Class_09_lab

Tessa Sterns PID: A18482353

Table of contents

The PDB is the main databank for protein structures.

```
library(readr)

ps.data <- read_csv("Data Export Summary.csv")
# if only using one function can write as
#'readr: :read_csv()`
```

```
sum(ps.data$Total)
```

```
[1] 243910
```

```
p.xray <- round((sum(ps.data$`X-ray`)/sum(ps.data$Total))*100, 2)
p.EM <- round((sum(ps.data$EM)/sum(ps.data$Total))*100,2)

Tot.p <- round((ps.data[1,9, drop = TRUE] / sum(ps.data$Total))*100,2)
```

> Q1: What percentage of structures in the PDB are solved by X-Ray and Electron Microscopy.

There have been 81.48% solved by X-ray and 12.22% solved by electron microscopy.

> Q2: What proportion of structures in the PDB are protein?

There are 86.05 % of the total data set are proteins.

> Q3: Type HIV in the PDB website search box on the home page and determine how many HIV-1 protease structures are in the current PDB?

There are 1150 structures of the HIV-1 protease in the current PDB.

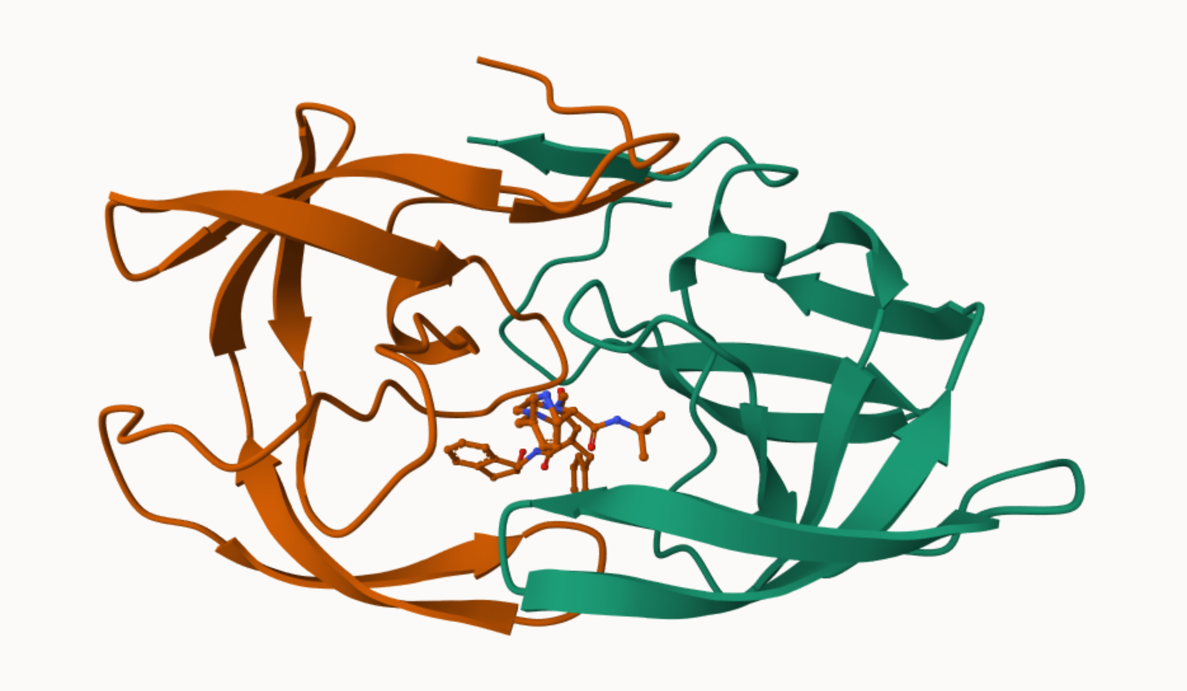
Exploring PDB structures

package for structural bioinformatics - bio3d

```
library(bio3d)

hiv <- read.pdb(file = "1hsg")
```

```
  Note: Accessing on-line PDB file
```

Using Mol* veiwer to explore 1HSG structure.



> Q4: Water molecules normally have 3 atoms. Why do we see just one atom per water molecule in this structure?

They are just showing the oxygen which is the larger and more electronegative atom in water. One reason for this is to make it easier to visualize. Similar to how we don't show all the molecules of water because if we did it would be difficult to see the protein.

> Q5: There is a critical "conserved" water molecule in the binding site. Can you identify this water molecule? What residue number does this water molecule have

This water molecule is HOH 308. It is participating in four hydrogen bonds, 2 with the ligand and 2 with the HIV-1 protease.

> Q6: Generate and save a figure clearly showing the two distinct chains of HIV-protease along with the ligand. You might also consider showing the catalytic residues ASP 25 in each chain and the critical water (we recommend "Ball & Stick" for these side-chains). Add this figure to your Quarto document.

A view of HIV-1 protease with catalytic residues in space filling model and the ligand MK1 as a ball and stick model.

