Supplementary Information

Supplementary Methods

Determination of the accuracy of secondary structure predictions. The PDB was searched

for single-stranded RNA structures > 20 nucleotides, released between Apr 2019–Feb 2020,

leading to a total of 17 X-ray crystal and cryo-electron microscopy structures ranging in size

from 31–867 nucleotides (Table S1). The total number of Watson-Crick pairs within each RNA

tertiary structure was counted from the reference secondary structure, which was retrieved using

the RNAPDBee server. The sequence used for structural determination was used as input for

Mfold (1), RNAfold (2), Sfold (3) and RNAstructure (4). Default parameters were used in all

cases. A score was given to each top prediction, according to the following formula: Score =

$[N_{corr} \times (N_{ref} - N_{incorr})] / N_{ref}^2$ (N_{ref}, number of Watson-Crick pairs in the reference structure;

N_{corr}, number of correct Watson-Crick pairs in the predicted model; N_{incorr}, number of Watson-

Crick pairs present in the model but not in the reference structure). The average of five scores for

each RNA was used to make Figure 5.

Supplementary Table

Table S1. Statistics for RNA structures used to generate Figure 5. RNAs are ranked by
increasing percentage of Watson-Crick pairs (5th column).

name	PDB ID	release date	length (nt)	% of nt in WC pairs	# WC bp in reference structure**	# correct WC bp of Mfold prediction	# incorrect WC bp of Mfold prediction	# correct WC bp of RNAfold prediction (MFE)	# incorrect WC bp of RNAfold prediction (MFE)	# correct WC bp of RNAfold prediction (centroid)	# incorrect WC bp of RNAfold prediction (centroid)	# correct WC bp of Sfold prediction	# incorrect WC bp of Sfold prediction	# correct WC bp of RNAstructure prediction	# incorrect WC bp of RNAstructure prediction	Average score
Pistol ribozyme	6UEJ	18-Dec-2019	51	23.5	6	0	10	0	14	0	6	0	11	0	10	
					SCORE	0.00		0.00		0.00		0.00		0.00		0.00
iMango-III fluorescent aptamer	6PQ7	31-Jul-2019	37	37.8	7	6	4	0	10	6	0	6	0	0	10	
					SCORE	0.37		0.00		0.86		0.86		0.00		0.42
Corn aptamer	6E80	31-Jul-2019	36	38.9	7	4	5	0	7	4	4	4	5	4	7	
					SCORE	0.16		0.00		0.24		0.16		0.00		0.11
pri-miRNA-449c-YdaO riboswitch fusion	6N5K	27-Nov-2019	125	46.4	29	26	9	22	15	12	24	23	11	6	33	
					SCORE	0.62		0.37		0.07		0.49		-0.03		0.30
U-A-U-rich RNA triple helix with 11 consecutive base triples	6SVS	1-Jan-2020	79	48.1	19	14	10	19	5	19	0	15	10	19	5	
					SCORE	0.35		0.74		1.00		0.37		0.74		0.64
Bacillus subtilis glyQS T-box riboswitch	6POM	20-Nov-2019	155	51.6	40	29	21	36	8	36	7	36	8	37	9	
					SCORE	0.34		0.72		0.74		0.72		0.72		0.65
L11-binding domain from E. coli 23S rRNA	6PRV	8-Jan-2020	58	51.7	15	13	5	13	5	13	5	13	5	13	5	
					SCORE	0.58		0.58		0.58		0.58		0.58		0.58
Group II intron retroelement	6MEC	14-Aug-2019	867	53.5	232	197	63	209	54	209	49	*	*	204	54	
					SCORE	0.62		0.69		0.71		-		0.67		0.67
Tetrahydrofolate riboswitch aptamer bound to 5-deazatetrahydropterin	6O57	18-Dec-2019	89	56.2	25	23	7	23	7	21	7	22	7	22	7	
					SCORE	0.66		0.66		0.58		0.67		0.63		0.64
T-box riboswitch discriminator	6PMO	20-Nov-2019	66	57.6	19	19	2	19	2	19	2	19	2	19	2	
					SCORE	0.89		0.89		0.89		0.89		0.89		0.89
phi29 pRNA domain II	6JXM	21-Aug-2019	97	57.7	28	27	2	27	2	26	2	27	2	26	1	
					SCORE	0.90		0.90		0.86		0.90		0.90		0.89
Manganese riboswitch	6NV2	2-Oct-2019	99	64.6	32	32	1	32	2	32	2	32	1	32	1	
					SCORE	0.97		0.94		0.94		0.97		0.97		0.96
HAV IRES domain V	6MWN	14-Aug-2019	92	65.2	30	26	6	26	6	26	5	26	6	26	6	
					SCORE	0.69		0.69		0.72		0.69		0.69		0.70
Mini tetraloop-tetraloop receptor	6DVK	26-Jun-2019	95	65.3	31	31	4	31	4	31	4	31	1	31	4	
					SCORE	0.87		0.87		0.87		0.97		0.87		0.89
Adenovirus-associated RNA	6OL3	3-Jul-2019	112	76.8	43	42	0	43	0	43	0	42	0	43	0	
					SCORE	0.98		1.00		1.00		0.98		1.00		0.99
SRS2 fragment of Rgs4 3' UTR	6HUR	17-Apr-2019	31	83.9	13	13	0	13	0	13	0	13	0	13	0	
					SCORE	1.00		1.00		1.00		1.00		1.00		1.00
helix 45 at 3' end of 12S rRNA	6AAS	5-Jun-2019	28	85.7	12	12	0	12	0	12	0	12	0	12	0	
					SCORE	1.00		1.00		1.00		1.00		1.00		1.00

Bibliography

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