AncPhore Manual

1. Introduction

AncPhore is a versatile tool for drug discovery, which is characterized by pharmacophore feature analysis and anchor pharmacophore steered molecules fitting and virtual screening. It involves ten types of pharmacophore features, including hydrogen bond donor, hydrogen bond acceptor, positively charged center, negatively charged center, metal coordination, aromatic ring, hydrophobic, and covalent bonding features, cation-π interaction, halogen bonding, and exclusion volumes (Figure 1). AncPhore can be used to generate a pharmacophore model for a ligand structure, an *apo*-protein structure, or a protein-ligand complex structure, and then used for virtual screening, target profiling, drug repositioning and so on.

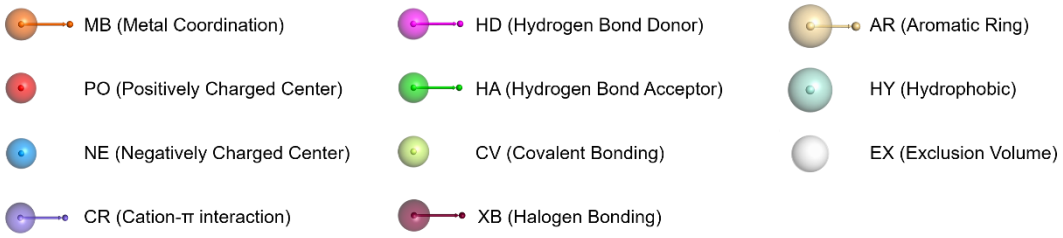

Figure 1. Graphic representation of eight types of pharmacophore features and exclusion volumes showed by a PyMol plugin.

2. Functions

(1) Pharmacophore feature analysis and model generation for a ligand structure, an *apo*-protein structure or a protein-ligand complex structure

(2) Ligand conformation generation by confab or genetic algorithm

(3) Ligand superimposition and similarity calculation according to their pharmacophore features

(4) Ligand fitting with a pharmacophore model

(5) High-throughput virtual screening by single-model or multi-model manner

3. Compatibility and Installing

Linux

AncPhore is expected to work on compatible 64-bit Linux systems, and it's tested and feasible on Centos (version 6, 7), Ubuntu (version 19.04, 20.04).

Windows

AncPhore current version has been tested on Windows 8/10 without any dependency.

Note: AncPhore old version(before 2021.11.10) must depend on C++ compiler suite which should be pre-installed. MinGW (https://osdn.net/projects/mingw/releases/) is recommended.

Installing

Click the downloaded AncPhore.zip and follow the instructions.

Note: Please send an email to ddtmlab_gbl@sina.com to obtain the unzipping password of the password-protected ZIP file.

4. Usage

Linux

Open the terminal and, if you installed AncPhore in the default location, type:

```
cd /home/AncPhore-centos7/bin
chmod +x AncPhore
./AncPhore --para parameter.txt
```

Windows

Open the Command Prompt after unzipping the package in your install location, type:

```
cd C:\Users\ibm\AncPhore-Window10\bin
./AncPhore.exe --para parameter.txt
```

See the parameters for details *via* type: *./AncPhore or .\AncPhore.exe*

Parameters

--para <parameter file (txt format)>

The parameter file contains all of the parameters listed below.

-p <input reference protein file (pdb format)>

--id <protein id when you generate pharmacophore model based on apo-protein without ligand, for example: 1jje >

--out <output path of pharmacophore model files based on apo-protein without ligand, for example: Example/Example_9/ >

-l <input reference ligand file (mol2/sd/sdf format)>

--refphore <generated reference pharmacophore (phore format)>

-p2 <input another protein file to match the reference protein (pdb format)>

-d <input database molecules (sd/sdf format)>

--dbphore <output generated database molecules pharmacophore (phore format)>

--mol <output aligned database molecules (sd/sdf format)>

--pdb <output aligned protein file (pdb format)>

--score <APScore (score format)>

--best <number of best molecules according to APScore, default 0.0>

CF <using confab algorithm to generate ligand conformations, which is the default (no need to assign value)>

