Supplementary Data

In silico site-directed mutagenesis informs species-specific predictions of chemical susceptibility derived from the Sequence Alignment to Predict Across Species Susceptibility (SeqAPASS) tool

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Summary

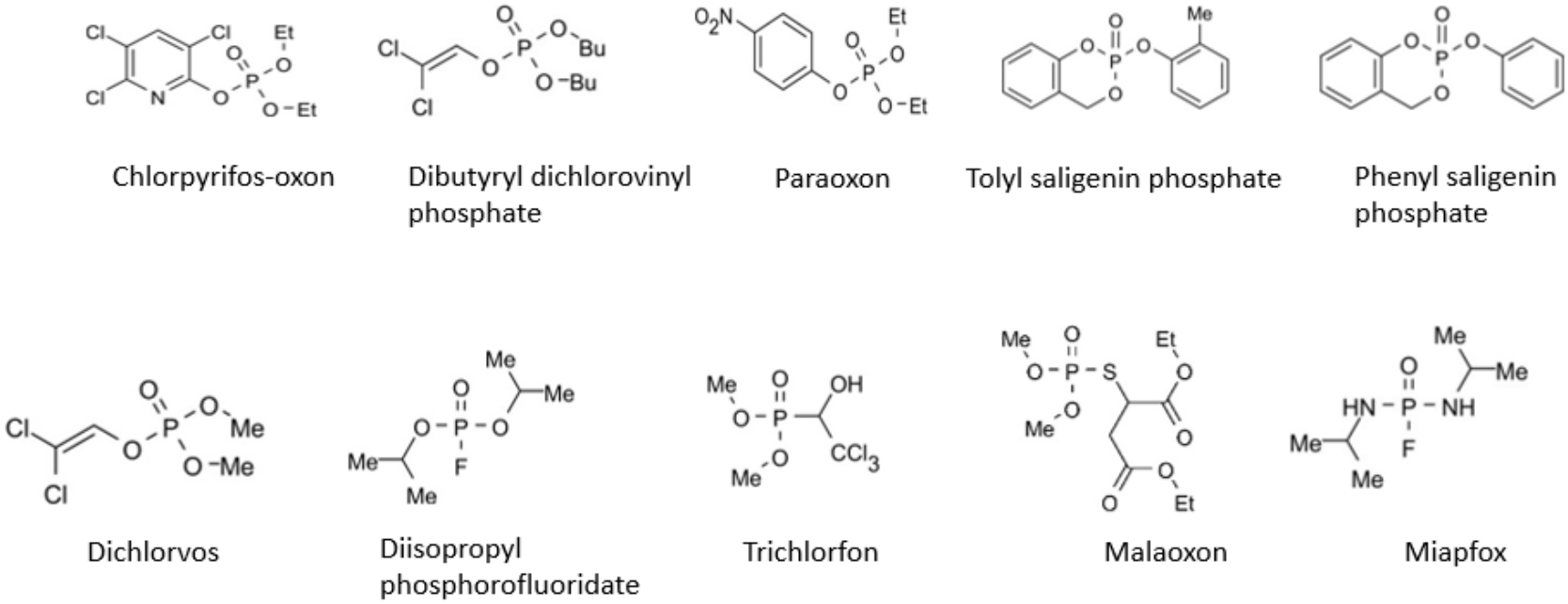

Figure S1 Test compounds for organophosphates (Lee & Barron 2015).

