Afpdb - An Efficient Protein Structure Manipulation Tool

This document is a tutorial on **Afpdb**. Throughout this tutorial, we will use two example antigen-antibody complex structures: 5cil.pdb and fake.pdb. 5cil.pdb contains three chains, L and H chains for the antibody light chain and heavy chain, respectively, and P chain for the antigen. fake.pdb contains two short chains, the L chain contains 4 residues including two with insertion codes and 3 missing residues, and the H chain contains two residues.

This document assumes you are using a Linux environment, however, we expect the notebook can run in other platforms if **Afpdb** has been installed.

```
from pathlib import Path
import os
pwd=Path(os.getcwd())
IN_COLAB=str(pwd)=="/content" # we are in Google Colab

if IN_COLAB:
    pwd=Path("/content/afpdb/tutorial")
    # remove local proxy setting
    os.environ["https_proxy"]=""
    os.environ["http_proxy"]=""
    os.environ["ftp_proxy"]=""
    # install afpdb
    if not os.path.isfile("INSTALL_AFPDB"):
        ! git clone git+https://github.com/data2code/afpdb.git && cd afpdb && pip install .
        ! touch INSTALL_AFPDB
    from IPython.display import Javascript
    display(Javascript('''google.colab.output.setIframeHeight(0, true, {maxHeight: 50000})'''))
    from IPython.display import HTML, display
    def set_css():
        display(HTML('''
          <style>
            pre {
                white-space: pre-wrap;
            }
          </style>
        '''))
    get_ipython().events.register('pre_run_cell', set_css)
else: # in a local jupyter notebook
    %reload_ext autoreload
    %autoreload 2
    # we assume afpdb has been preinstall

def install_pymol():
    try:
        import pymol2
    except Exception as e:
        if not IN_COLAB:
            print("Please install PyMOL first!")
        else:
            !pip install -q condacolab
            import condacolab
            condacolab.install()
            ! conda install conda-forge::pymol-open-source
            print("Colab does not have openssl 3.2.0, install it...")
            ! conda install conda-forge::openssl=3.2.0
            import pymol2

from afpdb.afpdb import Protein,util,RS,RL,ATS
import numpy as np
import pandas as pd
import re
# two example PDB files used in this tutorial
fn = pwd / "example_files/5cil.pdb"
fk = pwd / "example_files/fake.pdb"
```

Demo

To convince you *Afpdb* is effective and elegant, let us look at the following example, where we first identify contact residues in the antibody-antigen interface and then extract those residues into a separate PDB file. The structure display confirms the extraction was correct. These manipulations are not so straightforward using PyMOL or BioPython.

```
# load the ab-ag complex structure using PDB code name
p=Protein("5cil")

# show key statistics summary of the structure
p.summary()
```

```
Warning: residues with insertion code: H52A, H82A, H82B, H82C, H100A, H100B,
H100C, H100D, H100E, H100F, H100G, H100H, H100I, H100J, L27A
```

	Chain	Sequence	Length	#Missi Residue
0	H	VQLVQSGAEVKRPGSSVTVSCKASGGSFSTYALSWVRQAPGRGLEW...	220	

Notice that residue names do not start from "1" for chain "H" and chain "L" (see column "First Residue Name"), some residues in the CDR regions have insertion code (see column "#Insertion Code"), so that residue numbering goes from 1 to 211 for 212 residues on chain "L". It is good that learn that all residues have the complete set of backbone atoms "N,CA,C,O".

We can standardize residue naming:

```
print("Old P chain residue numbering:", p.rs("P").name(), "\n")
p.renumber("RESTART")
print("New P chain residue numbering:", p.rs("P").name(), "\n")
p.summary()
```

```
Old P chain residue numbering: ['671', '672', '673', '674', '675', '676',
'677', '678', '679', '680', '681', '682', '683']

New P chain residue numbering: ['1', '2', '3', '4', '5', '6', '7', '8', '9',
'10', '11', '12', '13']
```

	Chain	Sequence	Length	#Missi Residue
0	H	VQLVQSGAEVKRPGSSVTVSCKASGGSFSTYALSWVRQAPGRGLEW...	220	

```
# We can further remove the insertion code by
# renumber() returns a tuple, (Protein obj, old numbers)
p.renumber("NOCODE")[0].renumber("RESTART")
p.summary()
```

	Chain	Sequence	Length	#Missi Residue
0	H	VQLVQSGAEVKRPGSSVTVSCKASGGSFSTYALSWVRQAPGRGLEW...	220	
1	L	EIVLTQSPGTQSLSPGERATLSCRASQSVGNNKLAWYQQRPGQAPR...	212	

To predict the structure with AlphaFold, we can replace missing residues with Glycine.

```
print("Sequence for AlphaFold modeling, with the 20 missing residues replaced by Glycine:")
print(">5cil\n"+p.seq(gap="G")+"\n")
```

```
Sequence for AlphaFold modeling, with the 20 missing residues replaced by
Glycine:
>5cil
VQLVQSGAEVKRPGSSVTVSCKASGGSFSTYALSWVRQAPGRGLEWMGGVIPLLTITNYAPRFQGRITITADRSTSTAYL
```

```
# identify H,L chain residues within 4A to antigen P chain
binder, target, df_dist=p.rs_around("P", dist=4)

# show the distance of binder residues to antigen P chain
df_dist[:5]
```

	chain_a	resi_a	resn_a	resn_i_a	atom_a	chain_b	resi_b	resn_b	resn_i_b	a
90	P	437	6	6	OG1	H	97	98	98	
72	P	435	4	4	OD1	L	252	33	33	
21	P	433	2	2	N	L	314	95	95	
12	P	432	1	1	ND2	L	311	92	92	
93	P	438	7	7	ND2	H	107	108	108	

```
# create a new PDB file only containing the antigen and binder residues
p=p.extract(binder | "P")
# save the new structure into a local PDB file
p.save("binding.pdb")

# display the PDB struture, default is show ribbon and color by chains.
p.show(show_sidechains=True)
```

☹

The above script shows a few strength of Afpdbs:

- User friendly: We use "'Protein()'" to create a protein object, use "'save()'" to create a PDB file. We will explain both methods can accommodate multiple formats.
- Residue selection: We use "H:L" and "P" to select all Ab and Ag residues, we use `binder | ''P''` to Boolean combine selections. Afpdb supports a "contig" syntax to select residues with their human readable labels.
- Visualization: We here show how a project object can be visualized within Jupyter Notebook. We will explain how Afpdb integrates with PyMOL for more powerful 3D visualization and transfer the residues selections from Afpdb into PyMOL.
- AI ready: Afpdb was created to meet our own protein AI design needs. We show a brief examples of how Afpdb prepares an input for AlphaFold, and we will show more sophiscated AI use cases.

Fundermental Concepts

Internal Data Structure

Data Members

From the demo, we see how fast Afpdb analyses are, which is because Afpdb uses NumPy arrays for storing protein structure data, which is very different from most existing packages relying on the model-chain-residue-atom tree structure.

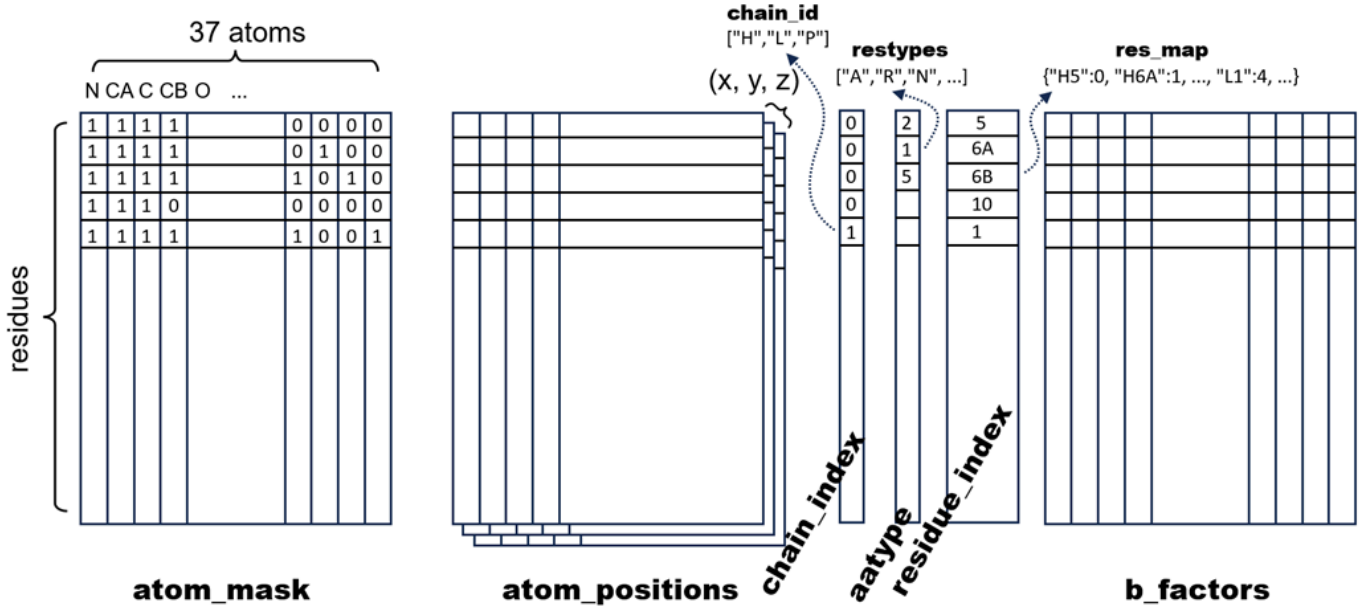
To better understand why **Afpdb** can manipulate protein structures efficiently, it is helpful to first discuss how the structure data in a PDB file is stored within the AlphaFold's Protein class. The following descriptions are mostly taken from DeepMind's alphafold/common/protein/protein.py file.

```
# Cartesian coordinates of atoms in angstroms. The atom types correspond to
# residue_constants.atom_types, i.e. the first three are N, CA, C.
atom_positions: np.ndarray  # [num_res, num_atom_type, 3]

# Amino-acid type for each residue represented as an integer between 0 and
# 20, where 20 is 'X'.
aatype: np.ndarray  # [num_res]

# Binary float mask to indicate presence of a particular atom. 1.0 if an atom
# is present and 0.0 if not. This should be used for loss masking.
atom_mask: np.ndarray  # [num_res, num_atom_type]

# Residue index as used in PDB. It is not necessarily continuous or 0-indexed.
residue_index: np.ndarray  # [num_res]
```

We modified residue_index to dtype "<U6" in order to be able to store residue id with possible insertion code. A residue_index was an integer in AlphaFold, in our extension, residue_index can include insertion code, e.g., "121", "122A", "112B". The maximum residue id can be 9999 if we allow insertion codes with two letters, otherwise, 99999 if all insertion codes are single letters.

```
# 0-indexed number corresponding to the chain in the protein that this residue
# belongs to.
chain_index: np.ndarray  # [num_res]

# B-factors, or temperature factors, of each residue (in sq. angstroms units),
# representing the displacement of the residue from its ground truth mean
# value.
b_factors: np.ndarray  # [num_res, num_atom_type]

# Chain names - this is added by us. DeepMind's original code does not memorize the chain name in the PDB file
# chain names became A, B, C, etc. afpdb keeps the original chain names in the PDB file
chain_id: np.ndarray # [num_chains]
```

Illustration



Notice the first axis of all ndarrays are the number of residues. The second axis can be either one or the number of possible non-hydrogen atom types (which is 37).

The above is AlphaFold's Protein class, developers access this data via Afpdb's Protein class, which is a wrapper of AlphaFold's protein class.

In Afpdb's Protein class, it contains an attribute called `data`, which points to an AlphaFold's Protein instance.

```
from afpdb import Protein
p=Protein(fk)
print(p.data.atom_positions[:, 1])
# outputs
# [[  6.30200005 -14.27600002 -18.36100006]
# [  9.34000015 -16.29100037 -17.33600044]
# [  9.67800045 -16.51799965 -13.59899998]
# [ 16.89500046 -17.8920002   -7.03000021]
# [ 25.96999931   6.87099981 -19.03800011]
# [ 25.11199951   9.9829998  -16.98600006]]
print(p.res_map)
# outputs
# {'L5': 0, 'L6A': 1, 'L6B': 2, 'L10': 3, 'H3': 4, 'H4': 5}
```

In the example above, we obtain the (X, Y, Z) coordinates of the CA atoms (i.e., we specify 1 for the 2nd axis of `atom_positions`) corresponding to the first six residues via accessing `p.data`.

To further speed up structure manipulations, Afpdb's Protein class maintains a dictionary called `res_map`, which maps the full residue id, in the format of {chain}{residue_index}, into its residue position. {chain}{residue_index} is the user-friendly format for identifying a specific residue,

e.g., "C122A" is the residue index "122A" on chain "C". This residue corresponds to an integer index in the NumPy arrays internally. Developers should remember to call `self._make_res_map()` to update this mapping dictionary, if a method requires the underlying mapping to be refreshed after changing the positions/types of residues for any location. The example above shows how the six full residue names are mapped to the numpy arrays.

For the rest of the tutorial, when we use the term "Protein" class, we refer to afpdb's Protein class, which can be imported using `from afpdb.afpdb import Protein`. We will no longer mention AlphaFold's Protein class to avoid confusion, infact, users do not need to directly access `p.data`, only developers need to be aware that `p.data` points to mulitple NumPy arrays behind the scene.

Example

Let us further explain the previous example and examine `p.data` numpy arrays using fake.pdb.

