Supplementary Information

- 2 Predicting Selective RNA Processing and Stabilization operons and their protein
- 3 stoichiometry via genome sequence

- 4 Yogendra Bhaskar^{1,3}, Xiaoquan Su¹, Chenggang Xu², Jian Xu^{1,3,*}
- ⁵ Single-Cell Center and CAS Key Laboratory of Biofuels and Shandong Key Laboratory of
- 6 Energy Genetics, Qingdao Institute of Bioenergy and Bioprocess Technology, Chinese
- 7 Academy of Sciences, Qingdao, Shandong, 266101, China
- ⁸ Key Laboratory of Chemical Biology and Molecular Engineering of Ministry of Education,
- 9 Institute of Biotechnology, Shanxi University, Taiyuan, Shanxi, 030006, China
- ³ University of Chinese Academy of Sciences, Beijing, 100049, China
- *Correspondence: Tel: +86 532 8066 2651; Fax: +86 532 8066 2654
- 12 E-mail address: xujian@qibebt.ac.cn

- 13 This file contains following materials:
- 1. Supplementary Figure.
- 15 2. Supplementary Tables.

Supplementary Figure

16

17

18

19

20

21

22

23

100 -**S**1 Percentage of intergenic Stem-loops S₂ **S**3 **S**4 75 -60 % cutoff 50 -25 -0 2500 5000 7500 10000 Number of Stem-loops

Figure S1: Selection of Stable stem-loops based on the four stability factors. Stable stem-loops were extracted from the millions of genome mapped stem-loops. Three stability factors for each stem-loop were calculated (Materials and Methods), and the stability factor which harbors the most number of intergenic stem-loops per 100 stem-loops was used with the 60% cutoff. Stability factor 4 (S₄) harbored the most number of intergenic stem-loops.

Supplementary Tables

Table S1. Bacterial genomes used in evaluating the SLOFE method.

Organism name	Genome size	RefSeq	Stable SLs	SRPS operons	Bi-cistronic operons
Ruminiclostridium cellulolyticum H10	4.07 mb	NC_011898.1	1441	53	11
Clostridium acetobutylicum ATCC 824	3.94 mb	NC_003030.1	2158	45	9
Clostridium thermocellum ATCC 27405	3.84 mb	NC_009012.1	1007	32	7
Bacillus subtilis Str. 168	4.22 mb	NC_000964.3	1829	42	11
Escherichia coli Str. K-12 substr. MG1655	4.64 mb	NC_000913.3	177	-	-

Table S2. Calculation of the normalized read-depth difference (NRD) for the predicted SRPS SLs in *Ccel.* The Normalized Read-depth Difference (NRD) data in cellulose, cellobiose and glucose carbon substrates from the dRNA-Seq study was used to calculate the difference in the read-depth of two neighboring genes flanked around the SLs. "Bi" denotes the bi-cistronic operon.

				Cellulos	e		Cellobiose			Glucose			
Stem-loop	Operon	ΔG	5' gene read- depth	3' gene read- depth	NRD	5' gene read-depth	3' gene read- depth	NRD	5' gene read-depth	3' gene read- depth	NRD	Max NRD	Remarks
SL_RS00005	1	-19.7	175	159	0.091429	2136	1695	0.206461	1429	874	0.388383	0.388383	non-SRPS
SL_RS00055	4-Bi	-18	221	183	0.171946	118	58	0.508475	94	57	0.393617	0.508475	SRPS
SL_RS00075	6	-14.1	17	49	-0.65306	140	84	0.4	181	100	0.447514	0.447514	non-SRPS
SL_RS00440	42	-18.4	656	17	0.974085	3094	74	0.976083	5422	268	0.950572	0.976083	SRPS
SL_RS00755	80	-23.2	795	90	0.886792	92	13	0.858696	130	43	0.669231	0.886792	SRPS
SL_RS01335	142	-25.2	967	132	0.863495	5526	1370	0.752081	6277	2499	0.60188	0.863495	SRPS
SL_RS01350	142	-14.7	430	168	0.609302	3505	1799	0.486733	8393	3972	0.526748	0.609302	SRPS
SL_RS01680	170-Bi	-16.2	147	1	0.993197	554	13	0.976534	850	19	0.977647	0.993197	SRPS
SL_RS01850	190	-15	34	59	-0.42372	205	307	-0.33224	197	446	-0.55829	-0.33224	non-SRPS
SL_RS02130	216	-24.4	-	-	-	-	-	-	-	-	-	0	-
SL_RS02230	228-Bi	-21.2	1858	1709	0.080194	16161	14943	0.075367	5538	5244	0.053088	0.080194	non-SRPS
SL_RS02395	237	-16.8	1326	1568	-0.15433	2192	1744	0.20438	4662	3320	0.287859	0.287859	non-SRPS
SL_RS02895	288	-19.9	-	-	-	-	-	-	-	-	-	0	-
SL_RS02990	295-Bi	-16.2	246	0	1	1043	8	0.99233	1393	13	0.990668	1	SRPS
SL_RS03180	314	-18.6	208	21	0.899038	733	41	0.944065	1036	226	0.781853	0.944065	SRPS
SL_RS03695	376	-23.5	24668	8503	0.655302	28656	14389	0.497871	3968	2052	0.482863	0.655302	SRPS
SL_RS03700	376	-26.8	85031	351	0.995872	14389	331	0.976996	2052	153	0.925439	0.995872	SRPS

