de.NBI and its Galaxy interface for RNA folding

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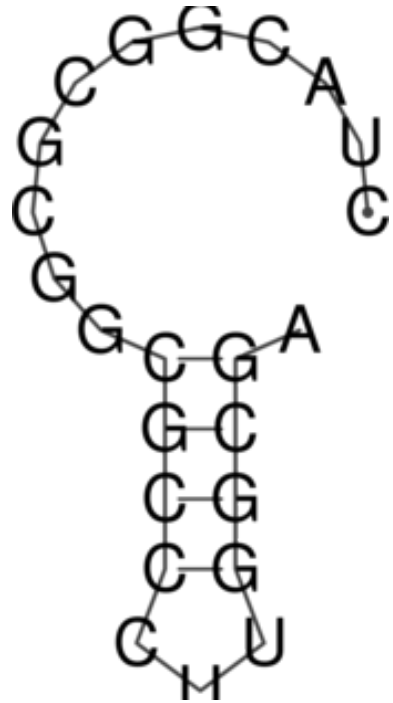
You can download the pdfs you will need today here
http://www.bioinf.uni-leipzig.de/~fall/RNA_folding_workshop-presentation.pdf
http://www.bioinf.uni-leipzig.de/~fall/Exercises.pdf

Goal: Use `RNAfold` to do a simple structure prediction.

- Upload the file `rna.fa` into your Galaxy session.
- Start `RNAfold` with standard parameters
- Look into the output

- CUACGGCGCGGCGCCCUUGGCGA
- .....((((...)))). (-5.00)



Goal: Use `RNAfold` to do a structure prediction using the partition function

- Start `RNAfold` using --partfunc
- Look into the output

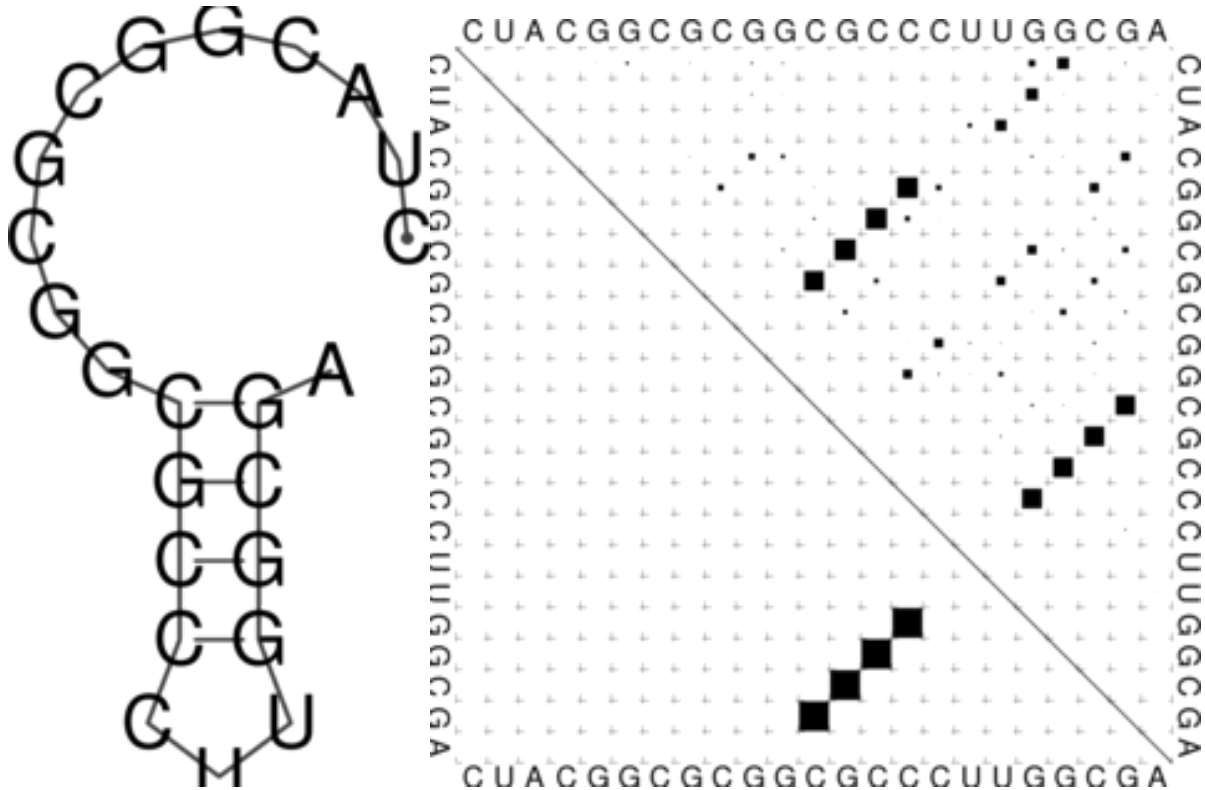
- CUACGGCGCGGCGCCCUUGGCGA
- MFE: .....((((...)))). (-5.00)
- PF: ....{,{{...||||...)}}}. [-5.72]

