

**SUPPLEMENTARY MATERIALS**

**Figure S1. Evolutionary distribution of organisms.**

**Figure S2. Schematic of universal conserved nucleotide element (uCNE) identification.**

**Table S1A. Eukarya Conserved Nucleotide Elements.**

**Table S1B. Archaea Conserved Nucleotide Elements.**

**Table S1C. Bacteria Conserved Nucleotide Elements.**

**Table S2. Summary of Conserved Nucleotide Element False Discovery Rates.**

**Table S3. Inter-subunit bridges involving 23S-28S rRNA CNEs.**

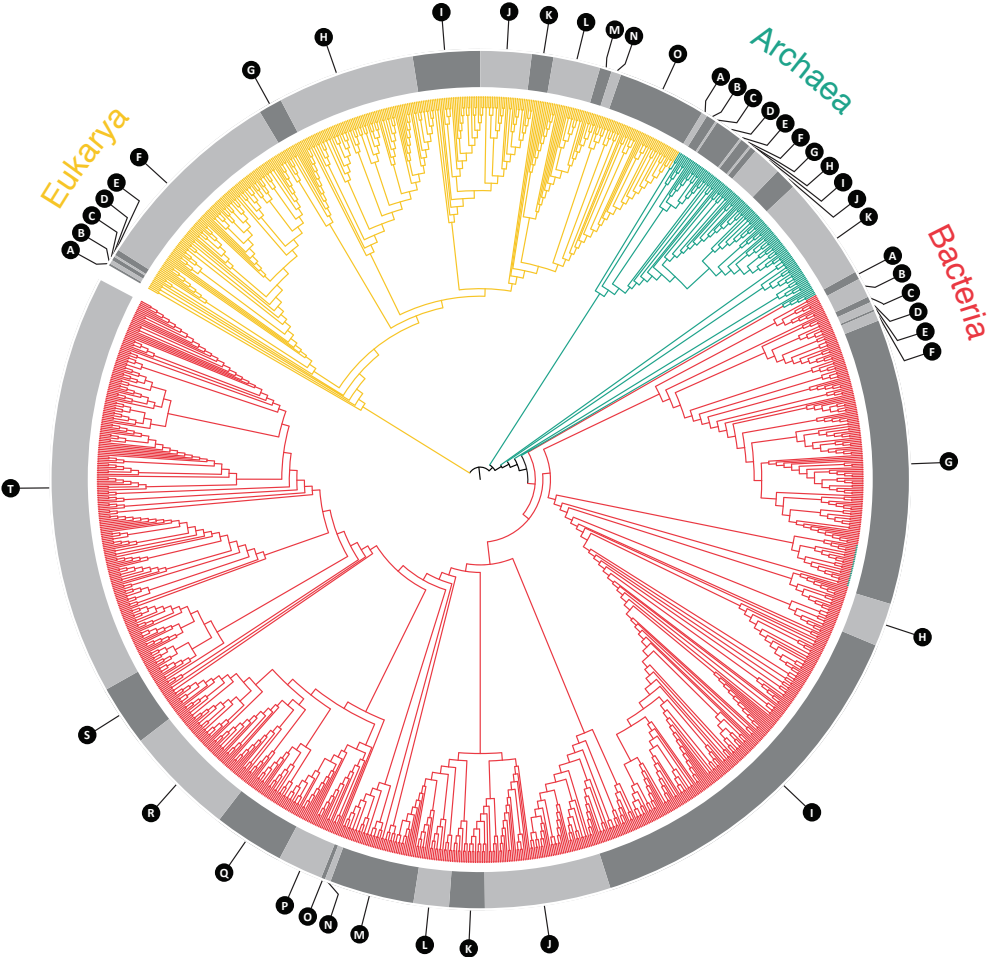
**Table S4. Cross domain conservation of eCNEs.**

**Table S5. Association of domain specific eCNEs with ribosome functions.**

**Figure S1. Evolutionary distribution of eukaryotic (yellow), archaeal (green), and bacterial (red) organisms.** The phylogenetic relationships of organisms whose 23S-28S rRNA sequences were included in FLORA. The cladogram was derived from the ARB/SILVA LSU Ref guide tree using the interactive tree of life (iTOL) software platform (<http://itol.embl.de/>). The number of species included for each group is indicated by the number in parentheses.

