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Physicochemical evaluation of eight test compounds

MMV_OSDD

Report #: MMV_OSDD_13_004

3 December, 2013

Quality Statement:

This non-GLP study was conducted using established techniques in accordance with the relevant guidelines and standard operating procedures (SOPs) of the Centre for Drug Candidate Optimisation, Monash University. This report accurately reflects the raw data obtained during the performance of this study.

The results described represent part of an on-going lead optimisation/drug discovery program. The study has been conducted to provide preliminary information regarding physicochemical properties of the candidate compound(s) and has utilised methods appropriate for the discovery stage.

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Study number(s):	MMV_OSDD_13_004 (kinetic solubility) MMV_OSDD_13_005 (lipophilicity)

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A. Study Objective

To evaluate the physicochemical properties of MMV668957, MMV668958, MMV669304, MMV670438, MMV670944, MMV671651, MMV672723 and MMV672727 which are represented by the structures depicted in Table 1.

B. Experimental Methods

a) Calculated physicochemical parameters using ChemAxon JChem software

Theoretical physicochemical values were determined using the calculators in the ChemAxon chemistry cartridge via JChem for Excel software. Parameters calculated and a brief explanation of their relevance is given below.

Molecular Weight (MW): Ideally, MW should be less than 500 for good membrane permeability.

Polar Surface Area (PSA): Calculated using a simplified 2-dimensional modelling approach, which has been validated against a more sophisticated 3-dimensional modelling strategy. The value has been calculated at pH =7.4, which takes ionisation of the molecule into account. It is usually accepted that PSA values of less than approximately 120 $Å^2$ will provide acceptable oral drug absorption and membrane permeability.

Freely Rotating Bonds: Number of single bonds that are not in a ring or constrained system and are not bound to a hydrogen atom. FRB should be less than or equal to 10 for good membrane permeability (See D. Veber et al, J. Med. Chem. 2002, 45, 2615-2623).

H Bond Donor / Acceptors: Number of hydrogen bond donors and acceptors gives an indication of the hydrogen bonding capacity of the molecule which is inversely related to membrane permeability. Ideally, the number of H-Bond donors should be less than 5 and the number of H-Bond acceptors should be less than 10.

pKa: Basic physicochemical measure of the acidity of a compound. In the context of drug development, the values themselves only indicate whether ionisation is likely to be relevant at physiological conditions.

b) Solubility Estimates using Nephelometry

Compound in DMSO was spiked into either pH 6.5 phosphate buffer or 0.01M HCl (approx pH 2.0) with the final DMSO concentration being 1%. Samples were then analysed via Nephelometry to determine a solubility range. (See C. D. Bevan and R. S. Lloyd, Anal. Chem. 2000, 72, 1781-1787).

c) LogD Measurement

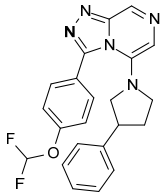
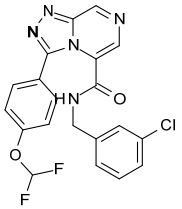
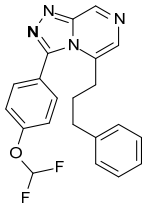
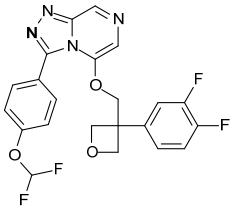
Partition coefficient values (LogD) of the test compounds were estimated by correlation of their chromatographic retention properties against the characteristics of a series of standard compounds with known partition coefficient values. The method employed is a gradient HPLC based derivation of the method developed by Lombardo (See F. Lombardo *et al*, J. Med. Chem. 2001, 44, 2490-2497).

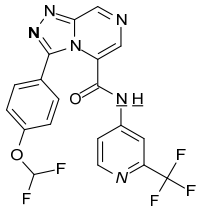
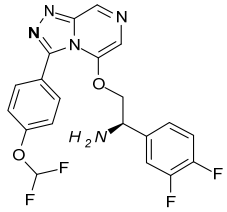
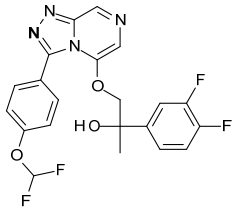
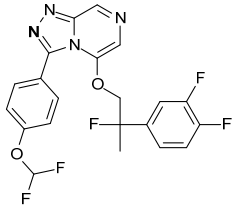
C. Results and Discussion

The results from the in silico screen for “drug-like” properties indicate that the molecular weight, polar surface area, freely rotatable bond and H-bond values of all eight compounds are within the ranges normally associated with “drug-like” compounds.

In general, the kinetic solubility and partitioning behaviours of these compounds would be considered moderate. MMV668957 showed an increase in solubility between pH 6.5 and 2.0, but only a minor reduction in LogD value between pH 7.4 and 3.0, suggesting a significant change in ionisation of the compounds between pH 3.0 and 2.0. This finding is contrary to the predicted pKa values for this compound and given the observed trends for the other compounds in this study most likely suggests ionisation of the pyrrolidine moiety under acidic conditions. In contrast, MMV671651 demonstrated a change in LogD value between pH 7.4 and 3.0, but little change in solubility between pH 6.5 and 2.0, suggesting a change in ionisation above pH 6.5, consistent with the predicted basic (pKa = 8.8) character of the compound. The remaining six compounds showed no change in solubility or LogD with change in pH.

Table 1: Summary of the physicochemical data for MMV668957, MMV668958, MMV669304, MMV670438, MMV670944, MMV671651, MMV672723 and MMV672727.

Compound	Batch #	Structure	MW	PSA (Å^2)	FRB	# H-Bond		pKa	LogD		Solubility (µg/mL)	
						Don	Acc		pH 3.0	pH 7.4	pH 2.0	pH 6.5
MMV668957	PCCBTAK-0075		407.42	55.6	5	0	6	1.8 – triazole (bold)	3.8	4.0	25 - 50	< 1.6
MMV668958	PCCBTAK-0075		429.81	81.4	6	1	7	1.6 – triazole (bold)	2.9	2.9	12.5 - 25	12.5 - 25
MMV669304	PCCBTAK-0127		380.39	52.3	7	0	5	1.8 – triazole (bold)	3.5	3.4	6.3 - 12.5	6.3 - 12.5
MMV670438	PCCBTAK-0234		460.38	70.8	7	0	9	1.7 – triazole (bold)	3.0	2.9	12.5 - 25	12.5 - 25

Compound	Batch #	Structure	MW	PSA (Å^2)	FRB	# H-Bond		pKa	LogD		Solubility (µg/mL)	
						Don	Acc		pH 3.0	pH 7.4	pH 2.0	pH 6.5
MMV670944	PCCBTAK-0275		450.32	94.3	6	1	8	9.4 – amide (acidic, underline) 2.1 – pyridine (italic) 1.4 – triazole (bold)	2.6	2.5	25 - 50	50 - 100
MMV671651	PCCBTAK-0284		433.36	89.2	7	2	8	8.8 – amine (italic) 1.7 - triazole (bold)	1.6	2.7	12.5 - 25	6.3 - 12.5
MMV672723	PCCBTAK-0367		448.37	81.8	7	1	9	1.7 - triazole (bold)	2.9	2.9	12.5 - 25	12.5 - 25
MMV672727	PCCBTAK-0371		450.36	61.5	7	0	7	1.7 - triazole (bold)	3.6	3.6	6.3 - 12.5	6.3 - 12.5