

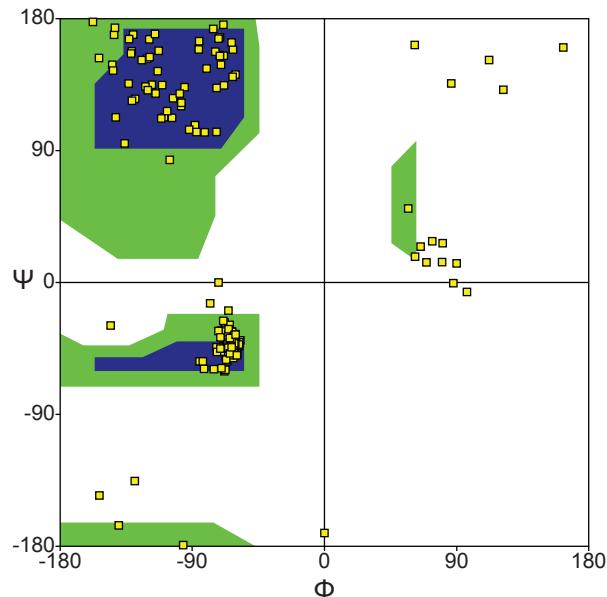
## Supplementary tables and figures

**Tab. S1.** Predicted protein expression levels at which growth is reduced to 50% of that of the wild type strain (EC<sub>50</sub>). N.E. indicates that the EC<sub>50</sub> is undefined because the model predicts that the gene is not essential for growth.

