Shapify: Pathways to SARS-CoV-2 frameshifting pseudoknot

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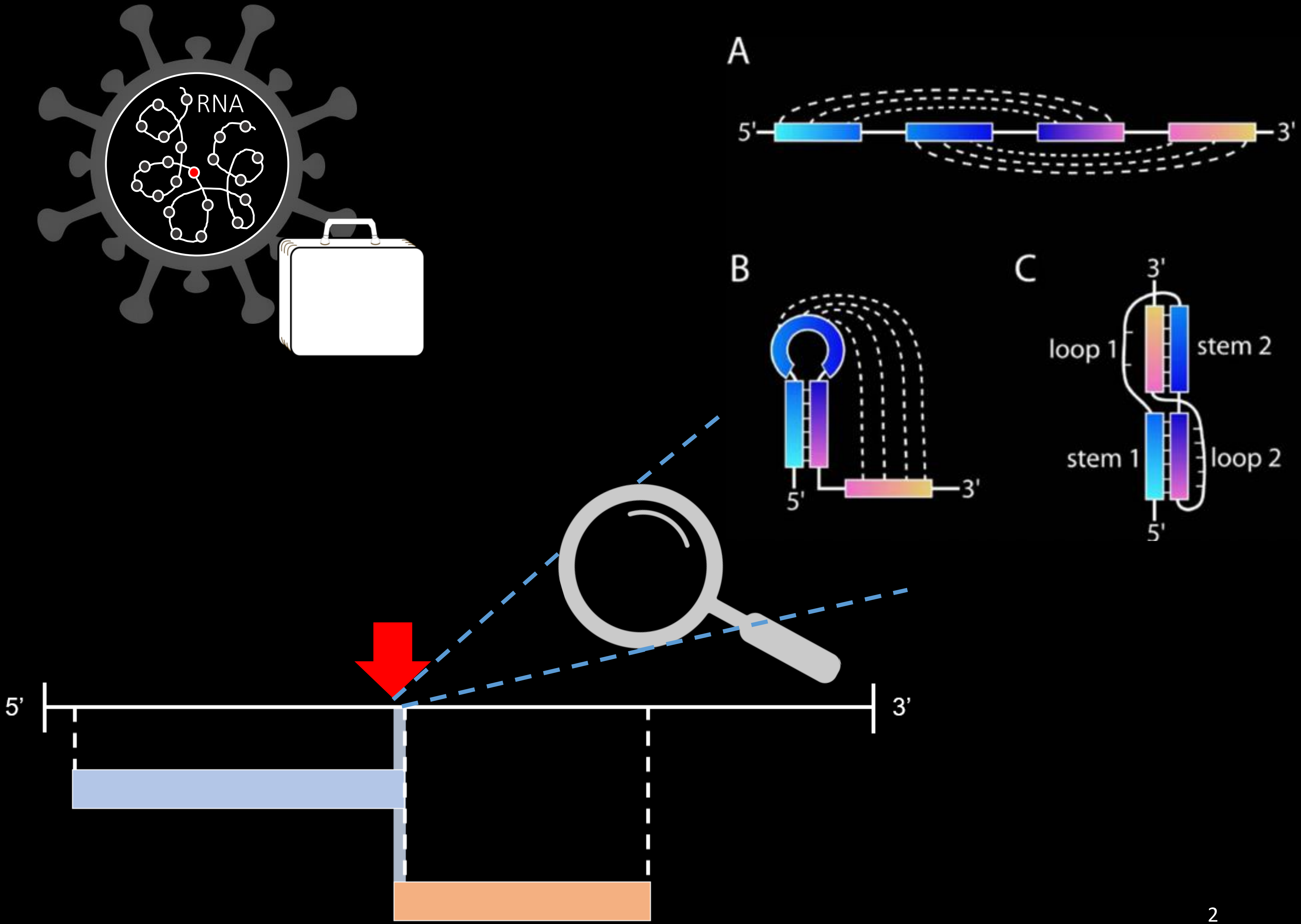
August 12, Benasque, Spain

