	–	–	–	–	–	–	–	–	–	–	$*$	π_{CU}	π_{GU}	π_{UU}
CU	–	–	–	–	–	–	–	–	–	–	–	–	π_{AU}	$*$	π_{GU}	π_{UU}
GU	–	–	–	–	–	–	–	–	–	–	–	–	π_{AU}	π_{CU}	$*$	π_{UU}
UU	–	–	–	–	–	–	–	–	–	–	–	–	π_{AU}	π_{CU}	π_{GU}	$*$

$n_k = 1$

$$
\begin{array}{c} \\ \mathbf{A}|A \\ \mathbf{C}|A \\ \mathbf{G}|A \\ \mathbf{U}|A \end{array}
\begin{array}{c}
\begin{array}{cccc} \mathbf{A}|A & \mathbf{C}|A & \mathbf{G}|A & \mathbf{U}|A \end{array} \\
\begin{pmatrix} * & \pi_{CA} & \pi_{GA} & \pi_{UA} \\ \pi_{AA} & * & \pi_{GA} & \pi_{UA} \\ \pi_{AA} & \pi_{CA} & * & \pi_{UA} \\ \pi_{AA} & \pi_{CA} & \pi_{GA} & * \end{pmatrix}
\end{array}
\qquad
\begin{array}{c} \\ \mathbf{A}|C \\ \mathbf{C}|C \\ \mathbf{G}|C \\ \mathbf{U}|C \end{array}
\begin{array}{c}
\begin{array}{cccc} \mathbf{A}|C & \mathbf{C}|C & \mathbf{G}|C & \mathbf{U}|C \end{array} \\
\begin{pmatrix} * & \pi_{CC} & \pi_{GC} & \pi_{UC} \\ \pi_{AC} & * & \pi_{GC} & \pi_{UC} \\ \pi_{AC} & \pi_{CC} & * & \pi_{UC} \\ \pi_{AC} & \pi_{CC} & \pi_{GC} & * \end{pmatrix}
\end{array}
$$

$$
\begin{array}{c} \\ \mathbf{A}|G \\ \mathbf{C}|G \\ \mathbf{G}|G \\ \mathbf{U}|G \end{array}
\begin{array}{c}
\begin{array}{cccc} \mathbf{A}|G & \mathbf{C}|G & \mathbf{G}|G & \mathbf{U}|G \end{array} \\
\begin{pmatrix} * & \pi_{CG} & \pi_{GG} & \pi_{UG} \\ \pi_{AG} & * & \pi_{GG} & \pi_{UG} \\ \pi_{AG} & \pi_{CG} & * & \pi_{UG} \\ \pi_{AG} & \pi_{CG} & \pi_{GG} & * \end{pmatrix}
\end{array}
\qquad
\begin{array}{c} \\ \mathbf{A}|U \\ \mathbf{C}|U \\ \mathbf{G}|U \\ \mathbf{U}|U \end{array}
\begin{array}{c}
\begin{array}{cccc} \mathbf{A}|U & \mathbf{C}|U & \mathbf{G}|U & \mathbf{U}|U \end{array} \\
\begin{pmatrix} * & \pi_{CU} & \pi_{GU} & \pi_{UU} \\ \pi_{AU} & * & \pi_{GU} & \pi_{UU} \\ \pi_{AU} & \pi_{CU} & * & \pi_{UU} \\ \pi_{AU} & \pi_{CU} & \pi_{GU} & * \end{pmatrix}
\end{array}
$$

Generally: $4^{n_k+1} \times 4^{n_k+1} \longrightarrow 4^{n_k}$ Submatrices

$$
\begin{array}{c}
 \\
\mathsf{A}|y_1,\ldots,y_{n_k} \\
\mathsf{C}|y_1,\ldots,y_{n_k} \\
\mathsf{G}|y_1,\ldots,y_{n_k} \\
\mathsf{U}|y_1,\ldots,y_{n_k}
\end{array}
\begin{array}{c}
\begin{array}{cccc}
\mathsf{A}|y_1,\ldots,y_{n_k} & \mathsf{C}|y_1,\ldots,y_{n_k} & \mathsf{G}|y_1,\ldots,y_{n_k} & \mathsf{U}|y_1,\ldots,y_{n_k}
\end{array} \\
\left(\begin{array}{cccc}
* & \pi_{\mathsf{C}|y_1,\ldots,y_{n_k}} & \pi_{\mathsf{G}|y_1,\ldots,y_{n_k}} & \pi_{\mathsf{U}|y_1,\ldots,y_{n_k}} \\
\pi_{\mathsf{A}|y_1,\ldots,y_{n_k}} & * & \pi_{\mathsf{G}|y_1,\ldots,y_{n_k}} & \pi_{\mathsf{U}|y_1,\ldots,y_{n_k}} \\
\pi_{\mathsf{A}|y_1,\ldots,y_{n_k}} & \pi_{\mathsf{C}|y_1,\ldots,y_{n_k}} & * & \pi_{\mathsf{U}|y_1,\ldots,y_{n_k}} \\
\pi_{\mathsf{A}|y_1,\ldots,y_{n_k}} & \pi_{\mathsf{C}|y_1,\ldots,y_{n_k}} & \pi_{\mathsf{G}|y_1,\ldots,y_{n_k}} & *
\end{array}\right)
\end{array}
$$

Generally: $4^{n_k+1} \times 4^{n_k+1} \longrightarrow 4^{n_k}$ Submatrices

$$
\begin{array}{c}
 \\
\mathsf{A}|y_1,\ldots,y_{n_k} \\
\mathsf{C}|y_1,\ldots,y_{n_k} \\
\mathsf{G}|y_1,\ldots,y_{n_k} \\
\mathsf{U}|y_1,\ldots,y_{n_k}
\end{array}
\begin{array}{c}
\begin{array}{cccc}
\mathsf{A}|y_1,\ldots,y_{n_k} & \mathsf{C}|y_1,\ldots,y_{n_k} & \mathsf{G}|y_1,\ldots,y_{n_k} & \mathsf{U}|y_1,\ldots,y_{n_k}
\end{array} \\
\left(\begin{array}{cccc}
* & \beta\pi_{\mathsf{C}|y_1,\ldots,y_{n_k}} & \alpha\pi_{\mathsf{G}|y_1,\ldots,y_{n_k}} & \beta\pi_{\mathsf{U}|y_1,\ldots,y_{n_k}} \\
\beta\pi_{\mathsf{A}|y_1,\ldots,y_{n_k}} & * & \beta\pi_{\mathsf{G}|y_1,\ldots,y_{n_k}} & \alpha\pi_{\mathsf{U}|y_1,\ldots,y_{n_k}} \\
\alpha\ \pi_{\mathsf{A}|y_1,\ldots,y_{n_k}} & \beta\pi_{\mathsf{C}|y_1,\ldots,y_{n_k}} & * & \beta\ \pi_{\mathsf{U}|y_1,\ldots,y_{n_k}} \\
\beta\pi_{\mathsf{A}|y_1,\ldots,y_{n_k}} & \alpha\ \pi_{\mathsf{C}|y_1,\ldots,y_{n_k}} & \beta\pi_{\mathsf{G}|y_1,\ldots,y_{n_k}} & *
\end{array}\right)
\end{array}
$$

