Introduction to Biopython

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(based on a slides by Stuart Brown, NYU)



Learning Goals

- Biopython as a toolkit
- Seq objects and their methods
- SeqRecord objects have data fields
- SeqIO to read and write sequence objects
- Direct access to GenBank with Entrez.efetch
- Working with BLAST results



Modules

- Python functions are divided into three sets
 - A small core set that are always available
 - Some built-in modules such as math and os that can be imported from the basic install (ie. >>> import math)
 - An number of optional modules that must be downloaded and installed before you can import them: code that uses such modules is said to have "dependencies"
 - Most are available in different Linux distributions, or via pypy.org using pip (the Python Package Index)
- Anyone can write new Python modules, and often several different modules are available that can do the same task



Download a file

 urllib() is a module that lets Python download files from the internet with the .urlretrieve method

- >>> import urllib
- >>>urllib.urlretrieve('https://raw.githubusercontent.co m/biopython/biopython/master/Tests/GenBank/NC_00 5816.fna', 'yp.fasta')





- Biopython is an integrated collection of modules for "biological computation" including tools for working with DNA/protein sequences, sequence alignments, population genetics, phylogenetics, molecular structures, etc.
- It also provides interfaces to common biological databases (e.g. GenBank) and to some common locally installed software (e.g. BLAST)



Biopython Tutorial

Biopython has a "Tutorial & Cookbook":

http://biopython.org/DIST/docs/tutorial/Tutorial.html

by: Jeff Chang, Brad Chapman, Iddo Friedberg, Thomas Hamelryck, Michiel de Hoon, Peter Cock, Tiago Antao, Eric Talevich, Bartek Wilczyński

from which, most of the following examples are drawn



Object Oriented Code

- Python implements oject oriented programming
- Classes bundle data and functionality

```
magic method" class MyClass:
                 """A simple example class"""
                 def __init__(self):
                     self.data = []
 private
                     self._priv = "fuggetaboutit"
                 def say hi(self):
                     return 'hello world'
                 def ___str___(self):
                     return "yoohoo"+str(self.data[:3]) +"..."
                            instantiation
    z = MyClass()
    z.data = [1, "foo", "bar", -5, 9]
    print(z) #???
```

The Seq object

- The Seq object class is simple and fundamental for a lot of Biopython work. A Seq object can contain DNA, RNA, or protein.
- It contains a string and a defined alphabet for that string.
- The alphabets are actually defined objects such as

IUPACAmbiquousDNA Or IUPACProtein

- Which are defined in the Bio. Alphabet module
- A Seq object with a DNA alphabet has some different methods than one with an Amino Acid alphabet

```
>>> from Bio.Seq import Seq
>>> from Bio.Alphabet import IUPAC
>>> my_seq = Seq('AGTACACTGGT', <u>IUPAC.unambiquous dna</u>)
>>> my_seq
Seq('AGTACACTGGT', IUPAC.unambiquous_dna())
>>> print(my_seq)
AGTACACTGGT
```

Seq objects have string methods

- Seq objects have methods that work just like string objects
- You can get the len() of a Seq, slice it, and count() specific letters in it:

```
>>> my seg = Seg('GATCGATGGGCCTATATAGGATCGAAAATCGC',
IUPAC.unambiguous_dna)
>>> len(my seq)
32
>>> print(my_seq[6:9])
TGG
>>> my seq.count("G")
9
```



Turn a Seq object into a string

- Sometimes you will need to work with just the sequence string in a Seq object using a tool that is not aware of the Seq object methods
- Turn a Seq object into a string with str()

```
>>> my_seq
Seq('GATCGATGGGCCTATATAGGATCGAAAATCGC',
IUPACUnambiguousDNA())
>>> seq_string=str(my_seq)
>>> seq_string
'GATCGATGGGCCTATATAGGATCGAAAATCGC'
```



