Fragment-based drug discovery by NMR. Where are the successes and where can it be improved?

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1.1. Supplementary Materials and Methods

1.2. Materials

FDA information for New Molecular Entities (NMEs) and original biologics were extracted from https://www.fda.gov/drugs/drug-approvals-and-databases/drug-trials-snapshots. For each entry, a literature search was conducted to determine whether small molecules (only) were derived from an FBDD-approach.

The list of molecules in various stages of clinical trials was reproduced from a number of web sources and published reviews[1–4]. Some web-based materials were extracted from a detailed analysis of the website Practical Fragments[5], where blog articles dating up to 31st December 2020 and a table as listed in 2015 and 2018 posts[6,7] provided the starting point for this study.

A literature review was conducted for each compound, filtering out only molecules in which NMR had been involved at some stage of the drug discovery process. Subsequently, the exact NMR technique used was noted wherever possible. Statistics were derived from publicly available

resources, including databases and web blogs, therefore, they could include errors, inaccuracies or be incomplete.

1.3. Methods

Fragments molecular structures were reproduced using our CcpNmr software ChemBuild[8]. OpenBabel or iBabel 3.6 was used to convert PDB and MOL2 to SMILES format[9,10]. Smiles were created in the canonical (xc) format; hydrogens and pH were excluded from the calculations of the various molecular properties. Molecular weights, polar surface areas and other properties were calculated using the online tools available at http://www.cheminfo.org.
A collection of scripts for analysing smiles and plotting molecular similarities were written in Python using the Pandas[11], Numpy[12], SciPy[13], Matplotlib[8] libraries. The Pybel[9] package was used for calculating the molecular fingerprints from SMILES and the Tanimoto coefficient[14]. Scripts and raw data are available in the Vuister Lab GitHub repository at https://github.com/VuisterLab/scripts.git.

PDB codes used:

- AZD-3839 and BACE-1: 4B05;
- BCL and ligands: 6O0L, 4LVT, 2YXJ;
- MCL1 and ligands: 6QXJ, 6QYK, 6QYL, 6QZ5, 6QZ7, 6QZ6, 6QZB, 6QYN, 6QZ8, 6QYP, 6QYO.

PubMed Central searching query (https://www.ncbi.nlm.nih.gov/pmc/):

(

("nuclear"[All Fields] AND "magnetic"[All Fields] AND "resonance"[All Fields]) OR

("nuclear magnetic resonance"[All Fields]) OR

("nmr"[All Fields])

)

AND

(

("fragment-based"[All Fields] AND "drug discovery") OR

("drug"[All Fields] AND "discovery"[All Fields]) OR

("drug discovery"[All Fields])

)

AND

("2015/1/01"[PubDate] : "2020/12/31"[PubDate])

1.4. Tables

Technique	Common abbreviation	Structural information	Dimension-ality	Observed	Labelling	Reference
Group Selective Saturation Transfer Difference	GS-STD	Intermolecular interactions between the ^{15}N labelled amino group of the target and the ligand	1D	Ligand	^{15}N	15
Structural information using Overhauser effects and Selective labelling (SOS) Saturation Transfer Difference in real-time	SOS-STD	Distance restraints between selectively labelled residues (>3) and the ligand	1D	Ligand	^{2}H	16
Group Epitope Mapping by Saturation Transfer Difference	GEM-STD	Quantification of atomic level interactions between the target and the ligand	1D	Ligand	None	17
Water-Ligand Observed via Gradient Spectroscopy	WaterLOGSY	Quantification of solvent-exposed protons of the ligand in complex with the target	1D	Ligand	None	18
Solvent Accessibility, Ligand binding, and Mapping of ligand Orientation by NMR Spectroscopy	SALMON	Indication of solvent-exposed protons of the ligand in complex with the target	1D	Ligand	None	19
Target Immobilized NMR Screening	TINS	Identification of the ligand binding site on the target	1D	Ligand	None	20
Spin Labels Attached to Protein Side chains as a Tool to Identify interacting Compounds	SLAPSTIC	Interactions between the target side chain atoms and the ligand	1D	Ligand	Para-magnetic labels	21
Ligand Proton Pseudocontact Shifts	PCS	Intermolecular restraints that enable the identification of the target binding site and ligand orientation	Various	Ligand / Target	Para-magnetic labels	22
Protein-observed ^{19}F NMR	PrOF	Orthosteric binding site identification	Various	Target	^{19}F	23
Interligand NOEs for Pharmacophore Mapping	INPHARMA	Interaction and orientation of two adjacent ligands in a target binding pocket	2D	Ligand	None	24
Inter-ligand nuclear Overhauser effect	ILOE	Interaction and orientation of two adjacent ligands in a target binding pocket	2D	Ligand	None	25
^{19}F chemical exchange saturation transfer (CEST)	^{19}F-CEST	Structural restraints between a ligand and the target for a complex in intermediate exchange	Various	Target	^{19}F	26
Nuclear Overhauser Effect	NOE	Various intra- and inter-molecular distances between ligand atoms and/or ligand-target atoms	Various	Ligand / Target	Various	27–29

Table 1
A list of NMR commonly used techniques for elucidating the structural properties of ligand-target binding events.

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