The following is the fake.pdb file, which contains 4-residue chain L and a 2-residue chain H.

```
MODEL     1
ATOM      1  N   GLU L   5       5.195 -14.817 -19.187  1.00  1.00           N
ATOM      2  CA  GLU L   5       6.302 -14.276 -18.361  1.00  1.00           C
ATOM      3  C   GLU L   5       7.148 -15.388 -17.731  1.00  1.00           C
ATOM      4  CB  GLU L   5       5.794 -13.368 -17.248  1.00  1.00           C
ATOM      5  O   GLU L   5       6.658 -16.231 -17.006  1.00  1.00           O
ATOM      6  CG  GLU L   5       6.934 -12.664 -16.494  1.00  1.00           C
ATOM      7  CD  GLU L   5       6.461 -11.817 -15.327  1.00  1.00           C
ATOM      8  OE1 GLU L   5       7.282 -11.138 -14.677  1.00  1.00           O
ATOM      9  OE2 GLU L   5       5.243 -11.804 -15.070  1.00  1.00           O
ATOM     10  N   ILE L   6A      8.444 -15.321 -17.934  1.00  1.00           N
ATOM     11  CA  ILE L   6A      9.340 -16.291 -17.336  1.00  1.00           C
ATOM     12  C   ILE L   6A      9.657 -15.849 -15.925  1.00  1.00           C
ATOM     13  CB  ILE L   6A     10.604 -16.433 -18.162  1.00  1.00           C
ATOM     14  O   ILE L   6A     10.192 -14.739 -15.685  1.00  1.00           O
ATOM     15  CG1 ILE L   6A     10.228 -16.847 -19.590  1.00  1.00           C
ATOM     16  CG2 ILE L   6A     11.540 -17.469 -17.523  1.00  1.00           C
ATOM     17  CD1 ILE L   6A     11.401 -17.319 -20.426  1.00  1.00           C
ATOM     18  N   VAL L   6B      9.339 -16.725 -14.982  1.00  1.00           N
ATOM     19  CA  VAL L   6B      9.678 -16.518 -13.599  1.00  1.00           C
ATOM     20  C   VAL L   6B     11.024 -17.188 -13.330  1.00  1.00           C
ATOM     21  CB  VAL L   6B      8.569 -17.028 -12.666  1.00  1.00           C
ATOM     22  O   VAL L   6B     11.242 -18.372 -13.679  1.00  1.00           O
ATOM     23  CG1 VAL L   6B      8.960 -16.919 -11.194  1.00  1.00           C
ATOM     24  CG2 VAL L   6B      7.268 -16.234 -12.927  1.00  1.00           C
ATOM     40  N   GLN L  10      15.587 -17.776  -7.649  1.00  1.00           N
ATOM     41  CA  GLN L  10      16.895 -17.892  -7.030  1.00  1.00           C
ATOM     42  C   GLN L  10      16.721 -18.330  -5.569  1.00  1.00           C
ATOM     43  CB  GLN L  10      17.616 -16.572  -7.093  1.00  1.00           C
ATOM     44  O   GLN L  10      16.270 -17.557  -4.746  1.00  1.00           O
ATOM     45  CG  GLN L  10      17.963 -16.094  -8.483  1.00  1.00           C
ATOM     46  CD  GLN L  10      18.781 -14.822  -8.460  1.00  1.00           C
ATOM     47  NE2 GLN L  10      20.052 -14.951  -8.083  1.00  1.00           N
ATOM     48  OE1 GLN L  10      18.284 -13.727  -8.786  1.00  1.00           O
ATOM    862  N   LEU H   3      27.368   6.440 -19.107  1.00  1.00           N
ATOM    863  CA  LEU H   3      25.970   6.871 -19.038  1.00  1.00           C
ATOM    864  C   LEU H   3      25.761   7.794 -17.840  1.00  1.00           C
ATOM    865  CB  LEU H   3      25.089   5.647 -18.873  1.00  1.00           C
ATOM    866  O   LEU H   3      25.979   7.398 -16.661  1.00  1.00           O
ATOM    867  CG  LEU H   3      25.225   4.606 -19.964  1.00  1.00           C
ATOM    868  CD1 LEU H   3      24.282   3.430 -19.748  1.00  1.00           C
ATOM    869  CD2 LEU H   3      24.962   5.190 -21.355  1.00  1.00           C
ATOM    870  N   VAL H   4      25.291   9.008 -18.090  1.00  1.00           N
ATOM    871  CA  VAL H   4      25.112   9.983 -16.986  1.00  1.00           C
ATOM    872  C   VAL H   4      23.672  10.399 -16.963  1.00  1.00           C
ATOM    873  CB  VAL H   4      25.976  11.257 -17.222  1.00  1.00           C
ATOM    874  O   VAL H   4      23.143  10.875 -17.973  1.00  1.00           O
ATOM    875  CG1 VAL H   4      25.686  12.319 -16.177  1.00  1.00           C
ATOM    876  CG2 VAL H   4      27.466  10.902 -17.258  1.00  1.00           C
TER     877      VAL H   4
```

```
ENDMDL
END
```

We first read in fake.pdb, there is a warning indicating there are residues with insertion codes: 6A and 6B. This alerts users that there may be duplicates in the integer portion of the residue ID, so it is a good habit not to rely the assumption that residue integer IDs are all unique. AlphaFold's original code did not support insertion codes. **Afpdb** made modifications to eliminate this limitation.

```
p=Protein(fk)
# show the sequences by chain
print(p.seq_dict())
# print the total number of residues
print("Total length:", len(p), "\n")
print("Residue labels:", p.rs().name(), "\n")
print("Their chain labels:", p.rs().chain())
```

```
Warning: residues with insertion code: L6A, L6B
{'L': 'EIVXXXQ', 'H': 'LV'}
Total length: 6

Residue labels: ['5', '6A', '6B', '10', '3', '4']

Their chain labels: ['L', 'L', 'L', 'L', 'H', 'H']
```

Our protein object p contains an attribute `data`, which points to AlphaFold's NumPy arrays. Let us examine the backend NummPy data within `p.data`. Here p.rs() is a "residue selection" that points to all residues. The residue selection, to be explained later, can be used to fetch the metadata for residues (their labels, chain names, amino acid types, etc).

```
g=p.data
# we used p.rs() to select ALL residues
# we used p.rs().name() to get all residue labels, which is stored in g.residue_index
print(g.residue_index, "\n")
# p.res_map maps each unique residue label to their row indices in the NumPy arrays.
print(p.res_map, "\n")
```

```
['5' '6A' '6B' '10' '3' '4']

{'L5': 0, 'L6A': 1, 'L6B': 2, 'L10': 3, 'H3': 4, 'H4': 5}
```

```
# we used p.rs().chain() to get the chain name of each residue
# chain information is stored in g.chain_index
print(g.chain_index, "\n")
# To obtain the list of chain names in the order of their appearance in the PDB file,
# we should use the chain_id() method instead
print(p.rs().chain(), "\n")
print(p.chain_id())
```

```
[0 0 0 0 1 1]

['L', 'L', 'L', 'L', 'H', 'H']

['L', 'H']
```

g.chain_index shows the first 4 residues belong to chain index 0, and the next 2 residues belong to chain index 1. AlphaFold uses BioPython to parse PDB files, Chains are numbered in the order of their appearance in the PDB file, so chain L is indexed as chain 0 and chain H is indexed as chain 1. It is important to remember that the best practice is to access data without assuming how chains appear in a certain order in the PDB or in Protein objects. It is safer to access structure or sequence data by their chain names.

What are the chain names for 0 and 1? AlphaFold's original code did not store chain names and chains were named as A, B, C, etc. afpdb have improved AlphaFold's code to add a new data member `chain_id()` into its Protein class.

Array `aatype` stores the identity of amino acids as defined in alphafold.common.residue_constants.py:

`restypes = ['A', 'R', 'N', 'D', 'C', 'Q', 'E', 'G', 'H', 'I', 'L', 'K', 'M', 'F', 'P', 'S', 'T', 'W', 'Y', 'V']`

So index 6 translates into "E".

There are a total of 6 residues. By assuming residues are numbered sequentially, we imply three residues (7, 8, and 9) are missing in the PDB file. Missing residues are not stored in the NumPy arrays; they can only be inferred based on the gaps in the integer portion of the residue IDs.

```
# afres contains protein structure constants defined by AlphaFold
import afpdb.myalphafold.common.residue_constants as afres
print(g.aatype, "\n")
[ afres.restypes[x] for x in g.aatype ]
```

```
[ 6  9 19  5 10 19]

['E', 'I', 'V', 'Q', 'L', 'V']
```

Users do not have to use afres, residue types can be extracted with the residue selection object:

```
# Notice three missing residues are represented as 3 "X", but they are not there in the protein structure
# notice sequences from different chains are separated by ":"
print(p.seq(), "\n")
# retrieve sequence information via residue selection
p.rs().aa()
```

```
EIVXXXQ:LV

['E', 'I', 'V', 'Q', 'L', 'V']
```

There are different types of metadata for residues. In Afpdb, we use the following convention:

- **resn** refers to the residue_index, a string with an optional insertion code without chain name.
- **resi** refers to the internal numpy array indices, e.g., the resi for resn = '6A' is 1.
- **resn_i** refers to the integer portion of the resn, resn_i for residue 6B is integer 6. `p.res_map` maps the unique residue name, (chain + **resn**), into **resi**.

Note: Early Afpdb users use `p.data_prt`, although this is still supported for now, please switch to use `p.data` going forward.

There are a total of 37 possible atom types when all 20 amino acid types are combined (not counting hydrogens), AlphaFold uses an array of 6 x 37 to store atom 3D coordinates (we have 6 residues in this example). Therefore, the atom_positions is a 3-dimensional NumPy array, where the last dimension is for (X, Y, Z) coordinates:

```
g.atom_positions.shape
```

```
(6, 37, 3)
```

No residue will have all 37 atoms, They all have N, CA, C, and O, but do not necessarily have CB, CG, etc. Therefore, AlphaFold's code uses more memory than needed to store the coordinates. As PDB data are small, trading extra memory to gain convenience and speed with NumPy arrays is wise. This is why implementing new computations in Afpdb can be done quickly, as no biolerplate loopings are required to navigation the model/chain/residue/atom tree in order to access coordinate data, like what is the required for using the Biopython API.

As not all array members are used, we thus need to know what atoms in the atom_positions array contain real data using a binary array atom_mask:

```
print(ATS.i('CA'), "\n")
print(g.atom_mask[:, ATS.i('CA')], "\n")
print(g.atom_positions[:, ATS.i('CA')], "\n")
print(g.atom_mask[:, ATS.i('CG')], "\n")
print(g.atom_positions[:, ATS.i('CG')], "\n")
```

```
1

[1. 1. 1. 1. 1. 1.]

[[  6.30200005 -14.27600002 -18.36100006]
 [  9.34000015 -16.29100037 -17.33600044]
 [  9.67800045 -16.51799965 -13.59899998]
 [ 16.89500046 -17.8920002   -7.03000021]
 [ 25.96999931   6.87099981 -19.03800011]
 [ 25.11199951   9.9829998  -16.98600006]]

[1. 0. 0. 1. 1. 0.]

[[  6.93400002 -12.66399956 -16.49399948]
 [  0.           0.           0.        ]
 [  0.           0.           0.        ]
 [ 17.96299934 -16.09399986  -8.4829998 ]
 [ 25.22500038   4.60599995 -19.9640007 ]
 [  0.           0.           0.        ]]
```

```
str(~ p.ats('CA'))
```

```
'N,C,CB,O,CG,CG1,CG2,OG,OG1,SG,CD,CD1,CD2,ND1,ND2,OD1,OD2,SD,CE,CE1,CE2,CE3,NE,
NE1,NE2,OE1,OE2,CH2,NH1,NH2,OH,CZ,CZ2,CZ3,NZ,OXT'
```

We refers to an atom with its name instead of memorizing its index in the arrays. ATS is the atom selection class to be introduced later. `ATS.i` method converts an atom name into its integer index. We also show a simple "~" operator to demonstrate selection objects can be Boolean manipulated.

We see that all six residues have CA (C_α) atom, but only the 1st, 4th, and 5th residues (E, Q, L) have CG (C_γ). So the coordinates for those atoms within atom_positions array, but with corresponding atom_mask set to 0 should be ignored (these atoms are not present in the PDB structure). Although atoms with coordinates at the origin are likely masked-out atoms, we should not rely on that assumption. In fact, when AlphaFold2 prediction starts, all atoms are placed at the origin -- so-called black hole conformation. Therefore, we should rely on the atom_mask array to determine the presence of atoms and assume the coordinates of masked out atoms can be any garbage numbers.

You might worry that the extra array elements for masked atoms will create computational overhead. In the Developers' Note section, we will explain methods that convert this sparse coordinate NumPy array into a dense coordinate NumPy array. Indeed, eliminating non-existing atoms further accelerate the computation and that is used in Afpdb's analyses.

Lastly and similarly, b_factors are also stored in an array at the atom level. Only elements with atom_mask equaling 1 have meaningful b factors.

```
g.b_factors.shape
```

```
(6, 37)
```

To summarize, for a given Afpdb Protein object p, we store all key PDB information into a few NumPy arrays: `atom_positions, atom_mask, residue_index, chain_index, b_factors,` and `chain_id` within p.data. We also have a helper dictionary `p.res_map`.

These arrays make structure manipulations a breeze. End users do not need to access these NumPy arrays, the architecture is for developers to understand.

Contig

Based on how afpdb uses ndarray to store structure data, we can access a few residues and a few atoms is as easily as accessing certain rows and columns in these NumPy arrays. To specify what residues are of interest, we use a string format called **contig**. Contig is a string format introduced by recent protein AI packages such as RFDiffusion and ProteinMPNN, we adopt and slightly improve the syntax as detailed below.

Single Residue

`RESIDUE = CHAIN RESIDUE_INDEX` without a space inbetween

In PDB, each residue is assigned an integer id. In some cases, a residue label (RESIDUE_INDEX) can be followed by an insertion code, which is an alphabet letter (two letters in very rare cases). This is because when insertions were made to a residue, such as residue 100 in the antibody CDR loops, people prefer to call the new residues 100A and 100B so that the rest of the residues do not need to be renumbered.

The residue label alone without the chain name is not unique, because the same residue label can appear on different chains. Therefore, to fully specify a residue, we include their chain name. For example, a single-residue contig can be H99, H100A. Remember `p.res_map` maps a residue contig into its internal array index.