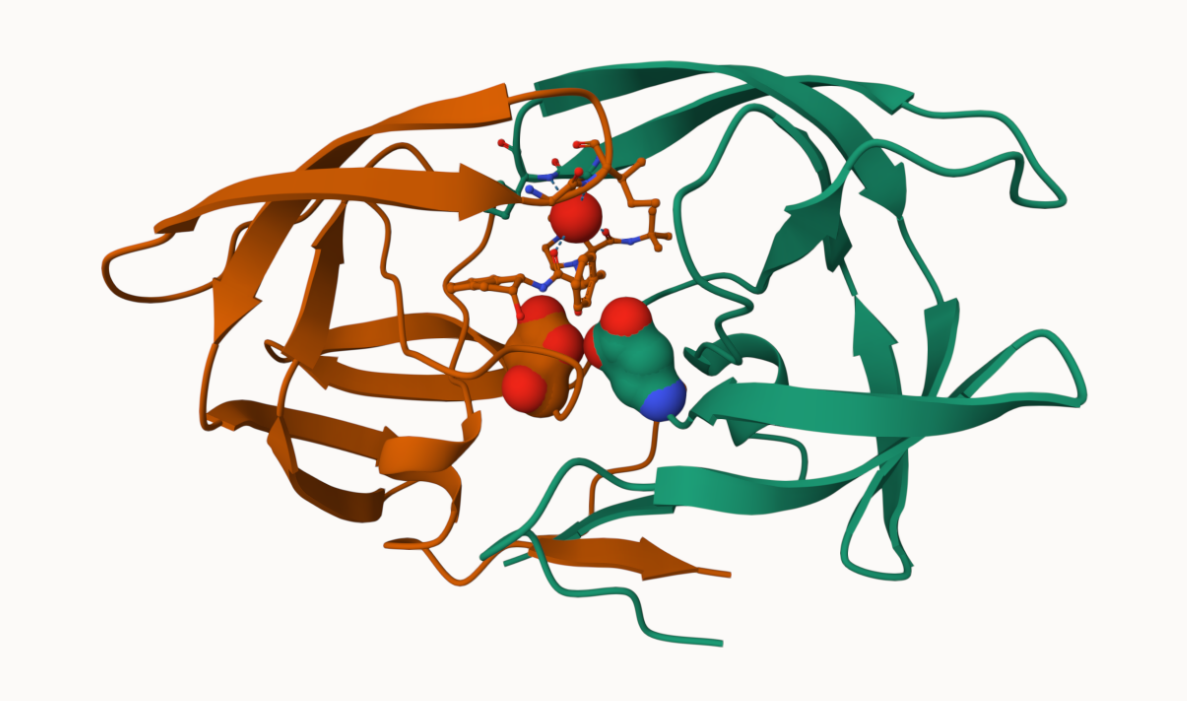


Figure 1: HIV-1 protease with catalytic D25 residues and catalytic H2O highlighted.

> Discussion Topic: Can you think of a way in which indinavir, or even larger ligands and substrates, could enter the binding site?

Because proteins are dynamic it is likely that when not bound by a ligand the HIV-1 protease is in a more open configuration and then conforms to fit the ligind upon interaction.

> Q7: [Optional] As you have hopefully observed HIV protease is a homodimer (i.e. it is composed of two identical chains). With the aid of the graphic display can you identify secondary structure elements that are likely to only form in the dimer rather than the monomer?

There is an antiparalel beta-sheet that is formed by a beta-strand from each of the monomers. Additionally the binding pocket itself rests in the middle of the two chains.

PDB objects in R

```
head(hiv$atom)
```

```
  type eleno elety  alt resid chain resno insert      x      y     z o     b
1 ATOM     1     N <NA>   PRO     A     1   <NA> 29.361 39.686 5.862 1 38.10
2 ATOM     2    CA <NA>   PRO     A     1   <NA> 30.307 38.663 5.319 1 40.62
3 ATOM     3     C <NA>   PRO     A     1   <NA> 29.760 38.071 4.022 1 42.64
4 ATOM     4     O <NA>   PRO     A     1   <NA> 28.600 38.302 3.676 1 43.40
5 ATOM     5    CB <NA>   PRO     A     1   <NA> 30.508 37.541 6.342 1 37.87
6 ATOM     6    CG <NA>   PRO     A     1   <NA> 29.296 37.591 7.162 1 38.40
  segid elesy charge
1  <NA>     N   <NA>
2  <NA>     C   <NA>
3  <NA>     C   <NA>
4  <NA>     O   <NA>
5  <NA>     C   <NA>
6  <NA>     C   <NA>
```