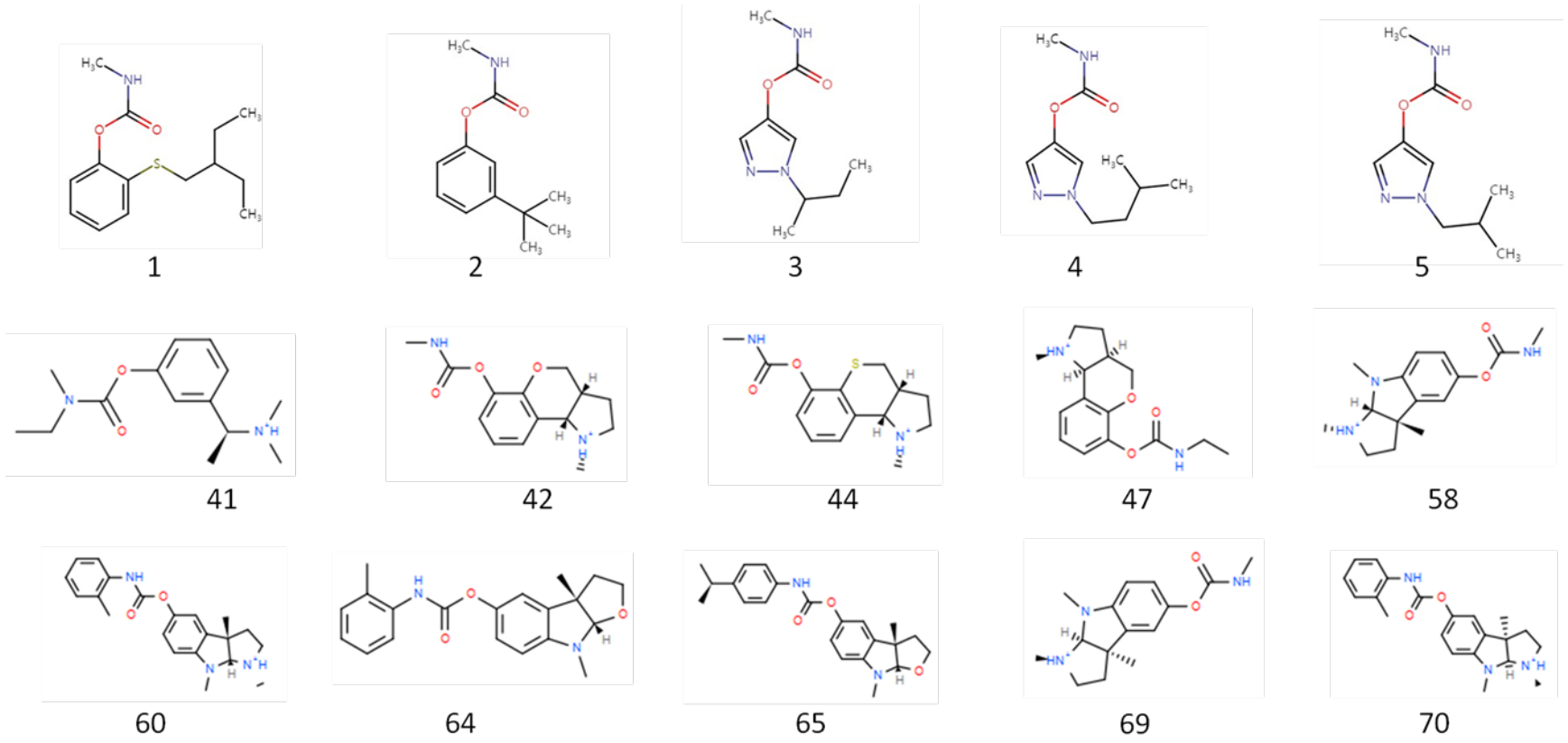

Figure S2 Test compounds for carbamates (Lee & Barron 2015; Temeyer et al 2014).