Note: Contig syntax cannot handle negative residue index. Residue index should not contain a negative integer. ***afpdb*** still can read such PDB files, but we should renumber the residues (introduced later) if we want to use the more advanced contig syntax.

Single Fragment

```
FRAGMENT = CHAIN[START ID][-][END ID]
```

To specify a continuous range of residues, we specify the start and end residues without the need to enumerate all residues inbetween. The chain name only occurs once in the beginning. The following are all valid contigs: H1-98, H-98, H98-, H, H98-100A, H98-100B, etc. H stands for the whole chain. H-98 has the start residue ID omitted, which means it starts from the first residue (which is not necessarily residue 1, as the numbering can start, e.g., from 5 in the PDB file fk.pdb). H98- covers 98 to the last residue. H98-100A does not include 100B. H98-100B includes both 100A and 100B. A single residue contig is a special fragment contig.

A chain can contain missing residues, e.g., the PDB structure may contain residues 92, 93, 98, 99, 100A, 100B, and 101, where the integer portion of the residue labels jumps from 93 to 98. We, therefore, assume residues 94-97 are missing. H92-99 will select all residues including 92, 93, 98, and 99, i.e., the contig is not broken simply because there maybe missing residues implied.

Multiple Same-Chain Fragments

To specify multiple fragments within the same chain:

```
IN-CHAIN_FRAGMENTS = FRAGMENT[,[START ID][-][END ID]]*
```

Chain letter should only appears once. E.g., H-5,10-15 covers residues from the beginning to residue 5 on chain H, followed by residue 10 to residue 15 on the same chain.

Note: Fragments may overlap, however, the only the unique residues end up in the residue selection object to be introduced later. We should avoid overlap, as it makes the contig string confusing.

Multiple Fragments

```
CONTIG = IN-CHAIN_FRAGMENTS[:[IN-CHAIN_FRAGMENTS]]*
```

We can specify multiple same-chain fragments by concatenating them with ":". An example contig is H-98:L:P2-5, which specifies the whole L chain, H residues from the beginning to residue ID 98, and includes residues in the P chain from 2 to 5. You can certainly describe same-chain fragments using the general contig syntax, e.g., H-5,10-15:L-10 is equivalent to H-5:H10-15:L-10. In the Demo section, we use "H:L" to select the whole Ab chains and "P" to select the whole antigen chain. Those are valid contig strings.

If there are multiple fragments within a chain, we should try to keep them together, so that these segments are not separated by segments from other chains. As mentioned later, `align()`, and `rmsd()` do not reorder the underlying residue selections in its arguments, as these methods requires the one-to-one residue mapping of the two lists of input residues. `extract()` methods do not reorder residues specified in the contig, however, it requires the all residues of the same chain stay together and ordered ascendingly in the contig string, otherwise, it will generate an error. This is because **Afpdb** makes a heavy use of a method called `chain_pos`, which assumes all residues colocated on the same chain appear sequentially. Do not worry too much as **Afpdb** code validates the input contig. Contig strings can be canonicalized, which is described later.

Special Keywords

None, "ALL" match all residues. "", "NONE", and "NULL" matches no residue.

TODO: We should patch the PDB parser, so that broken chains (if ever occurs), will be defragmented.

Note

As mentioned above, there are multiple ways of describing the same set of residues. Contig strings can be canonicalized using the `__str__` method of a residue selection object. `Print(rs)` or `str(rs)` will implicitly call the `__str__` method.

```
p=Protein(fk)
str(p.rs("L6B,5:L-6B"))
```

```
Warning: residues with insertion code: L6A, L6B
'L5-6B'
```

inplace

Many methods support a boolean argument called `inplace`, this is a practice copied from the well-known Python packaged called **Pandas**. `inplace=True` will modify the original protein object without allocating new memory, while `inplace=False` will leave the original object unmodified and return the modified structure as a new object.

```
p=Protein(fn)
# inplace=False by default
print(p.chain_id())
# q has the two chains switched
q=p.extract("H:L")
print(p.chain_id()[:2], q.chain_id(), p==q, "\n")

# p is modified if inplace=True
q=p.extract("H:L", inplace=True)
print(p.chain_id(), q.chain_id(), p==q)
```

```
['L', 'H', 'P']
['L', 'H'] ['H' 'L'] False

['H' 'L'] ['H' 'L'] True
```

Clone

If a method does not support inplace and you would like to keep the original object unmodified, you can first clone a copy:

```
p=Protein(fn)
print(p.chain_id(), "\n")
q=p.clone()
# we modify q in place
q.extract("P", inplace=True)
# q is modified
print(q.chain_id(), "\n")
# p is untouched
print(p.chain_id())
```

```
['L', 'H', 'P']

['P']

['L', 'H', 'P']
```

Selection

Selection is a powerful concept in PyMOL and we support both atom selections and residue selections. We consider the concept of selection a major feature missing in Biopython, which led to tedious residue/atom enumerations and made the code less readable and more error prone. **Afpdb** stores residues as rows and atoms as columns in the first two axes of the numpy arrays, a residue selection contains the row indices as its data member and an atom selection contains a column index as its data member. Therefore, a residue selection and an atom selection can be used to form a new numpy array with two axes. Notice the same atom selection is shared by all residues, which greatly simplify the usage and backend implementation. We do not support the selection that combines C_α from residue 1 and C_β from residue 2. We do not see this as a limitation in our practice.

Atom Selection

afpdb provides a class named `ATS` for an atom selection. `ATS` has an attribute `data`, which is a numpy array storing the atom column indices.

There are 37 unique atoms across all 20 amino acids. The syntax for users to select atoms is by providing an atom list, or as a comma-separate string. use either "N,CA,C,O" or ["N","CA","C","O"] for backbone atoms. All atom names are in upper case only.

Internally, atom_positions and atom_mask have 37 elements in their `axis=1` dimension, one per atom type. Therefore the atom list is represented as a numpy integer array internally. For "N,CA,C,O", they are represented as `ats.data = np.array([0, 1, 2, 4])`, so `atom_positions[:, ats.data]` will select the atom coordinates for the 4 backbone atoms. This is a key benefit of using numpy to store PDB structural data.

Note: We often use "ats" for an atom selection object, since the word "as" is a reserved word in python.

```
print(ATS("N,CA,C,O"), "\n")
print("ATS data member:", ATS("N,CA,C,O").data, "\n")
# Atoms are reorded in the selection object
print(ATS(["CA","C","N","O"]), "\n")
# Atoms are deduplicated in the selection object
print(ATS("CA,CA"), "\n")
# special keywords
print("ALL", ATS("ALL"), "\n")
print(None, ATS(None), np.all(ATS(None).data==ATS().data), "\n")
print("Empty string, NONE, NULL", ATS("").data, ATS("NONE").data, ATS("NULL").data, "\n")
```

```
N,CA,C,O

ATS data member: [0 1 2 4]

N,CA,C,O

CA

ALL
N,CA,C,CB,O,CG,CG1,CG2,OG,OG1,SG,CD,CD1,CD2,ND1,ND2,OD1,OD2,SD,CE,CE1,CE2,CE3,NE

None
N,CA,C,CB,O,CG,CG1,CG2,OG,OG1,SG,CD,CD1,CD2,ND1,ND2,OD1,OD2,SD,CE,CE1,CE2,CE3,NE
True
```

As shown above, the order of the atoms does not matter and dupliates are removed and column indices are sorted. We have also special keywords: None and "ALL" for selecting all atoms, "", "NONE", and "NULL" for an empty selection. You can also initialize an ATS object with a set/list/tuple/pandas.Series of atom strings, a numpy array, another ATS object. If a single integer is provided, it is treated as one atom index. Basically, ATS supports all sensible input formats.

If a method expect an ATS object as an argument, we can provide the value using any of the format that can be used to initialize an ATS object. For developers, the first thing the method should do is to cast the input argument into an ATS object using the following idiom:

```
ats=ATS(ats)
```

We consider it is a good practice to create an ATS object from a Protein object using `p.ats()`, so that we can avoid explicitly using the ATS class (one less class to memorize):

```
p=Protein()
ats=p.ats("N,CA,C,O")
print(ats, "\n")
print(p.ats([0,1,2,3]), "\n")
print(len(ats), "\n")
# not_full can be empty or less than 37 atoms
print(ats.is_empty(), ats.is_full(), ats.not_full(), "\n")
# method i() converts an atom name into its integer index
print(ATS.i("CA"), ATS.i("CB"), "\n")
# in operator
print("N" in ats, "CB" in ats, 2 in ats, "\n")

print(p.ats_not("N,CA,C,O"), "\n")
print(p.ats2str(ATS("N,C,CA,O")), "the same as", str(ATS("N,C,CA,O")), "\n")
```

```
N,CA,C,O

N,CA,C,CB

4

False False True

1 3

True False True

CB,CG,CG1,CG2,OG,OG1,SG,CD,CD1,CD2,ND1,ND2,OD1,OD2,SD,CE,CE1,CE2,CE3,NE,NE1,NE2,

N,CA,C,O the same as N,CA,C,O
```

We also support the set operations &, |, ~ (not), + (the same as |), - (a-b means in a but not in b). We strongly recommend to use these Boolean operators as they make the code easier to read:

```
ats1=ATS("N,CA,C,O")
ats2=ATS("C,O,CB")
print("or:", ats1 | ats2, "\nsame as +:", ats1+ats2, "\n")
print("and:", ats1 & ats2, "\n")
print("minus:", ats1 - ats2, "\n")
print("not:", ~ats1, "\nnot not:", ~~ats1, "\n")
```

```
or: N,CA,C,CB,O
same as +: N,CA,C,CB,O

and: C,O

minus: N,CA

not:
CB,CG,CG1,CG2,OG,OG1,SG,CD,CD1,CD2,ND1,ND2,OD1,OD2,SD,CE,CE1,CE2,CE3,NE,NE1,NE2,
not not: N,CA,C,O
```

Residue Selection

We use a contig string previously described to select residues. To create a residue selection use the `RS` class or preferrably the `p.rs()` method. A residue selection object contains a `data` member, which is literally an integer numpy.ndarray. E.g., rs.data=np.array([0,1,9]). Therefore, `atom_positions[rs.data][ ATS('N,CA,C,O').data]` will capture the 3D coordinates of the 4 backbone atoms for residue 1st, 2nd, and 10th.

For developers, `atom_positions[rs.data, ATS('N,CA,C,O').data]` does not work, as it pairs the residue id with the corresponding atom id. We could do `atom_positions[np.ix_(rs.data, ATS('N,CA,C,O').data)]` to select the subarray.

The following is an example where we only extract the backbone atoms from chain H and L and save them into a new PDB file.

```
p=Protein(fn)
p.extract("H-5:L-5", ats="C,CA,N,O", inplace=True)
p.save("bb.pdb")
print("\n".join(util.read_list("bb.pdb")[:15])+"\n...\n")
p.show(style="stick", show_sidechains=True)
```

```
HEADER    AFPDB PROTEIN                         11-Jun-24   XXXX
MODEL     1
ATOM      1  N   VAL H   1      32.582   3.997 -22.715  1.00  1.00           N
ATOM      2  CA  VAL H   1      31.113   4.261 -22.594  1.00  1.00           C
ATOM      3  C   VAL H   1      30.727   4.737 -21.206  1.00  1.00           C
ATOM      4  O   VAL H   1      30.948   4.033 -20.247  1.00  1.00           O
ATOM      5  N   GLN H   2      30.105   5.899 -21.101  1.00  1.00           N
ATOM      6  CA  GLN H   2      29.676   6.451 -19.826  1.00  1.00           C
ATOM      7  C   GLN H   2      28.215   6.827 -20.036  1.00  1.00           C
ATOM      8  O   GLN H   2      27.888   7.435 -21.037  1.00  1.00           O
ATOM      9  N   LEU H   3      27.368   6.440 -19.107  1.00  1.00           N
ATOM     10  CA  LEU H   3      25.970   6.871 -19.038  1.00  1.00           C
ATOM     11  C   LEU H   3      25.761   7.794 -17.840  1.00  1.00           C
ATOM     12  O   LEU H   3      25.979   7.398 -16.661  1.00  1.00           O
ATOM     13  N   VAL H   4      25.291   9.008 -18.090  1.00  1.00           N
...
```

☹

Because we only have backbone atoms, all residues (except Gly, which fake.pdb does not have) have missing atoms.

```
# no residue has missing backbone atoms
print("Missing backbone:", p.rs_missing_atoms(ats="N,CA,C,O"), "\n")
# they all have missing side chain atoms, ats defaults to None -- all atoms
print("Missing side chain:", p.rs_missing_atoms(), "\n")
```

```
Missing backbone:

Missing side chain: H:L
```

Similar to ATS, RS can be initiatlized by a contig str or special keywords (None, "ALL" for all residues and "", "NONE", "NULL" for an empty selection). RS can also be initialized with a set/list/tuple/pandas.Series, a numpy array, another RS object or RL object (explained later). If a single integer is provided, it is treated as one residue index. Basically, RS supports all sensible input formats.