Extract the sequence

```
pdbseq(hiv)
```

```
  1   2   3   4   5   6   7   8   9  10  11  12  13  14  15  16  17  18  19  20
"P" "Q" "I" "T" "L" "W" "Q" "R" "P" "L" "V" "T" "I" "K" "I" "G" "G" "Q" "L" "K"
 21  22  23  24  25  26  27  28  29  30  31  32  33  34  35  36  37  38  39  40
```

```
"E" "A" "L" "L" "D" "T" "G" "A" "D" "D" "T" "V" "L" "E" "E" "M" "S" "L" "P" "G"
 41  42  43  44  45  46  47  48  49  50  51  52  53  54  55  56  57  58  59  60
"R" "W" "K" "P" "K" "M" "I" "G" "G" "I" "G" "G" "F" "I" "K" "V" "R" "Q" "Y" "D"
 61  62  63  64  65  66  67  68  69  70  71  72  73  74  75  76  77  78  79  80
"Q" "I" "L" "I" "E" "I" "C" "G" "H" "K" "A" "I" "G" "T" "V" "L" "V" "G" "P" "T"
 81  82  83  84  85  86  87  88  89  90  91  92  93  94  95  96  97  98  99   1
"P" "V" "N" "I" "I" "G" "R" "N" "L" "L" "T" "Q" "I" "G" "C" "T" "L" "N" "F" "P"
  2   3   4   5   6   7   8   9  10  11  12  13  14  15  16  17  18  19  20  21
"Q" "I" "T" "L" "W" "Q" "R" "P" "L" "V" "T" "I" "K" "I" "G" "G" "Q" "L" "K" "E"
 22  23  24  25  26  27  28  29  30  31  32  33  34  35  36  37  38  39  40  41
"A" "L" "L" "D" "T" "G" "A" "D" "D" "T" "V" "L" "E" "E" "M" "S" "L" "P" "G" "R"
 42  43  44  45  46  47  48  49  50  51  52  53  54  55  56  57  58  59  60  61
"W" "K" "P" "K" "M" "I" "G" "G" "I" "G" "G" "F" "I" "K" "V" "R" "Q" "Y" "D" "Q"
 62  63  64  65  66  67  68  69  70  71  72  73  74  75  76  77  78  79  80  81
"I" "L" "I" "E" "I" "C" "G" "H" "K" "A" "I" "G" "T" "V" "L" "V" "G" "P" "T" "P"
 82  83  84  85  86  87  88  89  90  91  92  93  94  95  96  97  98  99
"V" "N" "I" "I" "G" "R" "N" "L" "L" "T" "Q" "I" "G" "C" "T" "L" "N" "F"
```

Q7: How many amino acid residues are there in this pdb object?

There are 198 amino acid residues in this pdb object.

```r
library(dplyr)
```

```

Attaching package: 'dplyr'

The following objects are masked from 'package:stats':

    filter, lag

The following objects are masked from 'package:base':

    intersect, setdiff, setequal, union
```

```r
non.p <- hiv$atom %>%
  filter(resid == "MK1")
table(non.p$resid)
```

```

MK1
 45
```

Q8: Name one of the two non-protein residues?

One of the non-protein residues is MK1 which is the ligind designed to inhibit the protease.

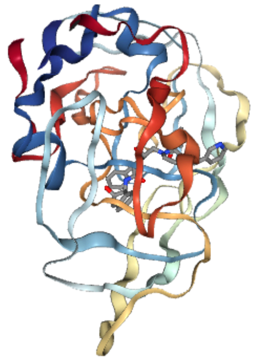
Q9: How many protein chains are in this structure?

There are two chains in this structure an alpha and a beta.

Need the **bio3dview** package installed in the console first.

```r
library(bio3dview)

view.pdb(hiv)
```

```
file:////private/var/folders/vr/dj7chnl94lv5g8p28r0c72b00000gn/T/Rtmp7fh4SE/file9ae42a4cb06/
```

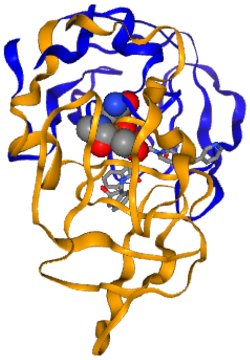


```
hiv1.Aseq <- pdbseq(trim.pdb(hiv, chain="A"))
```

Changing some settings

```
sel <- atom.select(hiv, resno=25)
view.pdb(hiv,
         highlight = sel,
         highlight.style = "spacefill",
         colorScheme = "chain",
         col = c("blue", "orange"),
         backgroundColor = "lightpink")
```

```
file:////private/var/folders/vr/dj7chnl94lv5g8p28r0c72b00000gn/T/Rtmp7fh4SE/file9ae36dfc2ef/
```



Predicting functional motions of a single structure

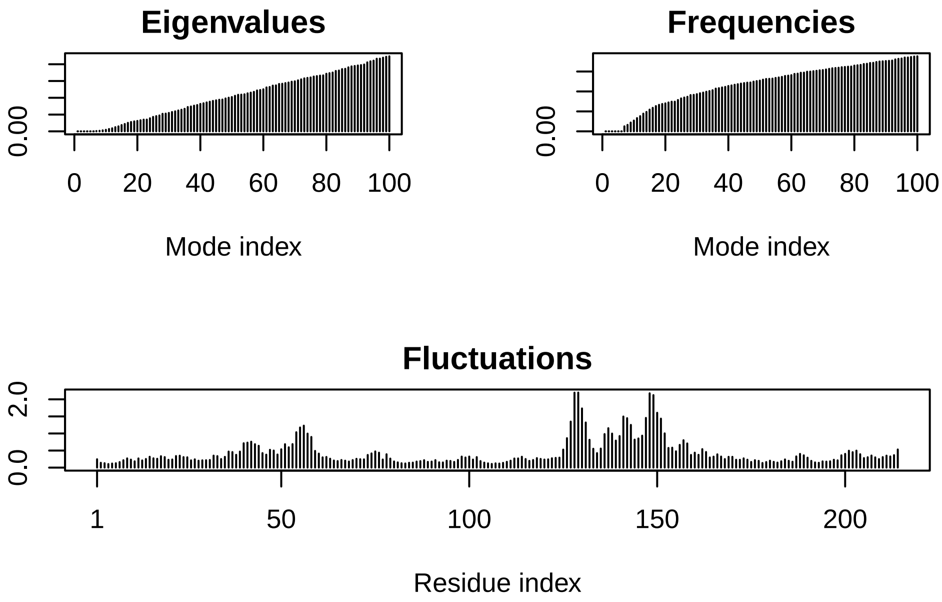
```r
adk <- read.pdb("6s36")
```

```
  Note: Accessing on-line PDB file
   PDB has ALT records, taking A only, rm.alt=TRUE
```

```r
#flexibility prediction
m <- nma(adk)
```

