

**Table S1 Oligonucleotide sequences used in this study**

| <b>Oligo Name &amp; Purpose</b>                         | <b>Sequence</b>  | <b>Reference</b> |
|---|--|------------------|
| CCHL.F-MfeI<br>(Construction of pRGK399)                | CTCAATTGATGGGTTTGTCTCCATCTGC                               | This study       |
| CCHL.R-HindIII<br>(Construction of pRGK399/<br>pRGK400) | CGAAGCTTGTGCATAGTTTTACGAGGTCCAAC                           | This study       |
| CCHL.F-His-NcoI<br>(Construction of pRGK400)            | CCAGCCATGGGTCATCATCATCATC<br>ACGTTTTGTCTCCATCTGCTCC        | This study       |
| pBAD-signal-right<br>(Construction of pRGK390)          | CACCATGAAAGCCGCTCTGGGGGCTG                                 | <i>a</i>         |
| pBADc4signal<br>(Construction of pRGK390)               | CGCAGCGCCCGCGGCCGCGAAACT                                   | This study       |
| cycS.F<br>(Construction of pRGK390)                     | CCCGCGGCCGCGATGGGTGATGTTGA<br>GAAAGGC                      | This study       |
| cycSHis.R<br>(Construction of pRGK390)                  | GCGGGTACCTTAGTGGTGGTGGTGGTG<br>GTGTTCAATAGTAGCTTTTTTCAGATA | This study       |
| cycS-c2 9-19.F<br>(Construction of pRGK401)             | TTCAACAAGTGCAAGACCTGCCACTC<br>GATCATCAAGGGAGGGCAAGCACAAG   | This study       |
| cycS-c2 2-8.R<br>(Construction of pRGK401)              | TTCTTTTTCGCCCTTCGCGGCGTCATCACC<br>CATCGCGGCCGCGAAAC        | This study       |
| cytc2:His L<br>(Construction of pRGK389)                | CTATGGTACCGAAGATCAGCCTCAGCC<br>TCACTGCCGCCACTGTC           | This study       |
| cytc2:His R<br>(Construction of pRGK389)                | CTATCTGCAGTTAGTGGTGGTGGTGGTG<br>GTGTTTCACGACCGAGGCCAGAT    | This study       |
| Tn5 Kan Fwd<br>(Verification of transductants)          | CGGTGCCCTGAATGAACTGC                                       | <i>b</i>         |
| Tn5 Kan Rev<br>(Verification of transductants)          | CGGCCACAGTCGATGAATCC                                       | <i>b</i>         |
| dsbA.F<br>(Verification of transductants)               | CGGGGAAGACTTACTGGCTGC                                      | This study       |
| dsbB.F<br>(Verification of transductants)               | AAACTGCGCACTCTATGCAT                                       | This study       |

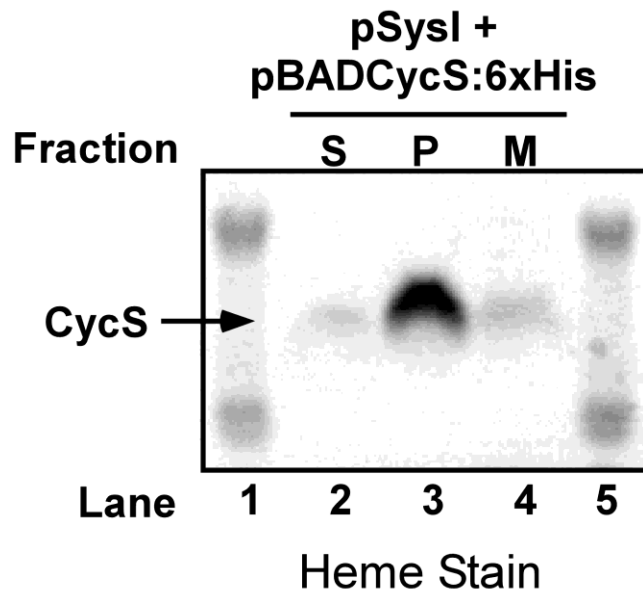
**Table S1 cont'd Oligonucleotide sequences used in this study**

| <b>Oligo Name &amp; Purpose</b>           | <b>Sequence</b>        | <b>Reference</b> |
|---|------------------------|------------------|
| dsbC.R<br>(Verification of transductants) | GGCGACGAAGTTGTATCTGTT  | This study       |
| dsbD.R<br>(Verification of transductants) | CGCTATTTTCCTCCGTCTTTCC | This study       |
| dsbG.F<br>(Verification of transductants) | CTCGACTTTTGCACTGACTG   | This study       |

---

<sup>a</sup>R.E. Feissner, C.L. Richard-Fogal, E.R. Frawley, J.A. Loughman, K.W. Earley, R.G. Kranz, Recombinant cytochromes *c* biogenesis systems I and II and analysis of haem delivery pathways in *Escherichia coli*, *Mol Microbiol*, 60 (2006) 563-577.

<sup>b</sup>K.A. Datsenko, B.L. Wanner, One-step inactivation of chromosomal genes in *Escherichia coli* K-12 using PCR products, *Proc Natl Acad Sci U S A*, 97 (2000) 6640-6645.



Supplemental Figure 1. Human cytochrome *c* (CycS) is matured in the *Escherichia coli* periplasm. Heme stain of SDS-PAGE of *E. coli* cellular fractions expressing pSys I (pRGK333) and pBADCycS:6xHis (pRGK390) where S=soluble (lane 2), P=periplasm (lane 3), and M=membrane (lane 4). Protein fractionation is detailed in Materials and Methods. Molecular weight standard proteins are shown in lanes 1 and 5. HoloCycS is indicated by an arrow.