Analysis, Comparison of Proteins

Andrew Torda, wintersemester 2008 / 2009, GST

From previous lectures

- we know about protein structures / coordinates
- we know how coordinates are collected

What kind of analysis would we like to do ?

- recognising common features
- classifying
  - (useful for prediction)

Philosophy

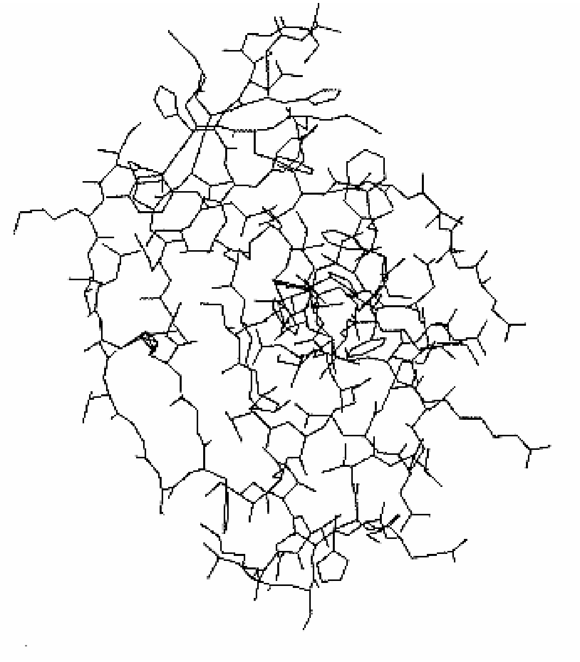
- ways to measure similarity between structures
- ways to find similar pieces / "motif"s
- most common motif ? Secondary structures

Next few weeks

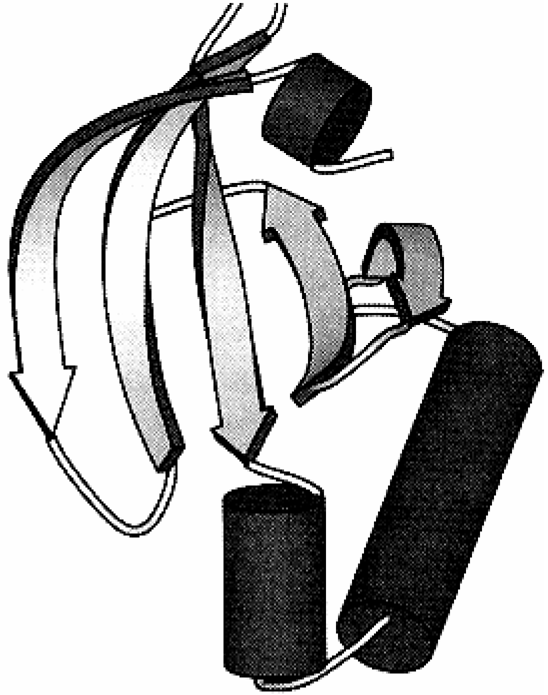
- Secondary structure definitions
- Classifying protein structures
- Domains
- Supersecondary structure
- Protein similarity – sequence versus structure
- Sequence space
- Classifications – hierarchical
- Classifications – other
- Comparison of proteins

- touching on evolution, alignments, …

Secondary Structure Recognition



from coordinates



assumes structures recognised

- how to define / recognise secondary structure ?

pictures from "Structural Bioinformatics", ed Bourne, PE and Weissig, H.

Defining Secondary Structure

- What do I want ?

