Expanding FTMap for the fragment-based identification of pharmacophore regions in ligand binding sites

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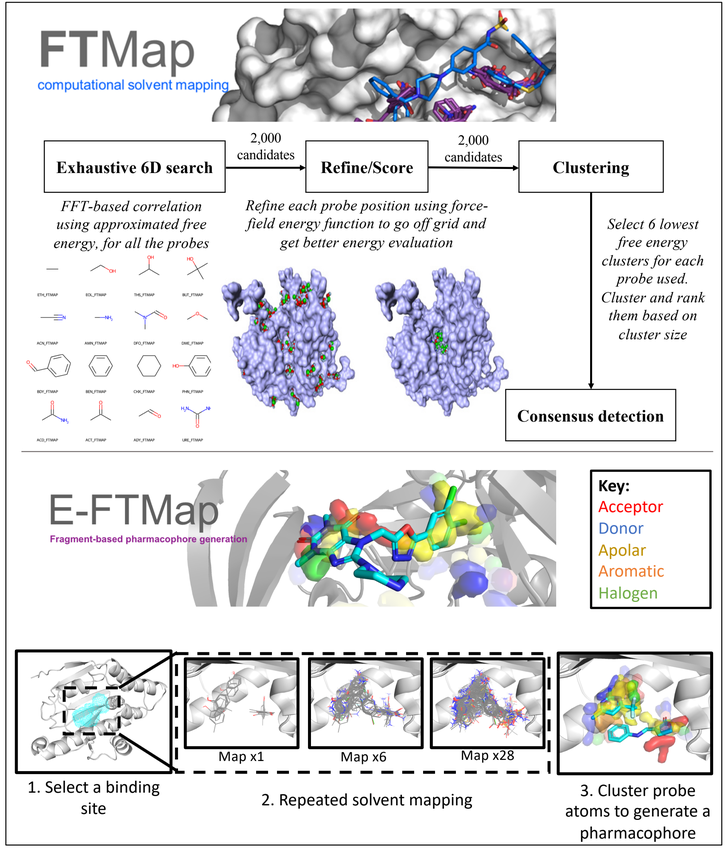
Abstract

In fragment-based drug discovery, the binding mode of a fragment bound to a hot spot is expected to be conserved as it is optimized into a larger ligand.[1,2] Therefore, predicting the locations of intermolecular interactions that are conserved in fragment-lead pairs is of great importance in the context of pharmacophore generation. To aid in the identification of pharmacophore regions in ligand binding sites we have developed E-FTMap, a computational solvent mapping algorithm which exhaustively maps binding sites with dozens of small organic probes, and identifies important interaction sites as atomic consensus sites (ACSs) where similar chemical groups bind. We validate E-FTMap against a set of 109 experimentally derived structures of fragment-lead pairs, finding highly ranked pharmacophore features that overlap with corresponding atoms in both fragment and lead compounds. Additionally, we compare mapping results to pharmacophores derived from ensembles of bound ligands, revealing that E-FTMap results tend to sample highly conserved protein-ligand interactions. Furthermore, we explore an application of E-FTMap in the context of virtual ligand screening by using mapping results to score and rank ligands on the basis of their binding affinity.

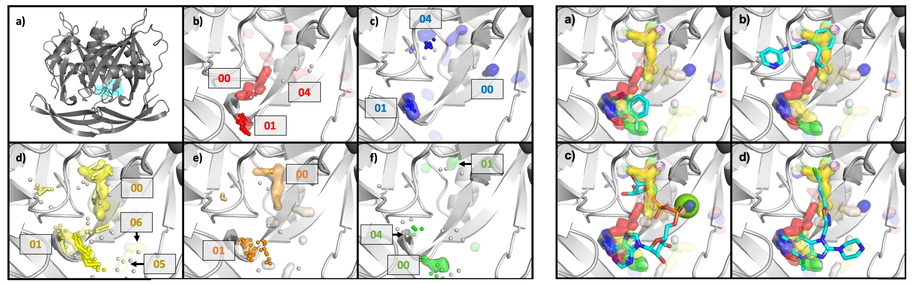
Methods



Comparing E-FTMap to ligand-derived pharmacophores

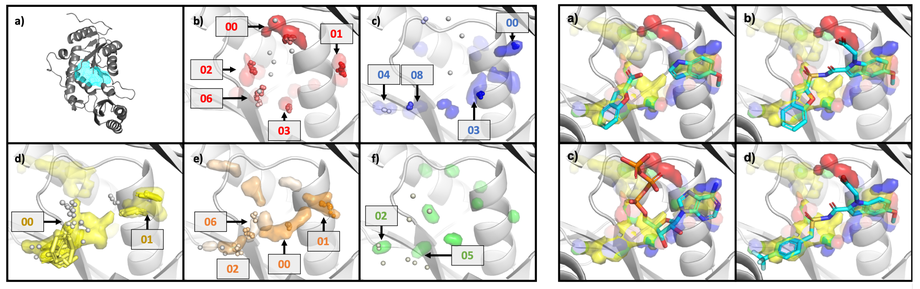
Background: To validate E-FTMap, we generated pharmacophores using *apo* receptor structures, and compared our results to pharmacophores based on ensembles of co-crystallized ligands extracted from homologous structures.

