

Supplemental Material to:

**Karine Lapouge, Remo Perozzo, Justyna Iwaszkiewicz,
Claire Bertelli, Vincent Zoete, Olivier Michielin,
Leonardo Scapozza, Dieter Haas**

**RNA pentaloop structures as effective targets of
regulators belonging to the RsmA/CsrA protein family**

2013; 10(6)

<http://dx.doi.org/10.4161/rna.24771>

www.landesbioscience.com/journals/rnabiology/article/24771/

SUPPLEMENTARY DATA:**Supplementary Table S1.** Theoretical binding free energies (ΔG) \pm SD for complex formation between RsmE and four RNA oligomers^a

Gene	RNA loop sequence ^b	ΔG_1 (kcal/mol)	ΔG_2 (kcal/mol)
<i>hcnA</i>	ACGGAU	-87.2 \pm 13.6	-69.3 \pm 14.5
<i>hcnA</i> Δ U	ACGGA-	-70.4 \pm 11.6	-60.7 \pm 11.7
<i>pltA</i>	AGGGA-	-81.5 \pm 13.8	-66.7 \pm 10.2
<i>pltA</i> +U	AGGGAU	-91.2 \pm 14.0	-85.2 \pm 12.2

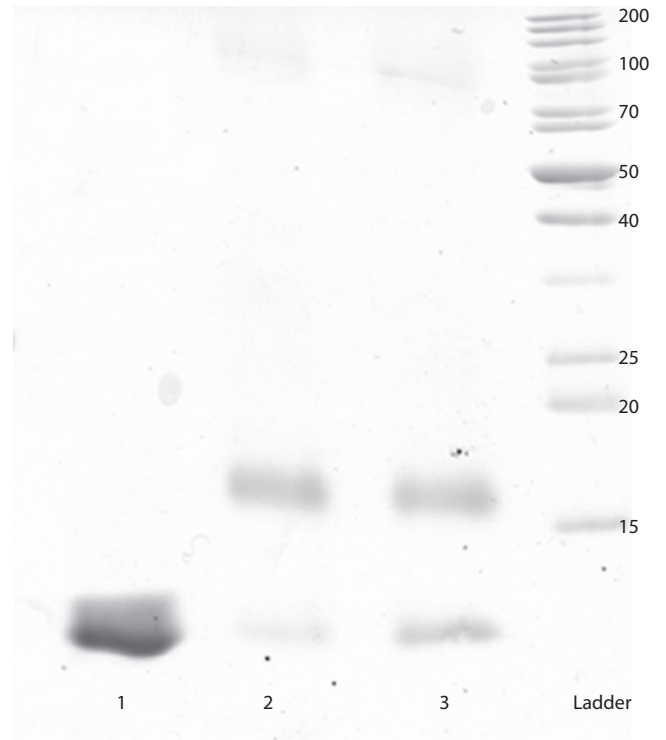
^a The ΔG values were obtained by the MM-GBSA method as described in Materials and Methods and are averaged over four molecular dynamic trajectories. The RNA oligomers having higher (ΔG_1) and lower (ΔG_2) affinities for RsmE were treated separately.

^b -: no nucleotide.

Supplementary Table S2. Oligonucleotides used in this study

Oligonucleotides	Sequence (5'→3')
6628	CCCATTCAATTTTTCGCGGATGAACCCAGCATG
6628rev	CTGGGTTTCATCCGCGAAAAATGAATGGGGTAC
9512	CCCATTCAATTTTTCACGGAGAACCCAGCATG
9512rev	CTGGGTTCTCCGTGAAAAATGAATGGGGTAC
9524	CCCAGTGCGCCTAACAGGGAGTGGGGCATG
9524rev	CCCCACTCCCTGTTAGGCGCACTGGGGTAC
9525	CCCAGTGCGCCTAACAGGGATGTGGGGCATG
9525rev	CCCCACATCCCTGTTAGGCGCACTGGGGTAC
9536	CTTCTGAAAAGAATGGAATCAAGAGGAGCATG
9536rev	CTCCTCTTGATTCCATTCTTTTCAGAAGGTAC
9537	CTTCTGAAAAGAATGGATCAAGAGGAGCATG
9537rev	CTCCTCTTGATCCATTCTTTTCAGAAGGTAC
10001	CCCATTCAATTTTTCAGGGAGAACCCAGCATG
10001rev	CTGGGTTCTGGGTGAAAAATGAATGGGGTAC
10002	CCCATTCAATTTTTCAGGAAGAACCCAGCATG
10002rev	CTGGGTTCTTCCTGAAAAATGAATGGGGTAC
10101	CCCAGTGCGCCTAACAAGGAGTGGGGCATG
10101rev	CCCCACTCCTTGTTAGGCGCACTGGGGTAC
10102	CCCATTCAATTTTTCGCGGACGAACCCAGCATG
10102rev	CTGGGTTTCGTCCGCGAAAAATGAATGGGGTAC

Supplementary Figure S1. RsmE cross-linking experiments. 12% sodium dodecyl sulfate polyacrylamide gel electrophoresis of RsmE cross-linking experiment: glutaraldehyde performed as described in ref 52. Lane 1 : RsmE (monomer) lanes 2 and 3 : different concentrations of RsmE treated with glutaraldehyde ladder: molecular weight markers.



Supplementary Figure S2. Correlation between mean computed ΔG values (Table 3) and experimentally obtained ΔG values (Table 2). The theoretical values were averaged over all trajectories; values for distinct RNA chains were treated separately and divided into groups of lower and higher ΔG observed per system. The assumption was made that the lower values represent the ΔG of the first RNA chain bound to the protein, and the higher ΔG values represent the second chain. Panel A shows the correlation between the lower theoretical ΔG values and the experimental ΔG_1 values; panel B shows the correlation between the higher theoretical ΔG values and the experimental ΔG_2 values. Purple star – *hcnA*; blue cross – *hcnA\Delta U*; green cross – *pltA*; blue square – *pltA+U*.