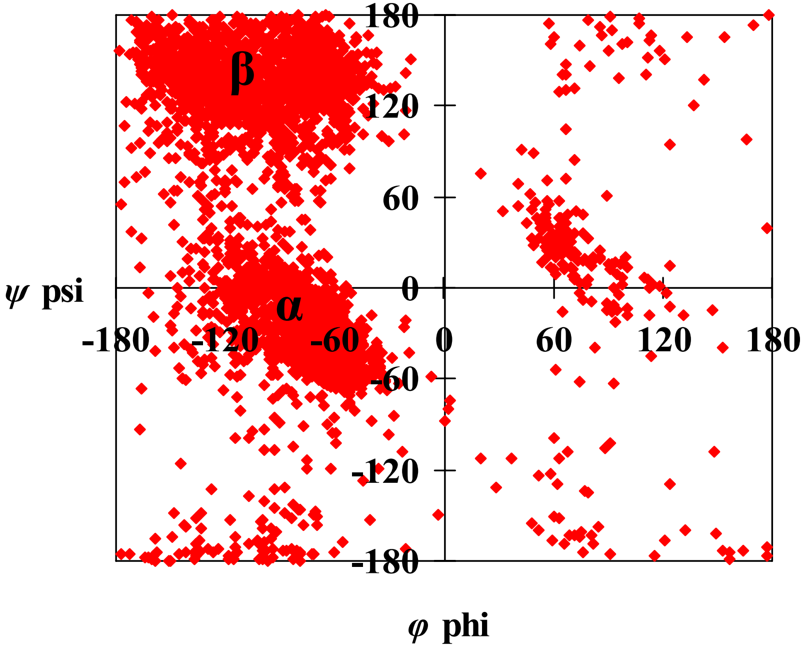
	1	2	3	4	5	6	7	8	9	10	…
residue	A	C	A	D	L	V	A	W	W	A	…
sec struct	-	H	H	H	H	H	-	E	E	E	…

- at each residue, label as to secondary structure type
  - no ambiguity
  - labels at residues – not between !
- I do not want probabilistic answers (more soon)
- remember not all residues are in recognisable α-helix or β-sheet

Secondary Structure From Coordinates

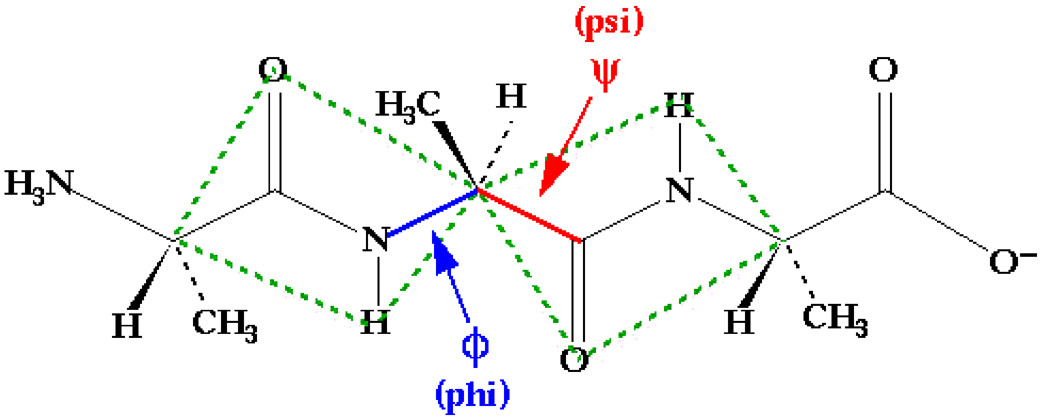
Start with α-helices

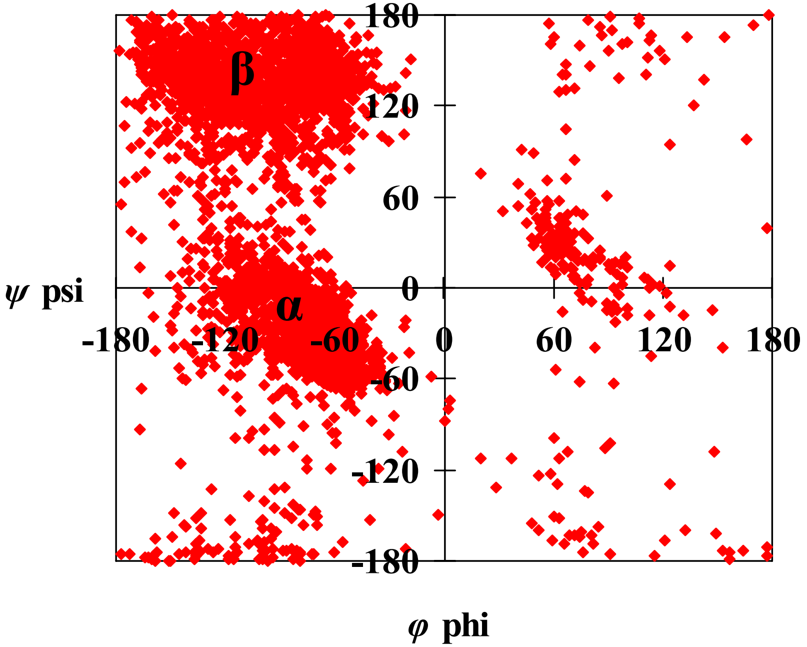
- what do we know
  - look like helices
  - 3.6 residues per turn
  - H-bond pattern
    - N residue *i* to *i*+4
  - residue backbone angles