Seq Objects have methods

- DNA Seq objects can .translate() to protein
 - With optional translation table and to_stop=True parameters

```
>>>coding_dna=Seq("ATGGCCATTGTAATGGGCCGCTGAAAGGGTGCCCGATAG",
IUPAC.unambiguous_dna)
>>> coding_dna.translate()
Seq('MAIVMGR*KGAR*', HasStopCodon(IUPACProtein(), '*'))
>>> print(coding_dna.translate(table=2, to_stop=True))
MAIVMGRWKGAR
```

Seq objects with a DNA alphabet have the reverse_complement() method:

```
>>> my_seq = Seq('TTTAAAATGCGGG', IUPAC.unambiguous_dna)
>>> print(my_seq.reverse_complement())
CCCGCATTTTAAA
```

 The Bio.SeqUtils module has some useful methods, such as GC() to calculate % of G+C bases in a DNA sequence.

```
>>> from Bio.SeqUtils import GC
>>> GC(my_seq)
46.875
```



Protein Alphabet

- You could re-define my_seq as a protein by changing the alphabet,
 which will totally change the methods that will work on it.
 - ('G','A','T','C' are valid protein letters)

```
>>> from Bio.SeqUtils import molecular_weight
>>> my sea
Seq('AGTACACTGGT', IUPACUnambiguousDNA())
>>> print(molecular_weight(my_seq))
3436, 1957
>>> my_seq.alphabet = IUPAC.protein
>>> my seq
Seq('AGTACACTGGT', IUPACProtein())
>>> print(molecular_weight(my_seq))
912.0004
```



SeqRecord Object

 The SeqRecord object is like a database record (such as GenBank). It is a complex object that contains a Seq object, and also annotation fields or features

```
.seq
```

.id

.name

.description

.letter_annotations

.annotations

.features

.dbxrefs



SeqRecord Example

http://biopython.org/wiki/SeqRecord

Extracting information from a SeqRecord

Lets look in detail at the well annotated SeqRecord objects Biopython creates from a
GenBank file, such as ls_orchid.gbk, which we'll load using the SeqIO module. This
file contains 94 records:



SeqIO and FASTA files

- SeqIO is the all purpose file read/write tool for SeqRecords
 - SeqIO can read many file types: http://biopython.org/wiki/SeqIO
- SeqIO has .read() and .write() methods
 - (do not need to "open" file first)
- It can read a text file in FASTA format
- In Biopython, fasta is a type of SeqRecord with specific fields
 - grab the file: NC 005816.fna, and saved it as a text file in your current directory

```
>>> from Bio import SeqIO
>>> gene = SeqIO.read("NC_005816.fna", "fasta")
>>> gene.id
'gi|45478711|ref|NC_005816.1|'
>>> gene.seq
Seq('TGTAACGAACGGTGCAATAGTGATCCACACCCCAACGCCTGAAATCAGATCCAGG...CTG', SingleLetterAlphabet())
>>> len(gene.seq)
9609
```



Multiple FASTA Records in one file

- The FASTA format can store many sequences in one text file
- SeqIO.parse() reads the records one by one
- This code creates a list of SeqRecord objects:



Seq-ing deeper

Seq: an object for containing sequence information. Basic sequence operations.

```
>>> from Bio.Seg import Seg
>>> dir(Seq)
['__add__', '__class__', '__contains ', ' delattr ',
'__dict__', '__dir__', '__doc__', '__eq__',
'__format__', '__ge__', '__getattribute__',
'__getitem__', '__gt__', '__hash__', '__init__',
'__le__', '__len__', '__lt__', '__module__', '_ ne ',
'__new__', '__radd__', '__reduce__', '__reduce_ ex ',
' repr ', ' setattr ', ' sizeof ', ' str ',
' subclasshook ', ' weakref ',
'_get_seq_str_and_check_alphabet', 'back_transcribe',
'complement', 'count', 'endswith', 'find', 'lower',
'lstrip', 'reverse complement', 'rfind', 'rsplit',
'rstrip', 'split', 'startswith', 'strip', 'tomutable',
'tostring', 'transcribe', 'translate', 'ungap', 'upper']
```