$$Q = \{Q_k | k = 1, \ldots, l\}$$

$$Q_k^{\gamma}(\mathbf{s}_k, \mathbf{y}) = \begin{cases} \gamma_{(\mathbf{s}_k,\mathbf{y})} \cdot Q_k(\mathbf{s}_k, \mathbf{y}) & \text{if } H(\mathbf{s}_k, \mathbf{y}) = 1 \text{ and } x_k \neq y_0 \\ -\sum\limits_{\substack{\mathbf{z} \in \mathcal{A}^{n_k+1} \\ \mathbf{z} \neq \mathbf{s}_k}} Q_k^{\gamma}(\mathbf{s}_k, \mathbf{z}) & \text{if } H(\mathbf{s}_k, \mathbf{y}) = 0 \\ 0 & \text{otherwise} \end{cases}$$

with $\mathbf{s}_k = (x_k, x_{i_1}, \ldots, x_{i_{n_k}}) \in \mathcal{A}^{n_k+1}$, where $\{i_1, \ldots, i_{n_k}\} = N_k$

$\mathbf{y} = (y_0, y_1, \ldots, y_{n_k}) \in \mathcal{A}^{n_k+1}$

$\gamma_{(\mathbf{s}_k,\mathbf{y})} > 0$ neighbourhood constraints,
e.g. as a function of energy values ΔG^0.

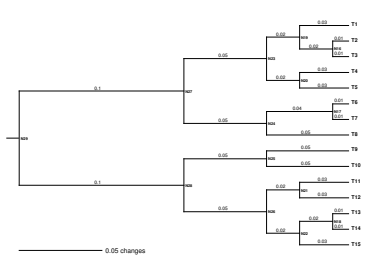
$Q_k(\mathbf{s}_k, \mathbf{y})$ rates given a chosen nullmodel

Normalisation:

$$d_k = -\sum_{\mathbf{z} \in \mathcal{A}^{n_k+1}} \pi_k(\mathbf{z}) \cdot Q_k(\mathbf{z}, \mathbf{z}) = 1.$$

SISSI SImulating Site-Specific Interactions

(Gesell, T. and von Haeseler, A. (2006) ,*Bioinformatics*, 22, 716-722)



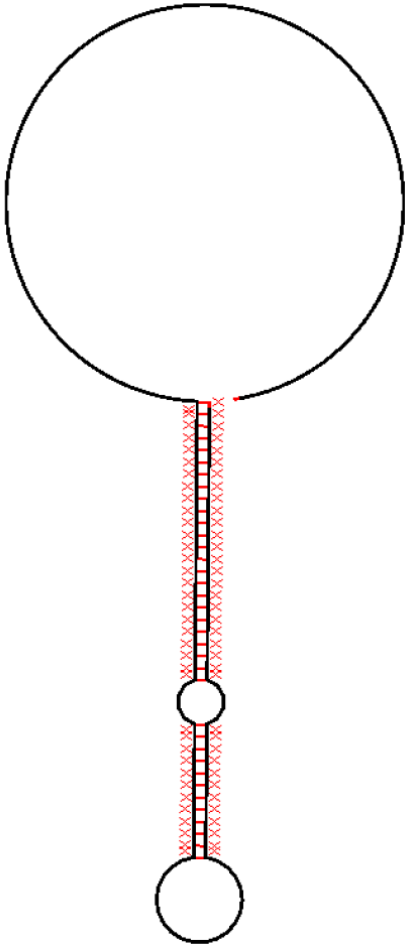
+

+

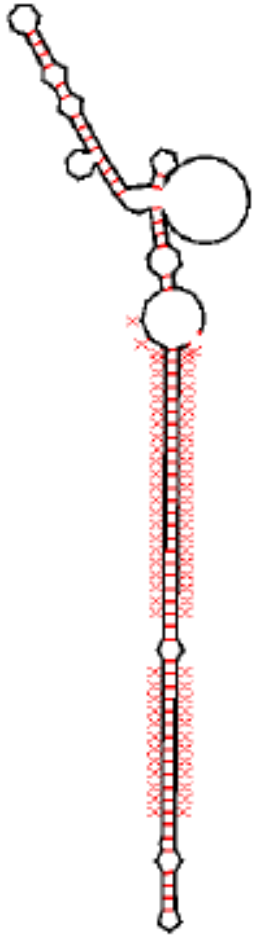
substitution models

```
 15 401
T1          AGACGGUCUGGUUGCGGGGGUGAUCACGACGAACGGUCGUGAUUGCCUUAGGCCGGUGGGCCUUGGUCAAGUCAGAUGAGCUC
T3          AGACGGUCUGGUUGCGGGGGUGAUUACGACGAACGGUCGUGAUUGCCUAAGGCCGGUGGGCCUUGGUCAAGUCGGAUGAGCUC
T2          AGACGGUCUGGUUGCGGGGGUGAUCACGACGAACGGUCGUGAUUGCCUAAGGCCGGUGGGCCUUGGUCAAGUCGGAUAAGCUC
T4          AGACGGUCUGGUUGCGGGGGUGAUCACGGCGAACGGUCGUGAUUGCCUACCGCAGGUGGGCCUAGGUCAAGUCGGAUGAGCUC
T5          AGACGGUCUGGUUGCGGGGGUGAUCACGACGAACGGUCGUGAUUGCCUAACGCAGGUGGGCCUAGGUCAAAUCGGACGAGCUC
T6          GGGCGGUCUGGUUAUGGGGGUGAUCACGGCGAACGGCCGUGAUGGCCUAAGGGAGGUUAGCCUGAGUUGAGUCGGAUUAGGUC
T7          GGGCGGUCUGGUUAUGGGGGUGAUCACGGCGAACGGCCGUGAUGGCCUAAGGGAGGUUGGCCUAAGUUGAGUCGGAUUAGGUC
T8          GGGCGGUCUGGUUAUGGGGGUCAUCACGGCGAACGGCCGUGAUGGCCUAAGGGAGGUUGGCCUAAGUUCAGUCGGAUUUGGUC
T9          CUAUGGUCUGGUUACGGGGGUGAUCAUGGCGGGCAGCCGUGAUUGCCGUGUGCAGGUGGGUUUAAGUUUAGUAGAAUUAGUGC
T10         CUAUGGUCUGGUUACGGGGGUGAUCAUGGCGGGCGCCCGUGAUCGCCGUGUGCAGGUGGGUCUAAUUUUAGUCGAAUUGGCGC
T11         CUAUGGCCUGGUUACGGGGGUGAUCAUGGUGGGCGGUCGUGAUUGCCGUGUGCAGGUGGGUCUAAGUUUAGGCGGAUUGGCGC
T12         CUAUGGUCUGGUUACGGGGGUGAUCAUGGUGGGCGGCCGUGAUUGCGGUGUGCAGAUGGGUCCAAGUUUAGGCGGAUUGGCGC
T13         CUAUGGUCUGGUUACGGGGGUGAUCACGGUGGGCGACCGUGAUUGCCCUGUGCAGGUGGGUCUAAGUUUAGGCGAAUUGGCGL
T14         CUAUGGUCUGGUUACGGGGGUGAUCAUGGUGGGCGGCCGUGAUUGGCCUGUGCAGGUGGGUCUAAGUUUAGGCGAAUUGGCGC
T15         CUAUGGUCUGGUCACGGGGGUGAUCAUGGUGAGCGGCCAUGAUUGCCCUGUGCAGUUGGGUCUAAGUUUAAGCGAAUUGGCGC
```