GA <using genetic algorithm to generate ligand conformations>

outConformer <assign whether to generate database file which contains multiple conformations firstly, default no (no need to assign value)>

--maxConformer <if you assigned the parameter "outConformer", you would assign the max generated conformations of molecules during alignment, default 5>

usedMultiConformerFile <if you use the database file containing multiple conformations for

virtual screening, you must assign "usedMultiConformerFile", default no (no need to assign value)> *onlyProteinBased <if you want to get pharmacophore models for apo-protein, you must assign "onlyProteinBased", default no (no need to assign value)>* *onlyLigandBased <if you want to get pharmacophore models for ligand structure, you must assign "onlyLigandBased", default no (no need to assign value)>*

5. PyMol Plugin Usage

The AncPhore-Plugin.py is included in AncPhore.zip.

Usage and Note:

(1) Open Pymol

(2) Open the tab "Plugin"，click the button "Plugin Manager"

(3) Click the button "Choose file" in the panel "Install New Plugin", import the file ancphore.py, installation completed

(4) Start the plug-in AncPhore: in the tab "Plugin", under the Legacy plugin option, find the AncPhore plug-in and click the button "Read pharmacophore" to read in the file (.phore), then view the pharmacophore model.

6. Examples

(1) Generate a pharmacophore model for a protein-ligand complex

Command: *./AncPhore --para Examples/Example_1/parameter.txt*
Parameters: *-p Examples/Example_1/input_1jje_protein.pdb* *-l Examples/Example_1/input_1jje_ligand.mol2* *--refphore Examples/Example_1/output_1jje_complex.phore*
Results:

output_1jje_complex.phore:

	A	B	C	D	E	F	G	H	I	J	K	L	M	N
1			Alpha	Weight	Factor	Coordinate.x	Coordinate.y	Coordinate.z	HasNormal	Normal.x	Normal.y	Normal.z	Label	AnchWeight
2	RefID	1JJE												
3	Features	MB	1	1.5	1	57.085	27.116	31.789	1	57.964	28.718	30.981	5	2
4		MB	1	1.5	1	57.085	27.116	31.789	1	55.225	27.372	32.811	5	2
5		MB	1	1.5	1	56.673	27.387	34.6	1	55.225	27.372	32.811	5	2
6		NE	1	1.5	1	57.4845	27.1855	35.278	0	0	0	0	0	1
7		HA	1	1.2	1	59.123	26.66	32.244	1	61.75	25.596	32.274	55	1
8		HA	1	1.2	1	58.296	26.984	35.956	1	57.617	29.339	37.169	55	1
9		HY	0.7	0.5	1	57.8447	23.6259	37.7348	0	0	0	0	0	1
10		EX	0.837	0.5	1	56.975	19.207	32.487	0	0	0	0	0	1
11	Exclusion volumes	EX	0.837	0.5	1	58.044	20.114	31.911	0	0	0	0	0	1
12		EX	0.837	0.5	1	55.595	19.586	31.963	0	0	0	0	0	1
13		EX	0.837	1	1	56.502	16.907	38.157	0	0	0	0	0	1
14		EX	0.837	0.5	1	58.476	18.607	37.042	0	0	0	0	0	1
15		EX	0.837	0.5	1	59.64	19.513	37.256	0	0	0	0	0	1
16		EX	0.837	0.5	1	60.307	19.781	38.417	0	0	0	0	0	1
17		EX	0.837	0.5	1	60.265	20.279	36.22	0	0	0	0	0	1
18		EX	0.837	0.5	1	61.319	20.68	38.157	0	0	0	0	0	1
19		EX	0.837	0.5	1	61.313	20.997	36.822	0	0	0	0	0	1
20		EX	0.837	0.5	1	60.021	20.41	34.852	0	0	0	0	0	1