Table S1 AChE *in silico* site-directed mutagenesis docking simulation validations.[a,b,c]

Compound	**Wild_different species** F326 & G153			**Wild_Sandfly**[d] C326 & G153			**Mutation** C326 & S153		
	Exp pIC_{50}	Classification	Cal pIC_{50}	Exp pIC_{50}	Classification	Cal pIC_{50}	Exp pIC_{50}	Classification	Cal pIC_{50}
Paraoxon	8.64	Inhibitor	8.98	8.54	Inhibitor	7.86	5.42	Inhibitor	7.86
Malaoxon	8.01	Inhibitor	8.41	8.36	Inhibitor	7.62	5.71	Inhibitor	5.11
Eserine	7.55	Inhibitor	7.24	8.50	Inhibitor	7.24	7.07	Inhibitor	7.24
Propoxur	5.84-6.89	Inhibitor	7.24	6.66	Inhibitor	7.24	2.37	Non-inhibitor	-
Carbofuran	7.60	Inhibitor	7.24	7.62	Inhibitor	7.24	3.91	Inhibitor	7.24
1	-	-	-	7.85	Inhibitor	7.24	3.63	Non-inhibitor	-
2	-	-	-	7.44	Inhibitor	7.24	3.91	Non-inhibitor	-
3	-	-	-	7.89	Inhibitor	7.07	6.63	Inhibitor	6.45
4	-	-	-	7.13	Inhibitor	7.24	5.32	Inhibitor	5.12
5	-	-	-	7.12	Inhibitor	7.24	5.67	Inhibitor	5.12

[a] Amino acids represented by acronyms is listed in Table 1.
[b] Acronyms: experimental (Exp), calculated (Cal), 50 % inhibition concentration (IC_{50}), -log of the IC_{50} (pIC_{50}).
[c] Exp pIC_{50} values published previously (Temeyer et al 2014).
[d] Sandfly (*Phlebotomus papatasi*).

Table S2 AChE *in silico* site-directed mutagenesis docking simulation validations continued.[a,b,c]

Compound	**pIC_{50}**		
	Exp_wild[d]	Cal_wild	Cal_F326Y
Paraoxon	8.54	8.980	8.189
Carbofuran	7.62	7.865	7.074
Propoxur	6.66	7.241	6.450

[a] Amino acids represented by acronyms is listed in Table 1.
[b] Acronyms: experimental (Exp), calculated (Cal), -log of the 50 % inhibition concentration (pIC_{50}).
[c] Exp pIC_{50} values published previously (Temeyer et al 2014).
[d] AChE from *Phlebotomus papatasi* (Temeyer et al 2014).

Table S3 AChE *in silico* site-directed mutagenesis docking simulation results.[a,b]

Compound	pIC_{50}						
	Experimental	**Predicted**					
	(wild)	wild	W117Y	W117A	G152A	A235S	W317A
Carbamate 41	5.52	7.24	6.45	6.45	7.07	7.07	7.07
Carbamate 42	7.50	7.24	7.29	7.29	7.24	7.24	7.24
Carbamate 44	8.09	7.86	8.08	8.08	7.24	7.24	7.24
Carbamate 47	6.38	6.45	6.45	6.45	7.28	6.45	7.07
Carbamate 58	7.55	7.86	7.29	7.29	7.24	7.24	7.24
Carbamate 60	8.00	7.57	7.57	7.57	4.23	7.57	7.57
Carbamate 64	7.89	7.57	7.57	7.57	5.04	7.57	7.57
Carbamate 65	5.41	5.04	5.04	5.04	4.23	5.04	5.04
Carbamate 69	5.00	5.64	8.08	8.08	7.24	7.24	5.83
Carbamate 70	5.26	5.83	7.57	7.57	5.04	7.57	7.57
Chlorpyrifos-oxon	9.47	9.60	8.98	8.98	9.60	9.60	9.60
Dibutyryl dichlorovinyl phosphate	9.03	8.75	8.75	8.75	8.75	8.75	6.23
Paraoxon	8.64	8.98	8.98	8.98	8.89	8.89	8.89
Tolyl saligenin phosphate	7.55	7.86	6.23	6.23	6.45	6.45	6.45
Phenyl saligenin phosphate	6.59	6.23	6.23	6.23	6.23	6.23	6.23
Dichlorvos	6.46	8.41	7.62	7.62	7.62	8.41	7.62
Diisopropyl phosphorofluoridate	6.72	5.90	7.62	7.62	5.11	5.11	5.11
Trichlorfon	5.92	5.90	5.12	5.12	5.90	5.90	5.90
Malaoxon	8.01	8.41	8.41	8.41	8.41	7.62	7.62
Miapfox	4.89	5.11	7.62	7.62	6.23	5.11	5.11

[a] Amino acids represented by acronyms is listed in Table 1.
[b] Acronym: -log of the 50 % inhibition concentration (pIC_{50}).