University of Victoria | COBRA
COMPUTATIONAL BIOLOGY RESEARCH AND ANALYTICS

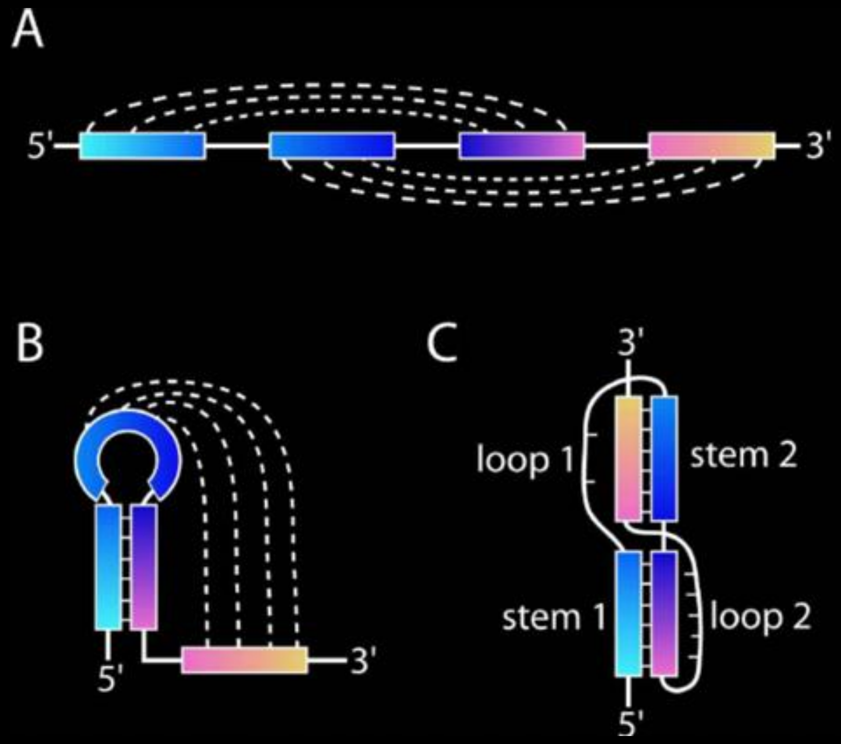

RNA
A
5'
3'
B
5'
3'
C
3'
loop 1
stem 2
stem 1
loop 2
5'
5'
3'

Goal

- Improve secondary structure prediction with SHAPE
- Contribute to treatments that disrupt replication [1-4]
  - Antisense RNA
  - Small molecules