Seq Source Code

```
def init (self, data, alphabet=Alphabet.generic alphabet):
       # Enforce string storage
        if not isinstance(data, basestring):
            raise TypeError("The sequence data given to a Seq object should "
                            "be a string (not another Seg object etc)")
        self._data = data
        self.alphabet = alphabet
def repr (self):
        """Returns a (truncated) representation of the sequence for debugging."""
        if len(self) > 60:
            # Shows the last three letters as it is often useful to see if there
           # is a stop codon at the end of a sequence.
            # Note total length is 54+3+3=60
            return "{0}('{1}...{2}', {3!r})".format(self. class . name ,
                                                    str(self)[:54],
                                                    str(self)[-3:],
                                                    self.alphabet)
        else:
            return '{0}({1!r}, {2!r})'.format(self.__class__.__name__,
                                              self. data,
                                              self.alphabet)
```



Seq Source Code

```
def transcribe(self):
    base = Alphabet._get_base_alphabet(self.alphabet)
    if isinstance(base, Alphabet.ProteinAlphabet):
        raise ValueError("Proteins cannot be transcribed!")
    if isinstance(base, Alphabet.RNAAlphabet):
        raise ValueError("RNA cannot be transcribed!")

if self.alphabet == IUPAC.unambiguous_dna:
        alphabet = IUPAC.unambiguous_rna
    elif self.alphabet == IUPAC.ambiguous_dna:
        alphabet = IUPAC.ambiguous_rna
    else:
        alphabet = Alphabet.generic_rna
    return Seq(str(self).replace('T', 'U').replace('t', 'u'), alphabet))
```



Create your own sequence class

Base class

```
from Bio. Seg import Seg
class MySeq(Seq):
    def ✓ repr (self):
         if len(self) > 60:
             # Shows the last three letters as it is often useful to see if there
             # is a stop codon at the end of a sequence.
             # Note total length is 54+3+3=60
             return "{0}('{1}---{2}', {3!r})".format(self.__class__.__name___,
                                                     str(self)[:54],
Derived class
                                                     str(self)[-3:],
                                  Was "..."
                                                     self.alphabet)
         else:
             return '{0}((1!r), (2!r))'.format(self. class . name ,
                                               self._data,
                                               self.alphabet)
```

- The new __repr__ overrides the old __repr__
- Everything else remains the same!
- MySeq is a derived class of Seq
- Also, MySeq is inherited from Seq



Direct Access to GenBank

- BioPython has modules that can directly access databases over the Internet
- The Entrez module uses the NCBI Efetch service
- Efetch works on many NCBI databases including protein and PubMed literature citations
- The 'gb' data type contains much more annotation information, but rettype='fasta' also works
- With a few tweaks, this code could be used to download a list of GenBank ID's and save them as FASTA or GenBank files:

```
>>> from Bio import Entrez
>>>Entrez.email = "you@iastate.edu"
>>> handle = Entrez.efetch(db="nucleotide", id="186972394", rettype="gb",
retmode="text")
>>> record = SeqIO.read(handle, "genbank")
```

>>> print(record)

ID: EU490707.1

Name: EU490707

Description: Selenipedium aequinoctiale maturase K (matK) gene, partial cds; chloroplast.

Number of features: 3

/sequence_version=1



These are sub-fields of the .annotations field

/source=chloroplast Selenipedium aequinoctiale

/taxonomy=['Eukaryota', 'Viridiplantae', 'Streptophyta', 'Embryophyta', 'Tracheophyta',

'Spermatophyta', 'Magnoliophyta', 'Liliopsida', 'Asparagales', 'Orchidaceae', 'Cypripedioideae',

'Selenipedium']

/keywords=['']

/references=[Reference(title='Phylogenetic utility of ycf1 in orchids: a plastid gene more variable than matK', ...), Reference(title='Direct Submission', ...)]