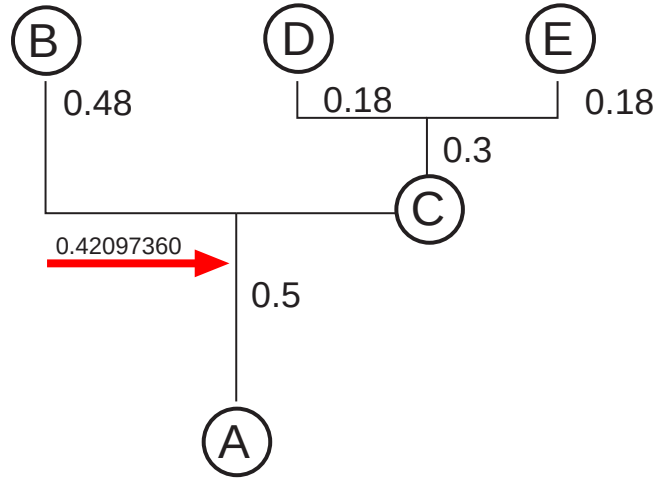
Phylogenetic RNAmovie



$\mathcal{N}$



mfe



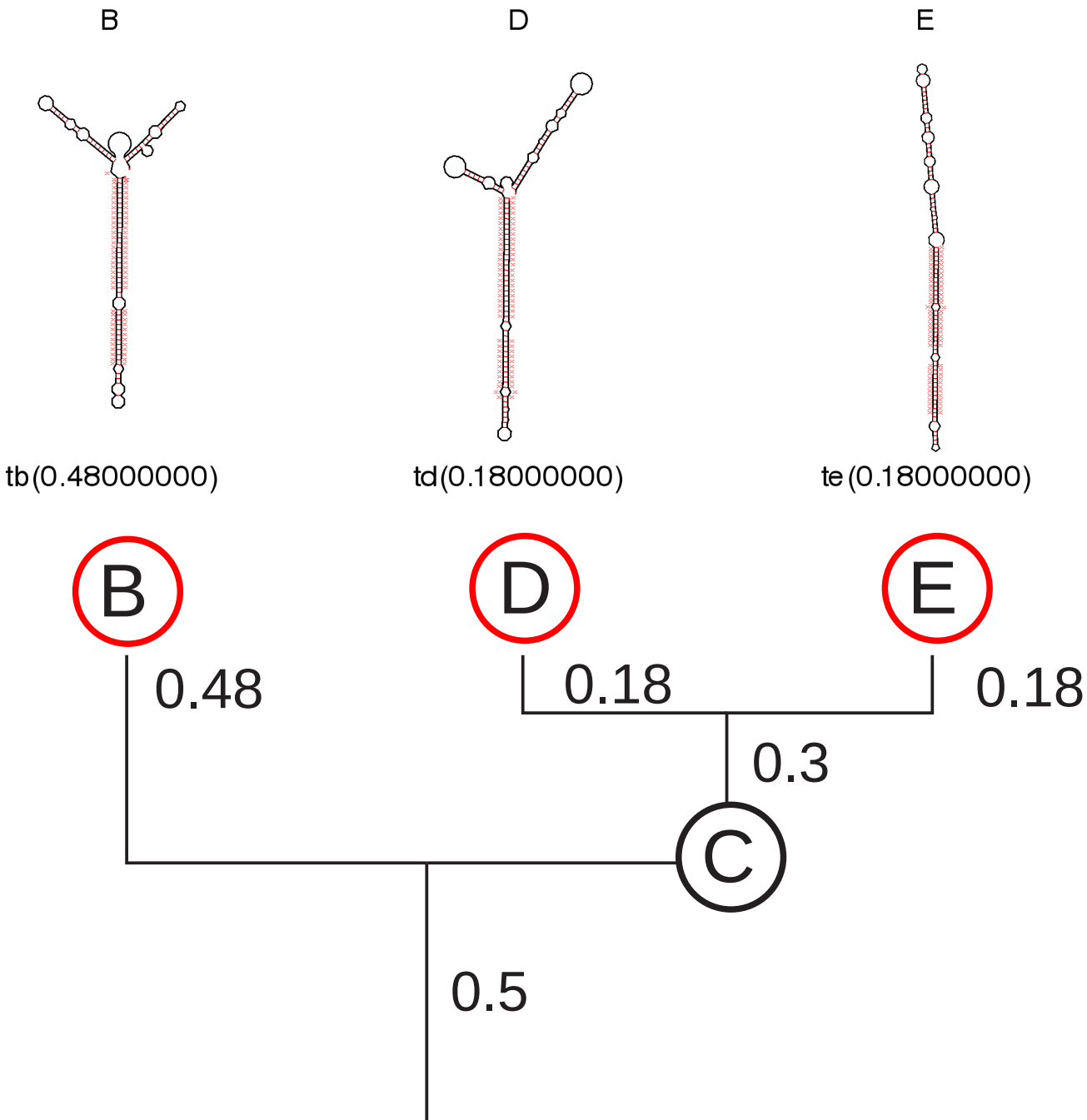
Movie

SISSI (Gesell, von Haeseler, 2006)

RNAfold (Hofacker et al., 1994)

RNAmovie (Evers, Giegerrich , 1999)