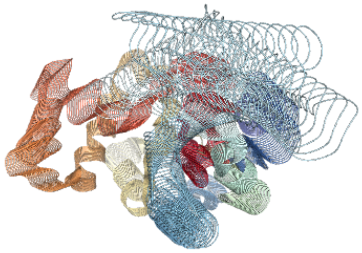
```
 Building Hessian...        Done in 0.009 seconds.
 Diagonalizing Hessian...   Done in 0.174 seconds.
```

```r
plot(m)
```



```r
# Molecular trajectory animation
mktrj(m, file="adk_m7.pdb")

view.nma(m, pdb=adk)
```

file:////private/var/folders/vr/dj7chnl94lv5g8p28r0c72b00000gn/T/Rtmp7fh4SE/file9ae1ee957b6/



Comparative structure analysis of Adenylate Kinase

> Q10. Which of the packages above is found only on BioConductor and not CRAN?

The 'BiocManager::install("msa")' package is not on CRAN, the use of ': :' indecates we are downloading a package from an outside library, and Bioc is a shortened form of Biocunductor.

> Q11. Which of the above packages is not found on BioConductor or CRAN?

The 'pak::pak("bioboot/bio3dview")' package.

> Q12. True or False? Functions from the pak package can be used to install packages from GitHub and BitBucket?

True

##Search and retrieve ADK structures

```r
aa <- get.seq("1ake_A")
```

```
Warning in get.seq("1ake_A"): Removing existing file: seqs.fasta
```

```
Fetching... Please wait. Done.
```

```r
aa
```

```
             1        .         .         .         .         .          60
pdb|1AKE|A   MRIILLGAPGAGKGTQAQFIMEKYGIPQISTGDMLRAAVKSGSELGKQAKDIMDAGKLVT
             1        .         .         .         .         .          60

            61        .         .         .         .         .          120
pdb|1AKE|A   DELVIALVKERIAQEDCRNGFLLDGFPRTIPQADAMKEAGINVDYVLEFDVPDELIVDRI
            61        .         .         .         .         .          120

           121        .         .         .         .         .          180
pdb|1AKE|A   VGRRVHAPSGRVYHVKFNPPKVEGKDDVTGEELTTRKDDQEETVRKRLVEYHQMTAPLIG
           121        .         .         .         .         .          180

           181        .         .         .   214
pdb|1AKE|A   YYSKEAEAGNTKYAKVDGTKPVAEVRADLEKILG
           181        .         .         .   214
```

```
Call:
  read.fasta(file = outfile)

Class:
  fasta

Alignment dimensions:
  1 sequence rows; 214 position columns (214 non-gap, 0 gap)

+ attr: id, ali, call
```

> Q13. How many amino acids are in this sequence, i.e. how long is this sequence?