SL_RS03710	376	-14.5	547	5705	-0.90411	550	4232	-0.8700	203	950	-0.78631	-0.78631	SRPS
SL_RS03715	376	-26.2	5705	50	0.991236	4232	100	0.976371	950	33	0.965263	0.991236	SRPS
SL_RS03740	376	-16.3	73	4	0.945205	133	6	0.954887	48	5	0.895833	0.954887	SRPS
SL_RS03930	391	-17.3	499	744	-0.32930	1296	1894	-0.31573	1652	2377	-0.30500	-0.30500	non-SRPS
SL_RS03960	391	-20.7	2725	130	0.952294	6440	578	0.910248	5536	993	0.820629	0.952294	SRPS
SL_RS04310	432-Bi	23.7	18	12	0.333333	116	42	0.637931	276	45	0.836957	0.836957	SRPS
SL_RS05015	495	-20	84	12	0.857143	26	8	0.692308	61	24	0.606557	0.857143	SRPS
SL_RS05150	511	-18.5	2721	133	0.951121	940	118	0.874468	122	32	0.737705	0.951121	SRPS
SL_RS05250	514	-26.1	880	394	0.552273	346	133	0.615607	77	54	0.298701	0.615607	SRPS
SL_RS05495	545	-22	260	3267	-0.92041	148	1093	-0.86459	67	1273	-0.94736	-0.86459	non-SRPS
SL_RS05655	566-Bi	-28.4	169	139	0.177515	814	503	0.382064	1194	681	0.429648	0.429648	non-SRPS
SL_RS05685	569	-17.9	2045	128	0.937408	94	3	0.968085	72	14	0.805556	0.968085	SRPS
SL_RS06165	617	-16.6	7	4	0.428571	39	11	0.717949	83	80	0.036145	0.717949	SRPS
SL_RS06175	617	-14.4	28	5	0.821429	48	42	0.125	84	80	0.047619	0.821429	SRPS
SL_RS06180	617	-18.4	5	3	0.4	42	15	0.642857	80	34	0.575	0.642857	SRPS
SL_RS06215	617	-24.7	7	3	0.571429	19	19	0	71	103	-0.31067	0.571429	SRPS
SL_RS06275	622	-18.2	62	7	0.887097	175	9	0.948571	147	16	0.891156	0.948571	SRPS
SL_RS06525	632	-16.8	-	-	-	-	-	-	-	-	-	0	-
SL_RS07065	693	-16.7	55	14	0.745455	421	261	0.380048	940	246	0.738298	0.745455	SRPS
SL_RS07075	693	-28.7	450	10	0.977778	4738	248	0.947657	878	111	0.873576	0.977778	SRPS
SL_RS07235	716	-19.6	93	10	0.892473	436	44	0.899083	870	131	0.849425	0.899083	SRPS
SL_RS07520	746	-24	3253	650	0.800184	1729	357	0.793522	2562	764	0.701795	0.800184	SRPS
SL_RS07530	746	-17.8	546	183	0.664835	247	100	0.595142	271	182	0.328413	0.664835	SRPS
SL_RS08285	813	-20.3	16	2	0.875	104	26	0.75	61	21	0.655738	0.875	SRPS
SL_RS08610	849-Bi	-28	279	105	0.623656	1481	636	0.57056	4551	1413	0.689519	0.689519	SRPS

SL_RS08720	863	-19.7	2136	36	0.983146	6982	197	0.971785	5963	226	0.9621	0.983146	SRPS
SL_RS09085	898	-15.5	1879	129	0.931346	41895	1795	0.957155	19769	759	0.961607	0.961607	SRPS
SL_RS09255	915	-16.2	41	3	0.926829	339	25	0.926254	417	51	0.877698	0.926829	SRPS
SL_RS10060	1000	-16.7	6219	223	0.964142	18300	2367	0.870656	24934	4374	0.824577	0.964142	SRPS
SL_RS10050	1000	-26.3	430	24	0.944186	2589	248	0.90421	4609	287	0.937731	0.944186	SRPS
SL_RS10295	1018	-15.4	139	214	-0.35046	440	649	-0.32203	580	786	-0.26208	-0.26208	non-SRPS
SL_RS10685	1052	-20	41638	1242	0.970171	79521	8757	0.889878	1191	54	0.95466	0.970171	SRPS
SL_RS10675	1052	-16.8	1599	2026	-0.21076	8982	17873	-0.49745	71	199	-0.64321	-0.21076	SRPS
SL_RS10860	1073	-16.2	-	-	-	-	-	-	-	-	-	0	-
SL_RS11420	1135-BI	-17.9	1528	238	0.844241	5991	910	0.848105	4475	1483	0.668603	0.848105	SRPS
SL_RS12550	1247	-18	38	18	0.526316	77	7	0.909091	215	59	0.725581	0.909091	SRPS
SL_RS12610	1254-Bi	-27	304	293	0.036184	537	1081	-0.50323	955	615	0.356021	0.356021	non-SRPS
SL_RS13360	1341-Bi	-22.4	62	11	0.822581	73	6	0.917808	393	28	0.928753	0.928753	SRPS
SL_RS13485	1354	-35.4	230	147	0.36087	481	467	0.029106	446	952	-0.53151	0.36087	non-SRPS
SL_RS13510	1358-Bi	-16.2	3	0	1	5	1	0.8	16	6	0.625	1	SRPS
SL_RS13525	1359-Bi	-27.5	105	4	0.961905	178	6	0.966292	307	8	0.973941	0.973941	SRPS
SL_RS13720	1382	-23	-	-	-	13	29	-0.55172	24	43	-0.44186	-0.44186	non-SRPS
SL_RS14235	1435	-23.3	-	-	-	-	-	-	-	-	-	0	-
SL_RS14390	1445	-18.4	-	-	-	-	-	-	-	-	-	0	-
SL_RS14525	1466-Bi	-16.2	-	-	-	139	1	0.992806	259	2	0.992278	0.992806	SRPS
SL_RS14630	1477-Bi	-16.7	13	39	-0.66666	123	39	0.682927	105	26	0.752381	0.752381	SRPS
SL_RS15510	1560	-18.4	161	74	0.540373	669	389	0.418535	686	477	0.304665	0.540373	SRPS
SL_RS15870	1600	-25.9	-	-	-	-	-	-	-	-	-	0	-
SL_RS17245	1745	-18.6	889	1962	-0.54689	4635	5332	-0.13072	6528	8018	-0.18583	-0.13072	SRPS

Table S3. Correlation between SLOFE-predicted transcript ratio and those experimentally measured for selected operons from *Ccel*, *Cace*, *Cthe* and *Bsub*. These operons have skewed transcript ratios as predicted by SLOFE.