Phylogenetic RNAmovie



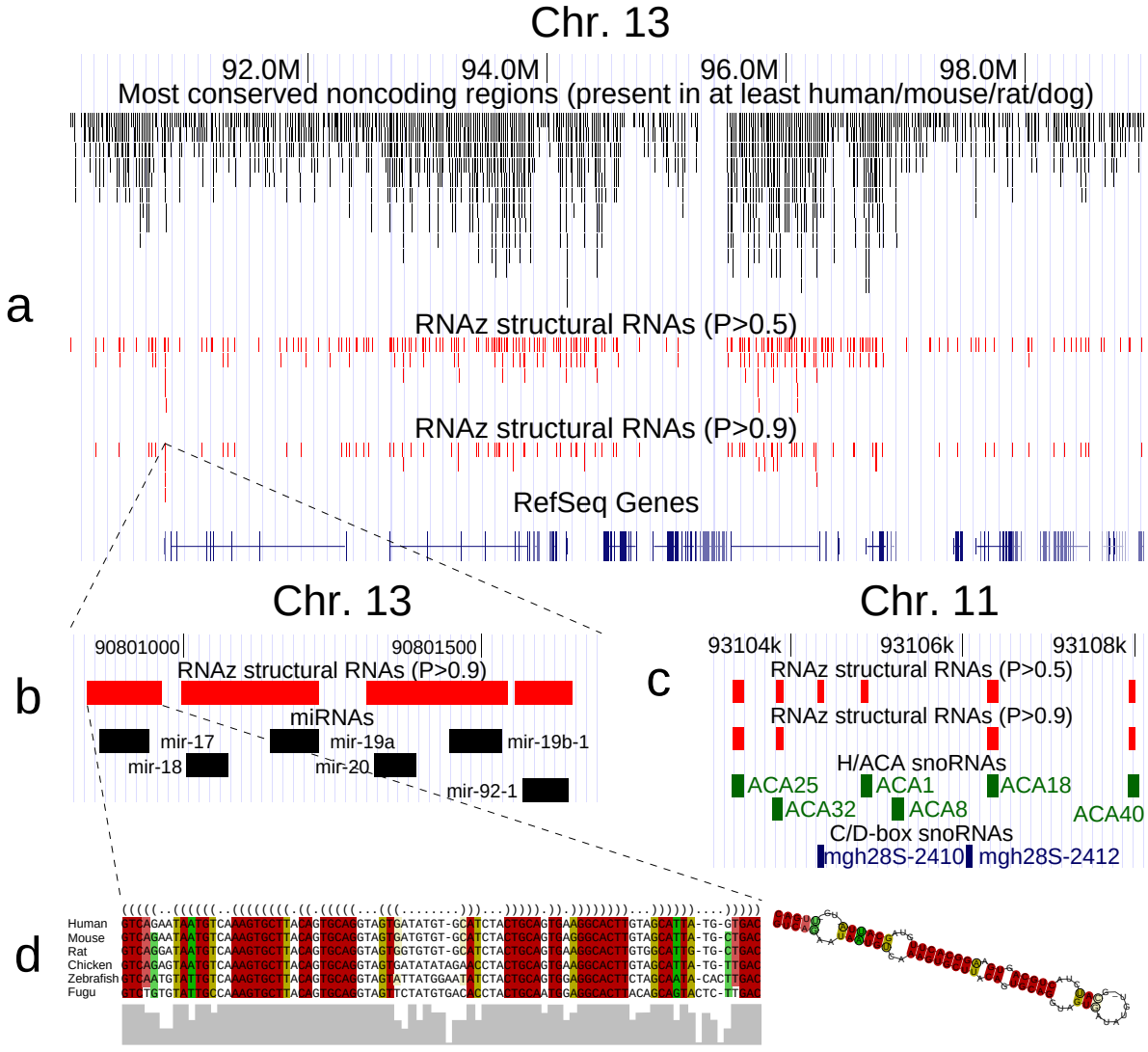
Applications of SISSI

- Energy landscape of RNA, Selection of RNA ...
- Performance of tree building method with dependencies
- Performance of structure prediction methods

Applications of SISSI

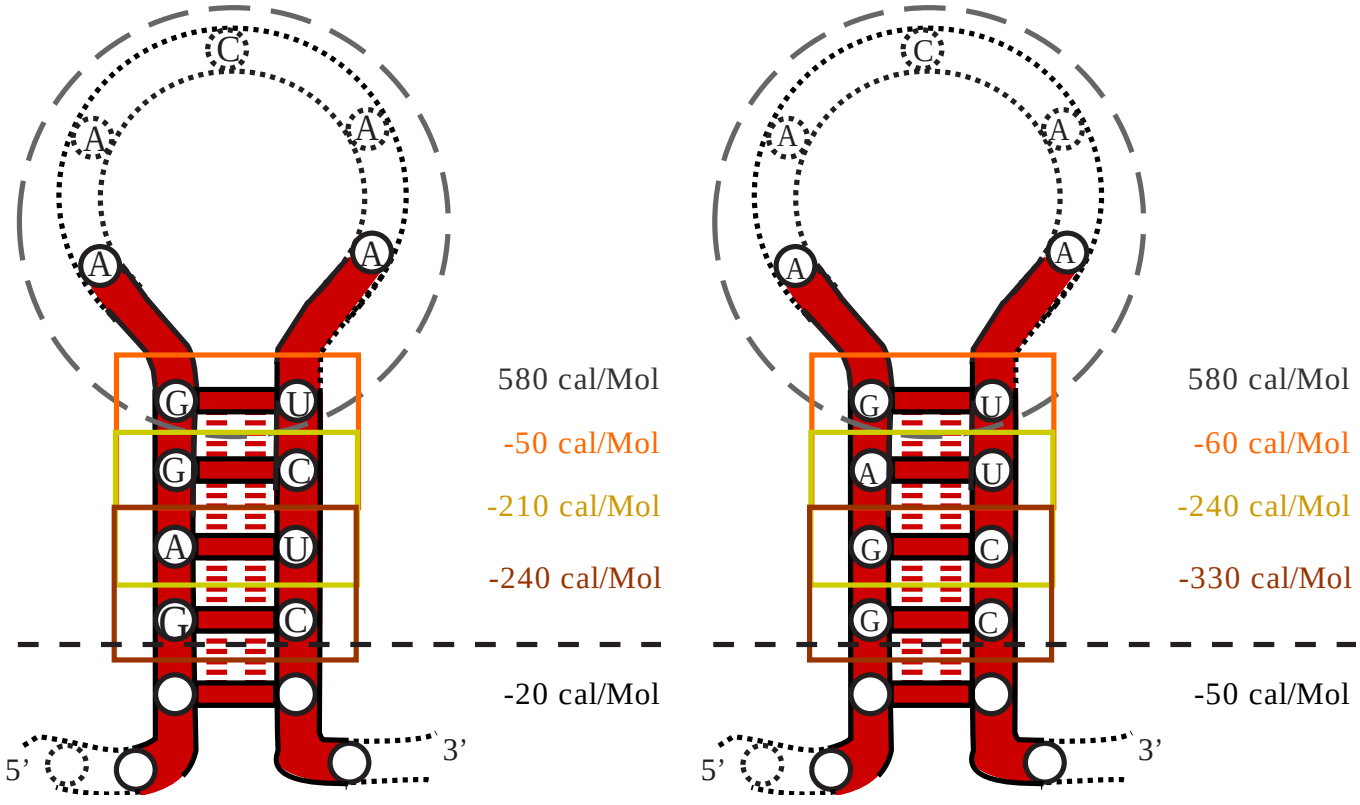
- Energy landscape of RNA, Selection of RNA ...
- Performance of tree building method with dependencies
- Performance of structure prediction methods
- Accurate Background Models for Comparative Genomic Screens: Applications to RNA Gene Prediction
  Joint work with Stefan Washietl