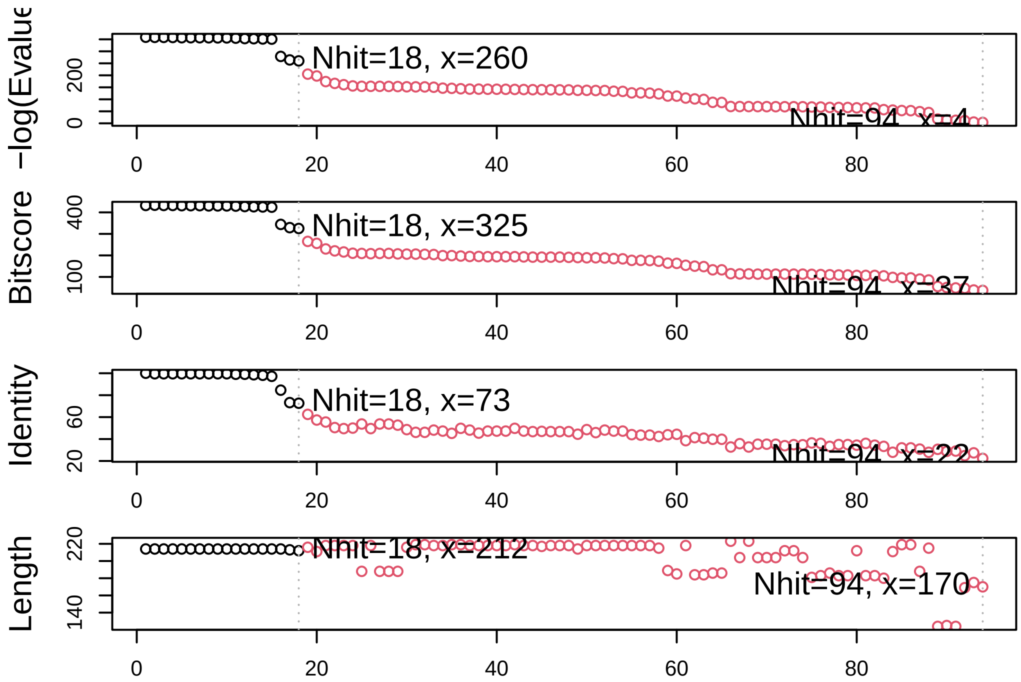
There are 214 amino acids in this sequence.

```
# Blast or hmmer search
b <- blast.pdb(aa)
```

```
 Searching ... please wait (updates every 5 seconds) RID = G9U8CRTJ014
 .........
 Reporting 94 hits
```

```
hits <- plot(b)
```

```
  * Possible cutoff values:    260 3
            Yielding Nhits:    18 94

  * Chosen cutoff value of:    260
            Yielding Nhits:    18
```



```r
head(hits$pdb.id)
```

```
[1] "1AKE_A" "8BQF_A" "4X8M_A" "6S36_A" "8Q2B_A" "8RJ9_A"
```

```r
# Download releated PDB files
files <- get.pdb(hits$pdb.id, path="pdbs", split=TRUE, gzip=TRUE)
```

```
Warning in get.pdb(hits$pdb.id, path = "pdbs", split = TRUE, gzip = TRUE):
pdbs/1AKE.pdb.gz exists. Skipping download

Warning in get.pdb(hits$pdb.id, path = "pdbs", split = TRUE, gzip = TRUE):
pdbs/8BQF.pdb.gz exists. Skipping download

Warning in get.pdb(hits$pdb.id, path = "pdbs", split = TRUE, gzip = TRUE):
pdbs/4X8M.pdb.gz exists. Skipping download

Warning in get.pdb(hits$pdb.id, path = "pdbs", split = TRUE, gzip = TRUE):
pdbs/6S36.pdb.gz exists. Skipping download

Warning in get.pdb(hits$pdb.id, path = "pdbs", split = TRUE, gzip = TRUE):
pdbs/8Q2B.pdb.gz exists. Skipping download
```

```
Warning in get.pdb(hits$pdb.id, path = "pdbs", split = TRUE, gzip = TRUE):
pdbs/8RJ9.pdb.gz exists. Skipping download

Warning in get.pdb(hits$pdb.id, path = "pdbs", split = TRUE, gzip = TRUE):
pdbs/6RZE.pdb.gz exists. Skipping download

Warning in get.pdb(hits$pdb.id, path = "pdbs", split = TRUE, gzip = TRUE):
pdbs/4X8H.pdb.gz exists. Skipping download

Warning in get.pdb(hits$pdb.id, path = "pdbs", split = TRUE, gzip = TRUE):
pdbs/3HPR.pdb.gz exists. Skipping download

Warning in get.pdb(hits$pdb.id, path = "pdbs", split = TRUE, gzip = TRUE):
pdbs/1E4V.pdb.gz exists. Skipping download

Warning in get.pdb(hits$pdb.id, path = "pdbs", split = TRUE, gzip = TRUE):
pdbs/5EJE.pdb.gz exists. Skipping download

Warning in get.pdb(hits$pdb.id, path = "pdbs", split = TRUE, gzip = TRUE):
pdbs/1E4Y.pdb.gz exists. Skipping download

Warning in get.pdb(hits$pdb.id, path = "pdbs", split = TRUE, gzip = TRUE):
pdbs/3X2S.pdb.gz exists. Skipping download

Warning in get.pdb(hits$pdb.id, path = "pdbs", split = TRUE, gzip = TRUE):
pdbs/6HAP.pdb.gz exists. Skipping download

Warning in get.pdb(hits$pdb.id, path = "pdbs", split = TRUE, gzip = TRUE):
pdbs/6HAM.pdb.gz exists. Skipping download

Warning in get.pdb(hits$pdb.id, path = "pdbs", split = TRUE, gzip = TRUE):
pdbs/8PVW.pdb.gz exists. Skipping download

Warning in get.pdb(hits$pdb.id, path = "pdbs", split = TRUE, gzip = TRUE):
pdbs/4K46.pdb.gz exists. Skipping download

Warning in get.pdb(hits$pdb.id, path = "pdbs", split = TRUE, gzip = TRUE):
pdbs/4NP6.pdb.gz exists. Skipping download
```

```
  |
  |                                                                      |   0%
  |
  |====                                                                  |   6%
  |
  |========                                                              |  11%
  |
  |============                                                          |  17%
  |
  |================                                                      |  22%
  |
  |===================                                                   |  28%
  |
  |=======================                                               |  33%
  |
  |===========================                                           |  39%
  |
  |===============================                                       |  44%
  |
  |===================================                                   |  50%
  |
  |=======================================                               |  56%
  |
  |===========================================                           |  61%
  |
  |===============================================                       |  67%
  |
  |===================================================                   |  72%
  |
  |======================================================                |  78%
  |
  |==========================================================            |  83%
  |
  |==============================================================        |  89%
  |
  |==================================================================    |  94%
  |
  |======================================================================| 100%
```