Operon ID	Organism	Predicted ratio	Correlation with transcript	Plot	Annotation
142	Clostridium cellulolyticum	1.00:1.00:0.58:0.58:0.58:0 .62:0.62:0.62	0.698		ATP synthase
376	Clostridium cellulolyticum	1.00:1.14:0.62:0.62:1.11:0 .69:0.69:0.69:0.69:0.69:0. 88:0.88	0.751		Cellulosome
693	Clostridium cellulolyticum	1.00:1.72:1.72:0.91:0.91:0 .91:0.91	0.556		ABC transporter
1000	Clostridium cellulolyticum	0:0:0:1.00:1.00:1.26	0.940		Two- component system
593	Clostridium acetobutylicum	1.00:1.00:0.53:0.53:0.53:0 .53:0.53:0.53:0.53:0.53:0. 53:0.82	0.701	0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	Cellulosome
1068	Clostridium acetobutylicum	0:0:1.45:1.45:1.00:1.00:1. 00:1.00:1.00:1.00:1.00:1.0	0.672		Cell division protein and lipoprotein
482	Clostridium thermocellum	1.00:1.00:0.97:0.97	0.672	100 100 100 100 100 100 100 100 100 100	Amino acid- binding protein
1135	Clostridium thermocellum	0:0:0:1.00:1.00:0.98	0.460		Restriction endonuclease Protein

-				
531	Clostridium thermocellum	1.00:1.09:1.09:1.09	0.802	Magnesium chelatase
679	Bacillus subtilis	1.00:0.90:0.90:0.90:0.90:0	0.773	ABC transporter
1491	Bacillus subtilis	0:0:0:0:1.12:1.12:1.01:1.0 1:1.00	0.762	Chaperone protein
1513	Bacillus Subtilis	0.8318:0.75:1.00	0.980	Mother cell lysis

35 Table S4. SLOFE-predicted ratios of the SRPS operons from *Ccel* (A), *Cthe* (B), *Cace* (C)

and **Bsub** (**D**). SLOFE predicted ratios for all the SRPS operons using ΔG of SLs.

37 **(A)** SLOFE-predicted ratios of the SRPS operons from *Ccel*.

1 4 1.00:0:0:0 4 2 1.00:0.89 6 3 1.00:1.00:1.00:1.00:1.00:1.00:1.00:0 80 4 1.00:0.94:0.94:0.94 142 8 1.00:1.00:0.58:0.58:0.58:0.62:0.62:0.62 170 2 1.00:0.96 190 4 1.00:1.00:1.5:1.5 216 3 1.00:0:0 228 2 1.00:0.91 237 3 1.00: 1.00:0 288 5 1.00:0.0:0:0 295 2 1.00:0.85	
6 3 1.00:1.00:1.354 42 9 1.00:1.00:1.00:1.00:1.00:1.00:1.00:0 80 4 1.00:0.94:0.94:0.94 142 8 1.00:1.00:0.58:0.58:0.58:0.62:0.62:0.62 170 2 1.00:0.96 190 4 1.00:1.00:1.5:1.5 216 3 1.00:0.0 228 2 1.00:0.91 237 3 1.00: 1.00:0 288 5 1.00:0:0:0:0	
42 9 1.00:1.00:1.00:1.00:1.00:1.00:1.00:0 80 4 1.00:0.94:0.94:0.94 142 8 1.00:1.00:0.58:0.58:0.58:0.62:0.62:0.62 170 2 1.00:0.96 190 4 1.00:1.00:1.5:1.5 216 3 1.00:0.0 228 2 1.00:0.91 237 3 1.00: 1.00:0 288 5 1.00:0:0:0:0	
80 4 1.00:0.94:0.94:0.94 142 8 1.00:1.00:0.58:0.58:0.58:0.62:0.62:0.62 170 2 1.00:0.96 190 4 1.00:1.00:1.5:1.5 216 3 1.00:0:0 228 2 1.00:0.91 237 3 1.00: 1.00:0 288 5 1.00:0:0:0:0	
142 8 1.00:1.00:0.58:0.58:0.58:0.62:0.62 170 2 1.00:0.96 190 4 1.00:1.00:1.5:1.5 216 3 1.00:0.0 228 2 1.00:0.91 237 3 1.00: 1.00:0 288 5 1.00:0:0:0:0	
170 2 1.00:0.96 190 4 1.00:1.00:1.5:1.5 216 3 1.00:0:0 228 2 1.00:0.91 237 3 1.00: 1.00:0 288 5 1.00:0:0:0:0	
190 4 1.00:1.00:1.5:1.5 216 3 1.00:0:0 228 2 1.00:0.91 237 3 1.00: 1.00:0 288 5 1.00:0:0:0:0	
216 3 1.00:0:0 228 2 1.00:0.91 237 3 1.00: 1.00:0 288 5 1.00:0:0:0:0	
228 2 1.00:0.91 237 3 1.00: 1.00:0 288 5 1.00:0:0:0:0	
237 3 1.00: 1.00:0 288 5 1.00:0:0:0:0	
288 5 1.00:0:0:0	
295 2 1.00:0.85	
314 3 1.00:0.989:0.989	
376 12 1.00:1.14:0.62:0.62:1.11:0.69:0.69:0.69:0.69:0.69:0.88:0.88	
391 24 1.00:1.00:1.00:1.00:1.00:1.00:1.00:1.00):1.00:1.
00:1.00:1.2:1.2:1.2:1.2:1.2:0:0	
432 2 1.00: 1.00	
495 3 1.00:0.805:0.805	
511 9 1.00:0:0:0:0:0:0:0	
514 11 1.00:1.00:1.00:1.00:1.00:1.00:0.74:0.74:0.74	
545 3 1.00: 1.00:0.95	
566 2 1.00:0:0	
569 3 1.00:0:0	
617 14 1.00:1.00:0.86:0.86:0.97:1.48:1.48:1.48:1.48:1.48:1.48:1.48:0:0	
622 4 1.00:1.00:0	
693 6 1.00:1.72:1.72:0.91:0.91:0.91	
716 3 1.00:0:0	
746 6 1.00:0.74:0.74:0.78:0.78	
813 7 1.00:1.00:1.00:1.17:1.17:1.17	