Maps of Structured ncRNAs in the Human Genome



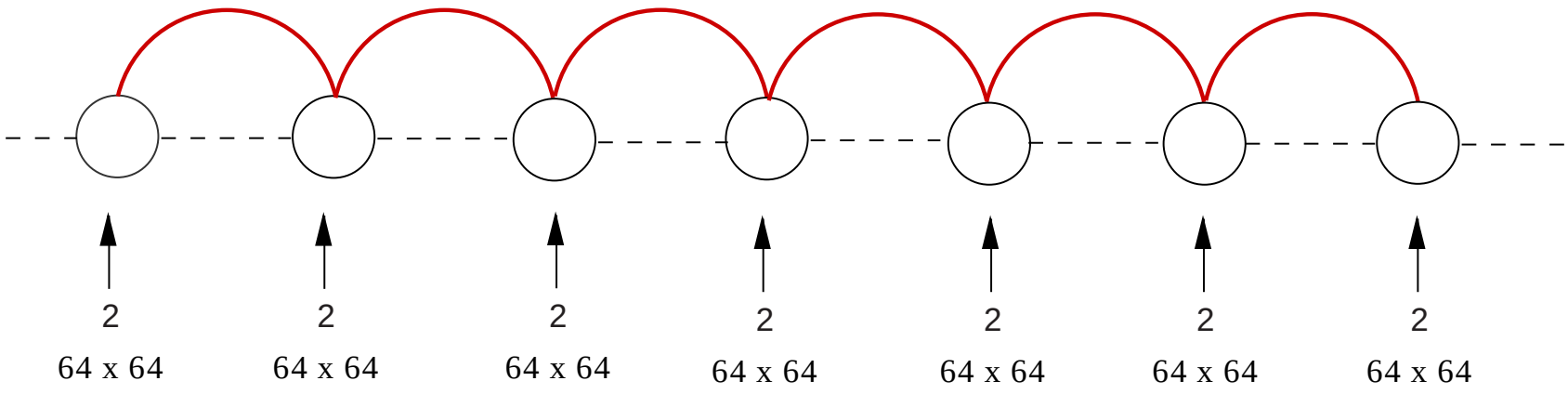
Washietl et. al., Nat. Biotech. 2005; 23:1383-90.

Dinucleotide Content Matters



How do we get dinucleotide controlled random alignments?

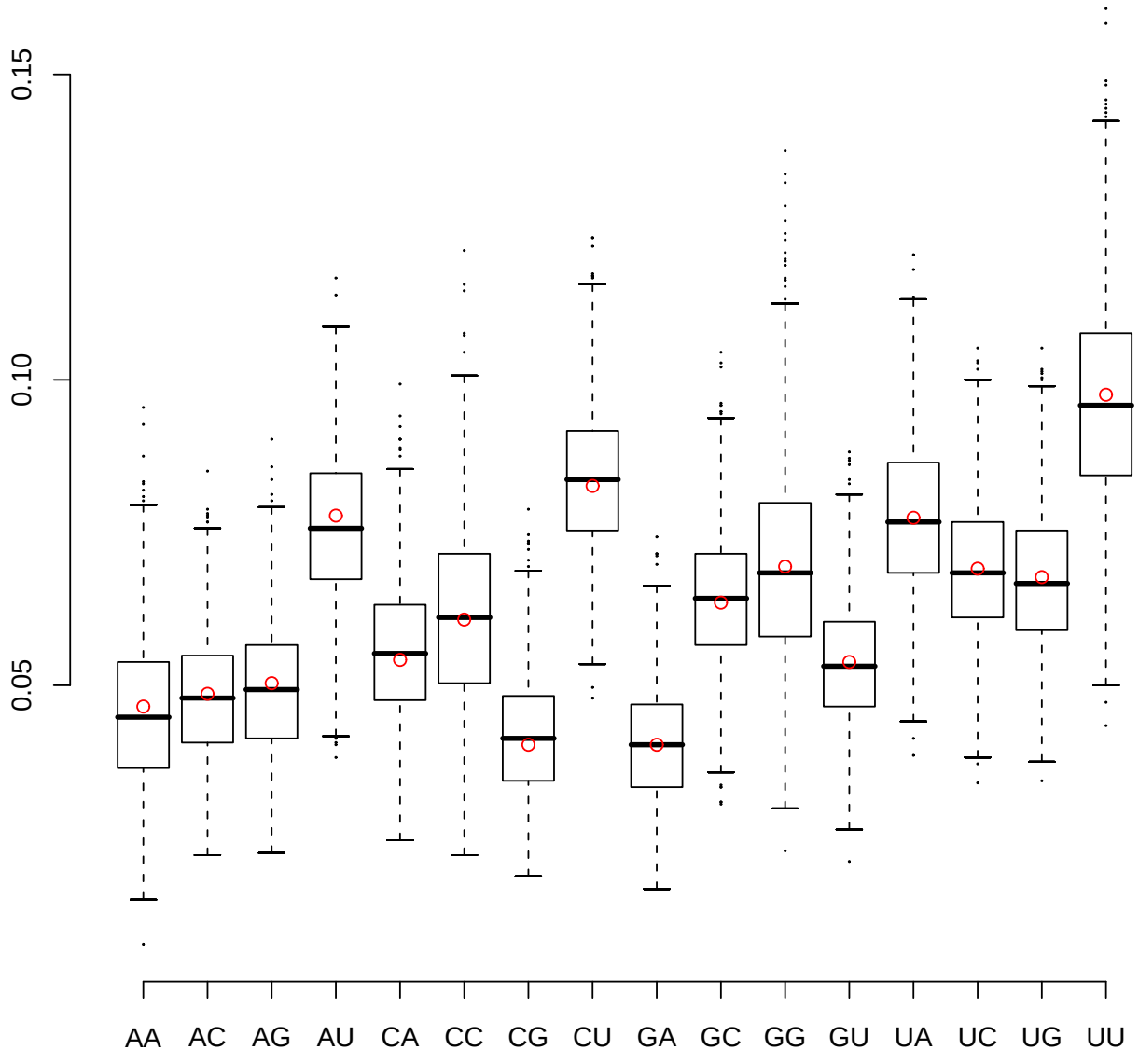
Neighbourhood System and Model for Dinucleotid Content



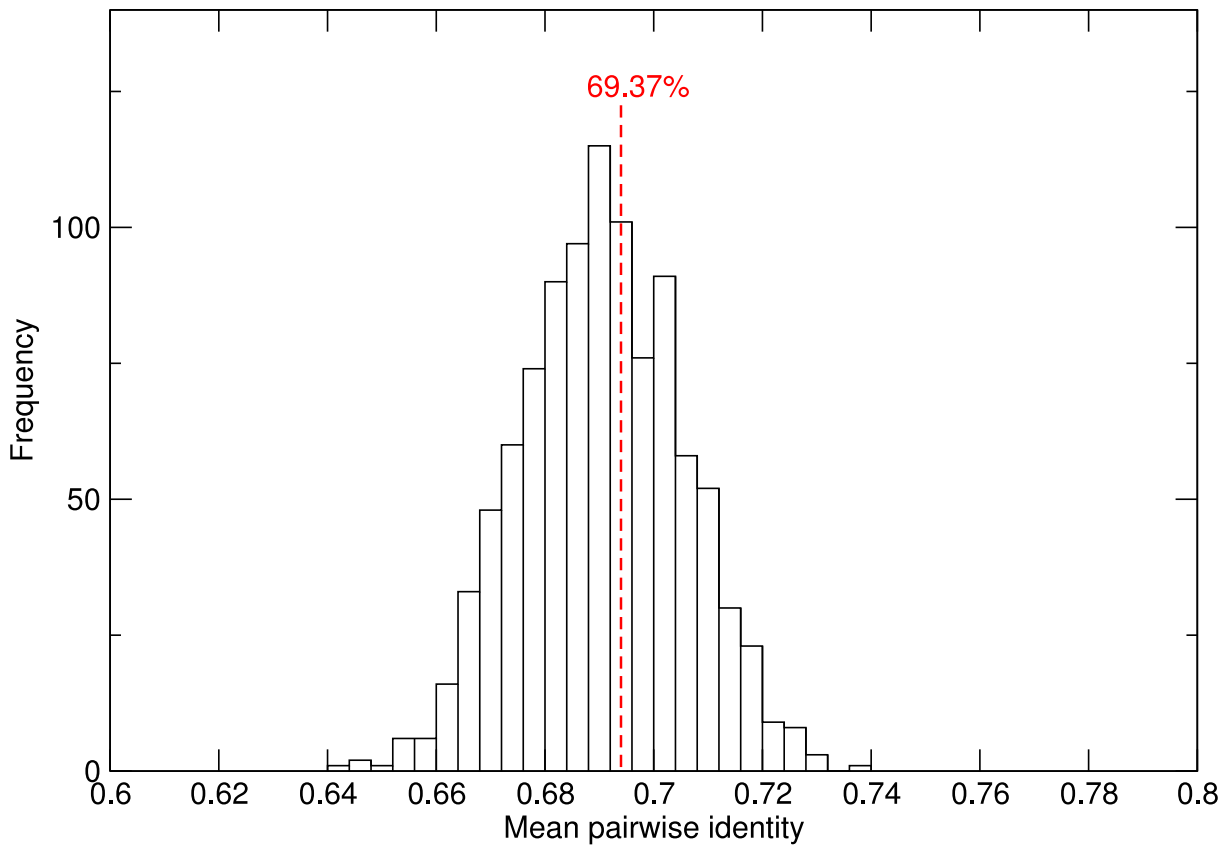
16 submatrices of the type:

$$
\begin{array}{c}
 \\
\mathrm{A}|x_{k-1},x_{k+1} \\
\mathrm{C}|x_{k-1},x_{k+1} \\
\mathrm{G}|x_{k-1},x_{k+1} \\
\mathrm{U}|x_{k-1},x_{k+1}
\end{array}
\begin{array}{c}
\begin{array}{cccc}
\mathrm{A}|x_{k-1},x_{k+1} & \mathrm{C}|x_{k-1},x_{k+1} & \mathrm{G}|x_{k-1},x_{k+1} & \mathrm{U}|x_{k-1},x_{k+1}
\end{array} \\
\begin{pmatrix}
* & \beta\pi_{\mathrm{C}|x_{k-1},x_{k+1}} & \alpha\pi_{\mathrm{G}|x_{k-1},x_{k+1}} & \beta\pi_{\mathrm{U}|x_{k-1},x_{k+1}} \\
\beta\pi_{\mathrm{A}|x_{k-1},x_{k+1}} & * & \beta\pi_{\mathrm{G}|x_{k-1},x_{k+1}} & \alpha\pi_{\mathrm{U}|x_{k-1},x_{k+1}} \\
\alpha\pi_{\mathrm{A}|x_{k-1},x_{k+1}} & \beta\pi_{\mathrm{C}|x_{k-1},x_{k+1}} & * & \beta\,\pi_{\mathrm{U}|x_{k-1},x_{k+1}} \\
\beta\,\pi_{\mathrm{A}|x_{k-1},x_{k+1}} & \alpha\pi_{\mathrm{C}|x_{k-1},x_{k+1}} & \beta\pi_{\mathrm{G}|x_{k-1},x_{k+1}} & *
\end{pmatrix}
\end{array}
$$