##Align and superpose structures

```r
pdbs <- pdbaln(files, fit = TRUE, exefile="msa")
```

```
Reading PDB files:
pdbs/split_chain/1AKE_A.pdb
pdbs/split_chain/8BQF_A.pdb
pdbs/split_chain/4X8M_A.pdb
pdbs/split_chain/6S36_A.pdb
pdbs/split_chain/8Q2B_A.pdb
pdbs/split_chain/8RJ9_A.pdb
pdbs/split_chain/6RZE_A.pdb
pdbs/split_chain/4X8H_A.pdb
pdbs/split_chain/3HPR_A.pdb
pdbs/split_chain/1E4V_A.pdb
pdbs/split_chain/5EJE_A.pdb
pdbs/split_chain/1E4Y_A.pdb
pdbs/split_chain/3X2S_A.pdb
pdbs/split_chain/6HAP_A.pdb
pdbs/split_chain/6HAM_A.pdb
pdbs/split_chain/8PVW_A.pdb
pdbs/split_chain/4K46_A.pdb
pdbs/split_chain/4NP6_A.pdb
   PDB has ALT records, taking A only, rm.alt=TRUE
.   PDB has ALT records, taking A only, rm.alt=TRUE
..   PDB has ALT records, taking A only, rm.alt=TRUE
.   PDB has ALT records, taking A only, rm.alt=TRUE
.   PDB has ALT records, taking A only, rm.alt=TRUE
.   PDB has ALT records, taking A only, rm.alt=TRUE
..   PDB has ALT records, taking A only, rm.alt=TRUE
..   PDB has ALT records, taking A only, rm.alt=TRUE
....   PDB has ALT records, taking A only, rm.alt=TRUE
.   PDB has ALT records, taking A only, rm.alt=TRUE
.   PDB has ALT records, taking A only, rm.alt=TRUE
..

Extracting sequences

pdb/seq: 1   name: pdbs/split_chain/1AKE_A.pdb
   PDB has ALT records, taking A only, rm.alt=TRUE
pdb/seq: 2   name: pdbs/split_chain/8BQF_A.pdb
   PDB has ALT records, taking A only, rm.alt=TRUE
pdb/seq: 3   name: pdbs/split_chain/4X8M_A.pdb
pdb/seq: 4   name: pdbs/split_chain/6S36_A.pdb
```

```
   PDB has ALT records, taking A only, rm.alt=TRUE
pdb/seq: 5   name: pdbs/split_chain/8Q2B_A.pdb
   PDB has ALT records, taking A only, rm.alt=TRUE
pdb/seq: 6   name: pdbs/split_chain/8RJ9_A.pdb
   PDB has ALT records, taking A only, rm.alt=TRUE
pdb/seq: 7   name: pdbs/split_chain/6RZE_A.pdb
   PDB has ALT records, taking A only, rm.alt=TRUE
pdb/seq: 8   name: pdbs/split_chain/4X8H_A.pdb
pdb/seq: 9   name: pdbs/split_chain/3HPR_A.pdb
   PDB has ALT records, taking A only, rm.alt=TRUE
pdb/seq: 10   name: pdbs/split_chain/1E4V_A.pdb
pdb/seq: 11   name: pdbs/split_chain/5EJE_A.pdb
   PDB has ALT records, taking A only, rm.alt=TRUE
pdb/seq: 12   name: pdbs/split_chain/1E4Y_A.pdb
pdb/seq: 13   name: pdbs/split_chain/3X2S_A.pdb
pdb/seq: 14   name: pdbs/split_chain/6HAP_A.pdb
pdb/seq: 15   name: pdbs/split_chain/6HAM_A.pdb
   PDB has ALT records, taking A only, rm.alt=TRUE
pdb/seq: 16   name: pdbs/split_chain/8PVW_A.pdb
   PDB has ALT records, taking A only, rm.alt=TRUE
pdb/seq: 17   name: pdbs/split_chain/4K46_A.pdb
   PDB has ALT records, taking A only, rm.alt=TRUE
pdb/seq: 18   name: pdbs/split_chain/4NP6_A.pdb
```

```r
# Vector containing PDB codes for figure axis
ids <- basename.pdb(pdbs$id)

# Draw schematic alignment
# Kept getting an error that the figure margins were too large, every time i tried to knit th
```