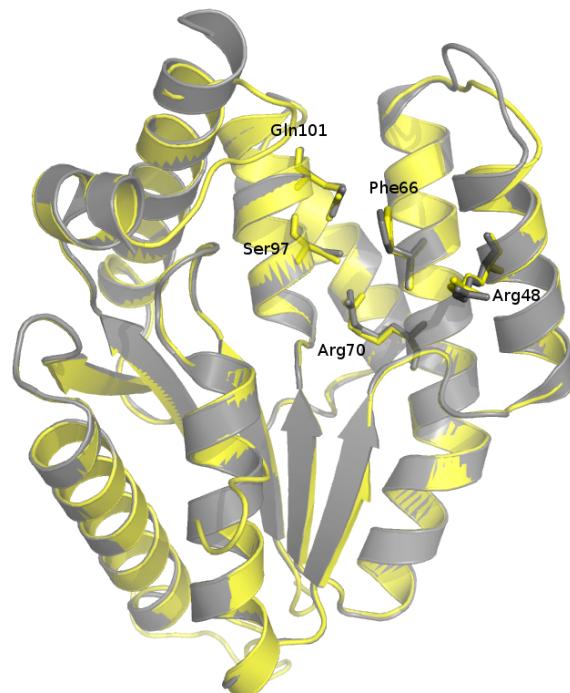
ID	Name	EC <sub>50</sub> (Rel)
MG006	Thymidylate kinase	5.00×10 <sup>-1</sup>
MG047	S-adenosylmethionine synthetase	1.77×10 <sup>-1</sup>
MG357	Acetate kinase	7.53×10 <sup>-2</sup>
MG023	Fructose-1,6-bisphosphate aldolase	6.93×10 <sup>-2</sup>
MG301	Glyceraldehyde-3-phosphate dehydrogenase, type I	4.92×10 <sup>-2</sup>
MG407	Enolase	4.00×10 <sup>-2</sup>
MG271	Dihydrolipoamide dehydrogenase	3.30×10 <sup>-2</sup>
MG272	Dihydrolipoamide acetyltransferase	3.30×10 <sup>-2</sup>
MG273	Pyruvate dehydrogenase component E1, beta subunit	3.30×10 <sup>-2</sup>
MG274	Pyruvate dehydrogenase component E1, alpha subunit	3.30×10 <sup>-2</sup>
MG034	Thymidine kinase	2.81×10 <sup>-2</sup>
MG276	Adenine phosphoribosyltransferase	1.92×10 <sup>-2</sup>
MG458	Hypoxanthine phosphoribosyltransferase	1.48×10 <sup>-2</sup>
MG299	Phosphate acetyltransferase	1.15×10 <sup>-2</sup>
MG431	Triosephosphate isomerase	9.99×10 <sup>-3</sup>
MG430	2,3-bisphoglycerate-independent phosphoglycerate mutase	9.79×10 <sup>-3</sup>
MG111	Glucose-6-phosphate isomerase	9.52×10 <sup>-3</sup>
MG145	Riboflavin biosynthesis protein RibF	9.02×10 <sup>-3</sup>
MG013	Methylenetetrahydrofolate dehydrogenase/methylenetetrahydrofolate cyclohydrolase	7.41×10 <sup>-3</sup>
MG245	5-formyltetrahydrofolate cyclo-ligase, putative	7.21×10 <sup>-3</sup>
MG037	Nicotinate phosphoribosyltransferase	4.84×10 <sup>-3</sup>
MG102	Thioredoxin-disulfide reductase	4.57×10 <sup>-3</sup>
MG383	NH(3)-dependent NAD <sup>+</sup> synthetase, putative	3.02×10 <sup>-3</sup>
MG330	Cytidylate kinase	2.21×10 <sup>-3</sup>
MG382	Uridine kinase	1.19×10 <sup>-3</sup>
MG058	Ribose-phosphate pyrophosphokinase	9.30×10 <sup>-4</sup>
MG342	NADPH-dependent FMN reductase domain protein	7.93×10 <sup>-4</sup>
MG128	Inorganic polyphosphate/ATP-NAD kinase, probable	6.04×10 <sup>-4</sup>
MG394	Serine hydroxymethyltransferase	2.32×10 <sup>-4</sup>
MG216	Pyruvate kinase	1.97×10 <sup>-4</sup>
MG434	Uridylate kinase	1.90×10 <sup>-4</sup>
MG107	Guanylate kinase	1.27×10 <sup>-4</sup>
MG038	Glycerol kinase	2.36×10 <sup>-5</sup>
MG053	Phosphoglucomutase/phosphomannomutase, putative	8.88×10 <sup>-16</sup>
MG066	Transketolase	8.88×10 <sup>-16</sup>
MG118	UDP-glucose 4-epimerase	8.88×10 <sup>-16</sup>
MG171	Adenylate kinase	0
MG215	6-phosphofructokinase	0
MG228	Dihydrofolate reductase	0
MG300	Phosphoglycerate kinase	0
MG396	Ribose 5-phosphate isomerase B	0
MG030	Uracil phosphoribosyltransferase	N.E.
MG039	FAD-dependent glycerol-3-phosphate dehydrogenase, putative	N.E.
MG049	Purine nucleoside phosphorylase	N.E.
MG050	Deoxyribose-phosphate aldolase	N.E.
MG051	Pyrimidine-nucleoside phosphorylase	N.E.
MG052	Cytidine deaminase	N.E.
MG063	1-phosphofructokinase, putative	N.E.
MG227	Thymidylate synthase	N.E.
MG240	Nicotinamide-nucleotide adenylyltransferase/conserved hypothetical protein	N.E.
MG259	Modification methylase	N.E.
MG264	Dephospho-CoA kinase	N.E.
MG265	Sugar phosphatase	N.E.
MG336	Cysteine desulfurase	N.E.
MG372	Thiamine biosynthesis/tRNA modification protein ThiI	N.E.
MG398	ATP synthase F1, epsilon subunit	N.E.
MG399	ATP synthase F1, beta subunit	N.E.
MG400	ATP synthase F1, gamma subunit	N.E.
MG401	ATP synthase F1, alpha subunit	N.E.
MG402	ATP synthase F1, delta subunit	N.E.
MG403	ATP synthase F0, B subunit	N.E.
MG404	ATP synthase F0, C subunit	N.E.

**Tab. S1.** (continued)

ID	Name	EC <sub>50</sub> (Rel)
MG405	ATP synthase F0, A subunit	N.E.
MG408	Methionine-S-sulfoxide reductase	N.E.
MG448	Methionine-R-sulfoxide reductase	N.E.
MG460	L-lactate dehydrogenase/malate dehydrogenase	N.E.
MG482	Holo-(acyl-carrier-protein) synthase	N.E.



**Fig. S1.** Ramachandran plot of the predicted *M. genitalium* Tmk structure shows that the predicted dihedral angles primarily fall within the most energetically favorable regions (colored blue and green).



**Fig. S2.** Superimposed structures of *M. genitalium* (yellow) and *S. aureus* (grey; PDB ID: 4QGG) Tmk shows that *M. genitalium* Tmk shares the same piperidinylthymine binding site (top-center; residues labeled according to *S. aureus*).