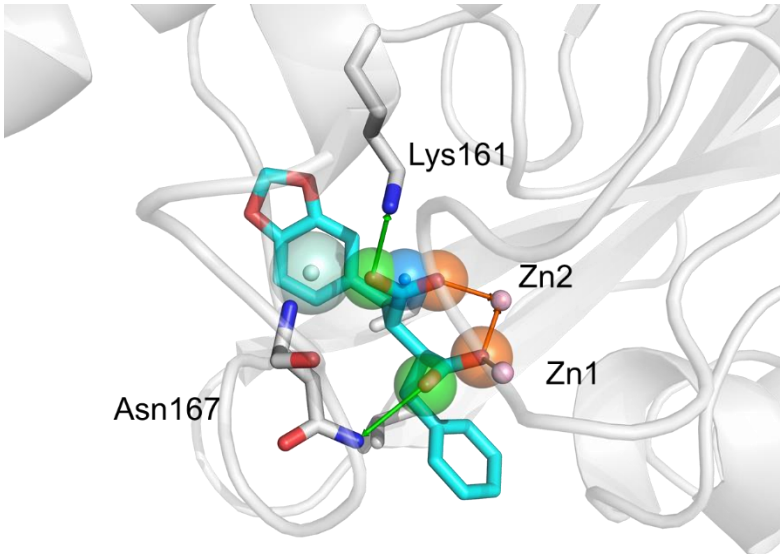

Figure 2. A pharmacophore model generated for a ligand-protein complex (PDB code: 1JJE).

(2) Generate a pharmacophore model for an *apo*-protein with ligand

Command:

./AncPhore --para Examples/Example_2/parameter.txt

Parameters:

-p Examples/Example_2/input_1jje_protein.pdb

-l Examples/Example_2/input_1jje_ligand.mol2

--refphore Examples/Example_2/output_1jje_onlyProtein.phore

onlyProteinBased

Results:

output_1jje_onlyProtein.phore

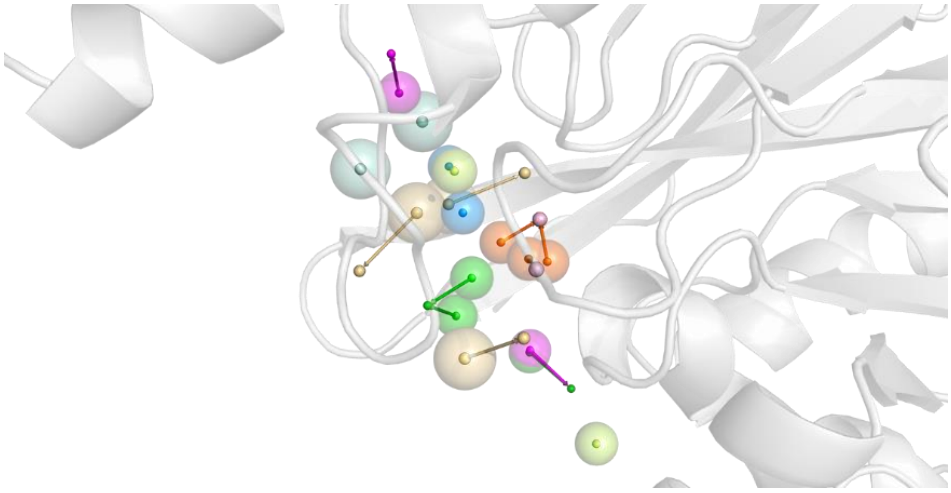

Figure 3. The result pharmacophore model based on *apo*-protein structure (PDB code: 1JJE).

(3) Generate a pharmacophore model for an *apo*-protein without ligand

Command: *./AncPhore --para Examples/Example_9/parameter.txt*
Parameters: *-p Examples/Example_9/input_1jje_protein.pdb* *--id 1jje* *--out Examples/Example_9/* *onlyProteinBased*
Results: *1) 1jje_1_refphore.phore* *2) 1jje_1_surface_residues.pdb*

(4) Virtual screening using one reference pharmacophore model

Command: *./AncPhore --para Examples/Example_4/parameter.txt*

Parameters:

--refphore Examples/Example_4/input_1jje_refphore.phore

-d Examples/Example_4/input_database_molecules.sd

--mol Examples/Example_4/output_aligned_db_molecules.sd

--dbphore Examples/Example_4/output_aligned_db_phore.phore

--best 10

--scores Examples/Example_4/output_aligned_db_score.score

Results:

1) output_aligned_db_molecules.sd

2) output_aligned_db_phore.phore:

	A	B	C	D	E	F	G	H	I	J	K	L	M
1	5n4s_ligand												
2	MB	1	1.5	1	56.9732	27.5191	31.1707	1	56.3425	29.2234	32.5804	5	1
3	MB	1	1.5	1	56.6385	27.2774	34.6266	1	54.9748	28.6694	33.862	5	1
4	NE	1	1.5	1	57.5069	27.1299	35.2497	0	0	0	0	0	1
5	HA	1	1.2	1	58.3753	26.9824	35.8729	1	59.8827	27.838	37.3848	5	1
6	HY	0.7	0.5	1	57.857	23.713	29.6918	0	0	0	0	0	1
7	HY	0.7	0.5	1	57.8484	23.4703	37.5413	0	0	0	0	0	1
8	$$$$												
9	6ew3_ligand												
10	MB	1	1.5	1	57.2623	27.0689	31.8796	1	56.1791	27.8128	29.9919	5	1
11	MB	1	1.5	1	56.4618	27.4596	34.8015	1	54.3654	28.3366	35.1562	5	1
12	NE	1	1.5	1	57.4254	27.2012	35.2478	0	0	0	0	0	1
13	HA	1	1.2	1	58.389	26.9428	35.694	1	59.7786	26.8518	37.5245	5	1
14	$$$$												
15	4pvt_ligand												
16	MB	1	1.5	1	57.2521	27.076	31.9569	1	56.0283	27.6997	30.1121	5	1
17	MB	1	1.5	1	56.4763	27.4523	34.7441	1	54.3206	28.2416	34.886	5	1
18	NE	1	1.5	1	57.4288	27.1988	35.222	0	0	0	0	0	1
19	HA	1	1.2	1	58.3813	26.9453	35.6999	1	59.7097	26.8042	37.5722	5	1
20	$$$$												

3) output_aligned_db_score.score:

	A	B	C	D	E	F	G	H	I
1	**dbId**	**dbEnergy**	**dbVolume**	**refId**	**refVolume**	**overlapVolume**	**resPharSize**	**APScore**	**APFitting**
2	5n4s_ligand	99.31	354.954	1JJE	348.184	233.672	6	0.8206	0.67
3	6ew3_ligand	141.9	179.781	1JJE	348.184	223.944	4	0.7422	0.7291
4	4pvt_ligand	142.4	179.781	1JJE	348.184	223.611	4	0.7396	0.7286
5	5y6e_ligand	107.7	316.92	1JJE	348.184	206.693	5	0.7178	0.6238
6	6hf5_ligand	61.61	406.81	1JJE	348.184	217.357	5	0.6967	0.6577
7	6d	70.23	242.779	1JJE	348.184	213.095	4	0.6882	0.6966
8	5mxr_ligand	68.38	375.311	1JJE	348.184	213.978	5	0.6793	0.6506
9	5mxq_ligand	94.08	343.812	1JJE	348.184	219.449	4	0.6759	0.7102
10	5k48_ligand	124.6	233.565	1JJE	348.184	209.74	4	0.6742	0.6778
11	Molecule	71.35	301.17	1JJE	348.184	213.1	4	0.6709	0.6959

(5) Virtual screening using multiple reference pharmacophore models

Command:

./AncPhore --para Examples/Example_5/parameter.txt

Parameters:

--refphore Examples/Example_5/input_multi_refphore.phore

-d Examples/Example_5/input_database_molecules.sd *--mol Examples/Example_5/output_aligned_db_molecules.sd* *--dbphore Examples/Example_5/output_aligned_db_phore.phore* *--best 10* *--scores Examples/Example_5/output_aligned_db_score.score*
Results: *1) output_aligned_db_molecules.sd* *2) output_aligned_db_phore.phore* *3) output_aligned_db_score.score*

