Supplementary data

Promising new antifungal treatment targeting chorismate synthase from *Paracoccidioides brasiliensis*

Franciele Abigail Vilugron Rodrigues-Vendramini[a], Cidnei Marschalk[b], Marina Toplak[c], Peter Macheroux[c], Patricia de Souza Bonfim-Mendonça[a], Terezinha Inez Estivalet Svidzinski[a], Flavio Augusto Vicente Seixas[b], Erika Seki Kioshima[a#]

[a]Department of Clinical Analysis and Biomedicine, Universidade Estadual de Maringá, Av. Colombo 5790, 87020-900 Maringá, PR, Brazil.

[b]Department of Technology, Universidade Estadual de Maringá, Av. Ângelo Moreira da Fonseca, 1800, 87506-370 Umuarama, PR, Brazil

[c]Institute for Biochemistry, Graz University of Technology, Petersgasse 12/2, 8010 Graz, Austria.

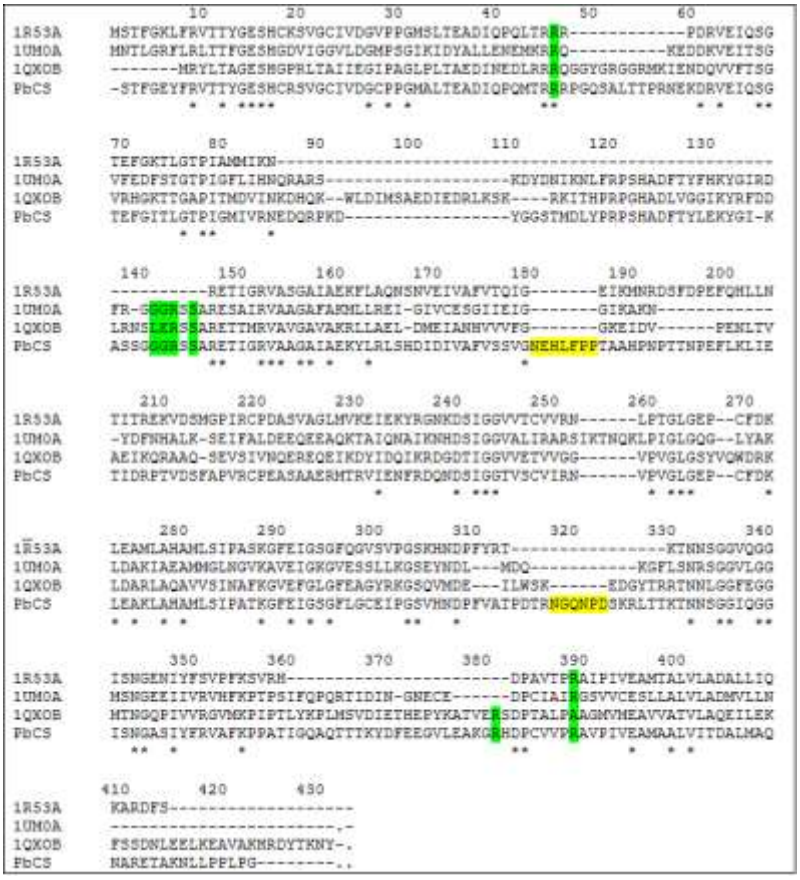

Figure S1. Alignment of the amino acid sequences from templates with *Pb*CS. The green square highlight residues that compose the 5-EPSP site in *P. brasiliensis*. Residues that do not have correspondence in the templates are marked in yellow. Conserved residues are represented by an asterisk and the ligands are represented by the points at the end of alignment.