849	2	0:1.00
863	5	0.845:1.00:1.00:1.00
898	7	0:0:0:0:1.00:1.00
915	2	1.00:0:0
1000	6	0.8859:0.8859:0.8859:1.00:1.00:1.26
1018	13	0:0:0:0:0:0:0:1.00:1.00:1.00:1.00:1.00:
1052	4	0.84:0.64: 1.00
1073	3	1.00: 1.00:0.96
1135	2	0:1.00
1247	3	1.00:0:0
1254	2	0:1.00
1341	2	1.60:1.00
1354	3	0.5649:0.5649:1.00
1358	2	1.00:0.96
1359	2	0.57:1.00
1382	7	0:0:1.00:1.00:1.00:1.00
1435	16	0:0:0:0:0:0:0:1.00: 1.00: 1.00: 1.00: 1.00: 1.00: 1.00: 1.00: 1.00
1445	5	1.59:1.59: 1.00: 1.00: 1.00
1466	2	1.00:0.96
1477	2	1.00:0.76
1560	3	0.56: 1.00: 1.00
1600	5	0: 1.00: 1.00: 1.00: 1.00
1745	4	1.00:0.658:0.658:0.658

(B) SLOFE-predicted ratios of the SRPS operons from Cthe.

# operon	# of genes	Ratio)
275	5	1:1:1:0.77:0.77	
357	3	1:1:0.96	
482	4	1:1:0.97:0.97	
531	4	1:1.09:1.09:1.09	
548	7	1:1:1:1:1:1.33	
552	4	1:1:1:0.74	
728	3	0:0:1	
747	3	1:1:0.46	
791	5	1:1:0.99:0.99:0.99	

794	5	0:0:1:1:1
804	23	0:1:1:1:1:1:0.74:0.74:0.74:0.65:0.65:0.98:0.98:0.98:0.98:0.98:0.98:0.98:0.74:0.74 4:0.74
806	7	0:0:0:0:1:1:1
938	3	0:0:1
957	8	0:0:0:0:1:1:1:1
1135	6	0:0:0:1:1:0.98
1209	3	0:0:1
1228	5	1:1:1:0:0
1353	5	1:1:1:0:0
1359	4	1:1:1:0.82
1395	8	1:1:1:0.97:0.97:0.97:0.97
1465	3	1:1:0.65
1487	6	1:0.93:0.93:0.93:0.93
1522	3	1:1.52:1.52
1536	6	1:1:1:0:0:0

42 (C) SLOFE-predicted ratios of the SRPS operons from *Cace*.

# operon	# of genes	Ratio
120	5	1.00:1.00:1.21:1.21:0
205	6	1.00:1.00:0:0:0:0
216	2	1.00:0.58
239	5	1.00:1.00:1.00:0.77:0.77
244	10	1.00:1.00:1.00:1.00:1.00:0.83:0.83:0.83:0.83:0.83
304	7	1.00:0.90:0.90:1.14:1.14:1.14:0
317	2	1.00:0.48
356	3	1.00:1.00:1.36
362	6	1.00:1.00:1.29:1.29:1.29
401	3	1.00:1.2013:1.20
466	2	1.00:1.39
481	4	1.00:0.89:1.24:0
593	9	1.00:1.00:0.53:0.53:0.53:0.53:0.53:0.53:0.53:0
614	2	1.00:1.12
633	2	1.00:0:0
635	3	1.00:1.00:0.80

673	2	1.00:1.24
715	8	1.00:1.00:1.00:1.00:1.00:1.00:0:0
730	10	1.00:1.7:1.7:1.7:1.7:1.7:1.7:1.7
738	5	1.00:1.13:1.03:1.03:1.03
789	6	1.00:1.00:1.10:1.10:1.10
849	3	0:0:1.00
909	11	1.00:1.00:1.00:1.00:1.00:1.00:1.00:1.00
910	2	0:1.00
943	2	1.00:0:0
944	17	1.00:1.00:1.00:1.00:1.00:1.00:1.00:1.00
949	9	1.00:1.00:1.00:1.00:1.02:1.02:1.20:1.20:
965	11	1.00:0.74:0.74:0.74:0.74:0.74:0.74:0.84:0.84:0.84
967	7	1.00:1.00:1.00:1.00:0:0:0
981	2	1.00:0.57
986	4	0:1.00:1.00:1.00
1008	26	1.00:1.16:1.16:1.16:1.16:1.16:1.16:1.16:
1006	20	1.14:1.14:1.14:1.14:1.14:1.14:1.14:1.14
1068	12	0:0:1.45:1.45:1.00:1.00:1.00:1.00:1.00:1.00:1.00:1.0
1090	6	0:1.02:1.21:1.21:1.38:1.00
1132	4	0:0:1.52:1.31
1248	9	0:0:1.00:1.00:1.00:1.00:1.00:1.00
1283	6	0:1.17:1.17:1.17:1.00:1.00
1336	3	1.00:1.00:1.08
1359	2	1.00:1.35
1362	2	1.00:1.01
1412	2	1.00:0:0
1454	5	1.00:1.09:1.09:0.82:0.82
		1.00:1.00:1.00:1.00:0.92:0.92:0.92:0.92:0.92:0.92:0.92:0
1519	35	0.92: 0.92
		1.03:1.03:1.03
1522	4	0:0:1.00:1.00
1537	5	1.00:1.05:1.05:1.05:1.05
1553	9	0:0:0:0:0.66:0.78:0.78:0.78:1.00
1566	4	1.00:1.00:1.29:1.29
1684	3	1.00:1.00:0.75

1749	2	1.00:0.96
1784	2	1.00:2.07
1793	5	1.00:1.00:1.00:1.00:1.16

(**D**) SLOFE-predicted ratios of the SRPS s from *Bsub*.