The partition function is a rough measure for the well-definedness of the MFE structure. The third line shows a condensed representation of the pair probabilities of each nucleotide, similar to the dot-bracket notation, followed by the ensemble free energy ($-kT * ln(Z)$) in kcal/mol

The partition function allows us to calculate the proportion of a certain structure in the ensemble.
Use RNAfold -p to get the ensemble free energy, which is related to the partition function via **F = -RT*ln(Q)**, for the unconstrained (Fu) and the constrained case (Fc), (use option -C), and evaluate **pc = exp((Fu - Fc)RT)** to get the desired probability.

- CUACGGCGCGGCGCCCUUGGCGA
- MFE: ..........((((...)))). (-5.00)
- PF: ....{,{{...||||...)}}}. [-5.72]
- CS: ..... { 0.00 d=4.66 }
- MEA: ......((...))((...))... { 2.90 MEA=14.79 }
- frequency of mfe structure in ensemble 0.311796; ensemble diversity 6.36

Pseudo bracket notation:
Here, the usual '(', ')', '.', represent bases with a strong preference (more than 2/3) to pair upstream (with a partner further 3'), pair down-stream, or not pair, respectively. '', '', and ',' are just weaker version of the above and '|' represents a base that is mostly paired but has pairing partners both upstream and downstream. In this case open and closed brackets need not match up.


C U A C G G C G C G G C G C C C U U G G C G A
C U A C G G C G C G G C G C C C U U G G C G A
C U A C G G C G C G G C G C C C U U G G C G A
C U A C G G C G C G G C G C C C U U G G C G A

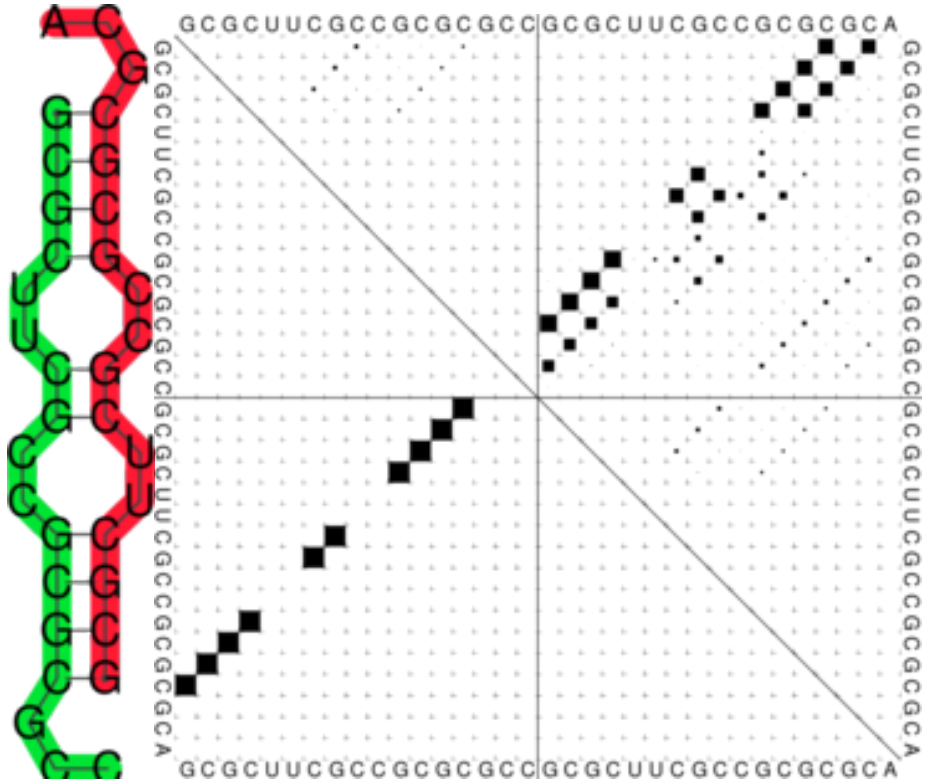
Goal: Use SHAPE-directed `RNAfold` to do a structure prediction

- Upload the file `rna.simple.shape` into your Galaxy session.
- Start RNAfold using the shape file and -shapeMethod=D
- Look into the output

Goal: Use `RNAcofold` to predict the cofolding of two sequences.