/accessions=['EU490707']

/data_file_division=PLN

/date=15-JAN-2009

/organism=Selenipedium aequinoctiale

/gi=186972394

Seq('ATTTTTACGAACCTGTGGAAATTTTTGGTTATGACAATAAATCTAGTTTAGTA...GAA', IUPACAmbiguousDNA())



BLAST

- BioPython has several methods to work with the popular NCBI BLAST software
- NCBIWWW.qblast() sends queries directly to the NCBI BLAST server. The query can be a Seq object, FASTA file, or a GenBank ID.

```
>>> from Bio.Blast import NCBIWWW
>>> from Bio import SeqIO
>>> query = SeqIO.read("test.fasta", format="fasta")
>>> result_handle = NCBIWWW.qblast("blastn", "nt", query.seq)
>>> blast_result = result_handle.read()
>>> blast_file = open("my_blast.xml", "w") # create an xml output file
>>> blast_file.write(blast_result)
>>> blast_file.close() # tidy up
>>> result handle.close()
```

XML

Extensible Markup Language
Used to encode documents in a form taht is human readable and machine readable

Note

To: Tove

From: Jani

Reminder

Don't forget me this weekend!



XML is Extensible

Note

To: Tove

From: Jani

Reminder

Don't forget me this weekend!

Note

To: Tove

From: Jani

Date: 2015-09-01 08:30

Don't forget me this weekend!



BLAST XML

```
MULTISPECIES: bacteriocin sakacin-P [Lactobacillus]
 Sequence ID: WP 004271264.1 Length: 61 Number of Matches: 1
 ▶ See 11 more title(s)
 Range 1: 1 to 61 GenPept Graphics
                                                                                     ▼ Next Match ▲ Previous Match
                   Expect Method
                                                                Identities
                                                                                 Positives
 122 bits(307) 1e-35 Compositional matrix adjust.
                                                              61/61(100%)
                                                                                61/61(100%) 0/61(0%)
                MEKFIELSLKEVTAITGGKYYGNGVHCGKHSCTVDWGTAIGNIGNNAAANWATGGNAGWN
MEKFIELSLKEVTAITGGKYYGNGVHCGKHSCTVDWGTAIGNIGNNAAANWATGGNAGWN
MEKFIELSLKEVTAITGGKYYGNGVHCGKHSCTVDWGTAIGNIGNNAAANWATGGNAGWN
 Sbjct 1
 Query 61
 Sbjct 61 K 61
Bownload ∨ GenPept Graphics
 MULTISPECIES: bacteriocin [Lactobacillus]
 Sequence ID: WP 056946897.1 Length: 61 Number of Matches: 1
 ▶ See 3 more title(s)
 Range 1: 1 to 61 GenPept Graphics
                                                                                     ▼ Next Match ▲ Previous Match
                                                                Identities
                                                                                 Positives
                                                                60/61(98%) 61/61(100%) 0/61(0%)
 120 bits(302) 8e-35 Compositional matrix adjust.
                MEKFIELSLKEYTAITGGKYYGNGVHCGKHSCTVDWGTAIGNIGNNAAANWATGGNAGWN
MEKFIELSLKEYTAITGGKYYGNGVHCGKYSCTVDWGTAIGNIGNNAAANWATGGNAGWN
MEKFIELSLKEYTAITGGKYYGNGVHCGKYSCTVDWGTAIGNIGNIGNNAAANWATGGNAGWN
 Query 61
Sbjct 61 K 61
Bownload → GenPept Graphics
 bacteriocin mundticin [Enterococcus pallens]
 Sequence ID: WP_010758561.1 Length: 62 Number of Matches: 1
 ▶ See 3 more title(s)
                                                                                     ▼ Next Match ▲ Previous Match
 Range 1: 1 to 61 GenPept Graphics
                                                                                  Positives
                    Expect Method
                                                                 Identities
 91.3 bits(225) 4e-23 Compositional matrix adjust. 45/61(74%) 50/61(81%) 0/61(0%)
                MEKFIELSLKEVTAITGGKYYGNGVHCGKHSCTVDWGTAIGNIGNNAAANWATGGNAGWN
M+ LS KE+ ITGGKYYGNG+ CGK+SC+VDWG AIG IGNNAAANWATGG AGWN
MQNIKALSAKELIEITGGKYYGNGLSCGKYSCSVDWGKAIGIIGNNAAANWATGGAAGWN
 Query 61
                    61
 Sbict 61 K
```