If a method expects an RS object as an argument, we should implement it in a way that users can provide the selection object using any of the data format that can be used to initialize an RS object. E.g., we can write `p.extract("H")`, `p.extract([0,1,2,3])`, `p.extract(np.array([2,4,6]))`, `p.extract((2,4,6))`, or `p.extract(p.rs("H"))`, etc. The contig is the most popular format we strongly recommend, as it is the most readable and error-proof. For developers, the first thing the method should do is to cast the input argument into an RS object using the following idiom:

```
rs=RS(self, rs)
```

Unlike ATS, RS requires a Protein object as its first argument in the initialization, as a residue selection will not be meaningful without being tied to a protein structure. For this reason, we often create an RS object from a Protein object using `p.rs()` instead of using `RS(protein_obj, selection_data)`

```
# residue selections
p=Protein(fn)
rs1=RS(p, "H1-3")
rs2=RS(p, "H3-5:L-3")
print("1st rs:", rs1)
print("2nd rs:", rs2, "\n")
# however, we prefer to create an RS object using Protein.rs() method
rs1=p.rs("H1-3")
rs2=p.rs("H3-5:L-3")
print("1st rs:", rs1)
print("2nd rs:", rs2, "\n")
# Protein also provides set operations on rs objects, however, try not the use these, as the Boolean operators (shown in the nex
print("and:", p.rs_and(rs1, rs2), "\n")
print("or:", p.rs_or(rs1, rs2), "\n")
# as we not H:L, the only residues left are P chain residues
print("not:", p.rs_not("H:L"), p.data.chain_index[p.rs_not("H:L").data], "\n")
# we can specify the residue universe using full_set, this can reduce the size of rs_not
# in the example below, we restrict the not selection within chain L
print("not with full set:", p.rs_not("L-100", rs_full="L"), "\n")
print("notin:", p.rs_notin(rs1, rs2), "\n")
print(p.rs2str(p.rs_or(rs1, rs2)))
```

```
1st rs: H1-3
2nd rs: L1-3:H3-5

1st rs: H1-3
2nd rs: L1-3:H3-5

and: H3

or: L1-3:H1-5

not: P [2 2 2 2 2 2 2 2 2 2 2 2]

not with full set: L101-111

notin: H1-2

L1-3:H1-5
```

In **Afpdb**, our convention is any method name starts with `rs_` is expected to return an RS object and any argument named `rs` or starts with `rs_` should take any residue selection formats whenever possible. The exception is `rs_seq()` returns a list of RS objects.

Residue selection behaves like a set, where residue indices within `rs.data` are uniqued and sorted. All sensible set operations apply, therefore, instead of using p.rs_or, p.rs_and, we prefer to use set operators directly as shown below:

```
p=Protein(fn)
# an unusal selection
rs1=p.rs("H1-3")
rs2=p.rs("H3-5:L-3")
print("and:", rs1 & rs2, "\n")
print("or:", rs1 | rs2, "same as", rs1+rs2, "\n")
print("not:", ~ p.rs("H:L"), "\n")
# ~ cannot take additional argument, so we need to use _not(), if rs_full needs to be specified
print("not with full set:", p.rs("L-100")._not(rs_full="L"), "\n")
print("notin:", rs1-rs2, "\n")
# inplace operations
rs1+=rs2
print(rs1, "\n")
rs1-=rs2
print(rs1, "\n")
```

```
and: H3

or: L1-3:H1-5 same as L1-3:H1-5

not: P

not with full set: L101-111

notin: H1-2

L1-3:H1-5

H1-2
```

```
# more
p=Protein(fn)
print(p.rs("").is_empty(), "\n")
print(p.rs("ALL").is_full(), "\n")
print(p.rs(None).is_empty(), p.rs(None).is_full(), "\n")
print(p.rs("H").is_full(), p.rs("H").is_empty(), "\n")

# to combine a list of selections
rs1=p.rs("H1-3")
rs2=p.rs("H3-5:L-3")
rs3=p.rs("H:P")
rs_list=[rs1, rs2, rs3]
print(RS._or(*rs_list), "\n")
# AND on the list
print(RS._and(*rs_list), "\n")

# only the first element of the list needs to be an RS object, the rest can be any format
# we need to extract the Protein object from the first RS object
print(RS._or(rs1, "H", "P"), "\n")

# residue lookup, convert a residue name to its internal AlphaFold residue order number
print(RS.i("GLY"), RS.i("Gly"), RS.i("G"), "\n")
```

```
True

True

False True

False False

L1-3:H:P

H3

H:P

7 7 7
```

Residule List

Residue indices within a RS object, `rs.data`, are unique and sorted. We sometimes need duplicate residue indices or require indices to be in specific order. Residule List (RL) class is the parent class of RS, which serves the purpose.

```
p=Protein(fn)
rl=p.rl("H3-5:L-3:H1-3")
print("Not sorted and not unique:", rl.data, "\n")
# compared to residue list
rs=p.rs("H3-5:L-3:H1-3")
print("Unique and ordered:", rs.data, "\n")
# cast between RS and RL
# notice we need to provide a protein object as the first arugment, when we use RS or RL to create the object
print(RS(rl.p, rl), "\n")

print("Sorted and unique:", p.rs(rl).data, "\n")
print(p.rl(p.rs("H3-5:L-3:H1-3")).data, "\n")
```

```
WARNING> contig should keep the segments for the same chain together H1-3, if
possible!
Not sorted and not unique: [113 114 115   0   1   2 111 112 113]

WARNING> contig should keep the segments for the same chain together H1-3, if
possible!
Unique and ordered: [  0   1   2 111 112 113 114 115]

L1-3:H1-5

Sorted and unique: [  0   1   2 111 112 113 114 115]

WARNING> contig should keep the segments for the same chain together H1-3, if
possible!
```

Residue list is handy for extracting key annotations for the residues, as demonstrated before:

```
p=Protein(fk)
rl=p.rl("H:L:H")
df=pd.DataFrame({"resi": rl.data})
df['chain']=rl.chain()
df['resn']=rl.name()
df['resni']=rl.namei()
df['aa']=rl.aa()
df['unique_name']=rl.unique_name()
df
```

```
Warning: residues with insertion code: L6A, L6B
WARNING> contig should keep the segments for the same chain together H, if
possible!
```

	resi	chain	resn	resni	aa	unique_name
0	4	H	3	3	L	H3
1	5	H	4	4	V	H4
2	0	L	5	5	E	L5
3	1	L	6A	6	I	L6A
4	2	L	6B	6	V	L6B
5	3	L	10	10	Q	L10
6	4	H	3	3	L	H3
7	5	H	4	4	V	H4

Next steps: Generate code with df View recommended plots

In the above example, we introduce a few terms:

- resi: the internal integer residue index used as the row index of backend numpy arrays
- chain: the chain name of the residue
- resn/name: the residue index, may include an insertion code, such as "6A" and "6B" in this example.
- resni: the integer part of the residue index without insertion code. This is integer type and can be used for sorting.
- aa: amino acid letter
- unique_name/contig: the unique residue name: {chain}{residue_index}

Later we will see residue list is also used in aligning two structures, as we would like to present the one-to-one mapping between residues.

Read/Write

One of our philosophy in designing Afpdb is to reduce the number of methods and arguments to bare minimum to reduce the burden of memorization. The convenience of Afpdb can be demonstrated by how we read/write protein structures. Regardless of what format a protein structure is represented, we use `Protein()` to create and use `save()` to export.

```
p=Protein(fn)
print(p.chain_id())
p.save("test.pdb")
out=util.unix("ls -l test.pdb")
```

```
['L', 'H', 'P']
-rw-r--r-- 1 root root 155520 Jun 11 04:09 test.pdb
```

The constructor can optionally take a contig string if you want to only read in a subset of the structure:

```
p=Protein(fn, contig="H:L")
print(p.chain_id(), "\n")
p=Protein(fn, contig="H-5:L-10")
print(p.seq(), "\n")

# however, for clarity, we recommend to separate the two operations, use contig in the extract() method explicitly.
p=Protein(fn)
print(p.extract("H:L").chain_id(), "\n")
p=Protein(fn)
print(p.extract("H-5:L-10").seq(), "\n")
```

```
['H' 'L']

VQLVQ:EIVLTQSPGT

['H' 'L']

VQLVQ:EIVLTQSPGT
```

The first argument in Protein() constructor can be quite flexible. Just be aware that only the first model is read, if the PDB file contains multiple models. AlphaFold was not designed to handle multiple conformations, afpdb can be made much more efficient by assuming only one conformation is being manipulated.

Below are a few examples:

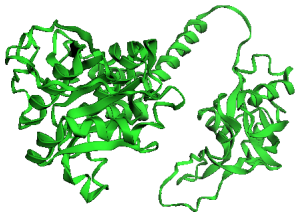
```
print("Load a local PDB file")
# support both .pdb and .ent extensions
print(fn)
p=Protein(fn)
print(p.chain_id(), "\n")

print("Create Protein from another afpdb.Protein object")
q=Protein(p)
print(q.chain_id(), "\n")

print("Create Protein from an afpdb.Protein.data object")
q=Protein(p.data)
print(q.chain_id(), "\n")

print("Create Protein from a BioPython Structure object")
b=p.to_biopython()
print(type(b))
q=Protein(b)
print(q.chain_id(), "\n")

print("Create Protein from a str containing the content of a PDB file")
s=p.to_pdb_str()
print("\n".join(s.split("\n")[:5])+"\n...")
q=Protein(s)
print(q.chain_id())
```

```
Load a local PDB file
/content/afpdb/tutorial/example_files/5cil.pdb
['L', 'H', 'P']

Create Protein from another afpdb.Protein object
['L' 'H' 'P']

Create Protein from an afpdb.Protein.data object
['L' 'H' 'P']

Create Protein from a BioPython Structure object
<class 'Bio.PDB.Structure.Structure'>
['L', 'H', 'P']

Create Protein from a str containing the content of a PDB file
HEADER    AFPDB PROTEIN                           11-Jun-24   XXXX
MODEL     1
ATOM      1  N   GLU L   1       5.195 -14.817 -19.187  1.00  1.00           N
ATOM      2  CA  GLU L   1       6.302 -14.276 -18.361  1.00  1.00           C
ATOM      3  C   GLU L   1       7.148 -15.388 -17.731  1.00  1.00           C
...
['L', 'H', 'P']
```

```
# more examples ...
print("Save as .cif, save Protein into a .cif file\n")
p=Protein(fn)
p.save("test.cif")
print("Create Protein from a local .cif file")
p=Protein("test.cif")
print(p.chain_id(), "\n")
print("Create with a PDB 4-letter code, fetch structure from PDB online.")
p=Protein("1crn")
print(p.seq(), "\n")
print("Create with a EMBL AlphaFold model code, fetch structure from EMBL online.")
p=Protein("Q2M403")
print(p.seq())
```

```
p.show()
```

```
Save as .cif, save Protein into a .cif file

    Create Protein from a local .cif file
    ['L' 'H' 'P']

    Create with a PDB 4-letter code, fetch structure from PDB online.
    TTCCPSIVARSNFNVCRLPGTPEAICATYTGCIIIPGATCPGDYAN

    Create with a EMBL AlphaFold model code, fetch structure from EMBL online.
    MVVVSLQCAIVGQAGSSFDVEIDDGAKVSKLKDAIKAKNATTITGDAKDLQLFLAKQPVEDESGKEVVPVYRPSAEEMKE
```



Sequence & Chain

Extraction

```
p=Protein(fn)
p.seq()
```

```
'EIVLTQSPGTQSLSPGERATLSCRASQSVGNNKLAWYQQRPGQAPRLLIYGASSRPSGVADRFSGSGSGTDFTLTISR
LEPEDFAVYYCQQYGQSLSTFGQGTKVEVKRTV:VQLVQSGAEVKRPGSSVTVSCKASGGSFSTYALSWVRQAPGRGLE
WMGGVIPLLTITNYAPRFQGRITITADRSTSTAYLELNSLRPEDTAVYYCAREGTTGDGDLGKPIGAFAHWGQGTLVTV
```

Notice the sequences from different chains are concatenated by colon (color in red) into one line:

EIVLTQSPGTQSLSPGERATLSCRASQSVGNNKLAWYQQRPGQAPRLLIYGASSRPSGVADRFSGSGSGTDFTLTISRLEPEDFAVYYCQQYGQSLS
TFGQGTKVEVKRTV:VQLVQSGAEVKRPGSSVTVSCKASGGSFSTYALSWVRQAPGRGLEWMGGVIPLLTITNYAPRFQGRITITADRSTSTAYLEL
NSLRPEDTAVYYCAREGTTGDGDLGKPIGAFAHWGQGTLVTVSS:NWFDITNWLWYIK

This is a standard format used to provide multimer sequence inputs to ColabFold and AlphaFold for protein structure prediction.

Here the order of the chain sequences is the same as how they occur in the PDB file. However, it is always safer to assume that chain order may change when we convert objects among different formats. Therefore, we strongly recommend users tie chain names with their sequences by using `seq_dict()` method instead:

```
p.seq_dict()
```

```
{'L':
'EIVLTQSPGTQSLSPGERATLSCRASQSVGNNKLAWYQQRPGQAPRLLIYGASSRPSGVADRFSGSGSGTDFTLTISRL
 'H':
```

Missing Residues

When there are missing residues in the structure, they will be represented as "X" in the sequence string:

```
p=Protein(fn)
q=p.extract("H1-10:H21-30")
print(len(q.data.residue_index), q.data.residue_index, "\n")
print("Number of residues in numpy arrays:", q.data.atom_positions.shape, "\n")
print(q.seq(), "\n")
print(q.seq_dict(gap="."), "\n")
print(q.seq().replace("X", "G"), "\n")
print(q.seq(gap="G"), "\n")
```

```
20 ['1' '2' '3' '4' '5' '6' '7' '8' '9' '10' '21' '22' '23' '24' '25' '26'
 '27' '28' '29' '30']

Number of residues in numpy arrays: (20, 37, 3)

VQLVQSGAEVXXXXXXXXXXCKASGGSFST

{'H': 'VQLVQSGAEV.........CKASGGSFST'}

VQLVQSGAEVGGGGGGGGGCKASGGSFST

VQLVQSGAEVGGGGGGGGGCKASGGSFST
```

The number of missing residues is determined based on residue numbering. In the above example, we first create a new object by taking the first ten residues from chain H, skipping 10 residues, then take another 10 residues. In the resultant object q, the 10-residue gap is preserved (residue index jumps from 10 to 21). Although the 10 missing residues have no corresponding elements in the internal numpy array representation or in the saved PDB file (the array only contains 20 residue in total, the discontinuity in the residue index allows us to figure out that 10 residues are missing). Therefore, `seq()` and `seq_dict()` outputs ten "X". You can change the display of "X" by the gap argument or suppress gap by "". For protein structure prediction, we often replace the missing residues by Glycine "G". If somehow you want to ignore missing residues, set `gap=''`.

```
p=Protein(fk)
print(p.data.residue_index, "\n")
print(p.seq_dict(), "\n")
print(p.seq_dict(gap=""))
```

```
Warning: residues with insertion code: L6A, L6B
['5' '6A' '6B' '10' '3' '4']

{'L': 'EIVXXXQ', 'H': 'LV'}

{'L': 'EIVQ', 'H': 'LV'}
```

When we determine missing residues, only the integer part of the residue ID is considered. In the above example, residue index 6 appears twice as 6A and 6B, followed by residue 10. We assume residues 7, 8, and 9 are missing.