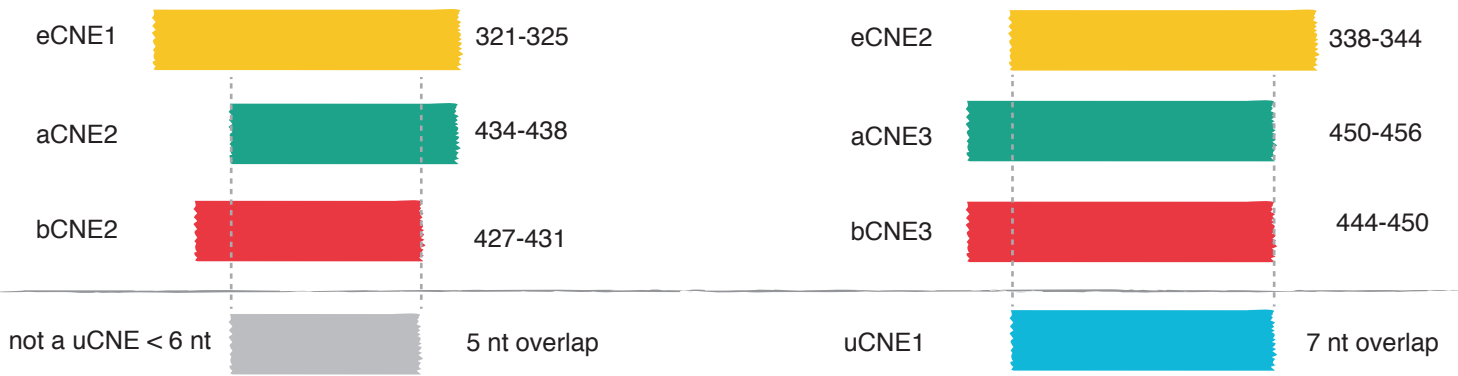
<b>EUKARYA</b>	<b>ARCHAEA</b>	<b>BACTERIA</b>
A Diplomonads (2)	A Methanobacteria (3)	A Aquificae (4)
B Parabasalids (2)	B Methanococci (5)	B Thermotogae (10)
C Amoebozoa (2)	C Korarchaeota (1)	C Deinococcus - Thermus (3)
D Rhizaria (1)	D Thermoprotei (14)	D Chloroflexi (4)
E Rhodophyta (3)	E Methanopyri (1)	E Dictyoglomi (1)
F Embryophyta (100)	F Thermococci (5)	F Fusobacteria (5)
G Porifera (12)	G Archaeoglobi (1)	G Firmicutes (145)
H Fungi (70)	H Thermoplasmata (3)	H Tenericutes (22)
I Alveolata (34)	I Thermoprotei (15)	I Cyanobacteria (186)
J Stramenophiles (26)	J Halobacteria (11)	J Actinobacteria (64)
K Mollusca (11)	K Methanomicrobia (34)	K Epsilonbacteria (18)
L Platyhelminthes (24)		L PVC – Planctobacteria (18)
M Chordata (6)		M Spirochaetes (43)
N Arthropoda(5)		N Acidobacteria (3)
O Nematoda (40)		O Synergistetes (2)
		P Deltaproteobacteria (23)
		Q Mitochondria (36)
		R Alphaproteobacteria (89)
		S Betaproteobacteria (56)
		T Gammaproteobacteria (179)

Figure S1.



**Figure S2. Schematic of universal conserved nucleotide element (uCNE) identification.** Using the ARB aligner, the intradomain CNEs are aligned by homologous position to measure the length of overlapping nucleotides (nt) in all three domains. Homologous sequences whose overlap is equal to 6 or more nucleotides are defined as uCNEs and their consensus sequence and conservation is measured. In the first scenario eCNE1, aCNE2, and bCNE2 share a 5 nt core sequence (coordinates on right) and thus are not considered to form a uCNE. In scenario two eCNE2, aCNE3, and bCNE3 share a 7 nt core element and after deriving its consensus sequence is classified as a uCNE greater than 90% conserved in all three domains.