#	#of genes	Ratio
40	6	1.00:1.00:1.00:1.00:0:0
44	5	1.00:1.00:1.00:1.00:1.23
47	7	1.00:1.00:0.64:0.64:0.64:0.64:0.71
49	31	1.00:1.00:1.00:1.00:1.00:0.91:0.91:0.91:
47	31	1: 0.91: 0.91: 0.91: 0.91: 0.91: 1.85: 1.85: 1.85: 1.85: 1.85: 1.85: 1.85: 1.85: 1.32: 1.32
130	3	1.00:0.93:0.93
136	3	1.00:1.00:0.90
200	4	1.00:1.00:1.00:0
341	3	1.00:1.07:1.07
361	12	1.00:1.00:1.00:1.06:1.06:1.06:1.06:1.06:
394	4	1.00:0:0:0
406	3	1.00:0.99:0.78
460	3	1.00:0:0
679	6	1.00:0.90:0.90:0.90:0.90:0
692	7	1.00:1.00:1.00:1.00:1.00:0
744	10	1.00:0.89:0.89:0.89:0.89:0.89:0.89:0.89
836	5	1.00:1.00:1.00:1.12:1.12
925	11	1.00:1.10:1.10:1.10:1.10:0.91:0.91:0:0:0
934	6	1.00:1.00:1.00:1.00:1.19:1.19
961	11	1.00:1.00:1.00:1.00:1.00:1.00:1.00:0.81:0.81
964	3	1.00:0:0
1242	3	0:0:1.00
1461	5	1.02:1.02:1.02:1.00
1491	9	0:0:0:0:1.12:1.12:1.01:1.00
1492	3	1.19:1.19:1.00
1513	3	0.83:0.75:1.00
1602	4	1.00:1.00:0.97:0.97
1672	4	0.87:1.00:1.00:1.00
1693	3	1.00:1.00:1.31
-		

1850	7	0:0.84:0.84:0.84:0.84:1.00
1894	5	0:0:0:0:1.00
1952	3	1.00:1.24:1.24
1956	4	0:0:0:1.00
1976	6	1.00:1.00:1.00:1.00:1.05:1.05
1990	13	1.00:1.00:1.00:1.00:1.00:0.80:0.80:0.80:
2016	8	0:0:0:0:0:1.00:1.00
2030	3	1.00:2.11:2.11
2279	8	1.00:1.00:1.00:1.00:0.64
2358	3	0:1.00:1.00
2363	4	0:0:0:1.00

Table S5. Pearson correlation coefficients between predicted ratio and experimentally measured ratio for the SRPS operons of *Ccel*, for each of the six methods (CAI, RCA, RCBS, MELP, Gene-order and SLOFE). Correlations with the experimentally measured abundance of transcripts (A) and proteins (B) were both shown.

51 (A) Correlations with the experimentally measured abundance of transcripts.

#Operon	# of gene	CAI	MELP	RCBS	RCA	Gene- order	SLOFE
1	4	0.164	0.060	-0.221	0.441	0.726	0.488
6	3	0.863	-0.945	0.916	0.028	-0.341	0.914
42	9	0.121	-0.465	-0.547	0.042	0.496	0.676
80	4	0.901	0.546	0.930	0.845	0.892	0.975
142	8	0.817	-0.682	0.555	0.804	0.349	0.714
190	4	-0.261	-0.826	-0.274	0.768	0.387	0.166
237	3	0.168	-0.894	-0.711	-0.415	-0.360	0.423
376	12	0.667	0.472	0.508	0.565	0.820	0.752
391	24	0.159	0.226	-0.271	0.288	0.070	0.579
511	9	0.591	-0.439	0.519	0.533	0.813	0.901
514	11	-0.368	-0.522	-0.770	-0.132	0.342	0.412
545	3	0.953	0.826	-0.962	0.232	-0.091	-0.780
569	3	0.698	0.017	0.258	0.288	1.000	0.969
617	14	-0.184	-0.201	-0.321	-0.335	-0.513	0.036
622	4	0.768	0.312	0.510	0.620	-0.410	0.525
693	6	0.615	0.182	0.182	0.294	0.228	0.381
716	3	-0.264	0.672	-0.697	0.807	0.925	0.989
746	6	-0.157	-0.318	-0.449	-0.285	0.916	0.737
813	7	0.240	-0.297	0.151	0.583	0.045	0.232
863	5	-0.040	-0.421	-0.357	0.334	-0.166	0.547

898	7	0.806	-0.054	0.049	0.808	0.414	0.857
1000	6	0.973	0.539	0.884	0.931	0.926	0.972
1018	13	0.367	0.443	-0.165	0.128	0.375	0.400
1052	4	0.974	0.639	0.765	0.735	0.906	0.040
1247	3	0.998	0.159	0.652	0.977	-0.194	0.194
1354	3	-0.882	0.349	0.699	-0.829	0.612	0.784
1382	7	0.276	0.375	-0.929	0.750	0.670	0.886
1560	3	0.985	0.246	0.182	0.910	0.472	0.962
1745	4	0.249	-1.000	-0.924	-0.395	0.860	0.638
314	3	-0.915	-0.914	-0.991	-0.994	0.666	0.826
495	3	0.995	-0.388	0.967	0.999	0.992	0.993
Avei	rage	0.364	-0.074	0.004	0.333	0.414	0.587

(B) Correlations with the experimentally measured abundance of proteins. Dash (-) denotes no data available.

