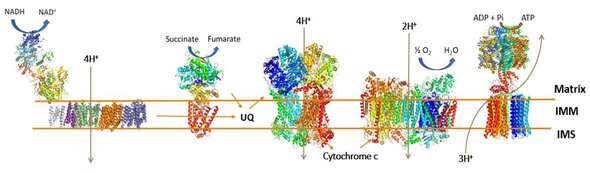
Development of QSAR models for the prediction of succinate-cytochrome c reductase inhibition

Alicia Rosell-Hidalgo, Anthony L. Moore and Taravat Ghafourian
Department of Biochemistry and Biomedicine, School of Life Sciences, University of Sussex, Brighton, BN1 9QG, UK. E-mail address: ar545@sussex.ac.uk

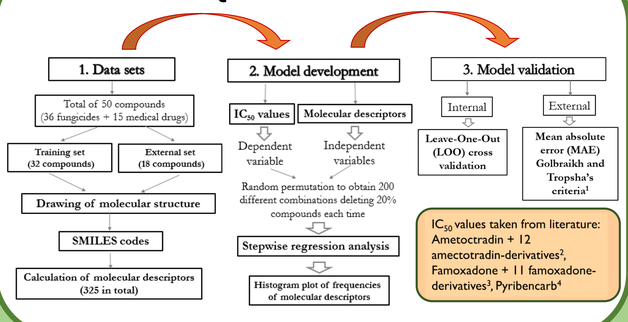
Introduction

Mitochondrial dysfunction can be the result of drug-induced toxicities. Given the structural and functional complexity of mitochondria, they can become the unintended off-target of some pharmaceutical drugs, and responsible, at least in part, for some of the undesirable side effects. In this study we focused on the mitochondrial electron transport chain (ETC), specifically in the activity of succinate-cytochrome c reductase (SCR). SCR plays a vital role in the ETC by catalysing the electron transfer reaction from succinate to cytochrome c and a disruption in this process could have serious consequences. This study was aimed at investigating the effects of several fungicides and pharmaceutical drugs on the activity of SCR to attempt to establish a relationship between their inhibitory activity and chemical characteristics through QSAR models.

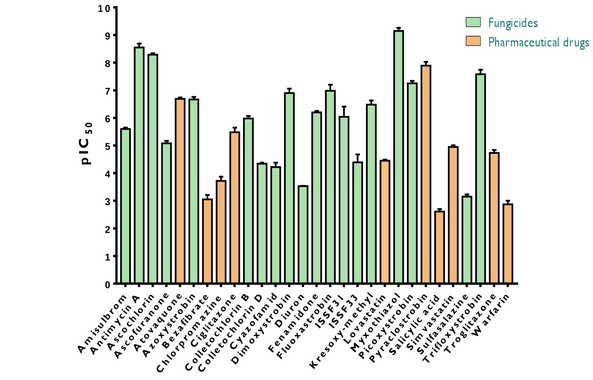


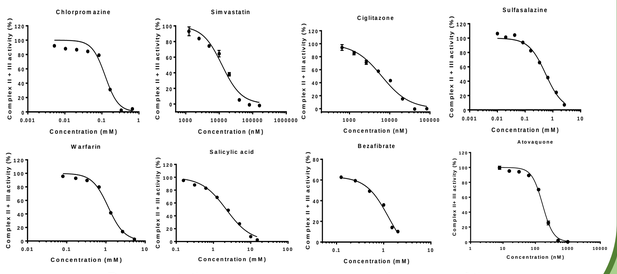
The activity of SCR was measured by monitoring the increase in absorbance of cytochrome c at 550 nm in a microplate spectrophotometer over a 4-min time-course at room temperature. Rat liver mitochondria (~3 µg/well) was added to an assay solution containing 200 mM sucrose, 25 mM KCl, 5 mM $MgCl_2$, 4 mM KH_2PO_4, 5 mM MOPS, 1 mM KCN, 1 mM ATP, 1 µM rotenone and 64 µM cytochrome c, pH 7.4. Reaction was started by the addition of 12.5 mM sodium succinate to each well.

QSAR workflow



Complex II + III enzymatic activity assays





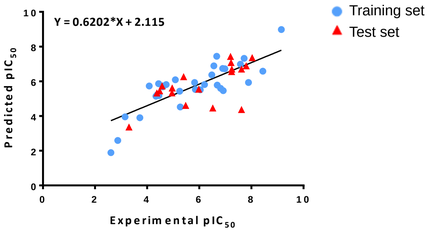
Representative dose-response curves for some of the pharmaceutical drugs tested

Conclusions

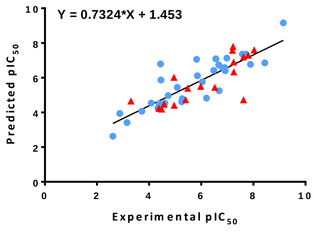
- ✓ The QSAR models highlighted the importance of several physicochemical and topological properties, including better inhibition by lipophilic molecules that are unionised in neutral pH (weaker acids or bases) and have conjugated double bonds (rather than single bond chains), with fewer rings.
- ✓ The QSAR models were successfully validated showing a good prediction accuracy in terms of ranking order of the IC_{50} values of the external compounds that were later tested in our laboratory.