**Figure S2.**



**uCNE1 in Eukarya**



**uCNE1 in Archaea**



**uCNE1 in Bacteria**



**uCNE1**  
conservation for consensus sequence CCGAUAG

- 97.7% conserved Eukarya
- 98.8% conserved Archaea
- 97.8% conserved Bacteria



Supplementary Table 1B.   Archaea conserved nucleotide elements		Structural filter				
		H. marismortui		S. solfataricus		
aCNE No.	aCNE sequence	Position	Length	IC Score	Length	IC Score
1	AAACAUCUUA	165	10	18.1	10	18.1
2	UAAUA	434	6	11.1	6	11.1
3	ACCGAUAG	449	8	15.2	8	15.2
4	CUGAAAAG	480	8	15.4	8	15.4
5	UGAAAC	517	6	11.4	6	11.4
6	CGAUCUA	773	7	13.4	7	13.4
7	CCAAUC	878	6	11.2	6	11.2
8	CUGGUUCCC	898	9	16.1	9	16.1
9	UCAAACUCCGAA	1002	12	22.8	12	22.8
10	GGUUAAGG	1092	8	14.3	8	14.3
11	CUAAGUG	1114	7	13.0	7	13.0
12	AGCAGCUA	1173	8	14.7	8	14.7
13	CGUAACAG	1195	8	14.5	9	15.9
14	UGGACC	1336	6	11.2	6	11.2
15	AUCCUG	1356	6	11.0	6	11.0
16	GGUCCUAAG	1450	9	16.1	9	16.1
17	GGUUAUUAUCC	1495	12	24.5	12	24.5
18	CGUAU	1592	6	11.1	6	11.1
19	UGAAAA	1651	6	11.1	6	11.1
20	CCGUACC	1671	7	13.1	7	13.1
21	AGGGAA	1739	6	11.0	6	11.0
22	UCGGCAAAU	1746	10	18.9	10	18.9
23	UAACU	1763	6	11.2	6	11.2
24	GUCGCA	1803	6	11.1	6	11.1
25	GACUGUUUAAU	1825	11	20.1	11	20.1
26	AACAU	1839	6	11.1	6	11.1
27	GGUACUAU	1947	9	16.8	9	16.8
28	ACCCUCUAAGGUAGC	1957	16	31.6	16	31.6
29	UACCUUGCC	1977	9	16.2	9	16.1
30	GCAUGAAU	2002	8	15.2	8	15.2
31	CACUGUCCC	2026	9	17.2	9	17.2
32	AAGACCC	2097	7	13.4	7	13.4
33	GAGCUUUACUGCA	2108	13	24.4	13	24.4
34	GCAGUU	2269	6	11.2	6	11.2
35	UGGGGC	2279	6	11.0	6	11.0
36	AGAAAA	2461	6	11.1	6	11.1
37	CUACCCC	2468	7	12.4	7	12.4
38	GGUAAC	2477	7	12.8	7	12.8
39	UUGCUACCUC	2523	10	18.6	10	18.6
40	GAUGUCG	2533	7	13.0	7	13.0
41	CCAUCCUG	2547	8	15.2	9	16.8
42	CAAGGGU	2570	7	13.4	7	13.4
43	CCUAUAAAAGG	2588	11	20.3	11	20.3
44	UGAGCUGGGUUUAGACCGUCGUGAGACAGGU	2605	31	58.3	31	58.3
45	UAGUACGAGAGGAAC	2685	15	28.0	15	28.0
46	GUUGUC	2726	6	11.2	6	11.1
47	GCUGAA	2774	6	11.4	6	11.4
48	GCAUCUAAGC	2781	10	19.7	10	19.7