#	# of gene	CAI	MELP	RCBS	RCA	Gene-order	SLOFE
1	4	0.547	0.534	-0.659	-0.715	0.748	0.524
6	3	-	-	-	-	-	-
42	9	0.328	-0.361	-0.284	0.403	0.282	0.983
80	4	0.971	0.705	0.966	0.940	0.951	1.000
142	8	0.092	-0.439	-0.138	-0.115	0.178	0.060
190	4	0.531	0.420	0.242	-0.199	-0.903	0.507
237	3	-0.610	-0.278	0.035	0.388	0.442	0.953
363	3	0.734	-0.475	-0.515	0.987	0.971	1.000
376	12	0.584	0.771	0.419	0.322	0.576	0.283
391	24	-0.052	0.216	-0.168	0.146	0.236	0.076
511	9	-	-	-	-	-	-
514	11	-0.555	-0.352	-0.818	-0.138	0.405	0.822

545	3	0.444	0.683	-0.417	0.997	0.971	0.500
569	3	0.853	0.228	-0.012	0.515	0.971	1.000
617	14	-	-	-	-	-	-
622	4	-	-	-	-	-	-
693	6	0.292	0.634	0.624	-0.022	0.653	0.484
716	3	-0.118	-0.555	-0.583	0.885	0.971	1.000
746	6	0.185	-0.200	-0.516	-0.038	0.339	0.096
813	7	-0.189	-0.536	-0.214	0.170	0.743	1.000
863	5	0.306	-0.548	-0.514	0.505	-0.379	0.145
898	7	-0.008	-0.770	-0.659	0.482	0.475	0.626
1000	6	0.704	0.318	0.520	0.541	0.890	0.925
1018	13	0.382	0.392	0.401	-0.030	0.740	0.712
1052	4	0.957	0.391	0.568	0.528	0.791	0.333
1247	3	-	-	-	-	-	-
1354	3	-	-	-	-	-	-
1382	7	-	-	-	-	-	-
1560	3	0.995	0.500	-0.092	0.763	0.693	1.000
1745	4	0.764	-0.541	-0.162	0.529	-0.669	0.262
Ave	rage	0.354	0.032	-0.086	0.341	0.482	0.621
					•		

Table S6. Pearson correlation coefficients between predicted ratio and experimentally measured ratio for the SRPS operons of *Cthe*, for each of the six methods (CAI, RCA, RCBS, MELP, Gene-order and SLOFE). Correlations with the experimentally measured abundance of transcripts were shown.

#	#of genes	CAI	MELP	RCBS	RCA	Gene-order	SLOFE
357	3	-0.818	-0.157	-0.018	-0.684	0.682	0.037
531	4	-0.423	-0.764	0.073	0.522	-0.832	0.803
548	7	0.127	-0.192	0.248	0.360	0.727	0.601
552	4	0.787	0.418	0.392	-0.532	-0.312	0.467
728	3	-0.582	-0.986	-0.612	-0.301	0.167	-0.075
747	3	-0.855	-0.515	-0.250	0.394	-0.780	0.907
791	5	0.219	-0.367	-0.234	0.216	0.053	-0.094
794	5	0.210	-0.109	0.634	0.307	0.798	0.711
804	23	-0.087	0.281	0.051	0.406	-0.366	0.171
806	7	-0.293	-0.441	-0.330	-0.248	-0.343	-0.359
938	3	-0.783	0.197	0.921	0.788	1.000	0.977
957	8	-0.059	0.147	0.392	-0.467	0.050	0.408
1135	6	-0.702	-0.527	-0.364	0.177	0.186	0.461
1209	3	0.999	0.019	0.283	0.648	-0.739	-0.556
1228	5	0.255	-0.824	-0.053	0.505	0.070	-0.021
1359	4	0.630	0.987	0.467	-0.291	0.300	-0.527
1395	8	-0.007	0.311	-0.132	-0.307	-0.890	0.886
1465	3	0.859	-0.900	-0.941	0.900	0.104	0.789
1487	6	0.035	0.161	0.118	0.157	0.587	0.387
1536	6	-0.193	0.308	0.002	-0.422	0.424	0.873
Av	erage	-0.034	-0.148	0.032	0.106	0.044	0.342

Table S7. Pearson correlation coefficients between predicted ratio and experimentally measured ratio for the SRPS operons of Cace, for each of the six methods (CAI, RCA, RCBS, MELP, Gene-order and SLOFE). Correlations with the experimentally measured abundance of transcripts were shown. 64

62

#	#of genes	CAI	MELP	RCBS	RCA	Gene-order	SLOFE
205	6	0.367	-0.377	-0.566	-0.555	0.658	0.742
239	5	-0.217	-0.793	-0.487	0.376	0.639	0.970
304	7	0.271	-0.508	-0.420	-0.496	-0.451	0.101
356	3	0.277	0.146	0.396	0.894	-0.982	0.816
362	6	0.777	-0.352	-0.077	-0.132	-0.749	0.740
401	3	0.457	-1.000	-0.489	-0.010	-0.692	0.498
481	4	0.687	0.739	0.791	0.381	-0.998	-0.459
593	9	0.190	-0.327	0.039	-0.300	0.484	0.702
635	3	-0.511	-0.923	-0.979	0.409	0.907	0.932
715	8	-0.202	-0.617	-0.216	-0.031	-0.405	0.153
730	10	0.090	0.080	-0.684	-0.393	-0.877	0.615
738	5	-0.579	-0.069	-0.311	-0.109	0.076	0.923
849	3	0.904	-1.000	0.595	0.594	0.916	0.793
909	11	0.281	-0.179	-0.272	-0.184	-0.490	0.214
965	11	0.700	0.543	0.324	-0.617	0.590	0.224
967	7	0.677	-0.417	-0.466	0.048	0.876	0.464
1008	26	-0.269	0.589	-0.194	0.312	0.197	-0.019
1090	6	0.406	0.045	0.412	0.694	-0.155	0.191
1132	4	0.058	0.970	0.459	-0.718	0.721	0.331
1248	9	-0.238	0.308	0.111	-0.690	-0.173	0.217
1283	6	0.372	0.411	0.314	0.090	-0.651	-0.545
1336	3	0.998	-0.931	0.073	0.961	0.083	0.320
1454	5	-0.119	-0.217	-0.309	-0.890	-0.228	0.046
1519	35	0.220	0.161	0.213	0.219	-0.098	-0.451