BLAST XML

Sbict 61 K 61

```
<Hit>
  <Hit num>3</Hit num>
  <Hit id>gi|498453073|ref|WP 010758561.1|</Hit id>
  <Hit def>bacteriocin mundticin [Enterococcus pallens] &gt;qi|486858092|qb|E0H91048.1| bacterioc
in mundticin [Enterococcus pallens ATCC BAA-351] &qt;qi|508242815|qb|E0U16245.1| bacteriocin mund
ticin [Enterococcus pallens ATCC BAA-351] &qt;qi|1105345489|qb|0JG79015.1| bacteriocin mundticin
[Enterococcus pallens]</Hit def>
  <Hit accession>WP 010758561/Hit accession>
  <Hit len>62</Hit len>
  <Hit hsps>
    <Hsp>
       <Hsp num>1</Hsp num>
       <Hsp bit-score>91.2781/Hsp bit-score>
       <Hsp score>225</Hsp score>
       <Hsp evalue>4.38515e-23/Hsp evalue>
       <Hsp query-from>1</Hsp query-from>
       <Hsp query-to>61</Hsp query-to>
       <Hsp hit-from>1</Hsp hit-from>
       <Hsp hit-to>61</Hsp hit-to>
       <Hsp query-frame>0</Hsp query-frame>
       <Hsp hit-frame>0</Hsp hit-frame>
       <Hsp identity>45</Hsp identity>
       <Hsp positive>50</Hsp positive>
       <Hsp gaps>0</Hsp gaps>
       <Hsp align-len>61</Hsp align-len>
       <Hsp qseq>MEKFIELSLKEVTAITGGKYYGNGVHCGKHSCTVDWGTAIGNIGNNAAANWATGGNAGWNK</Hsp_qseq>
       <Hsp hseq>MQNIKALSAKELIEITGGKYYGNGLSCGKYSCSVDWGKAIGIIGNNAAANWATGGAAGWNK</Hsp hseq>
       <Hsp midline>M+
                             LS KE+ ITGGKYYGNG+ CGK+SC+VDWG AIG IGNNAAANWATGG AGWNK</Hsp midline>
    </Hsp>
  </Hit hsps>
 bacteriocin mundticin [Enterococcus pallens]
 Sequence ID: WP 010758561.1 Length: 62 Number of Matches: 1
 ▶ See 3 more title(s)
                                               Range 1: 1 to 61 GenPept Graphics
           Expect Method
                                    Identities
                                              Positives
 91.3 bits(225) 4e-23 Compositional matrix adjust. 45/61(74%) 50/61(81%) 0/61(0%)
         MEKFIELSLKEVTAITGGKYYGNGVHCGKHSCTVDWGTAIGNIGNNAAANWATGGNAGWN
M+ LS KE+ ITGGKYYGNG+ CGK+SC+VDWG AIG IGNNAAANWATIG AGWN
MQNIKALSAKELIEITGGKYYGNGLSCGKYSCSVDWGKAIGIIGNNAAANWATIGAAGWN
 Query 61 K 61
```



BLAST

- BioPython has several methods to work with the popular NCBI BLAST software
- NCBIWWW.qblast() sends queries directly to the NCBI BLAST server. The query can be a Seq object, FASTA file, or a GenBank ID.

```
>>> from Bio.Blast import NCBIWWW
>>> from Bio import SeqIO
>>> query = SeqIO.read("test.fasta", format="fasta")
>>> result_handle = NCBIWWW.qblast("blastn", "nt", query.seq)
>>> blast_result = result_handle.read()
>>> blast_file = open("my_blast.xml", "w") # create an xml output file
>>> blast_file.write(blast_result)
>>> blast_file.close() # tidy up
>>> result handle.close()
```