- Upload the file `cofold.txt` into your Galaxy session. (Look at it)
- Start RNAcofold using cofold.txt with the --partfunc option
- Look at the output.

- GCGCUUCGCCGCGCGCC&GCGCUUCGCCGCGCGCA
- ((((..((..((((...&))))..))..))))... (-17.70)
- ((((..(,.((((,,.&))))..),.))))),,. [-18.26]
- frequency of mfe structure in ensemble 0.401754 , delta G binding= -3.95



Cofold can use concentrations of molecules for duplex prediction, but this is slow for longer sequences.

Goal: Use `RNAduplex` to predict *only* intermolecular base pairs of two sequences.

- Upload the file `duplex.txt` into your Galaxy session. (Look at it)
- Start RNAduplex using duplex.txt with standard parameter
- Look at the output.

RNAduplex does not use concentrations and neglects intramolecular interactions, faster but less reliable, good prefilter.

Goal: Use `RNAup` to test the `RNAduplex` result.

- Start RNAup using duplex.txt with --include_both
- Look at the output and compare it with the RNAduplex result.

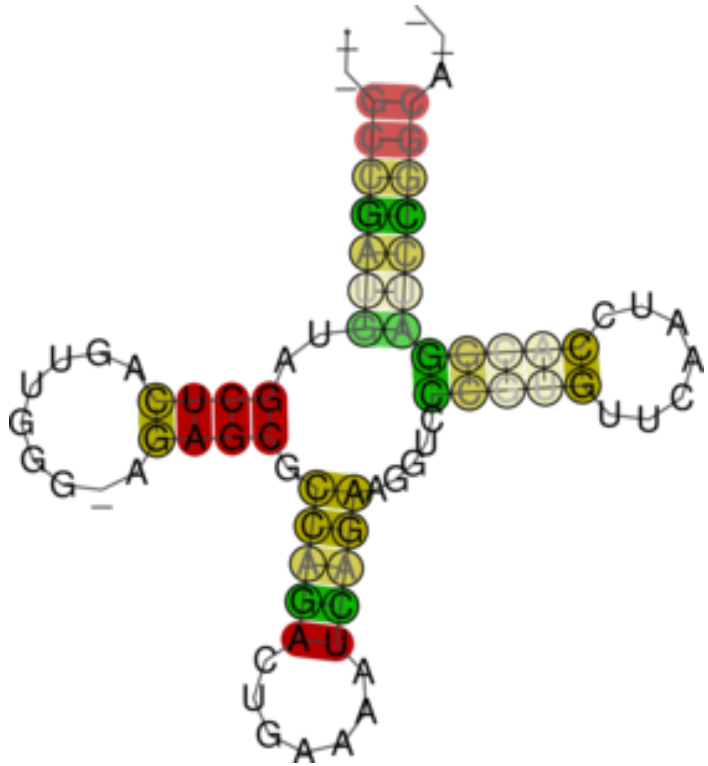
RNAup is also taking intramolecular interactions into account.

Goal: Use `RNAalifold` to predict the consensus structure

- Upload the clustal file `alifold.aln` into your Galaxy session.
- Edit the data type of alifold.aln to 'clustal'
- Use RNAalifold with the alifold.aln and --partfunc (Calculate partition function: 1)
- (Download the output) and look at it
- `Bonus`: Fold the sequences (alifold.fa) individually (RNAfold) and compare the results.

Goal: Use `RNAalifold` to predict *and visualize* the consensus structure

- Use RNAalifold with alifold.aln and --color and --aln
- (Download the output) and look at it



RNAalifold uses covariance information from sequence alignment to predict a consensus structure.

Goal: Use `RNAcode` to predict coding sequences in a MAF alignment.

- Upload the file `oskar.27way.rnacode.maf` into your Galaxy session.
- (Change its data type to maf)
- Use RNAcode with the maf file, --cutoff 0.05, --best_region, --best_hit, with GTF output
- (Download the output) and look at it

Goal: Use `RNAz`

- Upload the file `oskar.27way.rnaz.maf` into your Galaxy session.
- Use RNAz with the maf file
- (Download the output) and look at it