1522	4	0.116	-0.252	-0.656	0.687	-0.776	0.598
1537	5	-0.220	-0.518	0.030	0.439	-0.167	0.117
1553	9	0.431	0.497	0.304	-0.678	-0.553	-0.199
1566	4	0.396	-0.159	0.047	0.462	-0.678	-0.166
1684	3	0.774	0.308	0.453	-0.358	-0.833	-0.179
1793	5	-0.180	-0.238	-0.315	0.073	0.040	0.106
Average		0.23048	-0.136	-0.0627	0.01606	-0.1256	0.2931

Table S8. Pearson correlation coefficients between predicted ratio and experimentally measured ratio for the SRPS operons of *Bsub*, for each of the six methods (CAI, RCA, RCBS, MELP, Gene-order and SLOFE). Correlations with the experimentally measured abundance of transcripts (A) and proteins (B) were both shown.

(A) Correlations with the experimentally measured abundance of transcripts.

#	#of genes	CAI	MELP	RCBS	RCA	Gene-order	SLOFE
40	6	0.524	-0.435	-0.299	0.283	-0.356	0.325
47	7	0.094	-0.459	0.007	0.320	-0.027	0.800
49	31	0.077	-0.447	-0.586	0.077	0.064	0.011
130	3	0.848	-0.870	0.997	0.999	0.998	0.985
200	4	-0.814	-0.331	-0.208	0.171	0.334	0.725
361	12	0.678	-0.779	-0.582	0.695	-0.289	0.157
394	4	-0.488	-0.411	-0.045	0.042	0.958	0.975
406	3	0.263	0.227	0.806	0.622	-0.791	0.636
460	3	-0.913	-0.900	-0.999	-0.723	0.614	0.786
679	6	0.014	0.061	0.697	0.037	0.865	0.773
744	10	0.239	-0.553	-0.251	0.095	0.430	0.538
836	5	0.657	0.969	0.467	0.715	-0.635	0.904
925	11	-0.129	0.046	0.318	0.117	-0.706	-0.114
934	6	0.887	0.182	0.060	0.244	-0.124	0.353
961	11	0.482	-0.450	0.022	-0.233	-0.608	-0.378
964	3	-0.346	0.475	0.154	0.110	0.758	0.892
1513	3	0.695	-0.901	-0.878	0.903	0.739	0.980
1672	4	-0.170	0.573	0.448	-0.485	0.511	0.991
1693	3	0.934	0.970	0.780	-0.737	0.953	0.998
1850	7	-0.403	-0.772	-0.484	0.209	0.083	0.392
1894	5	-0.211	-0.886	-0.685	-0.364	0.934	0.759
1952	3	-0.985	-1.000	-0.928	0.437	-0.884	-0.950

1956	4	0.840	-0.860	-0.613	0.502	0.947	0.934		
1976	6	0.685	0.766	0.693	-0.522	-0.755	-0.389		
1990	13	-0.411	-0.600	-0.632	-0.301	0.389	0.322		
2363	4	-0.921	-0.992	-0.442	-0.738	-0.570	-0.349		
Average		0.082	-0.284	-0.084	0.095	0.147	0.464		

(B) Correlations with the experimentally measured abundance of proteins.

#	#of genes	CAI	MELP	RCBS	RCA	Gene-order	SLOFE
40	6	0.643	-0.331	-0.170	0.402	-0.252	0.336
47	7	-0.900	0.061	0.516	-0.843	-0.567	0.933
49	31	0.177	0.230	0.405	0.057	0.009	0.450
130	3	0.804	-0.906	0.988	0.999	0.989	0.996
200	4	0.642	0.984	0.901	0.266	-0.346	0.353
361	12	-0.339	-0.172	0.554	-0.606	-0.140	0.140
394	4	-0.330	-0.630	-0.304	-0.373	0.630	0.833
406	3	0.983	-0.779	-0.184	0.975	0.208	-0.419
460	3	0.785	0.804	0.426	-0.276	0.414	0.183
679	6	-0.146	-0.267	0.485	-0.319	0.949	0.370
744	10	-0.169	0.166	-0.401	-0.468	-0.240	-0.074
836	5	0.866	0.181	0.980	-0.119	-0.478	0.659
925	11	0.716	-0.326	-0.257	-0.239	-0.127	0.320
934	6	0.857	0.312	0.237	0.422	-0.367	0.509
961	11	0.685	-0.412	-0.384	0.155	0.550	0.516
964	3	-0.270	0.544	0.232	-0.021	0.704	0.854
1513		-0.857	0.982	0.724	-0.983	-0.539	-0.896
1672	4	-0.280	0.780	0.723	-0.850	-0.105	0.757
1693	3	0.972	0.932	0.853	-0.647	0.983	0.998

1850	7	0.539	-0.966	0.577	-0.359	0.914	0.370
1894	5	-0.185	-0.822	-0.786	-0.512	0.949	0.851
1952	3	0.447	1.000	0.623 -0.987		-0.195	0.572
1956	4	0.794	-0.907	-0.980	-0.028	0.596	0.334
1976	6	0.719	0.766	0.934	-0.828	-0.251	0.241
1990	13	0.302	0.304	0.154	-0.210	-0.214	0.136
2363	4	0.287	-0.106	0.990	-0.179	0.957	0.996
Avera	Average		0.055	0.301	-0.214	0.194	0.435

Table S9. Pearson correlation coefficients between predicted ratio and experimentally measured ratio for the bicistronic SRPS operons of *Ccel* (A), *Bsub* (B), *Cthe* (C), *Cace* (D) for each of the six methods (CAI, RCA, RCBS, MELP, Gene-order and SLOFE). Correlations with the experimentally measured abundance of transcripts and proteins were both shown for those SRPS operons where transcript and protein data are available, e.g., for *Ccel* and *Bsub*. Pearson correlation coefficients were calculated between the ratios predicted using these methods and the experimentally determined ratios at the transcript (or protein) level. NA denotes no data available.