Using Backbone Angles

Given coordinates, easy to calculate





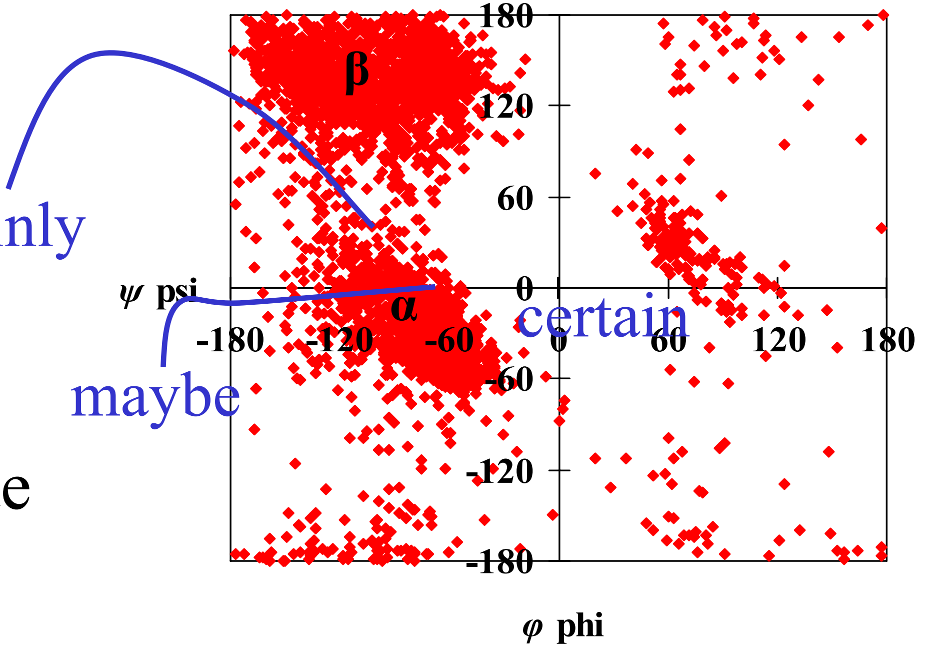
Problems

- what are my thresholds ?
- what if I see one residue with angle ?

Problems With Using Angles

- thresholds

- minimum number of residues
  - what if I see only one residue with perfect angles ?
  - not forming H bonds
  - need 3 or 4 residues