(6) Generate conformers for database molecules

Command: *./AncPhore --para Examples/Example_6/parameter.txt*
Parameters: *-d Examples/Example_6/input_database_molecules.sd* *--maxConformer 10* *--mol Examples/Example_6/output_conformers.sd* *outConformer*
Results: *output_conformers.sd*

(7) Use multi-conformation file for virtual screening

Command: *./AncPhore --para Examples/Example_7/parameter.txt*
Parameters: *--refphore Examples/Example_7/input_1jje_refphore.phore* *-d Examples/Example_7/input_multi_conformers.sd* *--mol Examples/Example_7/output_aligned_db_molecules.sd*

--dbphore Examples/Example_7/output_aligned_db_phore.phore

--best 1000

--scores Examples/Example_7/output_aligned_db_score.score

usedMultiConformerFile

Results:

1) output_aligned_db_molecules.sd

2) output_aligned_db_phore.phore

	A	B	C	D	E	F	G	H	I	J	K	L	M
1	5n4s_ligand												
2	MB	1	1.5	1	56.9732	27.5191	31.1707	1	56.3425	29.2234	32.5804	5	1
3	MB	1	1.5	1	56.6385	27.2774	34.6266	1	54.9748	28.6694	33.862	5	1
4	NE	1	1.5	1	57.5069	27.1299	35.2497	0	0	0	0	0	1
5	HA	1	1.2	1	58.3753	26.9824	35.8729	1	59.8827	27.838	37.3848	5	1
6	HY	0.7	0.5	1	57.857	23.713	29.6918	0	0	0	0	0	1
7	HY	0.7	0.5	1	57.8484	23.4703	37.5413	0	0	0	0	0	1
8	$$$$												
9	4pvt_ligand												
10	MB	1	1.5	1	56.6986	27.1298	31.8909	1	55.5596	28.2326	30.2247	5	1
11	MB	1	1.5	1	56.749	27.8755	34.7004	1	55.3588	29.67	35.071	5	1
12	NE	1	1.5	1	57.4863	27.1531	35.0669	0	0	0	0	0	1
13	HA	1	1.2	1	58.2237	26.4306	35.4334	1	59.5265	25.5802	37.1273	5	1
14	HY	0.7	0.5	1	58.0577	23.7447	29.3977	0	0	0	0	0	1
15	$$$$												
16	5y6d_ligand												
17	MB	1	1.5	1	57.0776	27.5275	31.3001	1	56.6406	29.1706	32.849	5	1
18	MB	1	1.5	1	56.6041	27.0761	34.5016	1	54.8219	28.3511	33.8031	5	1
19	NE	1	1.5	1	57.4976	26.974	35.1058	0	0	0	0	0	1
20	HA	1	1.2	1	58.391	26.872	35.71	1	59.8762	27.7699	37.2192	5	1

3) output_aligned_db_score.score

	A	B	C	D	E	F	G	H	I
1	5n4s_ligand	109.2	354.954	1JJE	348.184	233.672	6	0.8206	0.67
2	4pvt_ligand	138.2	179.781	1JJE	348.184	218.701	5	0.7515	0.687
3	5y6d_ligand	119	301.17	1JJE	348.184	217.788	5	0.7499	0.6625
4	5mxq_ligand	95.69	343.812	1JJE	348.184	221.128	5	0.7444	0.6765
5	6ew3_ligand	138.7	179.781	1JJE	348.184	224.21	4	0.7429	0.7308
6	6ew3_ligand	141.9	179.781	1JJE	348.184	223.945	4	0.7422	0.7291
7	4pvt_ligand	144.1	179.781	1JJE	348.184	223.611	4	0.7396	0.7286
8	4pvt_ligand	138.5	179.781	1JJE	348.184	213.51	5	0.7375	0.6699
9	6ew3_ligand	141.3	179.781	1JJE	348.184	221.716	4	0.7335	0.7225
10	5mxq_ligand	97.2	343.812	1JJE	348.184	213.309	5	0.7305	0.664

Thanks for use!

AncPhore Team

Department of Medicinal Chemistry

West China School of Pharmacy

Sichuan University

Chengdu 610041, China

Email: ddtmlab_gbl@sina.com