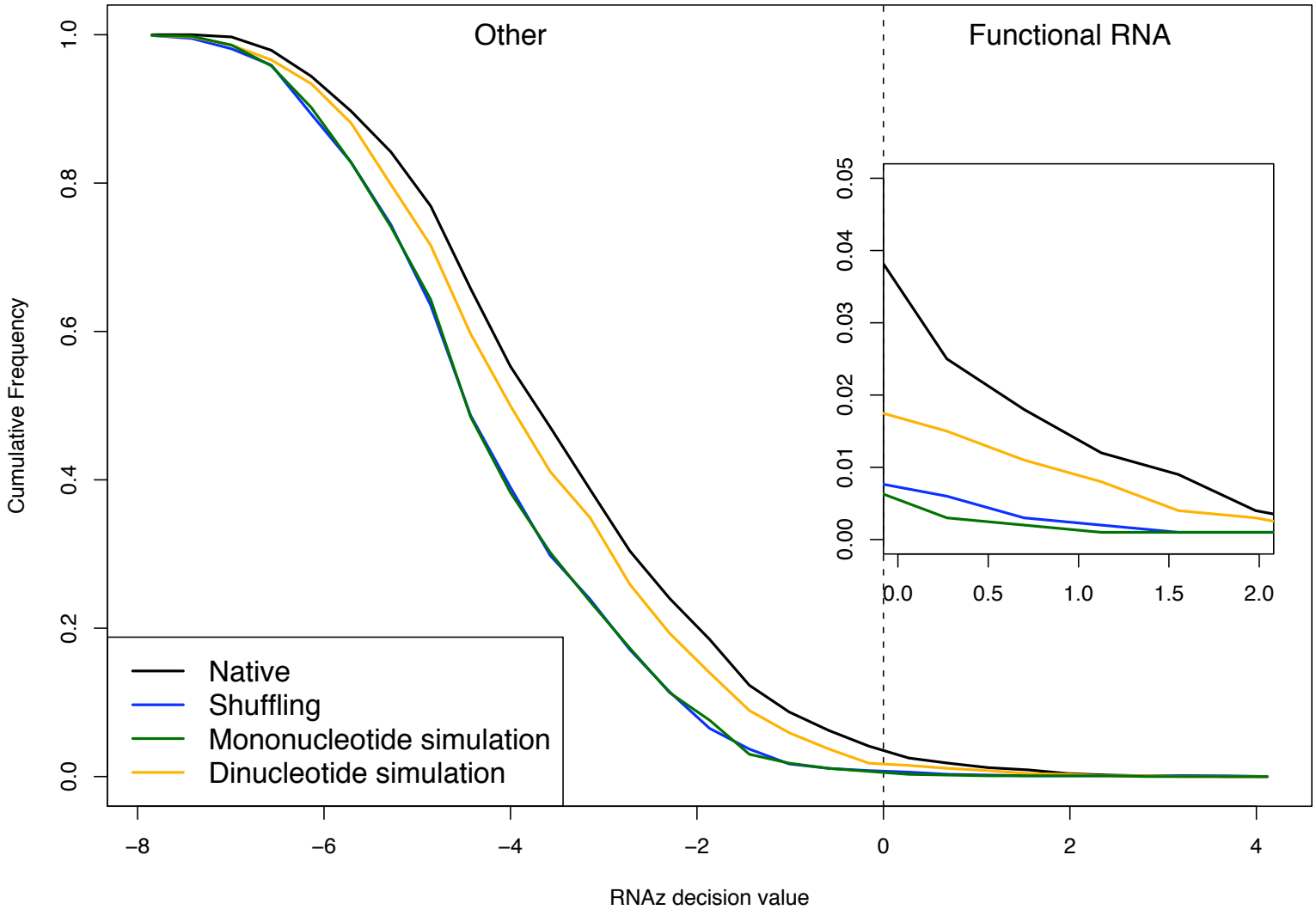
Results: Dinucleotide Content



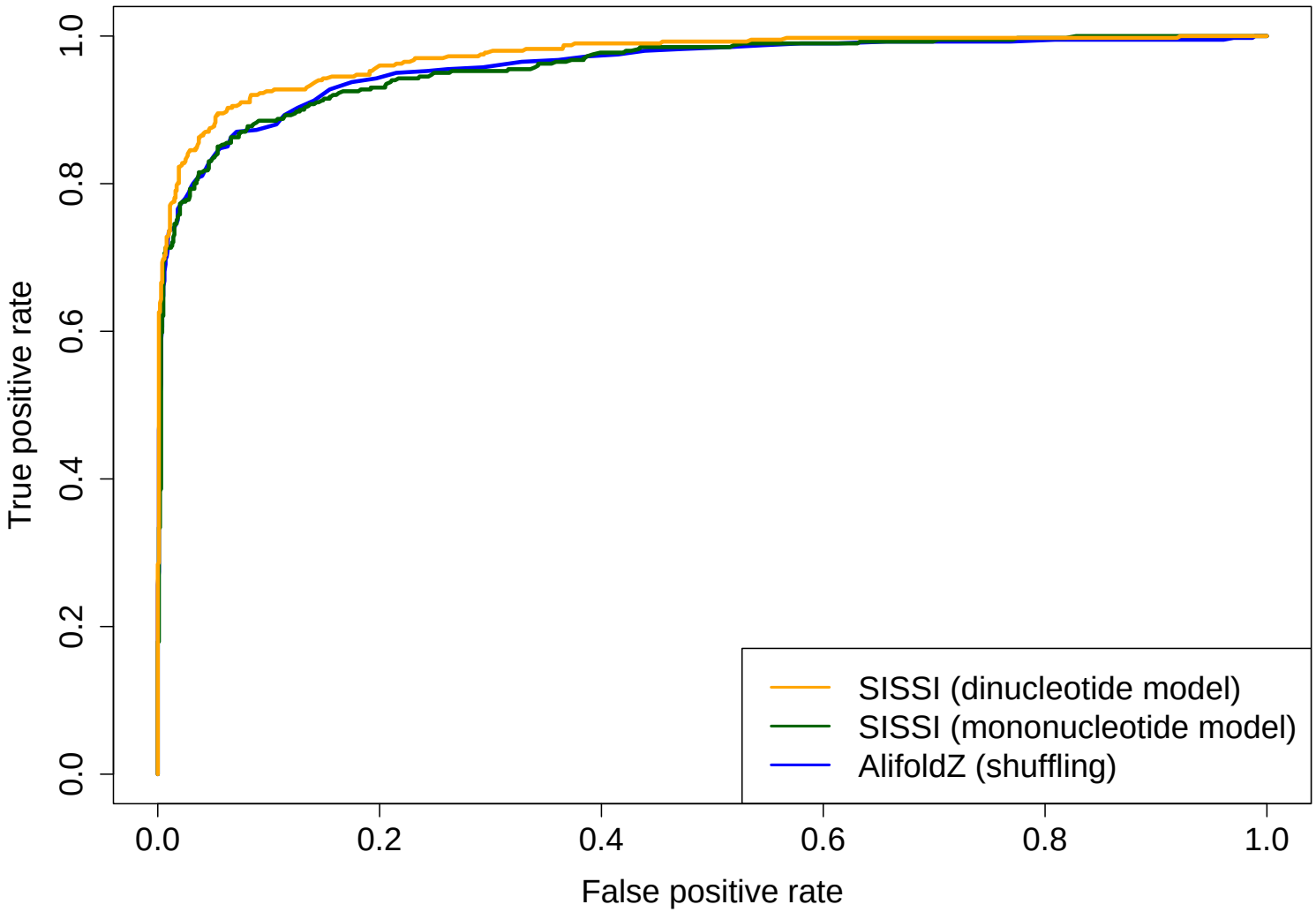
Results: Mean Pairwise Identity



Estimating False Positves for RNAz Gene Prediction



SISSIz: Dinucleotide Based RNA Gene Finder



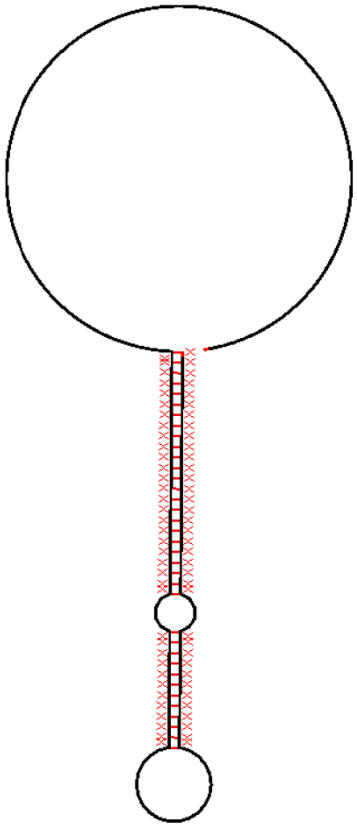
AlifoldZ:Washietl & Hofacker, J Mol Biol. 2004; 342:19-30

Summary

- We have presented a general framework and a model for arbitrary dependencies
- RNA gene predictions are biased by the genomic dinucleotid content.
- SISSI can be used to generate accurate null models giving more realistic estimation of false positives.
- SISSIz is introduced as a dinucleotide based gene finder (miRNA target search, transcription factor binding sites, ...)
- A phylogenetic definition of structure

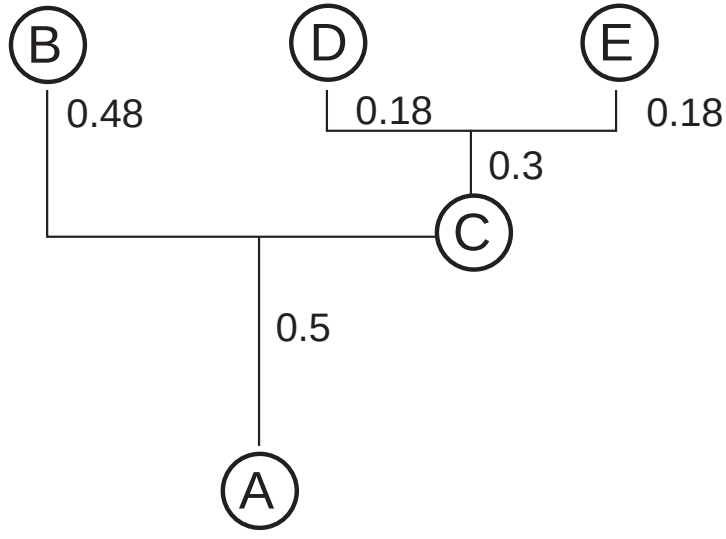
Future Directions in Phylogenetic Methods and Models