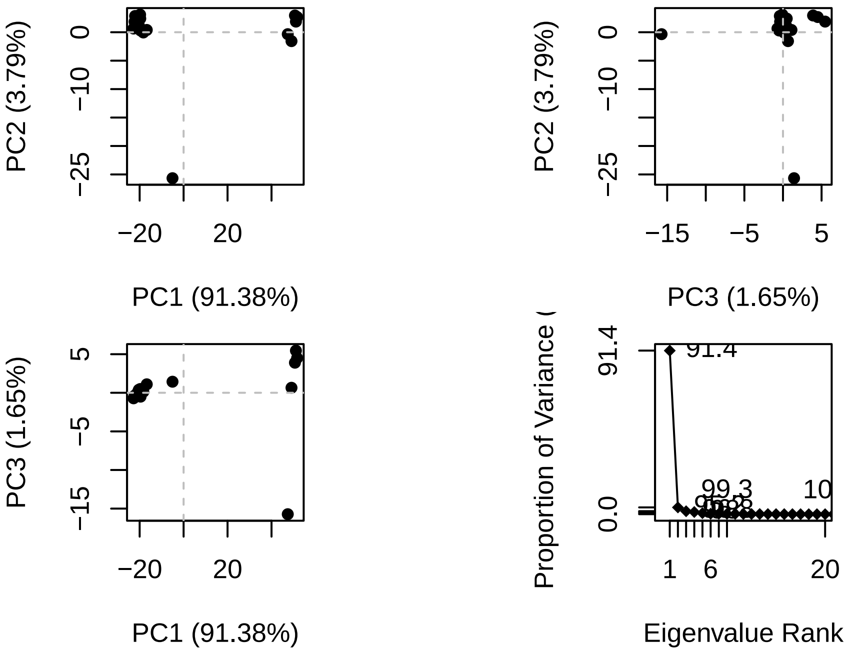
Annotate collected PDB structures

```r
anno <- pdb.annotate(ids)
unique(anno$source)
```

```
[1] "Escherichia coli"
[2] "Escherichia coli K-12"
[3] "Escherichia coli O139:H28 str. E24377A"
[4] "Escherichia coli str. K-12 substr. MDS42"
[5] "Photobacterium profundum"
[6] "Vibrio cholerae O1 biovar El Tor str. N16961"
```

Principal component analysis

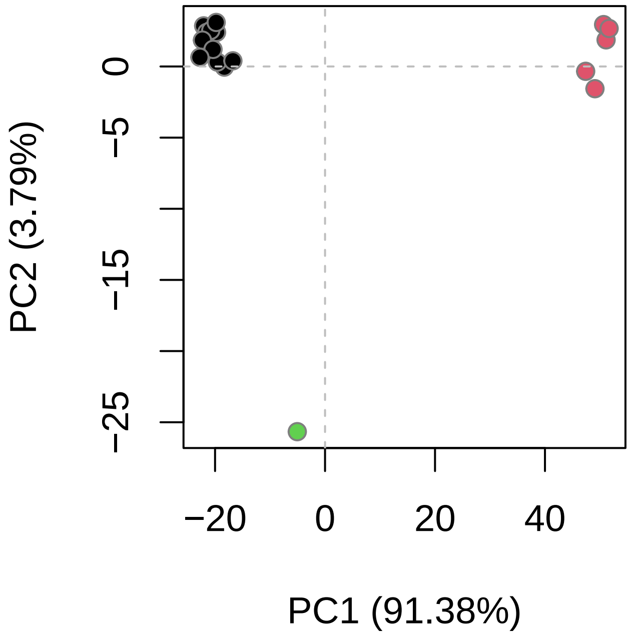
```
pc.xray <- pca(pdbs)
plot(pc.xray)
```



```
# Calculate RMSD
rd <- rmsd(pdbs)
```

```
Warning in rmsd(pdbs): No indices provided, using the 182 non NA positions
```

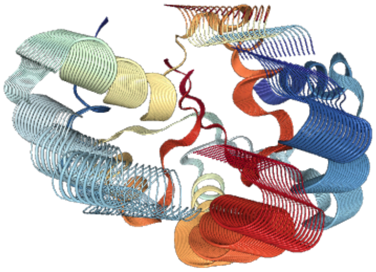
```
# Structure-based clustering
hc.rd <- hclust(dist(rd))
grps.rd <- cutree(hc.rd, k=3)

plot(pc.xray, 1:2, col="grey50", bg=grps.rd, pch=21, cex=1)
```