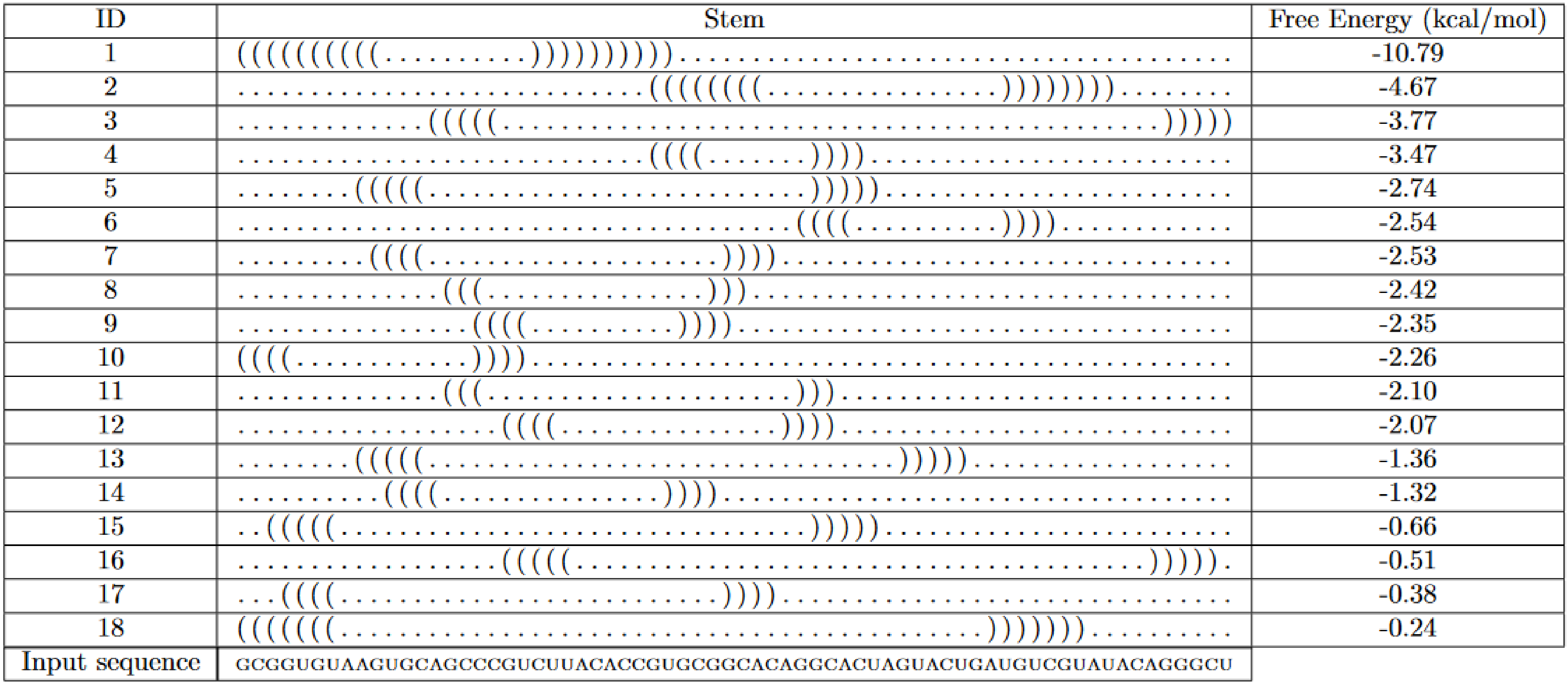
Hierarchical Folding Hypothesis

- Pseudoknot-free structure forms first (initial stem)
- Followed by additional pseudoknotted structure [5-10]



Method: Pseudoknot-free structures

- Calculate energetically favourable initial stems for the RNA sequence at the pseudoknot location [11]



ID	Stem	Free Energy (kcal/mol)
1	((((((((((..........))))))))))......................................	-10.79
2	............................((((((((................)))))))))........	-4.67
3	.............(((((...)))))	-3.77
4	............................((((.......))))..........................	-3.47
5	........(((((..........................)))))..........................	-2.74
6	......................................((((..........))))............	-2.54
7	.........((((....................))))...............................	-2.53
8	..............(((...............)))..................................	-2.42
9	................((((..........))))..................................	-2.35
10	((((............))))..	-2.26
11	..............(((.....................)))...........................	-2.10
12	..................((((...............))))..........................	-2.07
13	........(((((................................))))).................	-1.36
14	..........((((...............))))..................................	-1.32
15	..(((((................................)))))........................	-0.66
16	..................(((((.......................................))))).	-0.51
17	...((((..........................))))...............................	-0.38
18	(((((((..))))))))..........	-0.24
Input sequence	GCGGUGUAAGUGCAGCCCGUCUUACACCGUGCGGCACAGGCACUAGUACUGAUGUCGUAUACAGGGCU	

Method: Iterative H-Fold [12]