Note: Missing residues at the N or C terminals cannot be recovered. In addition, if the residue numbering in the full structure are not mumerically continous, e.g., both residues "6A" and "6B" were removed, we can only infer that one residue "6" is missing:

```
p=Protein(fk)
q=p.extract("L5,10:H")
print(q.data.residue_index, "\n")
print(q.seq(), "\n")
```

```
Warning: residues with insertion code: L6A, L6B
['5' '10' '3' '4']

EXXXXQ:LV
```

Length

The get the total number of residues (not counting missing residues), i.e, the number of rows in the backend numpy arrays, use:

```
p=Protein(fn)
print("Total residue counts:", len(p), "\n")
print("Residue counts per chains:", p.len_dict())
q=Protein(fk)
# missing residues are not counted as part of the length
print(q.seq(), "\n")
print(q.len_dict(), "\n")
```

```
Total residue counts: 250

    Residue counts per chains: {'L': 111, 'H': 126, 'P': 13}
    Warning: residues with insertion code: L6A, L6B
    EIVXXXQ:LV

    {'L': 4, 'H': 2}
```

Search

When we have a sequence fragment and would like to locate where it came from, use:

```
p=Protein(fn)
out=p.rs_seq('LTI')
for i,m in enumerate(out):
    print(f"Matched #{i+1}, location: {m}, sequence: {m.seq()}")
```

```
Matched #1, location: L74-76, sequence: LTI
Matched #2, location: H54-56, sequence: LTI
```

The name `rs_seq()` suggests the method performs sequence search and return residue selection (RS) objects. Since there can be multiple matches, it returns a list of RS objects. We will explain residue selection objects later, for now, we just need to know that a residue selection object can be printed as a contig string and its `seq()` method returns the corresponding sequence.

The two matches are in bold below:
EIVLTQSPGTQSLSPGERATLSCRASQSVGNNKLAWYQQRPGQAPRLLIYGASSRPSGVADRFSGSGSGTDFT***LTI***SRLEPEDFAVYYCQQYGQSLSTFGQGTK
VEVKRTV:
VQLVQSGAEVKRPGSSVTVSCKASGGSFSTYALSWVRQAPGRGLEWMGGVIPL***LTI***TNYAPRFQGRITITADRSTSTAYLELNSLRPEDTAVYYCAREGTTGDG
DLGKPIGAFAHWGQGTLVTVSS: NWFDITNWLWYIK

If we are only interested in sequence search within a list of chains, provide the chain name(s) list:

```
out=p.rs_seq('LTI', in_chains="H")
print(len(out), out[0], "\n")
```

```
1 H54-56
```

The sequence search pattern can be a regular expression, e.g., to match all charged residues:

```
out=p.rs_seq(r'[RHKDE]')
# concatenate the list of matched residue selection objects into one RS object, RS._or() will be explained later.
m=RS._or(*out)
print(m.seq())
```

```
EERRKRRRDRDREEDKEKREKRKRRERRDREREDREDDKHDK
```

Mutagenesis

To perform mutagenesis on a structure, we thread a new sequence onto an existing Protein object. As this method requires the installation of PyMOL, it throws an exception if PyMOL is not found.

When we specify the new sequence, we only specify the new residues for which there is coordinate information, i.e., missing residues should not occur in the new sequence. Chain L's sequence was EIVXXXQ, the target sequence is AIVD instead of AIVXXXD.

The following example requires PyMOL to be installed. The installation can take some time in Google Colab.

```
install_pymol()
p=Protein(fk)
print(p.seq(), "\n")
p.thread_sequence({"L":"AIVD", "H":"LG"}, "m.pdb")
```

```
Warning: residues with insertion code: L6A, L6B
EIVXXXQ:LV

Warning: residues with insertion code: L6A, L6B
MUTATE PyMOL> Old  L 5 GLU >>> New ALA
Selected!
 Mutagenesis: no rotamers found in library.
MUTATE PyMOL> Old  L 10 GLN >>> New ASP
Selected!
 Mutagenesis: no phi/psi, using backbone-independent rotamers.
 Mutagenesis: 9 rotamers loaded.
 Rotamer 9/9, strain=2.82
MUTATE PyMOL> Old  H 4 VAL >>> New GLY
Selected!
 Mutagenesis: no rotamers found in library.
m.pdb
Warning: residues with insertion code: L6A, L6B
{'L': 'AIVD', 'H': 'LG'}
Warning: residues with insertion code: L6A, L6B
###JSON STARTS
{"output_pdb": "m.pdb", "ok": true, "output_equal_target": false, "input":
{"L": "EIVQ", "H": "LV"}, "output": {"H": "LG", "L": "AIVXXXD"}, "target":
{"L": "AIVD", "H": "LG"}, "relax": false, "residues_with_missing_atom": "",
"mutations": [["L", 5, "GLU", "ALA"], ["L", 10, "GLN", "ASP"], ["H", 4, "VAL",
"GLY"]]}
###JSON END
{'output_pdb': 'm.pdb',
 'ok': True,
 'output_equal_target': False,
 'input': {'L': 'EIVQ', 'H': 'LV'},
 'output': {'H': 'LG', 'L': 'AIVXXXD'},
 'target': {'L': 'AIVD', 'H': 'LG'},
 'relax': False,
 'residues_with_missing_atom': '',
 'mutations': [['L', 5, 'GLU', 'ALA'
```

From the output JSON data, we see L5 was "E" and it is replaced with "A". L6A-L6B was not changed, therefore, their side chain coordinates remain the same. L10 is replaced by "D". On the chain H, H6 was "G" and is replaced by "V".

The new mutated residues have their side chain torsion angles generated by PyMOL. Therefore, it generally makes sense to relax the side chain, for example, with Amber. Relax is not supported by Afpdb due to its dependency on Amber. TODO: we might look into this more.

Mutation

If a chain name is in two proteins and the chain has the same number of residues, we can identify mutated residues by a residue-by-residue comparison. The example below identify three mutated residues:

```
p=Protein(fk)
# m.pdb was produced with the previous thread_seq() example
q=Protein("m.pdb")
print(p.seq(), q.seq(), "\n")
print(p.rs_mutate(q))
```

```
Warning: residues with insertion code: L6A, L6B
Warning: residues with insertion code: L6A, L6B
EIVXXXQ:LV LG:AIVXXXD

L5,10:H4
```

Chain Name and Order

In the fn PDB file, chains go from L, to H, to P. chain_id() will return chain names in the same order. `p.data.chain_index` stores the chain index. **Afpdb** expects all residues belong to the same chain are stored together without being interrupted by another chain.

```
p=Protein(fn)
print(p.chain_id(), "\n")
print(p.data.chain_index, "\n")
```

```
['L', 'H', 'P']

[0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1
 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1
 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1
 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 2 2 2 2 2 2 2 2 2 2 2 2 2]
```

Although we should not rely on the exact order of chains in the PDB file, we can neverthess reorder chains, if we wish:

```
p=Protein(fn)
q=p.data
print("Original chain order:", p.chain_id(), "\n")
print(p.seq(), "\n")
# notice the last 20 residues belong to chain L, then chain P in the q.chain_index NumPy array
print(q.chain_index[-20:], "\n")
print(q.residue_index[-20:], "\n")

p.reorder_chains(["P","L","H"])
q=p.data
print("After chain reorder:", p.chain_id(), "\n")
print(p.seq(), "\n")
# after ordering, # notice the last 20 residues belong to chain H in the q.chain_index NumPy array
print(q.chain_index[-20:], "\n")
print(q.residue_index[-20:], "\n")

# The method reorder_chains() is no longer needed, as we can use extract():
p=Protein(fn)
p.extract("P:L:H", inplace=True)
q=p.data
print("After chain reorder:", p.chain_id(), "\n")
print(p.seq(), "\n")
print(q.chain_index[-20:], "\n")
print(q.residue_index[-20:], "\n")
```

```
Original chain order: ['L', 'H', 'P']

EIVLTQSPGTQSLSPGERATLSCRASQSVGNNKLAWYQQRPGQAPRLLIYGASSRPSGVADRFSGSGSGTDFTLTISRLE

[1 1 1 1 1 1 1 2 2 2 2 2 2 2 2 2 2 2 2 2]

['120' '121' '122' '123' '124' '125' '126' '1' '2' '3' '4' '5' '6' '7' '8'
 '9' '10' '11' '12' '13']

After chain reorder: ['P', 'L', 'H']

NWFDITNWLWYIK:EIVLTQSPGTQSLSPGERATLSCRASQSVGNNKLAWYQQRPGQAPRLLIYGASSRPSGVADRFSGS

[2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2]

['107' '108' '109' '110' '111' '112' '113' '114' '115' '116' '117' '118'
 '119' '120' '121' '122' '123' '124' '125' '126']

After chain reorder: ['P' 'L' 'H']

NWFDITNWLWYIK:EIVLTQSPGTQSLSPGERATLSCRASQSVGNNKLAWYQQRPGQAPRLLIYGASSRPSGVADRFSGS

[2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2]

['107' '108' '109' '110' '111' '112' '113' '114' '115' '116' '117' '118'
 '119' '120' '121' '122' '123' '124' '125' '126']
```

Notice after reordering, the `chain_index` array goes from 0 to 2, and the chain_id list has also been updated.

The extract() method will error if the selection list contains duplicates or residues of the same chain are not ordered.

```
p=Protein(fn)
try:
    p.extract("H:H:L")
except Exception as e:
    print(e)

try:
    p.extract("H5-:H1")
except Exception as e:
    print(e)

try:
    p.extract("H1:L:H2")
except Exception as e:
    print(e)

# as_rl=False, all examples are fine, b/c the contig selection will be cast into an unordered/non-redundant residue selection
print("Treat as residue selection ...\n")
q=p.extract("H:H:L", as_rl=False)
q=p.extract("H5-:H1", as_rl=False)
q=p.extract("H1:L:H2", as_rl=False)
```

```
There are duplicate residues: [111 112 113 114 115 116 117 118 119 120 121 122
123 124 125 126 127 128
 129 130 131 132 133 134 135 136 137 138 139 140 141 142 143 144 145 146
 147 148 149 150 151 152 153 154 155 156 157 158 159 160 161 162 163 164
 165 166 167 168 169 170 171 172 173 174 175 176 177 178 179 180 181 182
 183 184 185 186 187 188 189 190 191 192 193 194 195 196 197 198 199 200
 201 202 203 204 205 206 207 208 209 210 211 212 213 214 215 216 217 218
 219 220 221 222 223 224 225 226 227 228 229 230 231 232 233 234 235 236]
Residues within chain H are not sorted in order.
WARNING> contig should keep the segments for the same chain together H2, if
possible!
Residues for chain ['H'] are not grouped together.
Treat as residue selection ...
```

Residue Labeling

The method `chain_pos()` is an important utility method. It returns the start and end NumPy indices of each chain, which can be used to access the corresponding numpy arrays. For our example structure, the complex consists of 3 chains. The array residue_index has 250 elements. The residues in chain L are stored as the first 111 elements in the numpy arrays, in Python, the indices are from 0 to 110.

Many methods relies on `chain_pos` to accelerate the computation, this is the reason why **Afpdb** assumes all residues of the same chain should be stored together within the backend numpy arrays.

```
p=Protein(fn)
print(p.chain_pos(), "\n")
q=p.data
print(q.residue_index[108:114], "\n")
print(q.chain_index[108:114])

# we should avoid using p.data, all can be done by residue selection
rs=p.rs(list(range(108, 115)))
print(rs.name(), "\n")
print(rs.chain())
```

```
{'L': [0, 110], 'H': [111, 236], 'P': [237, 249]}

['109' '110' '111' '1' '2' '3']

[0 0 0 1 1 1]
['109', '110', '111', '1', '2', '3', '4']

['L', 'L', 'L', 'H', 'H', 'H', 'H']
```

The chain_index for element 110 is the last residue of chain L, its residue name is '111' and chain id is 0, i.e., resn="111", resi=110, resn_i=111. Element 111 is the first residue of chain H. Its residue name is '1' and chain id is 1, i.e., resn="1", resi=111, resn_i=1.

If we only want to extract the coordinates for the 13 residues of chain P, we will use:

```
q.atom_positions[237:250].shape
```

```
(13, 37, 3)
```

Residue Renumbering

Residue index can be modified. There are a number of renumbering modes:

```
None: keep original numbering unmodified.
RESTART: 1, 2, 3, ... for each chain
CONTINUE: 1 ... 100, 101 ... 140, 141 ... 245
GAP200: 1 ... 100, 301 ... 340, 541 ... 645 # mimic AlphaFold gaps
You can define your own gap by replacing GAP200 with GAP{number}, e.g., GAP10
NOCODE: remove insertion code, residue 6A and 6B becomes 6, 7
```

```
p=Protein(fk)
q=p.data
print(q.chain_index, "\n")

p.renumber(None)
print(q.residue_index, "\n")

p.renumber('RESTART')
print(q.residue_index, "\n")

p.renumber('CONTINUE')
print(q.residue_index, "\n")

p.renumber('GAP200')
print(q.residue_index, "\n")

p.renumber('GAP10')
print(q.residue_index, "\n")

p.renumber('NOCODE')
print(q.residue_index)
```

```
Warning: residues with insertion code: L6A, L6B
[0 0 0 0 1 1]

['5' '6A' '6B' '10' '3' '4']

['1' '2A' '2B' '6' '1' '2']

['1' '2A' '2B' '6' '7' '8']

['1' '2A' '2B' '6' '207' '208']

['1' '2A' '2B' '6' '17' '18']

['1' '2' '3' '7' '8' '9']
```

NOCODE is used to remove insertion code, in case we need to feed a structure into a tool that cannot deal with insertion code. Notice "2A" and "2B" are renamed to "2" and "3" to avoid the collision, then residue "6" is renamed to "7" to preserve the three missing residues in the original naming.

