

Table S2. Unequal use of iso-synonymous codons in *S. cerevisiae*.

Iso-synonymous codon groups ¹	Codon	tRNA	aa	Perfect match ²	Stability ³	RSCU'
1*	GCT	AGC	Ala	1	1	0.717
	GCC	AGC	Ala	0	0	0.248
2*	GCA	TGC	Ala	1	1	0.030
	GCG	TGC	Ala	0	0	0.005
3*	CGT	ACG	Arg	1	1	0.174
	CGC	ACG	Arg	0	0	0.004
	CGA	ACG	Arg	0	1	0.003
4*	AAT	GTT	Asn	0	0	0.147
	AAC	GTT	Asn	1	1	0.853
5*	GAT	GTC	Asp	0	1	0.430
	GAC	GTC	Asp	1	1	0.570
6*	TGT	GCA	Cys	0	1	0.868
	TGC	GCA	Cys	1	1	0.132
7	GGT	GCC	Gly	0	1	0.929
	GGC	GCC	Gly	1	0	0.047
8*	CAT	GTG	His	0	1	0.263
	CAC	GTG	His	1	1	0.737
9	ATT	AAT	Ile	1	0	0.457
	ATC	AAT	Ile	0	1	0.519
10*	CTT	GAG	Leu	0	1	0.026
	CTC	GAG	Leu	1	1	0.005
11*	CTA	TAG	Leu	1	1	0.071
	CTG	TAG	Leu	0	1	0.008
12*	TTT	GAA	Phe	0	0	0.194
	TTC	GAA	Phe	1	1	0.806
13*	CCT	AGG	Pro	1	1	0.123
	CCC	AGG	Pro	0	0	0.011
14*	CCA	TGG	Pro	1	1	0.857
	CCG	TGG	Pro	0	0	0.009
15*	TCT	AGA	Ser	1	1	0.520
	TCC	AGA	Ser	0	1	0.347
16*	AGT	GCT	Ser	0	1	0.041
	AGC	GCT	Ser	1	1	0.027
17*	ACT	AGT	Thr	1	1	0.507
	ACC	AGT	Thr	0	1	0.442
18*	TAT	GTA	Tyr	0	0	0.138
	TAC	GTA	Tyr	1	1	0.862
19*	GTT	AAC	Val	1	1	0.549
	GTC	AAC	Val	0	1	0.416

¹ Iso-synonymous codons are recognized by the same tRNA species. The perfect-match rule states that perfect codon-anticodon matches are favored over imperfect matches. The stability rule assumes that a codon-anticodon interaction is stable if the codon has either 1 or 2 G/C and is otherwise unstable (Rocha 2004 *Genome Res* **14**, 2279). This rule claims that codons with stable interactions with anticodons are preferred. We denote a star (correct prediction) or cross (incorrect prediction) for cases where the predictions of the two rules are compatible with each other.

² Perfect codon-anticodon matches are denoted by 1 and imperfect matches are denoted by 0.

³ Stable codon-anticodon interactions are denoted by 1 and unstable interactions are denoted by 0.