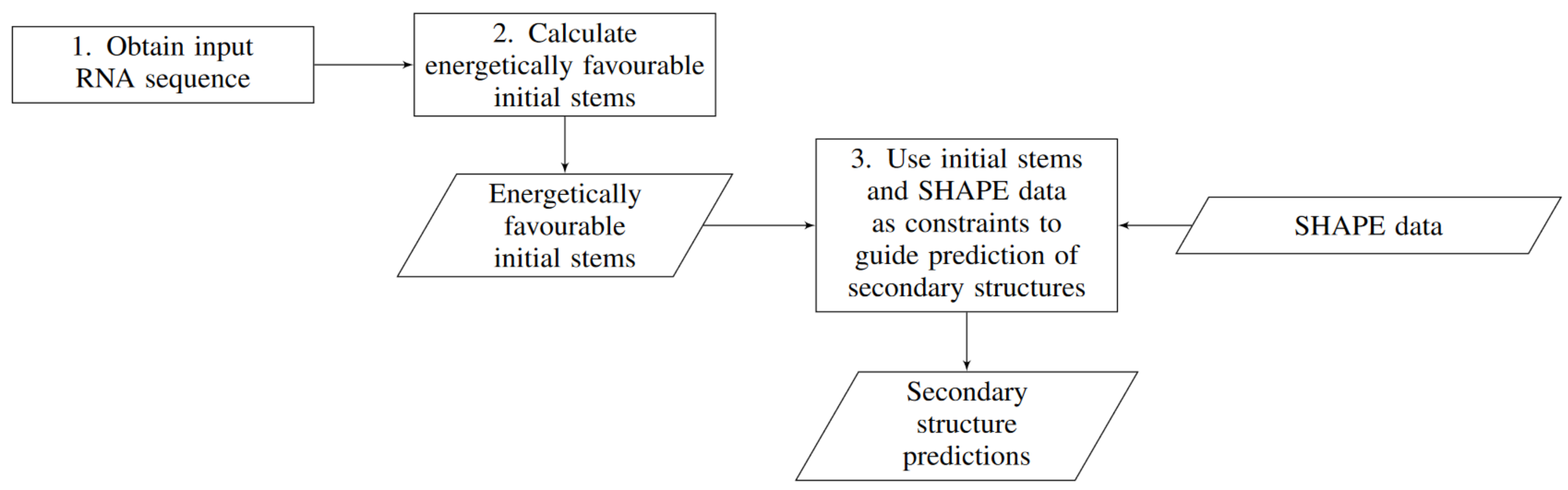
- Inputs: RNA sequence & pseudoknot-free structure
- Utilizes four biologically sound methods
- Allows minimal modification of input structure
- Energy minimization over the set of possible stems

Method: Shapify

- Follow the approach of ShapeKnots [13]

$$m\,(\log i + 1) + b$$

- Use three available SHAPE datasets for SARS-CoV-2 [14-16]