By default, `renumber()` returns a tuple `(p_object, old_number)`. `inplace=True` is the default, i.e., the returned object is the same input (but modified) object.

If we need a custom naming, e.g., we really want to remove the gaps for missing residues, we can provide our own custom list of residue names, using `resn()`. Without any argument, `resn()` is a read method that returns the current residue names. If providing an argument as a list or NumPy array, its length should match the number of residues.

```
p=Protein(fk)
q, old_resn=p.renumber("RESTART")
print("Old Name:", old_resn, "New Name:", q.resn(), "\n")
# restore the old residue names
q.resn(old_resn)
print(q.resn(), "\n")
# set custom residue names, we create new residue names as a continous integer without gap
q.resn([str(x) for x in range(1, len(q)+1)])
print(q.resn())
```

```
Warning: residues with insertion code: L6A, L6B
Old Name: ['5' '6A' '6B' '10' '3' '4'] New Name: ['1' '2A' '2B' '6' '1' '2']

['5' '6A' '6B' '10' '3' '4']

['1' '2' '3' '4' '5' '6']
```

Merge & Split Chains

Before AlphaFold Multimer was created, we had to use AlphaFold monomer model to predict multimer structures. The hack is to merge chains into one single chain, where we introduce a 200 gap in the residue indices between the original chain junctions to keep AlphaFold from treating the previous C- and the next N-terminals of two contiguous chains from being mistakenly treated as neighboring residues. This could be achieved by:

```
p=Protein(fn)
q, c_pos=p.merge_chains(gap=200, inplace=False)
print(q.seq_dict(gap="."), "\n")
print(c_pos, "\n")

r=q.split_chains(c_pos, inplace=False)
print(r.seq_dict())
```

```
{'L':
'EIVLTQSPGTQSLSPGERATLSCRASQSVGNNKLAWYQQRPGQAPRLLIYGASSRPSGVADRFSGSGSGTDFTLTISRL

{'L': [0, 110], 'H': [111, 236], 'P': [237, 249]}

{'L':
'EIVLTQSPGTQSLSPGERATLSCRASQSVGNNKLAWYQQRPGQAPRLLIYGASSRPSGVADRFSGSGSGTDFTLTISRL
'H':
```

As chains got merged into a single change, 200-residue gaps were introduced and shown as "." in the sequence output. We notice there is only one chain, where the first chain name in the original PDB file was used. The method returns the original `c_pos` object, which memorizes the chain structure before the merge.

After AlphaFold predicts the structure as a monomer, the output PDB file can then be used by split_chains to restore the original trimer structure. Here `c_pos` guides the method to split the monomer chain and restores the naming of the original chains.

If we send one sequence to AF without residue indices, we would actually need to replace the gap with glycines. The predicted structure will contain extra glycine linkers, those need to be removed before we can `split_chains()`. The extra glycines can be identified with `q.rsi_missing()`, as demonstrated later in the first AI use case. The idea is outlined below:

```
p=Protein(fn)
# for the sake of saving paper space, we use a gap of 20 residues
q, c_pos=p.merge_chains(gap=20, inplace=False)
seq=q.seq(gap="G")
# obtain the positions for the glycine linkers
linker_pos=q.rsi_missing()
print("Chain breaks:", c_pos, "\n")
print("Position of G:", linker_pos, "\n")
print("Send the following fasta sequence to AlphaFold monomer model for prediction:\n")
print(">af_pred\n"+seq, "\n")
print("AF prediction, generate pred.pdb ...\n")
print("Read the predicted structure:")
print("    pred=Protein('pred.pdb')\n")
print("Remove glycine linkers:")
print("    pred.extract(~ RS(pred, linker_pos), inplace=True)\n")
print("Restore three chains:")
print("    pred.split_chains(c_pos)\n")
```

```
Chain breaks: {'L': [0, 110], 'H': [111, 236], 'P': [237, 249]}

Position of G: [111 112 113 114 115 116 117 118 119 120 121 122 123 124 125 126
127 128
 129 130 257 258 259 260 261 262 263 264 265 266 267 268 269 270 271 272
 273 274 275 276]

Send the following fasta sequence to AlphaFold monomer model for prediction:

>af_pred
EIVLTQSPGTQSLSPGERATLSCRASQSVGNNKLAWYQQRPGQAPRLLIYGASSRPSGVADRFSGSGSGTDFTLTISRLE

AF prediction, generate pred.pdb ...

Read the predicted structure:
    pred=Protein('pred.pdb')

Remove glycine linkers:
    pred.extract(~ RS(pred, linker_pos), inplace=True)

Restore three chains:
    pred.split_chains(c_pos)
```

Geometry, Measurement, and Visualization

Select Neighboring Residues

For a given residue selection, `rs_around` selects neighboring residues, where their closest distance is within the specified `dist` argument. In the example below, we find all residues that are within 3.5Å around P chain residues. AlphaFold's NumPy arrays do not contain hydrogens. We compute distances among all possible atom pairs and use the minimum atom-atom distance as the final distance between two residues. In this example, it identifies 5 residues on chain L and 8 residues on chain H. The method does not consider intra-selection distances, i.e., it will not include residues that are already part of the seed selection. If you want to include seed residues, just use use "or" operator to combine two residue selections.

There are two returned values, the first is a new residue selection of the neighboring residues and the second is a dataframe object. The dataframe object provides the details about all residues pairs, the source is marked as "a" (chain P here) and the target is marked as "b" (chain H or L). resn_ stands for residue name, resn_i_ is for the integer part of the residue names without the insertion code and it is an integer type. resi_ is for the internal numpy array index (i.e., residue selection `rs.data`). The shortest distance between two residues is listed under the *dist* column and the atom pair that contributes to this shortest distance is listed as *atom_a* and *atom_b*. The dataframe is sorted by descending *dist* values.

```
p=Protein(fn)
rs_nbr, rs_seed, t=p.rs_around("P", dist=3.5)
print("Neighbors:", rs_nbr.name(), "\n")
print("Seeds:", rs_seed.name(), "\n")
t
```

```
Neighbors: ['33', '92', '93', '94', '95', '30', '56', '58', '98', '106', '107',
'108', '109']

Seeds: ['1', '2', '3', '4', '6', '7', '10']
```

	chain_a	resi_a	resn_a	resn_i_a	atom_a	chain_b	resi_b	resn_b	resn_i_b	a
23	P	242	6	6	OG1	H	208	98	98	
17	P	240	4	4	OD1	L	32	33	33	
7	P	238	2	2	N	L	94	95	95	
2	P	237	1	1	ND2	L	91	92	92	
27	P	243	7	7	ND2	H	218	108	108	
4	P	237	1	1	OD1	L	93	94	94	
14	P	238	2	2	NE1	H	166	56	56	
1	P	237	1	1	ND2	L	92	93	93	
31	P	246	10	10	NE1	H	216	106	106	
13	P	238	2	2	CE3	H	168	58	58	
5	P	237	1	1	OD1	L	94	95	95	
22	P	242	6	6	CG2	H	140	30	30	
16	P	239	3	3	CE2	L	94	95	95	
30	P	246	10	10	NE1	H	217	107	107	
15	P	239	3	3	CD2	L	93	94	94	
26	P	243	7	7	N	H	219	109	109	

Next steps: Generate code with t View recommended plots

To include the seed residues in the selection, use the following idiom:

```
# p.rs(t.resi_a) select all seed residues appears in the neighbor table
# resi_a contains the residue IDs for those on the P chain that interact with rs_nbr
rs_both = rs_nbr | rs_seed
print(rs_both)
```

```
L33,92-95:H30,56,58,98,106-109:P1-4,6-7,10
```

The distance table contains all residues pairs between seeds and their neighbors. If we only want to see one seed per neighbor, we define drop_duplicates = True:

```
rs_nbr, rs_seed, t2=p.rs_around("P", dist=3.5, drop_duplicates=True)
print(rs_seed, "\n")
print(rs_nbr)
t2
```

```
P1-4,6-7,10

L33,92-95:H30,56,58,98,106-109
```

	chain_a	resi_a	resn_a	resn_i_a	atom_a	chain_b	resi_b	resn_b	resn_i_b	a
23	P	242	6	6	OG1	H	208	98	98	
17	P	240	4	4	OD1	L	32	33	33	
7	P	238	2	2	N	L	94	95	95	
2	P	237	1	1	ND2	L	91	92	92	
27	P	243	7	7	ND2	H	218	108	108	
4	P	237	1	1	OD1	L	93	94	94	
14	P	238	2	2	NE1	H	166	56	56	
1	P	237	1	1	ND2	L	92	93	93	
31	P	246	10	10	NE1	H	216	106	106	
13	P	238	2	2	CE3	H	168	58	58	
22	P	242	6	6	CG2	H	140	30	30	
30	P	246	10	10	NE1	H	217	107	107	
26	P	243	7	7	N	H	219	109	109	

Next steps: Generate code with t2 View recommended plots

The reason we provide resn_i is to enable us to filter the output dataframe. The data type for resn is a string, as resn could contain insertion codes. If we are interested in rows corresponding to residues within H95-106, we cannot use the *resn* column for filtering. Instead, we can accomplish this with the *resn_i* column:

```
print("resn cannot be used for filtering, as it's string type, str greater than '95' will be all less than '116'")
t2=t[(t.chain_b=="H")&(t.resn_b>="95")&(t.resn_b<="106")]
t2
```

```
resn cannot be used for filtering, as it's string type, str greater than '95'
will be all less than '116'
```

	chain_a	resi_a	resn_a	resn_i_a	atom_a	chain_b	resi_b	resn_b	resn_i_b	atc

```
print("resn_i is an integer column")
t2=t[(t.chain_b=="H")&(t.resn_i_b>=95)&(t.resn_i_b<=106)]
t2
```

```
resn_i is an integer column
```

	chain_a	resi_a	resn_a	resn_i_a	atom_a	chain_b	resi_b	resn_b	resn_i_b	a
23	P	242	6	6	OG1	H	208	98	98	
31	P	246	10	10	NE1	H	216	106	106	

Next steps: Generate code with t2 View recommended plots

By default, all residue atoms are considered. However, we can restrict the distance measurement using an atom selection. We can also restrict the target residues using a residue selection `rs_within`.

```
p=Protein(fn)
rs=p.rs("P")
# select residues on chain H that have CA-CA distance within 8A.
rs_nbr, rs_seed, t=p.rs_around(rs, dist=8, rs_within="H", ats="CA")
print(rs_nbr)
t
```

```
H30-32,51,58,108-111
```

	chain_a	resi_a	resn_a	resn_i_a	atom_a	chain_b	resi_b	resn_b	resn_i_b	a
2	P	239	3	3	CA	H	221	111	111	
10	P	243	7	7	CA	H	219	109	109	
4	P	242	6	6	CA	H	141	31	31	
7	P	242	6	6	CA	H	140	30	30	
13	P	246	10	10	CA	H	218	108	108	
9	P	243	7	7	CA	H	218	108	108	
6	P	242	6	6	CA	H	219	109	109	
5	P	242	6	6	CA	H	142	32	32	
12	P	245	9	9	CA	H	140	30	30	
0	P	238	2	2	CA	H	168	58	58	
3	P	240	4	4	CA	H	221	111	111	
14	P	246	10	10	CA	H	219	109	109	
15	P	246	10	10	CA	H	140	30	30	
8	P	242	6	6	CA	H	161	51	51	
1	P	239	3	3	CA	H	142	32	32	
11	P	243	7	7	CA	H	220	110	110	
16	P	247	11	11	CA	H	218	108	108	

Next steps: Generate code with `t` View recommended plots

Display

To visualize a structure, please `save` it to a PDB file and use PyMOL. If you prefer to display the structure in an HTML file, use `util.save_string("my_protein.html", p.html())` and open it in a browser.

Within Jupyter Notebook, p.show() will display the structure as we do throughout this notebook.