Table S4 AChE *in silico* site-directed mutagenesis docking simulation results continued.[a,b]

Compound	pIC_{50}						
	Experimental	**Predicted**					
	(wild)	wild	F368A	F368W	F369L	F369S	F369W
Carbamate 41	5.52	7.24	6.45	6.84	6.45	6.45	7.07
Carbamate 42	7.50	7.24	7.24	7.01	7.24	6.45	7.07
Carbamate 44	8.09	7.86	7.24	7.01	7.24	7.24	7.24
Carbamate 47	6.38	6.45	6.45	6.22	7.07	6.45	6.45
Carbamate 58	7.55	7.86	7.24	7.63	7.01	7.01	7.01
Carbamate 60	8.00	7.57	7.57	6.76	6.45	6.45	4.23
Carbamate 64	7.89	7.57	7.57	7.34	6.45	6.45	4.23
Carbamate 65	5.41	5.04	5.04	5.04	6.45	6.45	4.23
Carbamate 69	5.00	5.64	7.24	5.12	6.45	6.43	6.45
Carbamate 70	5.26	5.83	7.57	4.76	6.45	5.04	4.23
Chlorpyrifos-oxon	9.47	9.60	8.98	8.89	8.48	8.89	8.48
Dibutyryl dichlorovinyl phosphate	9.03	8.75	8.75	9.37	7.34	6.23	7.62
Paraoxon	8.64	8.98	8.98	8.89	8.89	8.89	7.86
Tolyl saligenin phosphate	7.55	7.86	6.23	7.31	6.23	6.23	5.11
Phenyl saligenin phosphate	6.59	6.23	6.23	7.31	6.23	6.23	6.23
Dichlorvos	6.46	8.41	5.11	7.62	6.21	7.62	7.62
Diisopropyl phosphorofluoridate	6.72	5.90	5.11	5.11	5.11	5.11	7.62
Trichlorfon	5.92	5.90	5.90	5.90	5.90	6.23	5.90
Malaoxon	8.01	8.41	7.62	8.41	6.21	5.11	7.62
Miapfox	4.89	5.11	5.11	5.11	5.11	5.11	5.11

[a] Amino acids represented by acronyms is listed in Table 1.
[b] Acronym: -log of the 50 % inhibition concentration (pIC_{50}).

Table S5 EcR *in silico* site-directed mutagenesis docking simulation results.[a,b]

Mutant	Ligand	Relative Score[a]
D506E	E20	0.7
D506P	E20	14.2
T537A	E20	6.1
A592G	E20	0.3
A592F	E20	7.3
A592V	E20	0.9
N695A	E20	6.3

[a] Amino acids represented by acronyms is listed in Table 1.
[b] Binding free energy of mutant model relative to binding free energy of wild-type model.

References

Lee, S., Barron, M.G. (2015). Development of a 3D-QSAR model for acetylcholinesterase inhibitors using a combination of fingerprint, molecular docking, and structure-based pharmacophore approaches. *Toxicol. Sci.* **148(1)**, 60-70.

Temeyer, K.B., Tong, F., Totrov, M.M., Tuckow, A.P., Chen, Q., Carlier, P.R., Perez de Leon, A.A., Bloomquist, J.R. (2014). Acetylcholinesterase of the sand fly, Phlebotomus papatasi (Scopoli): construction, expression and biochemical properties of the G119S orthologous mutant. *Para. Vectors.* **7**, 577.