A Phylogenetic Definition of Structure



$\mathcal{N}$

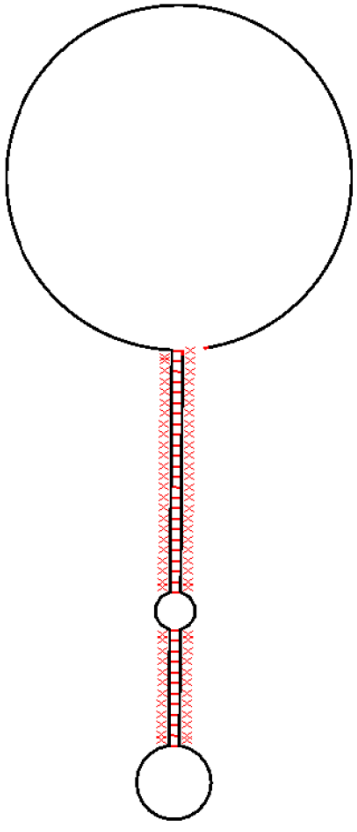
MODEL



Defining lineage specific neighbourhood systems

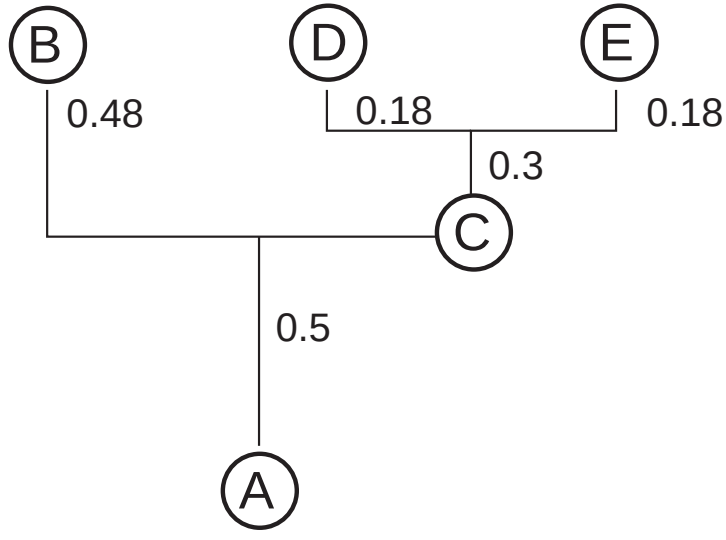
Future Directions in Phylogenetic Methods and Models

A Phylogenetic Definition of Structure



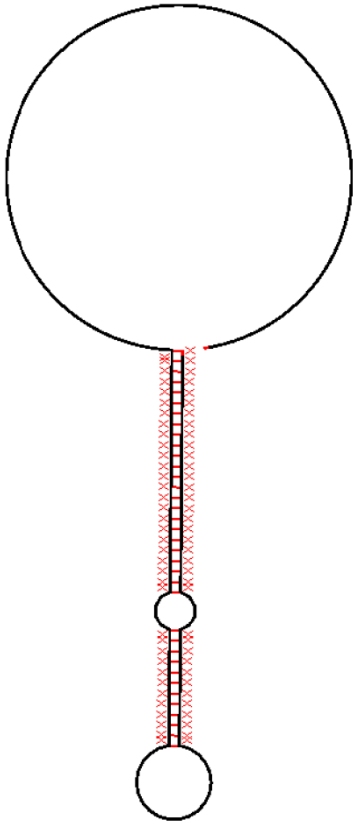
$\mathcal{N}$

$\Longrightarrow$ MODEL



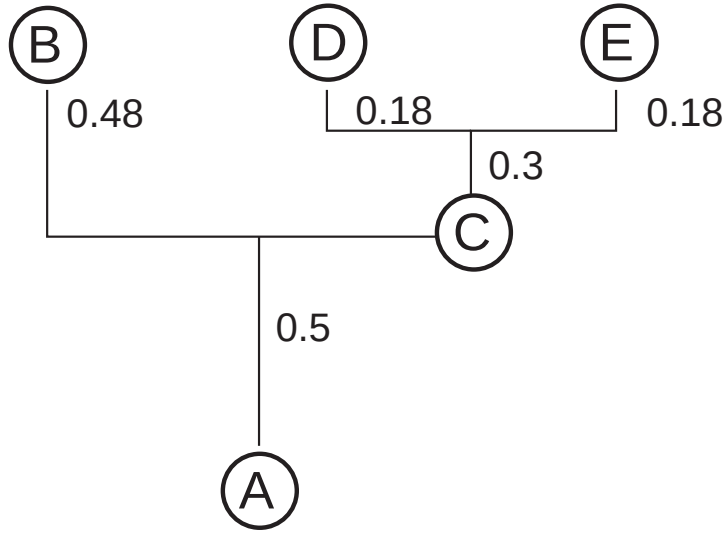
Future Directions in Phylogenetic Methods and Models

A Phylogenetic Definition of Structure

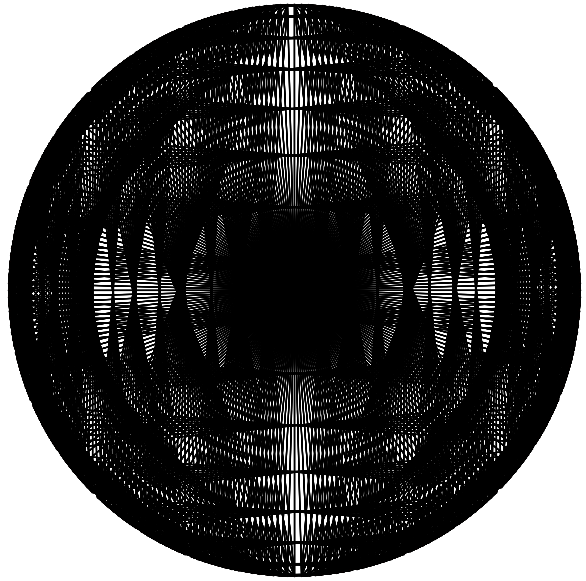


$\mathcal{N}$

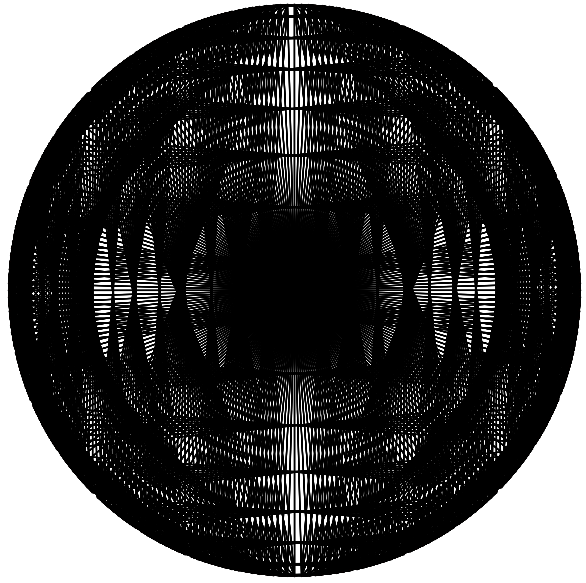
MODEL



...and Further Animations on the Sequence Space



...and Further Animations on the Sequence Space



28/29 April 2008: Discussion Meeting organised by Nick Goldman and Ziheng Yang at the Royal Society in London.

Acknowledgements

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Thank you for your attention!