The arguments for show() and html() are the same. `show_sidechains=False, show_mainchains=False, color="chain", style="cartoon", width=320, height=320)`

color can be: "chain", "lDDT", "b", "spectrum", "ss". style can be: "cartoon", "stick", "line", "sphere", "cross"

If colored by lDDT, the b-factors should be in the range of [0, 100]. If colored by "b", b-factors should be normalized to [0, 1].

```
p=Protein(fn)
util.save_string("my_protein.html", p.html())
print("You can also open my_protein.html in a browser to view the protein structure.")
print("We display the protein within Jupyter notebook below")
p.show(style="cartoon", color="ss")
```

```
You can also open my_protein.html in a browser to view the protein structure.
We display the protein within Jupyter notebook below
```

B-factors

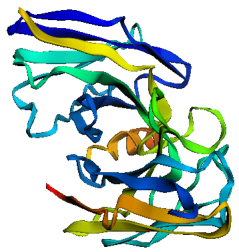
B-factor is an embedded attribute that can be repurposed to store other computation results. E.g., AlphaFold uses b-factors to store pLDDT for the structure prediction confidence. we can use b-factors to flag the CDR loop residues. PyMOL can then color residues by b-factors, which enables us to visualize the computational results in their structural context. In Afpdb, we currently read/write b-factors at the residue level, i.e., all atoms in a residue share the same b-factor value, as we have not yet encounter the need for displaying b-factors at the atom level.

```
p=Protein(fk)
# read b_factors as a numpy array
print(p.b_factors())
# set b_factors by providing a numpy array
import numpy as np
p.b_factors(np.random.random((len(p))), "\n")
```

```
Warning: residues with insertion code: L6A, L6B
[1. 1. 1. 1. 1. 1.]
array([0.12971284, 0.29491582, 0.19422936, 0.41865315, 0.90129086,
       0.85097538])
```

To color by b-factors, set `color = "b"` in the show() method. However, please normalize b-factor ndarray to be within [0, 1] to get the expected coloring effect (although show() method will clip the values, it does not normalize them).

```
p=Protein(fn)
n=len(p)
p.b_factors(np.sin(np.arange(n)/n*np.pi))
p.show(style="cartoon", color="b")
```



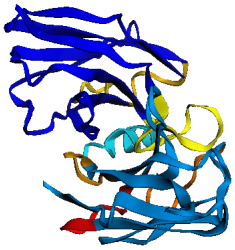
It may be more convenient to get or set b-factors for each chain using `b_factors_by_chain()` method.

```
# in this example, we color residues based on how far they are from the P chain
p=Protein(fn)
rs, rs_seed, dist=p.rs_around("P", dist=1e8)
# important to sort the residues by their internal indices, so that the order of distance matches the internal b-factor array
# we use drop_duplicates to make sure each residue only occur once (to its nearest seed residue)
dist.drop_duplicates("resi_b", inplace=True)
dist.sort_values("resi_b", inplace=True)
assert len(dist)==len(p.rs("H:L"))
d_min,d_max=dist.dist.min(), dist.dist.max()
# b factors should be in [0, 1], if we want to color by b-factos.
dist["dist"]=(dist["dist"]-d_min)/(d_max-d_min)
b={"P":0, "H":dist[dist.chain_b=="H"].dist.values, "L":dist[dist.chain_b=="L"].dist.values}
# set b_factors
p.b_factors_by_chain(b)
p.show(color="b")
```

We can also provide a residue selection for `b_factors()`.

```
# Use b-factor to mimic color by chain
p=Protein(fn)
# provide b-factors as an array
p.b_factors(np.zeros(len(p.rs("H"))), rs="H")
# a number is considered a special case
p.b_factors(0.5, rs="L")
p.b_factors(1, rs="P")
p.show(color="b")
```

```
# visualize CDRs
p=Protein(fn)
# CDR sequence (Chothia definition) light chains followed by heavy chain
CDR_L=['RASQSVGNNKLA', 'GASSRPS', 'QQYGQSLST']
CDR_H=['GGSFSTY', 'IPLLTI', 'EGTTGDGDLGKPIGAFAH']
# color the framework resdues
p.b_factors(1, "H")
p.b_factors(0.9, "L")
p.b_factors(0.8, "P")
for i,(seq,chain) in enumerate(zip(CDR_L+CDR_H, ["L"]*3+["H"]*3)):
    b=i/20
    # we select the CDR fragment by sequence search
    # as rs_seq returns a list of RS, we take the first element
    rs_cdr=p.rs_seq(seq, in_chains=[chain])[0]
    p.b_factors(b, rs=rs_cdr)
p.show(color="b")
```



We can leverage PyMOL to create more powerful visualizations, which will be described next.

PyMOL Interfac

PyMOL is a power structure visualization tool. **Afpdb** provides efficient and convenient programming interface for structure computation. A easy-to-use interface between Afpdb and PyMOL can help combine the strenght of both tools covering both structure analysis and visualization. The `PyMol()` method fills this role.

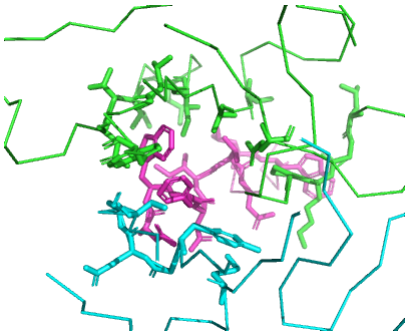
The method returns a PyMOL object, which has a `run` method enabling us to run any PyMOL commands.

```
# create a new PyMOL engine
pm=Protein().PyMOL()
pm.cmd(f"load {fn}, myobj")
p=Protein(fn)
rs_binder, rs_seed, t = p.rs_around("P", rs_within="H:L", dist=4)
# selecting all interface residues
rs_int = rs_binder | rs_seed
# convert Afpdb residue selection to a PyMOL selection command, where the selection object is named "myint"
rs_str = rs_int.str(format="PYMOL", rs_name="myint")
print(rs_str, "\n")

# We can also control the selection to CA only
print(rs_int.str(format="PYMOL", rs_name="myint_ca", ats="CA"), "\n")

pm.run(f"""
# color by chain
as ribbon, myobj
util.cbc
# defines a selection named myint
{rs_str}
show sticks, myint
# focus on the selection
zoom myint
""")
pm.run("""deselect; save mypm.png; save mypm.pse""")
# dispose the PyMOL object to save system resource
pm.close()
```

```
select myint, (chain L and resi 33+92-95+97) or (chain H and resi 30+32+46+49-
51+54+56-58+98+106-109) or (chain P and resi 1-7+9-10)

select myint_ca, ((chain L and resi 33+92-95+97) or (chain H and resi
30+32+46+49-51+54+56-58+98+106-109) or (chain P and resi 1-7+9-10)) and name CA

 util.cbc: color 26, (chain H)
 util.cbc: color 5, (chain L)
 util.cbc: color 154, (chain P)
 Save: Please wait -- writing session file
```

The generated mypm.pse is a PyMOL session file to visualize the binding interface in 3D.

We here also automatically save the image into a PNG file as below.



As shown in this example, we can send any PyMOL commands via the `run` method. The input can be a single line or multiple lines. We can use `#` to add comments. The computation, i.e., the identification of the interface residues, can be done in **Afpdb** using `rs_around`. A residue selection object's `str` method casts the selection into a PyMOL selection command:

`rs_str = rs_int.str(format="PYMOL", rs_name="myint")` turns the interface residues into the command that defines a selection object named "myint":

```
select myint, (chain L and resi 33+92-95+97) or (chain H and resi 30+32+46+49-51+54+56-58+98+106-109) or (chain P and resi 1-7+9-10)
```

If we only want to select CA atoms, we can do:

```
rs_str = rs_int.str(format="PYMOL", rs_name="myint", ats="CA")
```

This will produce:

```
select myint, ((chain L and resi 33+92-95+97) or (chain H and resi 30+32+46+49-51+54+56-58+98+106-109) or (chain P and resi 1-7+9-10)) and name CA
```

PyMOL command `show sticks, myint` and `zoom myint` shows the interface residues as sticks and zoom in to focus on them. We can also provide multiple PyMOL commands into one line with "; " as the concatenator. The example line is:

```
deselect; save mypm.png; save mypm.pse
```

Users can use all the PyMOL command they are already familiar with, but use **Afpdb** for the computational tasks that are not straightforward to

Distance

In a previous example, `rs_around` method already uses the distance measurement method `rs_dist` internally.

Given two residue groups, `rs_a` and `rs_b`, `rs_dist` computes the distance between all residue pairs. The distance is determined based on the two closest atoms. The output dataframe is sorted by distance and indicates which atom pair contributes to the distance measure.

```
p=Protein(fk)
print(p.seq_dict())
print(p.rs().name(), "\n")

t=p.rs_dist('L', 'H')
t
```

```
Warning: residues with insertion code: L6A, L6B
{'L': 'EIVXXXQ', 'H': 'LV'}
['5', '6A', '6B', '10', '3', '4']
```

	chain_a	resn_a	resn_i_a	resi_a	res_a	chain_b	resn_b	resn_i_b	resi_b	r
486	L	10	10	3	Q	H	3	3	4	
111	L	5	5	0	E	H	3	3	4	
201	L	6A	6	1	I	H	3	3	4	
291	L	6B	6	2	V	H	3	3	4	
488	L	10	10	3	Q	H	4	4	5	
115	L	5	5	0	E	H	4	4	5	
203	L	6A	6	1	I	H	4	4	5	
293	L	6B	6	2	V	H	4	4	5	

Next steps: Generate code with `t` View recommended plots

```
# we restricted the distance measurement to CA-CA distance, notice the distance are now longer
t=p.rs_dist('L', 'H', ats='CA')
t
```

	chain_a	resn_a	resn_i_a	resi_a	res_a	chain_b	resn_b	resn_i_b	resi_b	res
2	L	6A	6	1	I	H	3	3	4	
0	L	5	5	0	E	H	3	3	4	
6	L	10	10	3	Q	H	3	3	4	
4	L	6B	6	2	V	H	3	3	4	
3	L	6A	6	1	I	H	4	4	5	
7	L	10	10	3	Q	H	4	4	5	
1	L	5	5	0	E	H	4	4	5	
5	L	6B	6	2	V	H	4	4	5	

Next steps: Generate code with t View recommended plots

Instead of residue distance, we can also measure the distance between all-atom combinations. Our ultimate aim in the future is to add a column for interaction types, such as clash, electrostatic, hydrogen bonds, etc. Please contact us, if you have the knowledge in how to classify the interaction type of each atom pair.

```
# We list distances between all atom pairs of two residues
p=Protein(fk)
t=p.atom_dist('L6A', 'H3')
print(f"\n... total rows: {len(t)}\n")
t[:10]
```

```
Warning: residues with insertion code: L6A, L6B

... total rows: 64
```

	chain_a	resn_a	resn_i_a	resi_a	res_a	chain_b	resn_b	resn_i_b	resi_b	re
38	L	6A	6	1	I	H	3	3	4	
30	L	6A	6	1	I	H	3	3	4	
62	L	6A	6	1	I	H	3	3	4	
22	L	6A	6	1	I	H	3	3	4	
54	L	6A	6	1	I	H	3	3	4	
6	L	6A	6	1	I	H	3	3	4	
46	L	6A	6	1	I	H	3	3	4	
14	L	6A	6	1	I	H	3	3	4	
37	L	6A	6	1	I	H	3	3	4	
39	L	6A	6	1	I	H	3	3	4	

```
# We list distances between all backbone atom pairs of two residues
t=p.atom_dist('L6A', 'H3', ats=['CA','C','N','O'])
t
```

	chain_a	resn_a	resn_i_a	resi_a	res_a	chain_b	resn_b	resn_i_b	resi_b	re
13	L	6A	6	1	I	H	3	3	4	
15	L	6A	6	1	I	H	3	3	4	
14	L	6A	6	1	I	H	3	3	4	
12	L	6A	6	1	I	H	3	3	4	
9	L	6A	6	1	I	H	3	3	4	
1	L	6A	6	1	I	H	3	3	4	
11	L	6A	6	1	I	H	3	3	4	
5	L	6A	6	1	I	H	3	3	4	
8	L	6A	6	1	I	H	3	3	4	
10	L	6A	6	1	I	H	3	3	4	
3	L	6A	6	1	I	H	3	3	4	
0	L	6A	6	1	I	H	3	3	4	
2	L	6A	6	1	I	H	3	3	4	
7	L	6A	6	1	I	H	3	3	4	
4	L	6A	6	1	I	H	3	3	4	
6	L	6A	6	1	I	H	3	3	4	

Next steps: Generate code with t View recommended plots

RMSD

To measure the root mean square deviation (RMSD) between two residue selections, we first translate one molecule by 5Å, then call rmsd(). The method `rmsd` takes a target object, source residue list, target residue list, and optionally an atom selection.

```
p=Protein(fk)
q=p.translate([3.0,4.0,0.0], inplace=False)
print("Translation:", p.center(), q.center(), q.center()-p.center(), "\n")
# consider all matched atoms, rs='ALL' or rs=None mean all residues
print(q.rmsd(p, None, None), "\n")
# consider CA only
print(q.rmsd(p, ats='CA'))
```

```
Warning: residues with insertion code: L6A, L6B
Translation: [ 15.54949999  -8.0205001  -15.39166681] [ 18.54949999  -4.0205001
-15.39166681] [3. 4. 0.]

5.0

5.0
```

The two selections should have the same number of residues, unless one selection has only one residue. Otherwise, it will error. Make sure the order of residues in the two selections is what you want, as the distance is computed for each residue pairwise in order. If two residues are of different types, only the common atoms are used, which is enforced by the atom_mask array. If we have different residue types, we recommend users use `ats` to explicitly restrict the computation to the backbone atoms only, e.g., "N,CA,C,O" or "CA".

rmsd() and align() are two methods where the order of the residues in the input matters, therefore, argument rl_a and rl_b are Residue Selection (RL) instead of RS. The following example shows why it is important to make sure the residue orders should match:

```
p=Protein(fn)
print(p.chain_id(), "\n")
# protein p and q have different chain orders
q=p.extract("P:H:L")
print(q.chain_id(), "\n")

# The default behavior of using Residue List returns the right answer
print(p.rmsd(q, "H:L", "H:L", ats="CA"), "\n")

# However, if you pass in residue select, the result is wrong in this case, as residues in the RS has been reordered
print(p.rmsd(q, p.rs("H:L"), q.rs("H:L"), ats="CA"), "\n")
# this is because in object p, p.rs("H:L") standarize the residue index and put L residues in front of H residue
# in object q, p.rs("H:L") keeps H residues in front of L residues.
# Therefore, residue selection breaks the one-to-one mapping relationship between the two residue lists.
# This example is created to demonstrate why we need to be careful, if we use RS objects for the RL argument in rmsd() and align
```

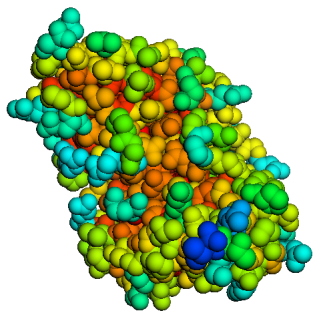
```
['L', 'H', 'P']

['P' 'H' 'L']

0.0

29.09069364318863
```

We often need to align the two structures first, before we take RMSD measurement. If we pass `align=True` into `rmsd()`, object `p` will then be first aligned to `q`, RMSD is then calculated. The `align()` method will be explained later.