Optional further visualization

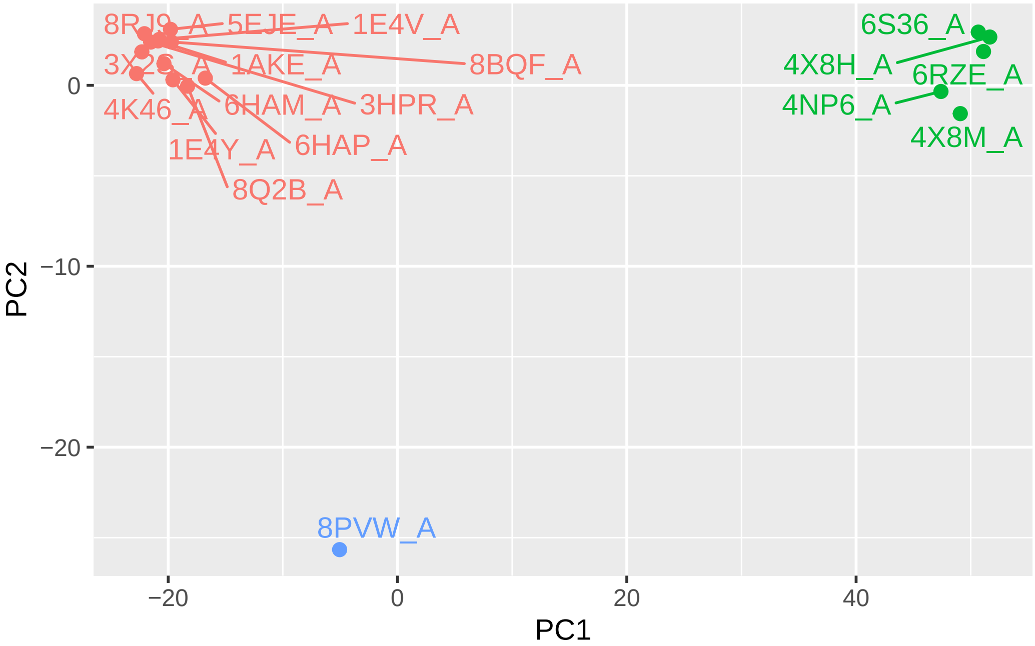
```
# Visualize first principal component
pc1 <- mktrj(pc.xray, pc=1, file="pc_1.pdb")
view.pca(pc.xray)
```

```
file:////private/var/folders/vr/dj7chnl94lv5g8p28r0c72b00000gn/T/Rtmp7fh4SE/file9ae346224b8/
```



PCA visualization with ggplot

```
library(ggplot2)
library(ggrepel)

df <- data.frame(PC1=pc.xray$z[,1],
                 PC2=pc.xray$z[,2],
                 col=as.factor(grps.rd),
                 ids=ids)

p <- ggplot(df) +
  aes(PC1, PC2, col=col, label=ids) +
  geom_point(size=2) +
  geom_text_repel(max.overlaps = 20) +
  theme(legend.position = "none")
p
```



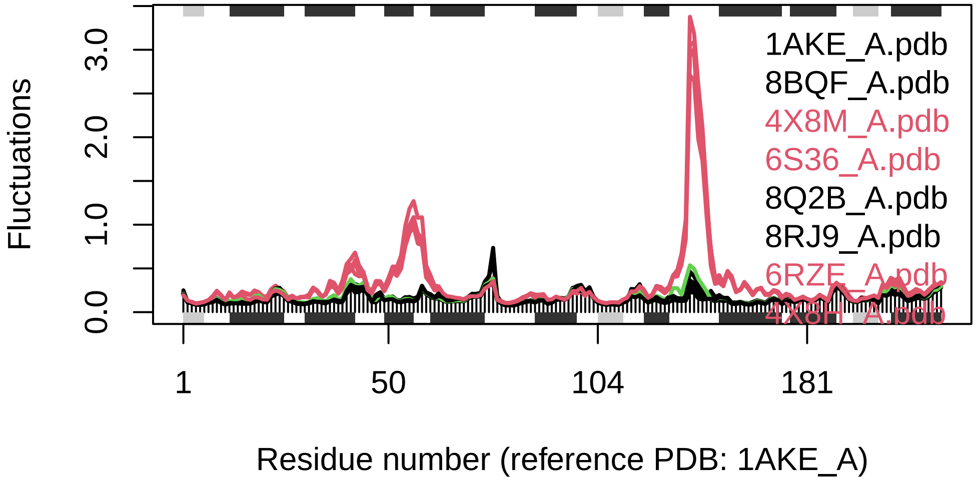
Normal mode analysis

```
modes <- nma(pdbs)
```

```
Warning in nma.pdbs(pdbs): 8BQF_A.pdb might have missing residue(s) in structure:
   Fluctuations at neighboring positions may be affected.


Details of Scheduled Calculation:
  ... 18 input structures
  ... storing 540 eigenvectors for each structure
  ... dimension of x$U.subspace: ( 546x540x18 )
  ... coordinate superposition prior to NM calculation
  ... aligned eigenvectors (gap containing positions removed)
  ... estimated memory usage of final 'eNMA' object: 40.6 Mb


  |
  |                                                                      |   0%
  |
  |====                                                                  |   6%
  |
  |========                                                              |  11%
  |
  |============                                                          |  17%
  |
  |================                                                      |  22%
  |
  |===================                                                   |  28%
  |
  |=======================                                               |  33%
  |
  |===========================                                           |  39%
  |
  |===============================                                       |  44%
  |
  |===================================                                   |  50%
  |
  |=======================================                               |  56%
  |
  |===========================================                           |  61%
  |
  |===============================================                       |  67%
  |
  |===================================================                   |  72%
  |
  |=======================================================               |  78%
  |
  |==========================================================            |  83%
  |
  |==============================================================        |  89%
  |
  |==================================================================    |  94%
  |
  |======================================================================| 100%
```

```
plot(modes, pdbs, col=grps.rd)
```

```
Extracting SSE from pdbs$sse attribute
```



> Q14. What do you note about this plot? Are the black and colored lines similar or different? Where do you think they differ most and why?

The lines are mostly similar except for a few key regions. I think this is due to the regions in which the protein undergoes a conformational change when in the pressence of a ligand.