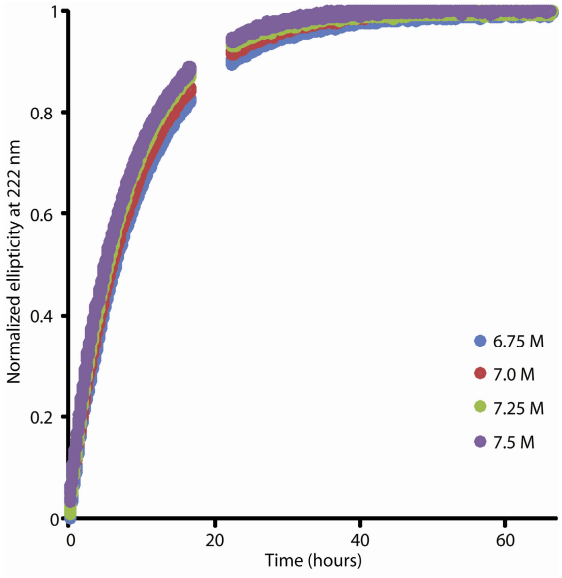
Cysteine-free Rop: a four-helix bundle core mutant has wild-type stability and structure but dramatically different unfolding kinetics

Sanjay B. Hari, Chang Byeon, Jason J. Lavinder, and Thomas J. Magliery

Figure S1. *Kinetics of unfolding of Rop variants at various concentrations of urea observed by CD spectroscopy*. The *k* values for each unfolding reaction were determined by fitting to a single exponential ($A_0\ e^{-kt}$). The missing points in the wild-type unfolding were because the sample had to be removed briefly during the very long experimental observation.

Wild-type (Cys/Cys)



Ala/Val

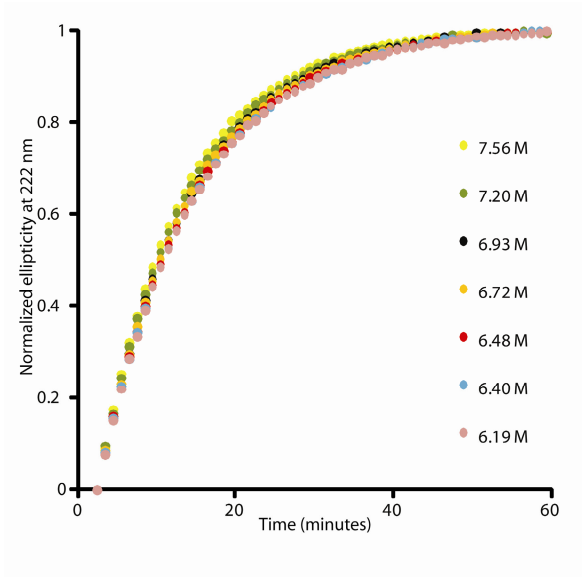


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Table S1. *Oligonucleotides used for Cys-free Rop mutant gene construction.* Primers 1 and 2 were used for all genes, while the third and fourth primers were specific to each variant. Bold lettering indicates the point mutation.

Primer	Sequence (5'→3')
1	ATGACCAAACAGGAAAAAACCGCCCTTAACATGGCCCGCTTTA TCAGAAGCCAGACATTAAC
2	CTGCCTGTTCATCCGCGTCCAGCTCGTTGAGTTTCTCCAGCAG CGTTAATGTCTGGCTTCTG
3Ala	GACGCGGATGAACAGGCAGATATC**GCG**GAATCGCTTCACGACC ACGCTGATGAGCTTTACCG
3Ser	GACGCGGATGAACAGGCAGATATT**AGC**GAATCGCTTCACGAC CACGCTGATGAGCTTTACCG
3Val	GACGCGGATGAACAGGCAGATATT**GTG**GAATCGCTTCACGAC CACGCTGATGAGCTTTACCG
4Ala	TCAGAGGTTTTCACCGTCATCACCGAAACGCGCGAG**CGC**ACT GCGGTAAAGCTCATCAGCG
4Ser	TCAGAGGTTTTCACCGTCATCACCGAAACGCGCGAG**GCT**GCT GCGGTAAAGCTCATCAGCG
4Val	TCAGAGGTTTTCACCGTCATCACCGAAACGCGCGAG**CAC**ACT GCGGTAAAGCTCATCAGCG
pMRpro	AATAATCCATGGCGCATCATCACCATCATCACGGCGGTGAAA ACCTGTATTTTCAGGGCACCAAACAGGAAAAAAC
pMRterm	AATAATGGATCCTCAGAGGTTTTCACCGTC
pACpro	AATAATAATCATATGACCAAACAGGAAAAAAC
pACterm	AATAATGGTACCTCAGAGGTTTTCACCGTC