The European Lead Factory: A Collaborative Approach to Drug Discovery

(1) ESC Chemistry: F. MacLeod, A. Morrison, E. Beck, P. Jones, J. Hewitt, M. Matheson, J. Schulz, J. Robinson, L. Robinson, J. Gillespie, M. Huggett, M. Rajamanickam; ESC CMC: A. Pannifer, V. Hamon; ESC Biology: S. McElroy, M. Speake, G. Baillie, E. Russell, J. Rainard, A. Porter, G. Pandarakalam, N. Clark, D. Tegazzini. (2) PPSC: H. Rutjes, C.A.A. van Boeckel, N. Smits, E. van Doornmalen, T. W. Lam, M. Bras. (3) C. S. Schofield; J. J. Brem; M. A. McDonough. (4) Leiden University: M. Van der Stelt, F. J. Janssen, P. P. Geurink, H. Ovaa, H. S. Overkleeft, A. C. M. van Esbroeck, M. P. Baggelaar, H. den Dulk.

(1) European Screening Centre Newhouse, Biocity Scotland, University of Dundee, Lanarkshire, UK, ML1 5UH. (2) Pivot Park Screening Centre, Kloosterstraat 9 5349 AB, Oss, NL. (3) Department of Chemistry, University of Oxford, 12 Mansfield Road, Oxford, OX1 3TA. (4) Department of Molecular Physiology, Leiden University, Leiden, NL.

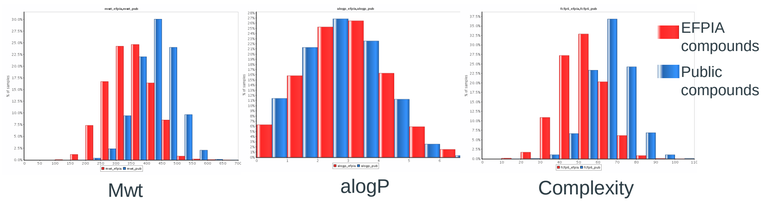
For further information contact; Fraser MacLeod, f.macleod@dundee.ac.uk

The European Lead Factory

The **European Lead Factory (ELF)** is a public-private drug discovery partnership consisting of 30 organisations throughout Europe that is funded by the Innovative Medicines Initiative (IMI). The goal of the ELF is to enable pre-competitive drug discovery by identifying and validating new biological targets that are amenable to small molecule intervention.

Joint European Compound Library (JECL)[1]

- The **JECL** is a high quality and diverse compound collection
- The collection is comprised of approximately 300 000 high quality, lead like compounds contributed by seven EFPIA partners
- A public compound collection (PCC) has been added to provide a further 200 000 bespoke compounds to the JECL

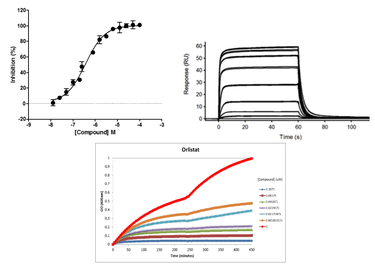


The European Screening Centre (ESC)[2]

- The ESC screens the target against the JECL to generate a Qualified Hit List (QHL) of up to 50 compounds
- Hit expansion activities are initiated by the medicinal chemistry team to provide an Improved Hit List (IHL)
- Compounds from the QHL or IHL may be used to gain crystallography, selectivity and ADME data
- The programme owner has exclusive rights to the structures and data generated for three years

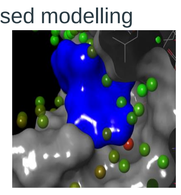
QHL Hit Characterisation[2]

- **Target activity:** Biochemistry, Cell-based assays
- **Target Engagement:** Biophysics: SPR, MST, TSA
- **Mode of Action:** Reversibility, Competitivity, Kinetics



Hit series Validation

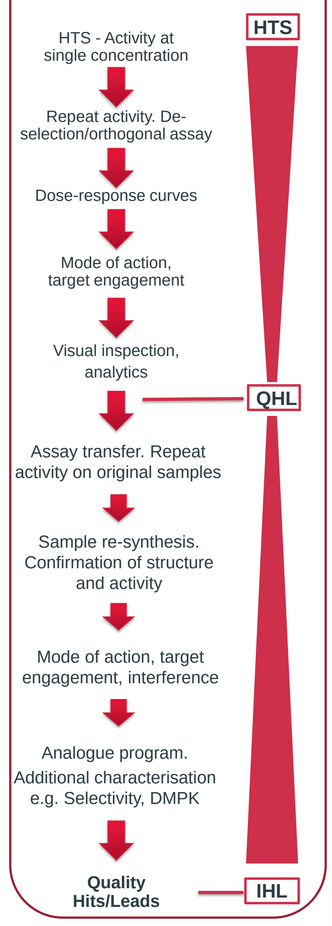
- Analogue design to establish SAR, improve potency, selectivity and drug-like properties
- Ligand & structure based modelling
- Cheminformatics
- Protein production
- X-ray crystallography