certainly not

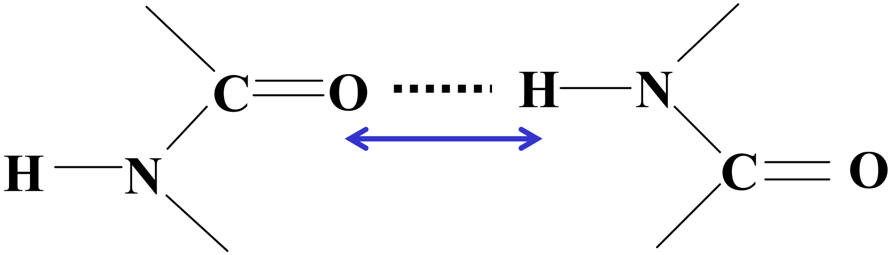
maybe

certain

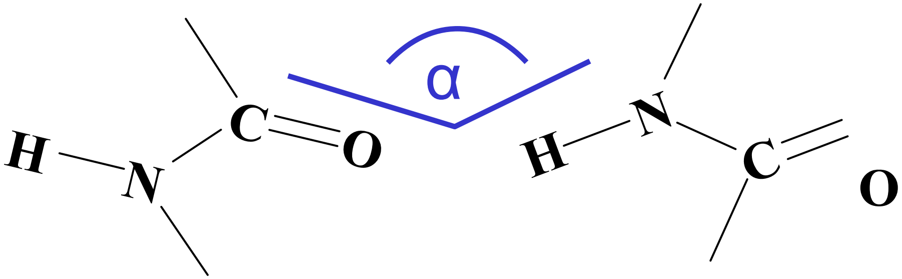
Maybe We Should Use H-Bonds

We have the coordinates

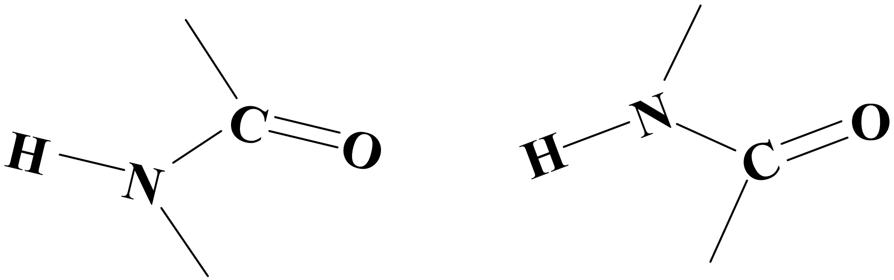
- should be easy to recognise all H bonds
- criteria ?
- distance r(ON) < ≈ 3.6 Å



- angle ? (≈ 120˚)



A practical definition



$$E = 332\, q_1 q_2 \left({}^{1}\!/\!_{r_{\mathrm{ON}}} + {}^{1}\!/\!_{r_{\mathrm{CH}}} - {}^{1}\!/\!_{r_{\mathrm{OH}}} - {}^{1}\!/\!_{r_{\mathrm{CN}}} \right)$$

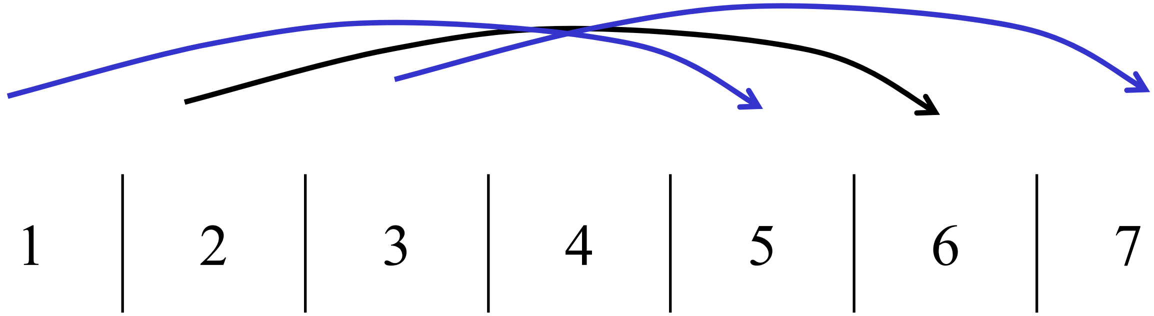
- require E < -3 (arbitrary !)
- note as r grows, E goes to 0

Kabsch, W. & Sander, C, Biopolymers, 22, 2577-2637, "Dictionary of Protein Secondary Structure…" (1983)

Problems with short helices

What if I see only 3 or 4 residues ?

- real helix has 2 H-bonds per residue
- what if I see one ?



Compromise

- call this a turn (only has one H bond)
- α-helix definition
  - at least two consecutive (4 residue) turns

Useful definitions (α-helix)

- recognise an H-bond $i,i+4$ (either
  - r_{ON} + angle α or
  - general distance formula

