Supplementary Material

Table S1. Base Appearance at the Second Codon

Species	1st letter			2nd letter			3rd letter		
Base	*O*	*E*	*Z*	*O*	*E*	*Z*	*O*	*E*	*Z*
H. sapiens									
A	21.9	26.8	-12.4	26.8	31.4	-11.0	15.5	19.2	-10.6
T	13.6	16.9	-9.9	14.8	26.3	-29.6	20.6	22.3	-4.5
G	49.3*	31.8	42.7	18.6	18.9	-1.0	37.7*	28.7	22.6
C	15.2	24.6	-24.7	39.8*	23.4	43.8	26.2	29.9	-9.1
D. melanogaster									
A	25.4	27.5	-5.3	30.0	32.7	-6.5	17.8*	15.9	6.0
T	20.7*	16.5	12.5	19.0	25.9	-17.7	21.3*	19.7	4.3
G	34.5*	31.1	8.2	17.3	17.9	-1.6	30.1	31.7	-3.9
C	19.5	24.9	-14.0	33.8*	23.6	26.8	30.8	32.7	-4.5
C. elegans									
A	31.6*	30.1	4.5	28.8	32.7	-11.5	25.1	28.8	-11.3
T	21.6*	20.4	4.2	19.9	28.7	-26.5	30.4	31.4	-3.2
G	28.3	29.5	-3.5	18.4*	15.7	10.2	20.5	20.0	1.7
C	18.5	20.1	-5.3	32.9*	22.9	32.5	24.1*	19.7	14.9
A. thaliana									
A	22.6	29.3	-23.2	31.3	31.7	-1.4	21.4	25.1	-13.8
T	13.1	20.1	-27.6	15.4	28.2	-45.0	33.3	33.0	0.9
G	56.9*	31.7	85.6	16.4	17.4	-4.3	34.3*	22.5	44.3
C	7.4	18.9	-46.5	36.9*	22.6	53.9	11.1	19.3	-32.8
O. sativa									
A	18.4	23.6	-9.3	25.0	26.4	-2.3	15.5	15.7	-0.5
T	9.6	16.3	-13.8	14.4	24.7	-18.3	18.1	18.7	-1.2
G	53.5*	35.6	28.6	21.1	22.0	-1.6	42.3*	31.4	18.1
C	18.4	24.5	-10.8	39.5*	26.9	21.6	24.1	34.2	-16.3
S. cerevisiae									
A	31.7	33.5	-2.9	26.6	35.2	-14.1	28.3	29.3	-1.6
T	26.0*	22.1	7.3	21.1	28.3	-12.4	36.4*	32.7	6.1
G	29.4	28.3	1.9	15.4	14.2	2.6	16.3	18.7	-4.9
C	12.9	16.2	-6.9	37.0*	22.4	27.4	19.0	19.3	-0.5

S. pombe									
A	29.7	29.4	0.4	29.5	33.0	-5.0	25.8	27.1	-2.0
T	23.7	22.6	1.6	17.0	28.7	-17.4	43.2*	39.8	4.7
G	32.9*	29.1	5.5	16.5*	14.8	3.1	17.5	16.5	1.8
C	13.8	18.8	-8.6	37.0*	23.4	21.6	13.5	16.6	-5.6

The observed (*O*) and expected (*E*) fractions of each base at the first, second and third letters in the second codon are given in percentage. The biases are evaluated by the *Z*-test (see Methods). An asterisk shows the value significantly larger than the expectation ($p < 0.1\%$).

Table S2. Position-dependent Base Biases

Codon Number	A	T	G	C
H. sapiens				
E	19.2	22.3	28.7	29.9
2	15.5	20.6	37.7*	26.2
12	14.8	17.7	35.4*	32.1*
-4	21.5*	23.2	27.2	28.1
-2	18.4	18.8	25.8	37.0*
A. thaliana				
E	25.1	33.0	22.5	19.3
2	21.4	33.3	34.3*	11.1
4	28.2*	30.6	22.8	18.5
37	23.3	32.8	19.2	24.7*
-40	26.9*	29.6	25.6*	17.9
-2	21.3	35.4*	20.1	23.1*

The fraction of each base at the third letter in a given codon position and the expected value (*E*) are shown in percentage. An asterisk shows the value significantly larger than the expectation ($p < 0.1\%$).