**Supplementary Table S6.** Models for evolutionary gain and loss of single codons at 3' splice sites resulting from splicing changes caused by single nucleotide substitutions.

|  |  |  |
| --- | --- | --- |
| **Description** | **Pattern of splicing/sequence1** | **Required/favorable conditions2** |
| **Exon contraction** | **/... ⇒ .../** |  |
| Upstream NAG creation | N1BGN2AG/ ⇒ N1AG/N2AG  | N1 > N2 as -3 base |
| Upstream NAG creation | N1AHN2AG/ ⇒ N1AG/N2AG | N1 > N2 as -3 base |
|  |  |  |
| **Exon expansion** | **.../ ⇒ /...** |  |
| Cryptic NAG activation | N1AG/N2AG ⇒ N1BGN2AG/  | N1 > N2 as -3 base |
|  | N1AG/N2AG ⇒ N1AHN2AG/  | N1 > N2 as -3 base |
| -3 base preference reversal | N1AG/N2AG ⇒ N1’AGN2AG/ | N1 > N2 > N1’ as -3 base |
|  |  |  |
| **Partial contraction** | **.../ ⇒ /.../** |  |
| Upstream NAG creation | N1BGN2AG/ ⇒ N1AG/N2AG/  | N1,N2 not too different as -3 base  |
| Upstream NAG creation | N1AHN2AG/ ⇒ N1AG/N2AG/ | N1,N2 not too different as -3 base |
| Upstream -3 base strengthening  | N1AGN2AG/ ⇒ N1’AG/N2AG/ | N1 < N2 ≈ N1’ as -3 base |
|  |  |  |
| **Partial expansion** | **/... ⇒ /.../** |  |
| Downstream NAG creation | N1AG/N2BG ⇒ N1AG/N2AG/ | N1,N2 not too different as -3 base  |
| Downstream NAG creation | N1AG/N2AH ⇒ N1AG/N2AG/ | N1,N2 not too different as -3 base |
| Upstream -3 base weakening | N1AG/N2AG ⇒ N1’AG/N2AG/ | N2 ≈ N1’ < N1 as -3 base |
|  |  |  |
| **Alternative expansion** | **/.../ ⇒ /...** |  |
| Downstream NAG loss | N1AG/N2AG/ ⇒ N1AG/N2BG | N1,N2 not too different as -3 base  |
| Downstream NAG loss | N1AG/N2AG/ ⇒ N1AG/N2AH | N1,N2 not too different as -3 base  |
| **Alternative contraction** | **/.../ ⇒ .../** |  |
| Upstream NAG loss | N1AG/N2AG/ ⇒ N1BGN2AG/ | N1,N2 not too different as -3 base  |
| Upstream NAG loss | N1AG/N2AG/ ⇒ N1AHN2AG/ | N1,N2 not too different as -3 base  |

1For each major category of change (bold headings in first column), the second column diagrams the effect on splicing, e.g., /... **⇒** .../ indicates a 3 base shift downstream in the location of the 3' splice site, .../ **⇒** /.../ indicates a change from constitutive splicing to alternative splicing at both the original 3' splice site and a site 3 bases upstream. Below each diagram is a sequence motif consisting of specific bases (A, G), degenerate positions (N, indicating any base), or partially degenerate positions (B = “not A” = a C, G or T, H = “not G”, etc.).

**2**The third column lists conditions that are expected to favor each type of change (based on -3 base preferences shown in Fig. 3B).