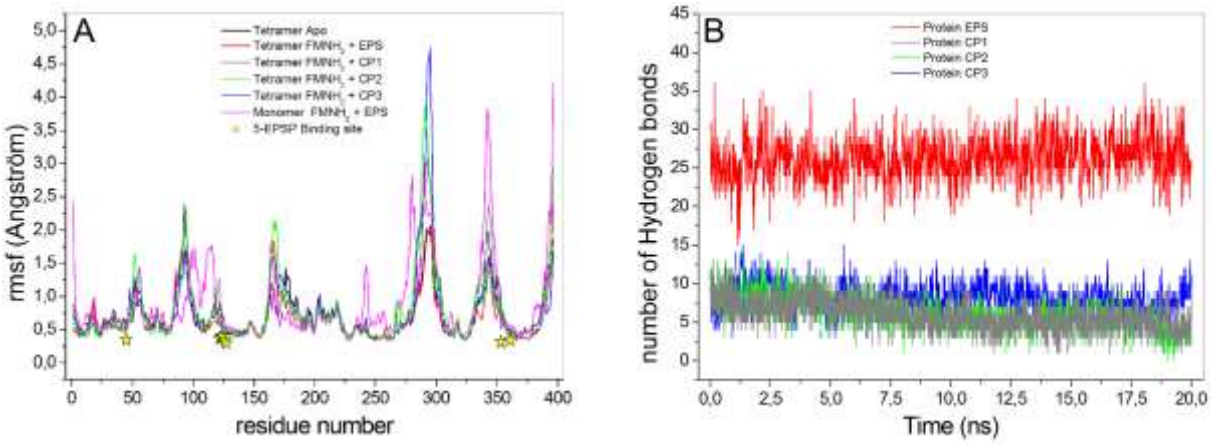

Figure S2. A) Root-mean-square fluctuation (rmsf) of Cα atoms from *Pb*CS residues. The yellow stars mark the position of the active site residues described in **Table S1**. B) Average number of hydrogen bonds formed between the protein and ligand along the simulation. *Pbrasiliensis*CS structures: The A chain bonded to $FMNH_2$ + 5-EPSP (**pink**), tetramer apo (**black**), tetramer bonded to $FMNH_2$ + 5-EPSP (**red**), tetramer bonded to $FMNH_2$ + CP 1 (**gray**), tetramer bonded to $FMNH_2$ + CP2 (**green**), and tetramer bonded to $FMNH_2$ + CP3 (**blue**). The root-mean-square deviation (A) and radius of gyration (B) were obtained from the oscillation of the protein main chain atoms relative to the full minimized structure.

Table S1. Contact frequency up to 4 Å between *Pb*CS and the evaluated ligands.

Residue	EPSP	CP1	CP2	CP3	Residue	EPSP	CP1	CP2	CP3
Ser-15	–	0.24	–	0.12	Tyr-110	–	0.23	–	–
Arg-45	**0.99**	0.67	**0.73**	**0.80**	Ala-118	–	0.08	–	–
Arg-46	0.21	–	0.29	0.01	Ser-119	–	0.56	–	0.35
Ser-50	–	0.19	–	–	Ser-120	–	0.59	0.17	0.22
Leu-52	–	0.20	0.10	–	Gly-121	0.55	0.23	0.32	0.20
Thr-53	0.32	0.58	0.46	0.26	Gly-122	**0.78**	0.34	**0.74**	**0.83**
Thr-54	0.42	0.43	0.46	0.45	**Gly-123**	**0.97**	0.67	**0.99**	**0.90**
Pro-55	0.45	0.31	0.26	0.23	Arg-124	**0.99**	**1.00**	0.60	0.53
Asn-57	–	–	0.22	0.24	Ser-125	0.52	**0.76**	–	–
Glu-58	–	0.09	–	0.10	Ser-126	**0.97**	**0.99**	0.65	0.49
Asn-84	0.01	0.04	0.11	0.27	Ala-127	**0.73**	0.61	0.43	0.41
Asp-86	0.22	0.03	0.09	0.38	Thr-128	0.12	–	–	0.24
Gln-87	–	–	0.19	0.28	Thr-130	–	0.36	0.50	0.32
Asp-91	–	–	–	0.10	Ile-131	0.09	0.06	0.02	0.24
Tyr-92	0.10	–	0.05	–	Pro-258	–	0.15	–	–
Gly-93	–	–	–	0.12	Ala-330	–	–	0.06	0.39
Gly-94	–	–	–	0.40	Thr-331	0.08	0.19	0.44	0.48
Ser-95		0.20	0.11	0.05	Lys-351	–	–	0.02	0.09
Thr-96	–	–	–	0.06	Gly-352	0.31	0.26	0.28	0.48
Asp-98	–	0.24	–	–	**Arg-353**	**1.00**	**0.97**	**0.95**	**0.99**
Arg-102	0.25	0.37	–	**0.88**	His-354	–	0.27	0.59	0.07
Ser-104	–	–	0.24	–	Asp-355	0.04	0.55	0.52	0.21
His-105	0.25	0.7	0.69	0.64	Val-358	–	–	–	0.05
Ala-106	–	0.27	0.28	–	**Arg-361**	**0.93**	**0.72**	**0.95**	**1.00**
Asp-107	–	0.24	–	0.36	**$FMNH_2$**	**1.00**	**1.00**	**1.00**	**1.00**

Obs: only contacts with significant frequently ($p < 0.05$) are presented plementary Table Y.