

## Supporting Information

Table 1: Bacterial strains, plasmids, genes and primers.

| strain  | Relevant characteristics   | Reference or source     |
|---|--|-------------------------|
| <i>E. coli</i> BL21                             | DE3 allowing over-expression of desired protein under IPTG inducible T7 promoter | Novagen                 |
| <b>Plasmids</b>                                 |  |                         |
| pET28a <sup>+</sup> :: <i>chrR</i> <sup>a</sup> | <i>E. coli chrR</i> (GenBank accession no. <a href="#">DQ989184</a> )            | [Ackerley et al., 2004] |
| <b>primers</b>                                  |  |                         |
| <i>FchrR</i> <sup>b</sup>                       | 5' – CGCGGGGG <u>CATATG</u> TCTGAAAAATTGCAGGT - 3'                               | [Ackerley et al., 2004] |
| <i>RchrR</i> <sup>c</sup>                       | 5' – TTTGGGATCCTTAGATCTTAACTCGCTGAA - 3'   | [Ackerley et al., 2004] |
| FA120N  | 5' – GTATTGATTCAGACCAGCTCAATGGGCGTGATTGG– 3'                                     | This research           |
| RA120N  | 5' – CCAATCACGCCATTGAGCTGGTCTGAATCAATAC-3'                                       | This research           |
| FN128Y  | 5' – TTGGCGGCGCGCTGTGAGTATCACCTGCGCCAGA– 3'                                      | This research           |
| RN128Y  | 5' – TCTGGCGCAGGTGATACTGACAGCGCGCGCCCAAT -3'                                     | This research           |
| FN160T  | 5' – GTTGATCCGCAAACCGGAGAAGTGATTGA– 3'   | This research           |
| RN160T  | 5' – ATCAATCACTTCTCCGTTTGC GGATCAAC -3'  | This research           |
| RL175G  | 5' – TTA ACTCGCTGAATAAACTCACCAAATGCGGTCA<br>ATTGCCCGGTCAGGTG -3'                 | This research           |
| F Y128N   | 5' –TTGGCGGCGCGCTGTCAGAATCACCTGCGCCAGA– 3'                                       | This research           |
| RY128N  | 5' –TCTGGCGCAGGTGATTCTGACAGCGCGCGCCCAAT -3'                                      | This research           |
| FA44V   | 5' –GTTACCATCCATTGTCGACATTCCCT – 3'  | This research           |
| RA44V   | 5' – AGGGAATGTCGACAATGGATGGTAAC– 3'  | This research           |
| FG150S  | 5' – GAATTTATGGGCAGCGTGATTCAG– 3'  | This research           |
| RG150S  | 5' – CTGAATCACGCTGCCATAAAATTC– 3'  | This research           |
| FQ153H  | 5' – GTGATTCATAACAAAGTTGATCCG – 3'   | This research           |
| RQ153H  | 5' – CGGATCAACTTTGTTATGAATCAC – 3'   | This research           |
| FN154T  | 5' – GTGATTCAGACCAAAGTTGATCCG – 3'   | This research           |
| RN154T  | 5' – CGGATCAACTTTGGTCTGAATCAC – 3'   | This research           |
| RQ184H  | 5' – GTTAACTCGGTGAATAAACTCACCAAATG– 3'   | This research           |
| FV120A  | 5' – GTATTGATTCAGACCAGCTCAATGGGCGCGATTGG– 3'                                     | This research           |
| R V120A   | 5' – CCAATCGCGCCCATTGAGCTGGTCTGAATCAATAC -3'                                     | This research           |
| F N154T- Q153H                                  | 5' – GTGATTCATACCAAAGTTGATCCG– 3'  | This research           |
| RN154T-Q153H                                    | 5' – CGGATCAACTTTGGTATGAATCAC -3'  | This research           |

<sup>a</sup> Protein accession number in PubMed data base.

<sup>b</sup> Bold underline indicates *Nde* I restriction site.

<sup>c</sup> Bold underline indicates *BamH* I restriction site.

Table 2: Expected fitness contribution (transformed) of mutations, for various values of *m*

| <b>m</b> | <b>-1.5</b> | <b>-1.3</b> | <b>-1.1</b> | <b>-0.9</b> | <b>-0.7</b> | <b>-0.5</b> | <b>-0.3</b> | <b>-0.1</b> |
|----------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|
| A44V     | -0.455      | -0.379      | -0.303      | -0.226      | -0.150      | -0.0741     | 0.0021      | 0.0783      |
| D103G    | -0.691      | -0.625      | -0.560      | -0.494      | -0.429      | -0.363      | -0.298      | -0.232      |
| V120A    | 0.307       | 0.282       | 0.256       | 0.231       | 0.206       | 0.180       | 0.155       | 0.129       |
| Y128N    | 1.944       | 1.931       | 1.918       | 1.905       | 1.892       | 1.878       | 1.865       | 1.852       |
| G150S    | -0.492      | -0.452      | -0.412      | -0.372      | -0.332      | -0.291      | -0.251      | -0.211      |
| Q153H    | 0.111       | 0.156       | 0.202       | 0.247       | 0.293       | 0.338       | 0.383       | 0.429       |
| N154T    | -0.422      | -0.379      | -0.337      | -0.294      | -0.251      | -0.208      | -0.166      | -0.123      |
| T160N    | 0.160       | 0.161       | 0.162       | 0.163       | 0.164       | 0.165       | 0.165       | 0.166       |
| Q175L    | -0.782      | -0.742      | -0.701      | -0.661      | -0.620      | -0.580      | -0.540      | -0.499      |
| Q184H    | -0.422      | -0.379      | -0.337      | -0.294      | -0.251      | -0.208      | -0.166      | -0.123      |
| K187T    | -0.340      | -0.317      | -0.287      | -0.246      | -0.228      | -0.194      | -0.165      | -0.144      |