1. Obtain input RNA sequence

2. Calculate energetically favourable initial stems

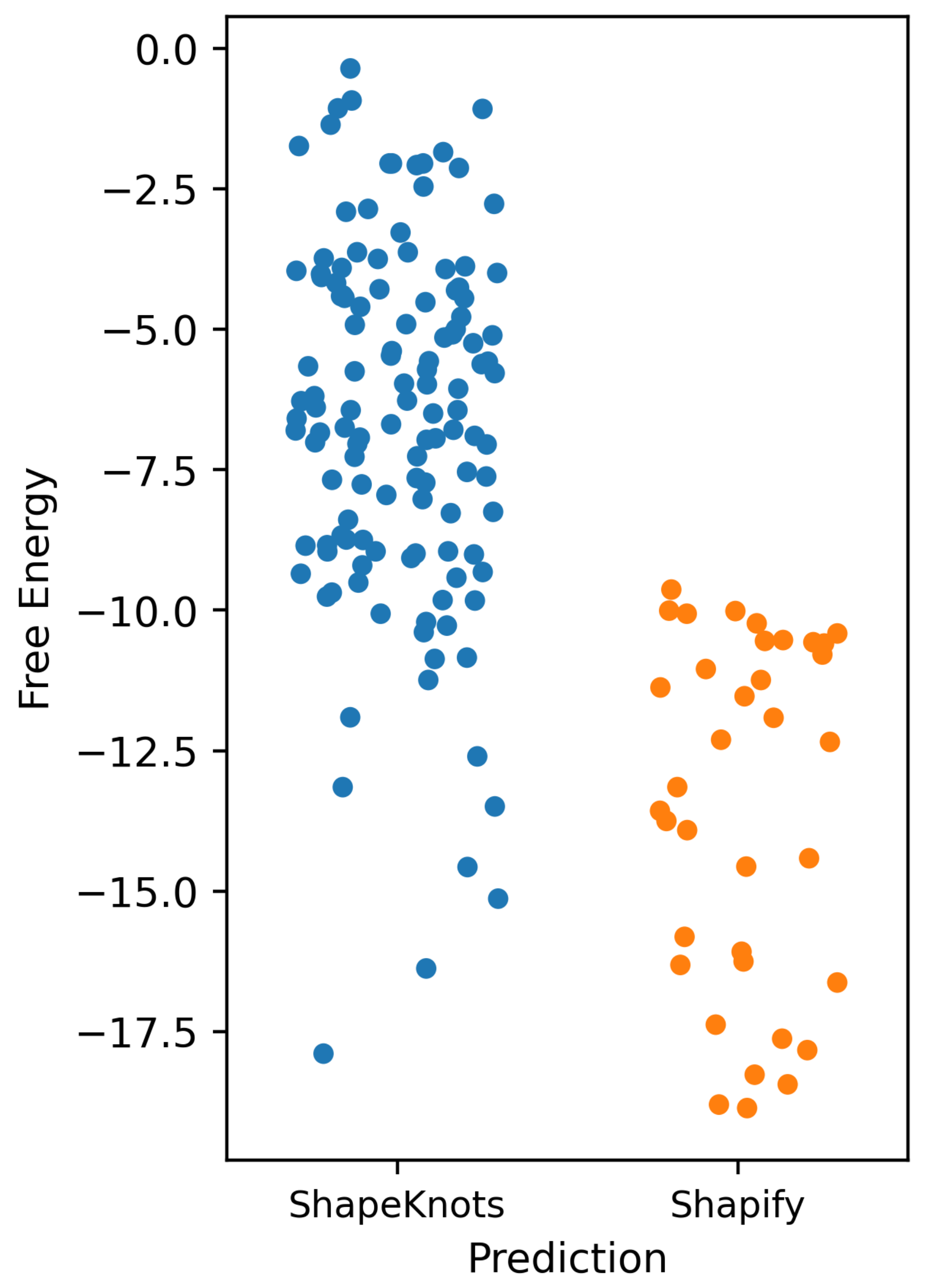
Energetically favourable initial stems

3. Use initial stems and SHAPE data as constraints to guide prediction of secondary structures

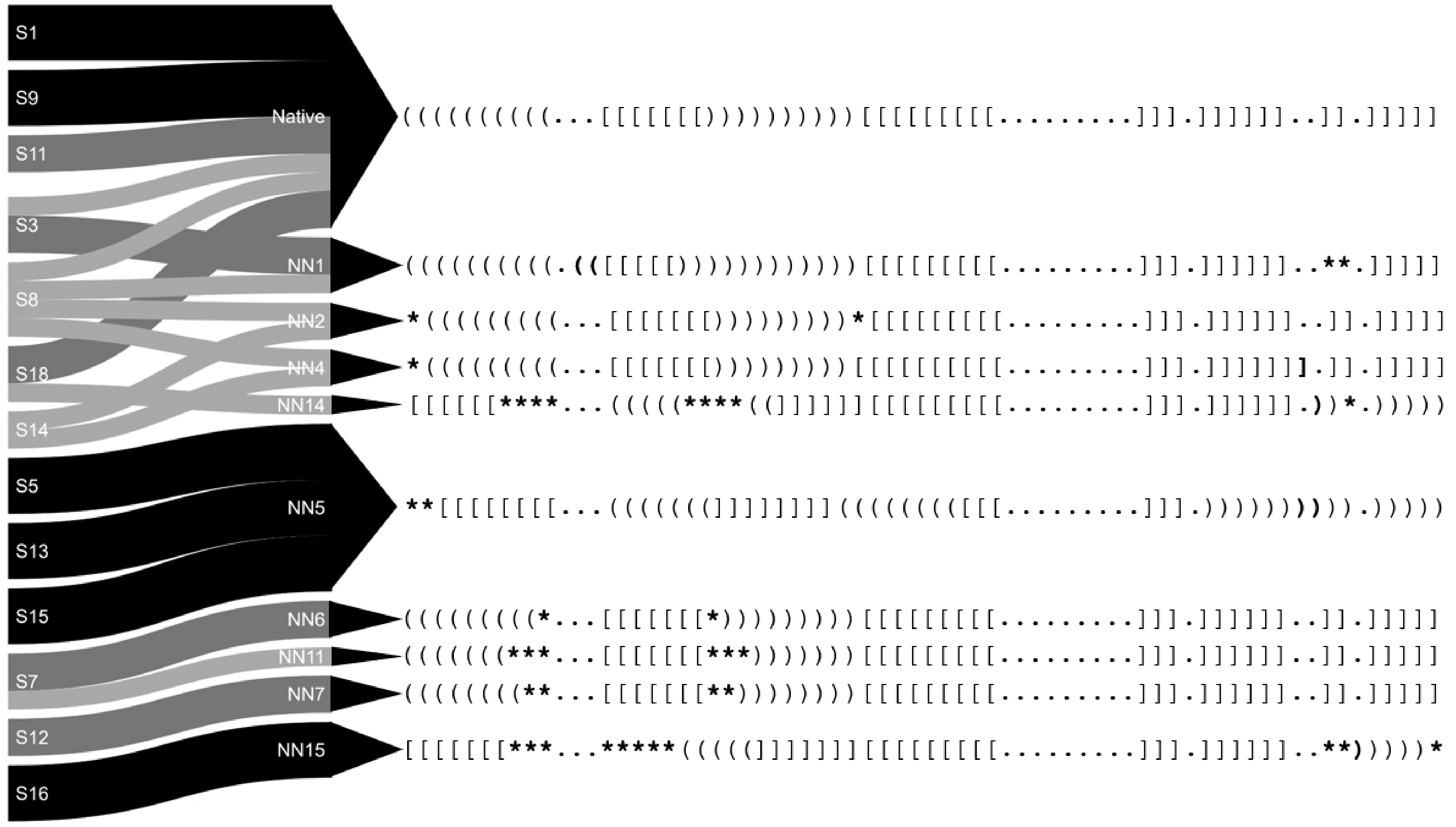
SHAPE data

Secondary structure predictions

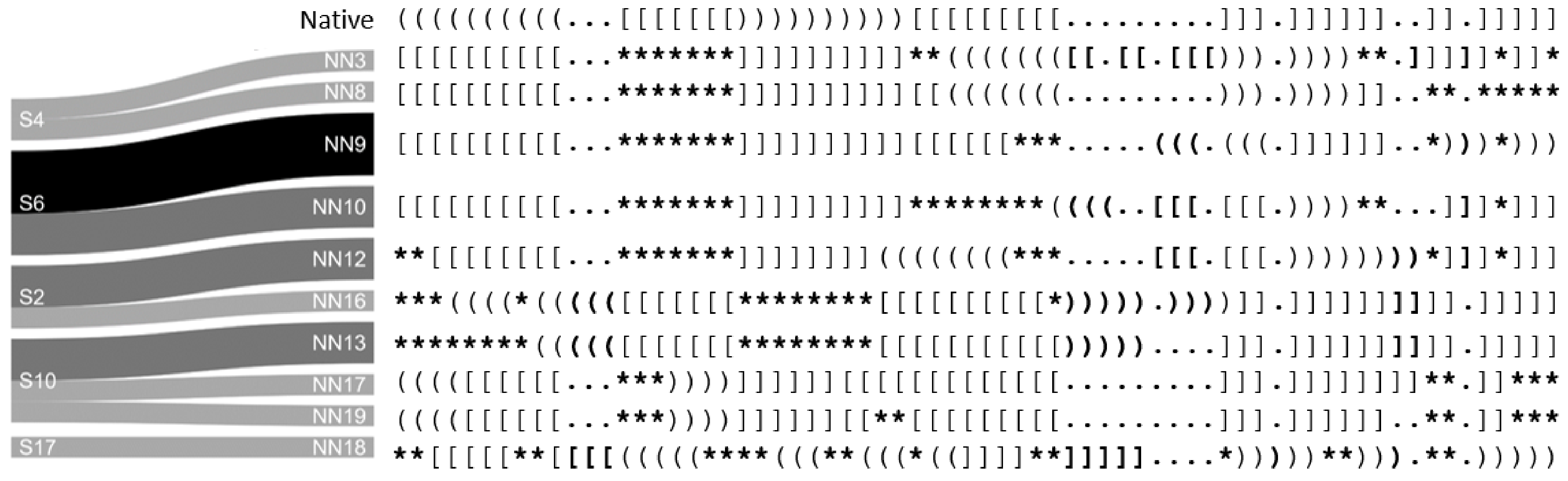
Results


0.0
−2.5
−5.0
−7.5
−10.0
−12.5
−15.0
−17.5
Free Energy
ShapeKnots
Shapify
Prediction

Native pathway redundancy



Less redundancy, less convergence



Future Work

- Partition function for pseudoknotted structures
    - Allow more diverse class of structures
    - Condition on pseudoknot-free structure

Thank you!

References

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