Supplementary Table 1C.  Bacteria conserved nucleotide elements		Structural filter				
		E. coli		C. ramosum		
bCNE No.	bCNE sequence	Position	Length	IC Score	Length	IC Score
1	UGAAACAUCU	193	10	19.3	10	19.3
2	CUAAAU	426	6	11.1	6	11.1
3	ACCGAUAG	443	8	15.0	8	15.0
4	AGUACCGU	457	8	15.1	8	15.1
5	CCUUUUG	564	7	13.4	7	13.4
6	ACCCGAA	670	7	13.2	7	13.2
7	UGAUCUA	683	7	13.2	7	13.2
8	CCGAAC	731	6	11.3	6	11.3
9	AUAGCUGGUUCUC	802	14	26.0	14	26.0
10	AGCACU	863	6	11.5	6	11.5
11	CAAACUC	908	7	12.8	7	12.8
12	GAAGCAGCCA	1068	10	18.4	10	18.4
13	GCGUAAUAGCUCACUG	1091	16	29.9	16	29.9
14	UGAGUA	1263	6	11.1	6	11.1
15	CCUAAG	1350	6	11.5	6	11.5
16	CGUACC	1600	6	11.2	6	11.2
17	ACCGACAC	1610	8	15.2	8	15.2
18	AGGAACUCGGC	1665	11	20.2	11	20.2
19	CCGUAAAUUCGG	1685	12	22.6	12	22.6
20	GACUGUUUA	1772	9	17.8	9	17.8
21	AAAAACACAG	1783	10	18.6	10	18.6
22	ACGCCUGCCCGGU	1829	13	24.1	13	24.1
23	AAGCCC	1889	6	11.2	6	11.2
24	AACGGC	1900	6	11.3	6	11.2
25	CCGUAAUAUAACGGUCCUAAGGUAGCGAAAUCCUUGUCGG	1908	42	81.1	42	81.1
26	GUAAGUUCGACCUGCACGAA	1950	21	39.1	21	39.1
27	ACUGUCUC	1989	8	15.9	8	15.9
28	AAAGACCCCGU	2058	11	20.1	11	20.1
29	ACCUUUACU	2071	9	17.5	9	17.5
30	UCUAAC	2195	6	11.3	6	11.3
31	CAGUUUG	2240	7	12.4	7	12.4
32	UGGGGC	2249	6	11.2	6	11.2
33	GCCUCCAA	2259	9	16.4	9	16.4
34	GUAACGGA	2271	8	15.1	8	15.1
35	UUGACUG	2343	7	12.9	7	12.9
36	UAGUGAUCCG	2387	10	19.6	10	19.6
37	UCGCUCAACG	2419	10	18.9	10	18.9
38	GAUAAAAG	2429	8	14.8	8	14.8
39	GGGAUAACAGGCUGAU	2445	16	30.1	16	30.1
40	CACAUCGACG	2475	10	18.4	10	18.4
41	GUUUGGCACCUCGAUGUCGGCUCAUC	2490	26	51.8	26	51.8
42	CAUCCUGGG	2517	9	16.1	9	16.1
43	GUCCCAAGGGU	2536	11	20.7	11	20.7
44	GGCUGUUCGCC	2549	11	22.1	11	22.1
45	UUAAAG	2562	6	11.5	6	11.5
46	GAGCUGGGUUA	2576	12	22.4	12	22.4
47	GAACGUCGUGAGACAGUUCGGUCCCUAUCU	2588	30	56.7	30	56.7
48	CUAGUACGAGAGGACC	2652	16	29.9	16	29.9
49	GCAUCUAA	2751	8	14.7	8	14.7



Supplementary Table 2. | Summary of Conserved Nucleotide Element False Discovery Rates

<b>Length</b>	<b>Euk CNE</b>	<b>Arc CNE</b>	<b>Bac CNE</b>	<b>Univ CNE</b>
6	0.549517241	0.279764706	0.38428	0.116222222
7	0.288321429	0.197225806	0.229435897	0.053789474
8	0.198208333	0.141666667	0.156363636	0.025882353
9	0.158944444	0.097666667	0.110615385	0.0175
10	0.108258065	0.070923077	0.072818182	0.0086
11	0.066785714	0.047555556	0.050125	0.00475
12	0.040518519	0.031428571	0.033166667	0.0032
13	0.030666667	0.03	0.0248	0.002
14	0.020833333	0.023	0.017555556	0.000666667
15	0.015454545	0.0185	0.01225	0
16	0.013578947	0.02	0.008	0
17	0.011052632	0.022	0.0104	0
18	0.008210526	0.017	0.008	0
19	0.00775	0.01	0.0068	0
20	0.006933333	0.01	0.0064	0
21	0.006	0.007	0.0048	0
22	0.005333333	0.006	0.006	0
23	0.005846154	0.004	0.004	0
24	0.006727273	0.003	0.004	0
25	0.006363636	0.003	0.004	0
26	0.006	0.003	0.004	0
27	0.005818182	0.002	0.004	0
28	0.005454545	0.002	0.004	0
29	0.005454545	0.002	0.002	0
30	0.004727273	0.002	0.001333333	0

Supplementary Table 3. | Inter-subunit bridges involving 23S-28S rRNA CNEs

Bridge*	uCNE	eCNE
B1a***	X	X
B2a	uCNE 12, 13	eCNE 38
B2b	uCNE 13	eCNE 36, 38
B2c	X	eCNE 36
B3	uCNE 20	eCNE 38, 39
B4***	X	eCNE 7
B5**	uCNE 4, 15	X
B6	uCNE 4	X
B7a	X	eCNE 36, 38

\* Positions of bridges taken from Yusupov et al. (2001) for bacteria and Ben-Shem et al. (2010, 2011) for yeast.

\*\* Bridge B5 has shifted from its position in bacteria upward to sites in an expansion segment region in yeast.

uCNE5 is directly adjacent to nucleotides utilized for bridge B5 in bacteria (Yusupov et al 2001), but uCNE5 (within eCNE35) is directly adjacent to nucleotides utilized for bridge B3 in yeast (Ben-Shem et al. 2011).

