Supporting Information

Fan et al. 10.1073/pnas.1717621115

N A C



Fig. S1. (Upper) Scheme of chimera design. Red represents ACT domain from TmaDAH7PS, orange represents CM domain from GspDAH7PS, blue represents catalytic domain of TmaDAH7PS, and green represents catalytic domain of GspDAH7PS. (Lower) Alignment of wild-type parent proteins and two protein chimeras. Black arrow identifies the fusion site.



Fig. S2. DAH7PS activity of the parent and chimeric proteins in the presence of 1 mM Phe. Wild-type TmaDAH7PS and ^{Tma}ACT-^{Gsp}DAH7PS showed inhibition with Phe.



Fig. S3. Thermostability of the four proteins in the presence or absence of 1 mM Phe.

() <



Fig. S4. Analysis of key interactions associated with allostery. Key residues involved in interactions between regulatory and catalytic domains are identified in *Tma*DAH7PS (*A*) and *Gsp*DAH7PS (*B*). (C) Partial alignment of type Iβ DAH7PS sequences, showing segments of the catalytic barrel. Black rectangles highlight the location of key barrel residues identified in *A* and *B*. Figure was formatted with Weblogo3.

<

Table S1. Primers designed to amplify the genes encoding for the chimeras

DCD	cton
r U N	step

PNAS PNAS

PCR step	^{Tma} ACT- ^{Gsp} DAH7PS	^{Gsp} CM- ^{Tma} DAH7PS	
PCR amplifying regulatory domain	5^{\prime} GCTTCGAAGGAGATAGAACCATGATTGTCGTTTTGAAACCC 3^{\prime}	5'GCTTCGAAGGAGATAGAACCATGGGTAATGAACGTCT- GGAT 3'	
and linker region with overlap	5'GACGAAATATTGATTGCCGTTTCCTATCTTCACATCT 3'	$5^\prime \texttt{CGCTATGATGGTGAAATACTGATTGCCATCACCAAT}\ 3^\prime$	
PCR amplifying	5'gtgaagataggaaacggcaatcaatatttcgtcatggg 3'	5'AATCAGTATTTCACCATCATAGCGGGACCG 3'	
catalytic domain with overlap	5 ' GGGGACCACTTTGTACAAGAAAGCTGGGTCTTATGCACGAACAAACTGACG 3 '	$5^{\prime} \mbox{GGGGACCACTTTGTACAAGAAAGCTGGGTCTTAATTC-ACCTTCACCCCCAG} 3^{\prime}$	
Generic PCR fusing the above	5 ' GGGGACAAGTTTGTACAAAAAAGCAGGCTTCGAAGGAGATAGAACCATG 3 '	$5^{\prime} \text{GGGGACAAGTTTGTACAAAAAAGCAGGCTTCGAAGGA-} \\ \text{GATAGAACCATG} 3^{\prime}$	
products	5'ggggaccactttgtacaagaaag 3'	5'ggggaccactttgtacaagaaag 3'	

Table S2. Structural parameters from SAXS profile of each chimera in the absence or presence of Tyr or prephenate

Structural parameter	TmaACT-GspDAH7PS (apo)	TmaACT-GspDAH7PS (tyr)	^{Gsp} CM- ^{Tma} DAH7PS(apo)	^{Gsp} CM- ^{Tma} DAH7PS (prephenate)
l(0), cm ⁻¹ *	0.12 ± 0.003	0.116 ± 0.002	0.408 ± 0.002	0.397 ± 0.003
R _g , Å*	33.96 ± 0.08	32.71 ± 0.08	36.89 ± 0.2	34.76 ± 0.2
l(0), cm ⁻¹ †	0.12 ± 0.001	0.116 ± 0.001	0.408 ± 0.001	0.397 ± 0.001
D _{max} , Å	104.16	101.35	120.71	118.43
R _g , Å [†]	34.04 ± 0.2	32.75 ± 0.2	36.91 ± 0.2	34.77 ± 0.2
Porod volume estimate, Å ³	226,364	222,091	274,596	258,918
Fit with crystal/model structure, $\chi^{2\ddagger}$	1.08	0.7	0.78	1.33

*From pair distribution function. Data presented as mean \pm SD. *From Guinier plot. Data presented as mean \pm SD.

‡From CRYSOL.