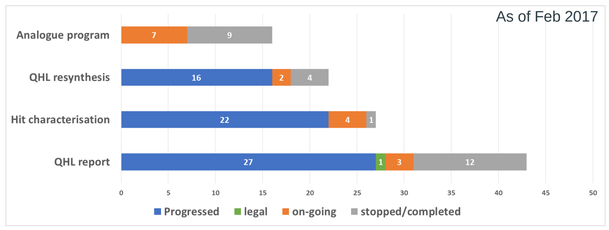
Validated Hit Series

- Proven chemical structure
- Target activity and specificity
- Emerging and progressive SAR
- Properties favourable for optimisation

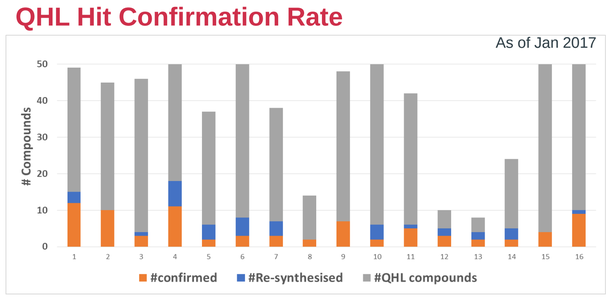
Overview of Process



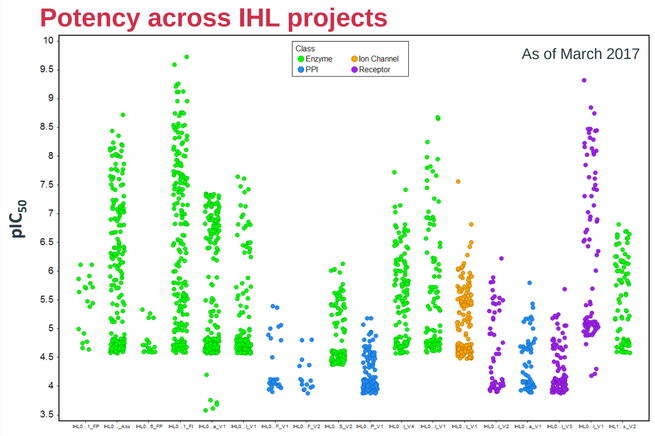
IHL Program Progression



QHL Hit Confirmation Rate

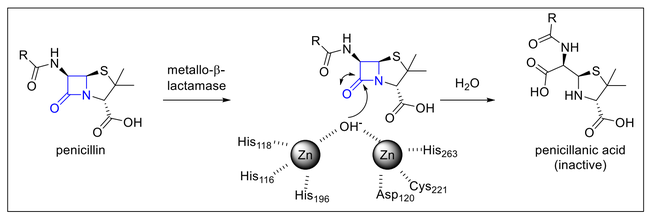


Potency across IHL projects



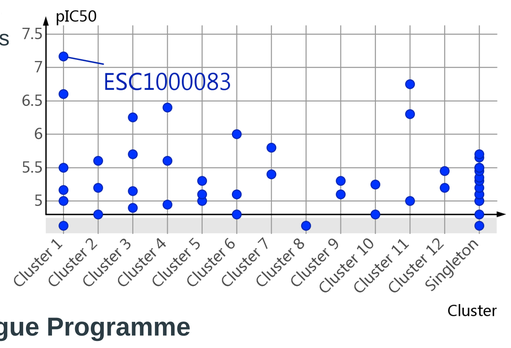
Case Study 1: New Delhi Metallo-β-Lactamase 1 (NDM-1)

- Antibiotic resistance represents a major threat to global healthcare
- Metallo-β-lactamases (MBLs) are zinc dependent hydrolases that catalyse the hydrolysis of β-lactam antibiotics
- NDM-1 is capable of hydrolysing antibiotics of last resort including the carbapenems
- Objective was to discover novel inhibitors of NDM-1
- Building in cross-MBL against VIM and IMP was also highly desirable



Screening

- QHL contained 50 compounds that consisted of 28 structural clusters -15 were singletons
- Eighteen compounds were selected for resynthesis - 13 confirmed activity
- Orthogonal SPR and ^{19}F NMR assays confirmed binding



ESC1000083
NDM-1 pIC_{50} **7.5**
SPR pK_D **7.7**
LE **0.51**
lipE **4.7**
AlogP **4.26**
LogD **2.8**
HBA / HBD **3 / 2**
PSA **53**

QHL to Improved Hit List

- **ESC100083** prioritised for further SAR
- 137 analogues prepared
- **Picomolar** inhibitors of **NDM-1** developed
- Broad-based MBL activity observed
- Activity improvements of **100-fold** *vs.* NDM-1 and VIM-2 and **1000-fold** *vs.* IMP-1
- Protein crystallography confirmed binding mode
- Schofield and co-workers working with IMI ENABLE (European Gram-negative Antibacterial Engine) to develop these early stage compounds towards clinical trials

For further information contact; Christopher Schofield, christopher.schofield@chem.ox.ac.uk