QSAR models

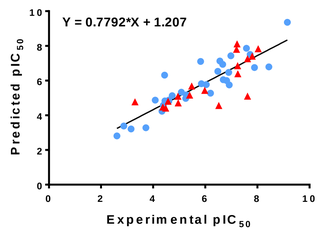
QSAR model 1: pIC_{50} = 2.808 - 0.271 b_max1len + 0.064 PEOE_VSA+3 - 0.746 Log S (pH = 7.40)



QSAR model 2: pIC_{50} = 2.384 + 2.119 Neutral form (pH = 7.4) - 0.679 Log S - 0.495 Number of rings (size 6)



QSAR model 3: pIC_{50} = 2.826 + 1.680 Neutral form (pH = 7.4) - 0.943 Log S - 0.741 Number of rings (size 6) - 0.274 b_max1le



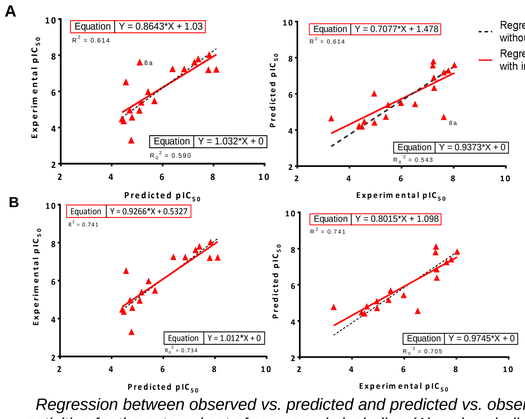
b_max1len: length of the longest single bond chain
PEOE_VSA+3: sum of the van der Waals surface area (Å²) of all the atoms where the partial charge is in the range [0.15, 0.20).
Log S (pH = 7.4): aqueous solubility at pH 7.4
Neutral form (pH = 7.4): fraction of compounds that is not ionised at pH 7.4

Validation of QSAR models 1, 2 and 3

		Model 1	Model 2		Model 3	
Training set	N	32	32		32	
	R^2	0.68	**0.77**		**0.82**	
	F	19.5	31.1		31.4	
	s	0.97	0.82		0.73	
	MAE (100 % data)	0.75	**0.55**		**0.50**	
	LOO Q^2	0.57	**0.68**		**0.73**	
Test set	N	18	18		18	
		with outliers	without outliers	with outlier	without outlier	with outlier	without outlier
	MAE (100% data)	0.875	0.653	0.683	**0.553**	0.635	**0.524**
	R^2	0.414	0.766	0.621	**0.793**	0.614	**0.741**
	R_0^2	0.382	0.746	0.597	0.792	0.590	0.734
	$R'_0{}^2$	0.110	0.537	0.552	0.763	0.543	0.705
	k	1.041	1.005	1.045	1.024	1.031	1.012
	k'	0.929	0.981	0.937	0.965	0.949	0.974
	$(R^2 - R'_0{}^2 / R^2)$	0.733	0.298	0.110	0.038	0.116	0.049
	$(R^2 - R_0^2 / R^2)$	0.077	0.026	0.038	0.002	0.040	0.009
	R_m^2	0.278	0.547	0.503	0.716	0.50	0.649
	$\Delta R_m{}^2$	0.094	0.230	0.009	0.050	0.01	0.046

QSAR validation: the selected model (3)

Among all three models, model 3 showed the best statistical quality and predictive abilities (R^2=0.82, Q^2=0.73)



Regression between observed vs. predicted and predicted vs. observed activities for the external set of compounds including (A) and excluding (B) the outlier (compound 8a).

Acknowledgements

This work is funded by a University of Sussex PhD studentship and supported by a grant from the Biotechnology and Biological Sciences Research Council (BBSRC).

References

1. Golbraikh, A. and Tropsha, A., *Beware of q2!* Journal of Molecular Graphics and Modelling, 2002. **20**(4): 269-276.
2. Zhu, X., *et al*., *Ametoctradin is a potent Qo site inhibitor of the mitochondrial respiration complex III.* J Agric Food Chem, 2015. **63**(13): 3377-86.
3. Wang, F., *et al*., *Design, syntheses, and kinetic evaluation of 3-(phenylamino)oxazolidine-2,4-diones as potent cytochrome bc(1) complex inhibitors.* Bioorg Med Chem, 2011. **19**(15): 4608-15.
4. Kataoka, S., *et al*., *Mechanism of action and selectivity of a novel fungicide, pyribencarb.* Journal of Pesticide Science, 2010. **35**(2): 99-106.