Solvent-Accessible Surface Area (SASA)

We often would like to exclude some chains during the surface area computation. Although we can use `extract()` to take the chains we like, the `in_chains` argument provides more convenience. For example, if all chains are included in the antibody-antigen complex, CDR binding residues will not be accessible to water. But if the antigen chain is excluded, contact CDR residues will be exposed. Computing the different of SASA values with different in_chains is another popular way of identifying interface residues in addition to using `rs_around`.

```
p=Protein(fn)
# consider all chains, H:L:P, so the interface residues between H:L and P are mostly buried with low SASA values
t_HLP=p.sasa()
sasa=p.sasa().SASA.values
p.b_factors(sasa/sasa.max())
p.show(style="sphere", color="b")
```



```
# Get residue surface of a few L-chain residues
t_all=t_HLP[(t_HLP.chain=="L")][90:105]
t_all
```

	chain	resn	resn_i	resi	SASA
90	L	91	91	90	1.071459
91	L	92	92	91	1.071459
92	L	93	93	92	19.422432
93	L	94	94	93	61.171949
94	L	95	95	94	41.911208
95	L	96	96	95	37.959864
96	L	97	97	96	3.214377
97	L	98	98	97	12.076282
98	L	99	99	98	17.390160
99	L	100	100	99	0.000000
100	L	101	101	100	124.880067
101	L	102	102	101	14.201306
102	L	103	103	102	2.142918
103	L	104	104	103	103.554748
104	L	105	105	104	3.214377

Next steps: Generate code with t_all View recommended plots

```
print("\nnotice residue L92 only has a small surface area 1.071459\n")
# ignore antigen, CDR contact residues are now exposed with high SASA values
t_HL=p.sasa("H:L")
t_ag=t_HL[(t_HL.chain=="L")][90:105]
t_ag
```

```
notice residue L92 only has a small surface area 1.071459
```

	chain	resn	resn_i	resi	SASA
216	L	91	91	90	1.071459
217	L	92	92	91	36.329227
218	L	93	93	92	33.896076
219	L	94	94	93	78.605526
220	L	95	95	94	80.936500
221	L	96	96	95	37.959864
222	L	97	97	96	8.044890
223	L	98	98	97	12.076282
224	L	99	99	98	17.390160
225	L	100	100	99	0.000000
226	L	101	101	100	124.880067
227	L	102	102	101	14.201306
228	L	103	103	102	2.142918
229	L	104	104	103	103.554748
230	L	105	105	104	3.214377

Next steps: Generate code with t_ag View recommended plots

```
print("\nnotice residue L92 now has a large surface area 36.3292\n\n")
t_ag['DELTA_SASA']=t_ag['SASA'].values-t_all['SASA'].values
t_ag
```

```
notice residue L92 now has a large surface area 36.3292
```

	chain	resn	resn_i	resi	SASA	DELTA_SASA
216	L	91	91	90	1.071459	0.000000
217	L	92	92	91	36.329227	35.257768
218	L	93	93	92	33.896076	14.473644
219	L	94	94	93	78.605526	17.433577
220	L	95	95	94	80.936500	39.025291
221	L	96	96	95	37.959864	0.000000
222	L	97	97	96	8.044890	4.830513
223	L	98	98	97	12.076282	0.000000
224	L	99	99	98	17.390160	0.000000
225	L	100	100	99	0.000000	0.000000
226	L	101	101	100	124.880067	0.000000
227	L	102	102	101	14.201306	0.000000
228	L	103	103	102	2.142918	0.000000
229	L	104	104	103	103.554748	0.000000
230	L	105	105	104	3.214377	0.000000

Next steps: Generate code with t_ag View recommended plots

```
binder, rs_seed, t=p.rs_around(p.rs("P"), rs_within="L", dist=4)
print("Interface residues can also be identified with rs_around directly:", binder, "\n")
# the two methods are quite consistent with each other
```

```
Interface residues can also be identified with rs_around directly: L33,92-95,97
```

The example below uses `SASA` to identify the Ab-Ag interface residues.

```
p=Protein(fn)
print(p.chain_id())
# surface area of the whole complex
t=p.sasa()
all_L=t[t.chain=="L"]['SASA'].values
all_H=t[t.chain=="H"]['SASA'].values
all_P=t[t.chain=="P"]['SASA'].values
one_piece=np.concatenate((all_L, all_H, all_P))

# surface area of the antibody alone
t=p.sasa("H:L")
ab_L=t[t.chain=="L"]['SASA'].values
ab_H=t[t.chain=="H"]['SASA'].values
# surface area of the antigen alone
t=p.sasa("P")
ag_P=t['SASA'].values

two_pieces=np.concatenate((ab_L, ab_H, ag_P))

# delta ASAS for each chain
dASAS=two_pieces-one_piece
# normalize to [0, 1]
dASAS/=dASAS.max()
# color by delta surface area, the interface residues are highlighted
p.b_factors(1-dASAS)
p.show(color="b")
```

```
['L', 'H', 'P']
```

This demonstrates that we can either use rs_around to select interface residues, or calculate $\Delta ASAS$.

Secondary Structures - DSSP

Method `dssp` is a wrapper on BioPython's `Bio.PDB.DSSP` API, which requires the pre-installation of the DSSP package as described in https://en.wikipedia.org/wiki/DSSP_(algorithm).

We can install DSSP with:

```
conda install sbl::dssp
```

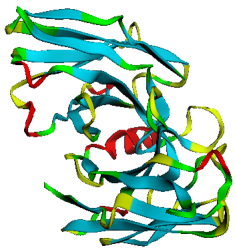
The secondary structure codes are:

- G = 3-turn helix (310 helix). Min length 3 residues.
- H = 4-turn helix (α helix). Minimum length 4 residues.
- I = 5-turn helix (π helix). Minimum length 5 residues.
- T = hydrogen bonded turn (3, 4 or 5 turn)
- E = extended strand in parallel and/or anti-parallel β-sheet conformation. Min length 2 residues.
- B = residue in isolated β-bridge (single pair β-sheet hydrogen bond formation)
- S = bend (the only non-hydrogen-bond based assignment).
- C = coil (residues which are not in any of the above conformations).

In the simplified mode, we map the codes into a - alpha helix, b - beta sheet, and c - coil.

```
# BioPython relies on DSSP command line program to compute secondary structures
if IN_COLAB:
    if not os.path.isfile("INSTALL_DSSP"):
        ! conda install sbl::dssp
        ! touch INSTALL_DSSP

p=Protein(fn)
# Biopython relies on command line DSSP program, it does not automatically detect the version, so we provides a locate_dssp() me
# locate_dss() does this for us
dssp_settings=Protein.locate_dssp()
print("DSSP command line and its version:", dssp_settings)
ss=p.dssp(simplify=True, **dssp_settings)
print(ss, "\n")
# set helix to 1, otherwise 0
for k,v in ss.items():
    b=np.zeros(len(v))+0.5 # default to turns
    b[np.array(list(v))=="a"]=0
    b[np.array(list(v))=="b"]=0.8
    b[np.array(list(v))=="c"]=0.3
    ss[k]=b
p.b_factors_by_chain(ss)
p.show(color="b")
```

```
DSSP command line and its version: {'DSSP': '/usr/local/bin/mkdssp',
'dssp_version': '4.2.2.1'}
{'L': '---bb-cccbbbb-cc--bbbbbbbccPPaaa-bbbbbb-ccc--bbbbbcccb--ccc-
ccbbbbbbccbbbbbbcc--aaa-cbbbbbb-cccc-bb---bbbbb----', 'H': '-bbbbPPPbbb-cc--
bbbbbbbbcc-cccc-bbbbbb-ccc-bbbbbbbbaaacbbbb-aaacccbbbbbbccccbbbbbb-c--aaa-
bbbbbbbbbbccccccc-bbbbbbbb---bbbbb--', 'P': '-aaaaaaaaaaa-'}
```



Internal Coordinate

To obtain the bond length (for backbone) and bond angles:

```
p=Protein(fk)
t=p.internal_coord(rs="L")
t
```

```
Warning: residues with insertion code: L6A, L6B
chain break at GLN  10  due to MaxPeptideBond (1.4 angstroms) exceeded
```

	chain	resn	resn_i	resi	aa	-1C:N	N:CA	CA:C	phi	psi	
0	L	5	5	0	E	NaN	1.483377	1.532696	NaN	127.190158	
1	L	6A	6	1	I	1.313512	1.449593	1.512209	-83.955214	121.806014	1
2	L	6B	6	2	V	1.325801	1.438910	1.527408	-92.621830	NaN	1
3	L	10	10	3	Q	NaN	1.451717	1.535136	NaN	NaN	

Next steps: Generate code with t View recommended plots

resi is the internal residue_index, and *resn* is residue name. The bond lengths are "-1C:N" (previous C to current N), N-CA, and CA-C, followed by all bond angles.

Notice ϕ and ω angle for the first residue is NaN, as there is no peptide bond for the first residue. Similarly, ψ angle is not defined for the last residue.

The peptide bond length is NaN between residues 6B and 10, as there are missing residues. If we do want to know how far these two residues are, we can modify the default MaxPeptideBond from 1.4 to a bigger value (which can be useful for imperfect structures output by diffusion methods):

```
p=Protein(fk)
t=p.internal_coord(rs="L", MaxPeptideBond=1e6)
t
```

```
Warning: residues with insertion code: L6A, L6B
```

	chain	resn	resn_i	resi	aa	-1C:N	N:CA	CA:C	phi	psi
0	L	5	5	0	E	NaN	1.483377	1.532696	NaN	127.190158
1	L	6A	6	1	I	1.313512	1.449593	1.512209	-83.955214	121.806014
2	L	6B	6	2	V	1.325801	1.438910	1.527408	-92.621830	176.913905
3	L	10	10	3	Q	7.310299	1.451717	1.535136	-165.152326	NaN

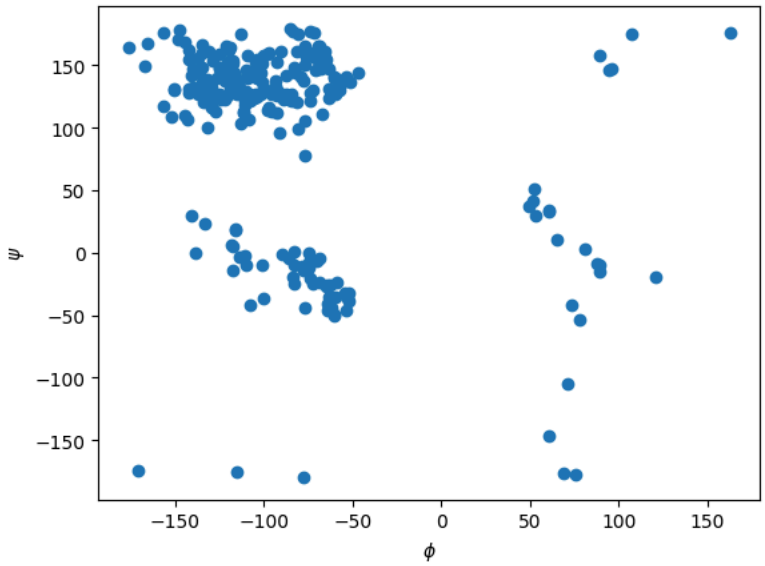
Next steps: Generate code with t View recommended plots

To run the example below, make sure you have matplotlib installed. If not, run in the shell:

```
pip install matplotlib
```

```
p=Protein(fn)
t=p.internal_coord()
#t.display()
from matplotlib import pyplot as plt
plt.scatter(t.phi, t.psi)
plt.xlabel('$\\phi$')
plt.ylabel('$\\psi$')
print("Ramachandran Plot")

#TODO: add the boundaries for popular engles, similar to the countours in https://www.mathworks.com/help/bioinfo/ref/ramachandra
```

```
Ramachandran Plot
```



Protein Object Manipulation

Move Objects

We can recenter, translate, and rotate an object.

```
p=Protein(fk)
print("Original center:", p.center(), "\n")
q=p.center_at([3.0,4.0,5.0], inplace=False)
print("New center after recentering:", q.center(), "\n")
q=p.translate([1.,0.,-1.], inplace=False)
print("New center after translate:", q.center(), "\n")
# rotate 90 degrees around axis [1,1,1]
print("RMSD before rotation:", q.rmsd(p, None, None, "CA"), "\n")
# we only rotate 5 degrees
q=p.rotate([1,1,1], 5, inplace=False)
print("Old center:", p.center(), "New center:", q.center(), "\n")
print("RMSD after rotation:", q.rmsd(p, ats="CA"), "\n")
# Warning, rotate is done using the origin as the center, that's why the rotation dramatically increase RMSD in the example abov
# most of the time, it's only meaningful if we center the molecule before rotation
rc=p.center()
q=p.center_at([0,0,0], inplace=False)
# rotate 5 degrees
q.rotate([1,1,1], 5, inplace=True)
# restore the center to the old center
q.center_at(rc, inplace=True)
# now the RMSD is much smaller
print("RMSD (center before rotate):", q.rmsd(p, None, None, "CA", align=True), "\n")
```

```
Warning: residues with insertion code: L6A, L6B
Original center: [ 15.54949999  -8.0205001  -15.39166681]

New center after recentering: [3. 4. 5.]

New center after translate: [ 16.54949999  -8.0205001  -16.39166681]

RMSD before rotation: 1.4142135623730951

Old center: [ 15.54949999  -8.0205001  -15.39166681] New center: [ 15.10944354
-6.44301224 -16.52909822]
```

To reset the position of an object, use `reset_pos()`. This will place the object center at the origin, as well as align its three PCA axes along Z, X, and Y axis, respectively.

```
p=Protein(fn)
print("Old center:", p.center(), "\n")
# rotation around a random axis direction with a random angle
p.rotate(np.random.random(3), np.random.random()*180)
p.show()
p.reset_pos()
p.show()
print("New center:", p.center())
```

```
Old center: [ 20.15256404  -5.12843199 -15.77199195]
```

```
New center: [-1.58451030e-14  1.81756832e-14 -1.21307409e-14]
```