\*\*\* Bridges B1a and B4 are not clustered with the other rRNA-containing bridges

Supplementary Table 4 | Cross-domain conservation of eCNEs

eCNE No.	Euk Position	Euk Percent	Arc Position	Arc Percent	Bac Position	Bac Percent
1	319	98.0	432	87.6	425	92.7
2	338	97.8	450	94.3	444	89.6
3	347	98.1	459	73.5	453	83.5
4	635	98.3	619	73.3	562	47.4
5	800	96.9	760	82.8	670	98.0
6	815	96.5	775	69.3	685	66.5
7	844	97.7	804	57.3	714	32.5
8	872	98.8	832	66.0	741	39.0
9	907	97.2	868	68.2	776	68.7
10	950	97.7	911	56.1	819	46.6
11	992	98.7	951	79.2	858	89.5
12	1001	97.7	960	64.5	866	62.1
13	1036	97.3	995	56.1	900	46.3
14 <sup>DS</sup>	1112	99.0	1040	33.2	942	46.4
15	1124	98.2	1052	67.4	954	47.1
16 <sup>DS</sup>	1135	98.2	1063	38.9	966	40.7
17	1150	96.9	1078	67.3	981	57.4
18	1221	97.2	1150	86.6	1047	73.9
19	1229	99.0	1158	64.5	1055	54.6
20	1238	98.7	1167	58.9	1064	63.4
21	1251	98.7	1180	74.8	1077	64.5
22	1291	96.6	1220	65.6	1117	60.5
23 <sup>DS</sup>	1430	97.8	1352	45.5	1250	48.7
24	1442	97.9	1364	53.0	1262	45.5
25	1491	96.0	1414	72.1	1310	69.3
26	1529	97.9	1452	89.1	1349	85.6
27 <sup>DS</sup>	1682	96.4	1558	4.9	1456	23.6
28	1790	97.7	1649	76.2	1565	64.7
29	1829	91.6	1671	74.4	1599	70.4
30	1839	96.8	1681	58.1	1609	57.5
31	1864	93.5	1707	53.4	1634	42.6
32	1894	99.1	1739	92.1	1664	83.3
33	1912	96.8	1757	80.6	1682	81.6
34 <sup>DS</sup>	1931	98.2	1776	33.8	1701	45.2
35	2125	96.9	1821	80.5	1768	76.5
36	2184	98.6	1879	64.9	1826	70.6
37 <sup>DS</sup>	2222	97.3	1917	11.8	1871	0.0
38	2230	98.8	1926	72.3	1888	74.1
39	2300	98.2	1996	77.8	1958	66.2
40 <sup>DS</sup>	2341	96.7	2037	43.9	1999	34.3
41	2389	99.5	2086	73.0	2048	60.5
42	2483	96.5	2211	60.8	2169	69.9
43 <sup>DS</sup>	2507	96.4	2234	46.0	2192	42.9
44	2607	98.7	2270	87.0	2240	82.2
45	2615	99.2	2278	96.8	2248	97.4
46	2625	97.8	2288	51.3	2258	34.5
47	2648	97.0	2311	71.9	2280	65.1
48	2673	95.6	2336	62.5	2305	60.6
49	2709	97.1	2374	61.6	2343	77.6
50 <sup>DS</sup>	2717	94.3	2383	0.0	2352	27.5
51	2748	96.6	2415	68.0	2382	54.7
52	2794	99.5	2460	76.5	2427	66.3
53	2830	98.1	2496	66.7	2465	68.9
54	2913	98.2	2578	65.1	2548	64.0
55	2932	99.4	2597	76.1	2567	68.4
56	3017	98.7	2685	97.8	2653	92.8
57	3111	90.2	2780	80.7	2750	70.3

DS = domain specific conservation

Supplementary Table 5. | Correlation of domain-specific eCNEs with ribosome functions

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<b>ds eCNE No.</b>	<b>Possible Function</b>
14	tunnel; contacts eukaryotic-specific ribosomal protein L29e
16	tunnel; contacts eukaryotic-specific ribosomal protein L29e
23	tunnel
27	unknown
34	abuts expansion segment 27L
37	contacts eukaryotic-specific ribosomal protein L36e
40	tunnel
43	E site tRNA ; abuts expansion segment 31L
50	contacts eukaryotic-specific ribosomal protein L29e

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