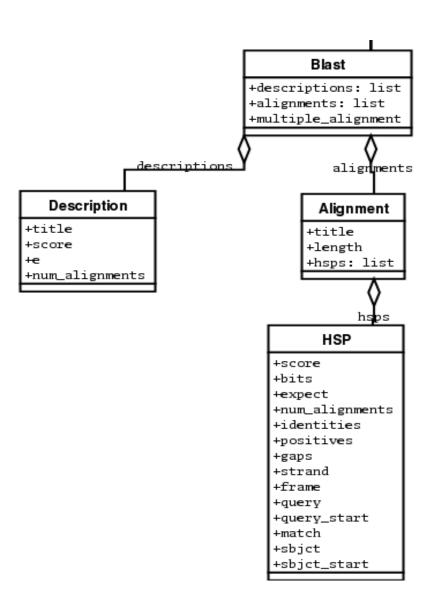
Parse BLAST Results

- It is often useful to obtain a BLAST result directly (local BLAST server or via Web browser) and then parse the result file with Python.
- Save the BLAST result in XML format
 - NCBIXML.read() for a file with a single BLAST result (single query)
 - NCBIXML.parse() for a file with multiple BLAST results (multiple queries)

```
>>> from Bio.Blast import NCBIXML
>>> handle = open("my_blast.xml")
>>> blast_record = NCBIXML.read(handle)
>>> for hit in blast_record.descriptions:
... print(hit.title)
... print(hit.e)
>>> handle.close()
```



BLAST Record Object





View Aligned Sequence

```
>>> from Bio.Blast import NCBIXML
    >>> handle = open("my blast.xml")
    >>> blast record = NCBIXML.read(handle)
    >>> for hit in blast_record.alignments:
             for hsp in hit.hsps:
                  print hit.title
                  print hsp.expect
                  print (hsp.query[0:75] + '...')
                  print(hsp.match[0:75] + '...')
                  print(hsp.sbjct[0:75] + '...')
qi|731383573|ref|XM 002284686.2| PREDICTED: Vitis vinifera cold-regulated 413
plasma membrane protein 2 (LOC100248690), mRNA
2.5739e-53
ATGCTAGTATGCTCGGTCATTACGGGTTTGGCACT-CATTTCCTCAAATGGCTCGCCTGCCTTGCGGCTATTTAC...
1111 1 11 111 111
                     ATGCCATTAAGCTTGGTGGTCTGGGCTTTGGCACTACATTTCTTGAG-TGGTTGGCTTCTTTTTGCTGCCATTTAT...
```



Many Matches

- Often a BLAST search will return many matches for a single query (save as an XML format file)
- NCBIXML.parse() can return these as BLAST record objects in a list, or deal with them directly in a for loop.



Get a file by FTP in Python

```
>>> from ftplib import FTP
>>> host="ftp.sra.ebi.ac.uk"
>>> ftp=FTP(host)
>>> ftp.login()
'230 Login successful.'
ftp.cwd('vol1/fastq/SRR020/SRR020192')
'250 Directory successfully changed.'
>>> ftp.retrlines('LIST')
-r--r-- 1 ftp
                 ftp 1777817 Jun 24 20:12 SRR020192.fastq.gz
'226 Directory send OK.'
>>> ftp.retrbinary('RETR SRR020192.fastq.gz', open('SRR020192.fastq.gz',
'wb').write)
'226 Transfer complete.'
>>> ftp.quit()
'221 Goodbye.'
```

Learning Objectives

- Biopython is a toolkit
- Seq objects and their methods
- SeqRecord objects have data fields
- SeqIO to read and write sequence objects
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- Working with BLAST results