щ	# of	RCA		CAI		M	ELP	R	RCBS		Gene order		SLOFE	
#	# genes	Gene	Protein	Gene	Protein	Gene	Protein	Gene	Protein	Gene	Protein	Gene	Protein	
4	2	-	NA	-	NA	-	NA	-	NA	0.388225	NA	0.614764	NA	
170	2	-0.051	-0.039	-0.010	-0.008	-0.096	-0.074	-0.050	-0.039	0.421	0.323	0.034	0.026	
228	2	0.617	0.499	0.563	-0.459	0.106	-0.027	0.337	0.915	0.062	-0.016	0.308	-0.080	
295	2	-0.078	-0.591	0.020	0.016	0.714	0.563	0.077	0.061	0.403	0.318	0.131	0.104	
566	2	-0.067	-0.004	0.879	0.057	-0.463	-0.030	0.258	0.017	0.114	0.562	0.067	0.952	
849	2	0.315	0.117	0.142	0.053	0.072	0.027	0.122	0.045	0.853	0.436	0.504	0.738	
915	2	-0.093	NA	0.119	NA	0.731	NA	0.001	NA	0.533	NA	0.903	NA	
1135	2	0.139	0.116	0.128	0.107	0.443	0.372	0.171	0.143	0.680	0.571	0.868	0.967	
1254	2	-0.348	-0.070	-0.734	-0.033	0.101	0.242	0.255	0.096	0.033	0.728	0.020	0.810	
1466	2	-0.063	NA	0.032	NA	-0.031	NA	0.009	NA	0.460	NA	0.037	NA	
1477	2	0.198	NA	-0.021	NA	-0.472	NA	0.876	NA	-0.960	NA	0.531	NA	
Aver	age	0.052	0.004	0.102	-0.038	0.100	0.153	0.187	0.177	0.272	0.417	0.365	0.502	

⁽A) Correlations with the experimentally measured abundance of transcripts and proteins for bi-cistronic operons in *Ccel*.

(B) Correlations with the experimentally measured abundance of transcripts and proteins for bi-cistronic operons in Bsub.

#	# ofRCA		CA	CA	ΛI	MELP		RCBS		Gene order		SLOFE	
#	genes	Gene	Protein	Gene	Protein	Gene	Protein	Gene	Protein	Gene	Protein	Gene	Protein
1	2	-0.100	-0.039	-0.044	-0.017	0.753	0.516	0.212	0.083	-0.463	-0.839	-0.273	-0.703
280	2	0.104	0.025	-0.053	-0.013	0.195	0.816	0.588	0.141	-0.424	-0.565	-0.250	-0.957
620	2	-0.271	-0.704	-0.026	-0.066	-0.459	-0.177	-0.670	-0.575	0.463	0.178	0.844	0.456
893	2	-0.498	-0.111	0.006	0.001	-0.264	-0.843	-0.948	-0.235	0.412	0.541	0.921	0.205
1212	2	-0.374	-0.097	-0.246	-0.064	-0.220	-0.843	0.025	0.006	0.603	0.432	0.356	0.732
1411	2	-0.048	-0.108	-0.031	-0.070	-0.130	-0.289	-0.045	-0.099	0.955	0.472	0.454	0.993
1463	2	-0.083	-0.087	0.011	0.012	0.773	0.810	0.193	0.202	0.356	0.373	0.603	0.632
1635	2	-0.008	-0.004	-0.002	-0.001	0.203	0.097	0.151	0.072	0.891	0.538	0.526	0.911
1768	2	-0.026	-0.024	0.020	0.018	-0.053	-0.050	-0.095	-0.089	0.612	0.573	0.701	0.656
1934	2	-0.034	-0.036	0.035	0.037	-0.873	-0.821	-0.155	-0.165	0.513	0.546	0.869	0.924
2144	2	-0.190	0.240	-0.058	0.073	-0.964	0.762	-0.487	0.616	-0.755	0.597	0.227	-0.287
Ave	erage	-0.121	-0.071	-0.029	-0.004	-0.155	-0.055	-0.118	-0.023	0.297	0.280	0.472	0.370

(C) Correlations with the experimentally measured abundance of transcripts for bi-cistronic operons in *Cthe*.

#	# of gene	RCA	CAI		RCE	S	MELP	Gene- order		SLOFE
236	2	-0.012	-0.009		-0.01	8	-0.145	0.669)	0.883
569	2	-0.567	0.373		0.04	2	0.010	-0.029)	0.990
699	2	0.540	-0.148		-0.22	21	0.078	-0.153	3	-0.090
1265	2	0.052	0.243		0.41	7	0.755	-0.347	7	0.301
1289	2	0.181	-0.	.021	0.93	0	0.225	-0.527	7	-0.311
1380	2	-0.146	0.	194	-0.77	70	0.227	-0.595	5	-0.351
1518	2	0.486	-0.	-0.055		8	0.176	0.125	i	0.074
Average	0.076	0.082		0.178		0.189		-0.122		0.214

(**D**) Correlations with the experimentally measured abundance of transcripts for bi-cistronic operons in *Cace*.

# Operon	# of gene	Gene- order	MELP	RCA	CAI	RCBS	SLOFE
216	2	-0.104	0.069	0.418	0.817	0.318	-0.123
466	2	-0.758	0.356	-0.063	0.037	0.442	0.933
614	2	0.725	-0.829	-0.006	-0.079	-0.211	-0.200
633	2	0.877	0.381	-0.088	-0.160	0.067	0.518
943	2	0.061	-0.539	-0.321	0.101	0.313	0.036
981	2	-0.626	0.552	0.005	-0.051	0.098	0.721
1362	2	0.165	0.984	-0.144	0.553	-0.415	0.148
1412	2	-0.776	-0.355	0.025	-0.245	0.365	-0.458
1749	2	0.259	0.157	0.190	-0.353	0.963	0.330
Aver	Average		0.064	0.070	0.076	0.108	0.210