Analogue Programme

ESC1000083
pIC_{50}
NDM-1 **7.5**
VIM-2 **6.4**
IMP-1 **4.8**
LE = **0.51**
LipE = **4.70**

ESC1000382
pIC_{50}
NDM-1 **8.1**
VIM-2 **8.4**
IMP-1 **7.8**
LE = **0.48**
LipE = **3.44**

ESC1000406
pIC_{50}
NDM-1 **9.3**
VIM-2 **8.4**
IMP-1 **7.8**
LE = **0.47**
LipE = **5.77**

Cellular Profiling

MIC values (mg/L) of meropenem against *Klebsiella pneumonia* in the presence and absence of RamA

Klebsiella pneumonia	NDM-1	+RamA	VIM-1	+RamA	IMP-1	+RamA
DMSO	64	32	4	16	16	16
ESC1000382 (25mg/L)	0.125	<=0.063	<=0.063	<=0.063	0.25	0.25

DMPK

	Kinetic Solubility^	Measured LogD	MLM*	MLH*	HLM*
ESC1000382	94	1.53	2.8	2.1	1.6

^ Micromolar
* Clearance, ml/min/g liver

Case Study 2: Diacylglycerol Lipase-α (DAGL-α)

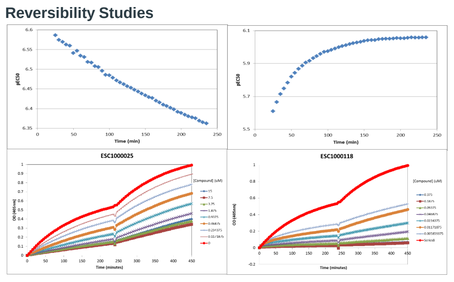
- DAGL-α is a serine hydrolase that hydrolyses diacylglycerol into the endocannabinoid 2-achidonylglycerol (2-AG) in the central nervous system
- Enzyme inhibition was hypothesized to have therapeutic benefit for obesity, metabolic disorders and neurodegenerative disorders
- No potent, selective inhibitors of DAGL-α had been described and were required to validate the target

Screening

- 302655 compounds screened in primary assay
- QHL contained 46 compounds consisting of 30 structural clusters, of which 20 were singletons
- Orthogonal ABPP profiling used to assess potency and serine hydrolase selectivity in mouse brain proteome

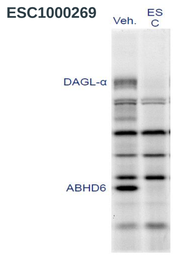
Target inhibition
para-Nitrophenyl butyrate activity assay

Reversibility Studies

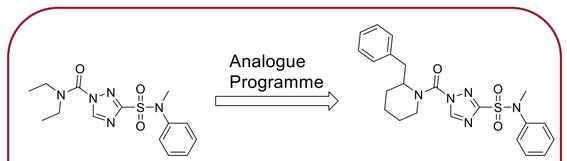


Orthogonal ABPP profiling used to assess potency and serine hydrolase selectivity in mouse brain proteome

ESC1000269

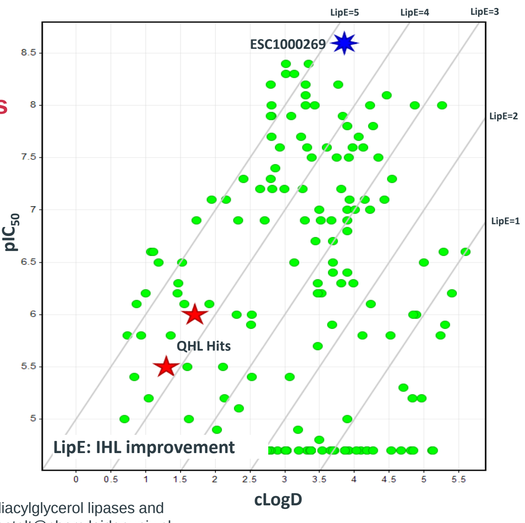


QHL to Improved Hit List: Triazole Urea Series



ESC1000032
Irreversible Inhibitor
IC_{50} 1.2 μM
MWt 337, cLogD 1.7, HBA 8
Off-targets = 3

ESC1000269
Irreversible Inhibitor
IC_{50} 3 nM
MWt 439, cLogD 3.85, HBA 8
Off-targets = 2
active in-vivo



For further information see PhD thesis: Discovery of novel inhibitors to investigate diacylglycerol lipases and α/β hydrolase domain 16A, F. J. Janssen, M. Van der Stelt, 2016. Email: m.van.der.stelt@chem.leidenuniv.nl

References

1. J. Besnard, P. Jones, A. Hopkins, A. Pannifer, " The Joint European Compound Library: boosting precompetitive research." *Drug Discov. Today*. **2015**, *20*, 181; 2. P. Jones, S. McElroy, A. Morrison, A. Pannifer, "The importance of triaging in determining the quality of output from high-throughput screening." *Future Med. Chem*. **2015**, *7*, 181.

The research leading to these results has received support from the Innovative Medicines Initiative Joint Undertaking under grant agreement n' 115489, resources of which are composed of financial contribution from the European Union's Seventh Framework Programme (FP7 / 2007-2013) and EFPIA companies' in-kind contribution.