Example 1: Mapping of NUDT5



Left: E-FTMap probe atom clusters (surface) overlaid with ligand-derived atomic clusters (licorice). Larger clusters are shown in a more vibrant color, smaller clusters are shown in a duller shade.
Right: Bound ligands overlaid with E-FTMap results. a) Fragalysis fragment x1212, b) Fragalysis fragment x1024, c) ADP Ribose and Mg^{2+} (PDB: 2DSC), and d) TH5427, a potent inhibitor with an IC50 of 29 nM (PDB: 5NWH)

Example 2: Mapping of Pantothenate Synthetase



Left: E-FTMap probe atom clusters (surface) overlaid with ligand-derived atomic clusters (licorice). Larger clusters are shown in a more vibrant color, smaller clusters are shown in a duller shade.
Right: Ligands overlaid with E-FTMap results. a) Two bound fragments (PDB: 3IMC, 3IME), b) a ligand linking the fragment hits (PDB: 3IVX), c) bound ATP (PDB: 2A84), and d) an optimized ligand with an IC50 of 240 nM (PDB 4MUK).

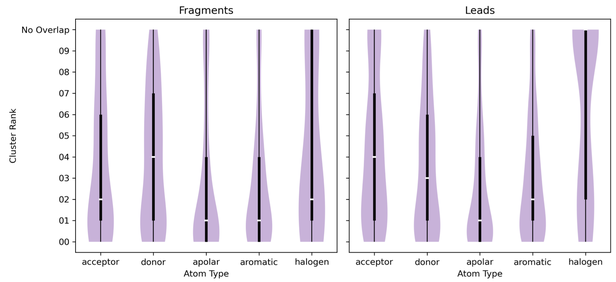
Overlap metrics for five benchmark systems

Protein	PDB[a]	# Ligs.[b]	Atom Type					
			Overlap	Acceptor	Donor	Apolar	Aromatic	Halogen
NUDT5	6GRU_AB	46	acs_lig	0.57	0.32	0.90	0.46	0.69
			lig_acs	0.96	0.93	0.78	0.86	0.61
Pantothenate Synthetase	3COV_B	38	acs_lig	0.84	0.47	0.70	0.61	0.21
			lig_acs	0.97	0.87	0.94	0.93	0.56
p38α MAPK	1R39_A	186	acs_lig	0.66	0.64	0.86	0.70	0.58
			lig_acs	0.60	0.87	0.72	0.71	0.56
Androgen Receptor	2AM9_A	31	acs_lig	0.59	0.33	0.55	0.58	0.39
			lig_acs	0.91	1.00	0.92	0.93	0.81
Nsp3- Mac1	7KR0_A	353	acs_lig	1.00	0.99	1.00	0.95	0.68
			lig_acs	0.96	0.66	0.77	0.77	0.59

[a]The PDB accession code and chain(s) which was mapped with E-FTMap. [b]The number of co-crystallized ligands used in the ligand–derived pharmacophore

Benchmarking on fragment-lead pairs

Background: We validate E-FTMap by mapping *apo* structures in a benchmark set containing crystal structures of 109 fragment–lead pairs.[3-5] Then we compare the overlap of predicted pharmacophore features with bound ligands.

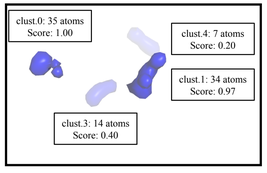


Above: Violin plots showing the ranks of E-FTMap clusters that overlap with fragment and lead atoms. The strongest cluster has rank 00, and the weakest cluster has rank 09.

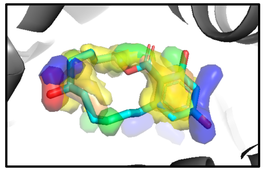
Using E-FTMap to score ligands

Background: Beyond providing a qualitative guide for identifying pharmacophore regions in ligand binding sites, we have begun developing a method which uses E-FTMap to score and rank ligands on the basis of their binding affinity.

1. Associate a score with each pharmacophore feature



2. Score ligands based on overlap with E-FTMap features



$$Lig\ Score = w_1 Score_{Don} + w_2 Score_{Acc} + w_3 Score_{Apo} + w_4 Score_{Aro} + w_5 Score_{Hal}$$

PDBBind w/ exact Kd reported and ≥ 10 structures
43 unique UNIPROT IDs 1,364 structures
↓
Map structure with weakest ligand bound
↓
Superimpose all bound ligands, and rank by E-FTMap score
↓
Correlate scores with $-\log(K_d)$

R_p correlating E-FTMap score with $-\log(K_d)$ for each protein in the PDBBind benchmark set

	Unweighted	Weighted
R <= 0	7	3
0 < R <= 0.3	13	8
0.3 < R <= 0.5	9	9
0.5 < R	14	23

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