  = turn
- two successive turns

  = minimal α-helix
- more overlapping helices

  = longer helix

- all we have done is an α-helix

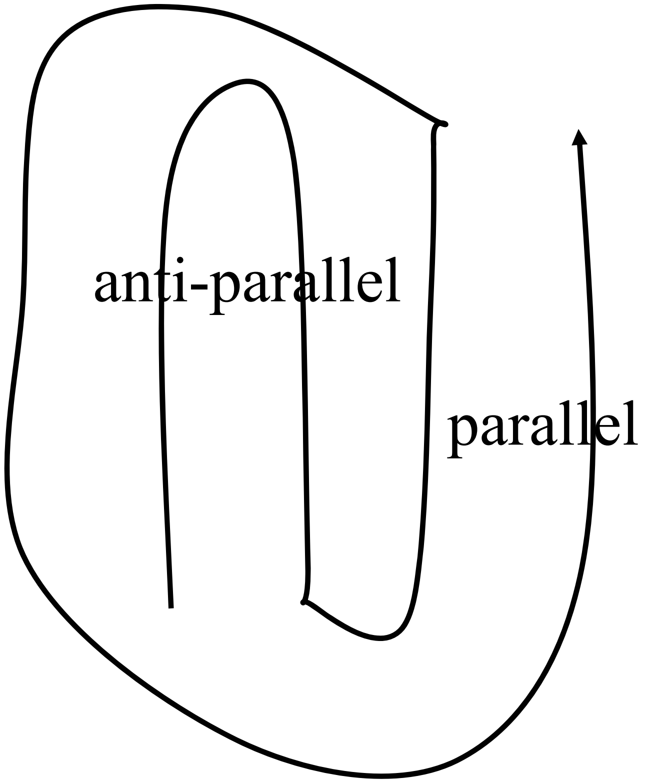
a β-strand / sheet

- much more difficult
  - parallel versus anti-parallel
  - H-bond neighbours not known
    - 5-109
    - 6-110
    - 7-111 … parallel

    or

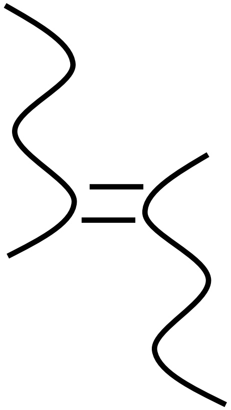
    - 5-109
    - 6-108
    - 7-107…anti-parallel

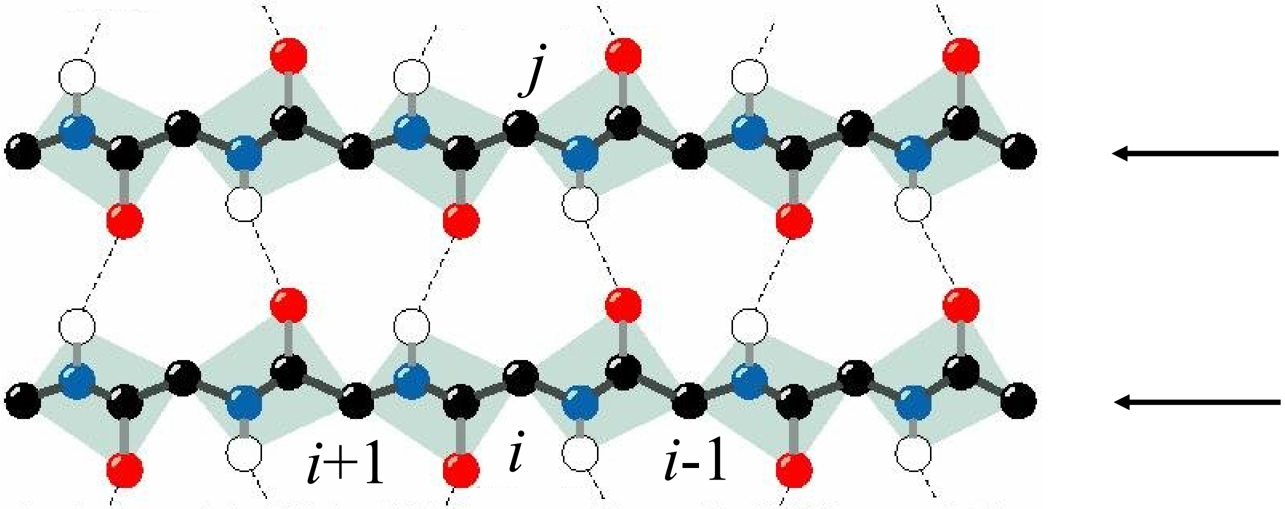
- formalise this



Defining a β-sheet

- start with a bridge
- parallel bridge
  - H-bond (*i*-1, *j*) & H-bond (*j*, *i*+1) or
  - H-bond (*j*-1, *i*) & H-bond (*i*, *j*+1)





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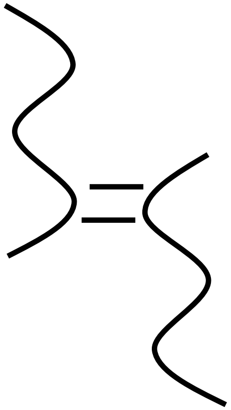
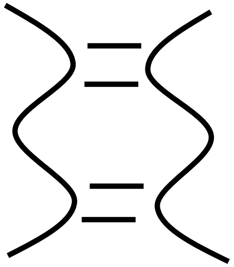


- ladder = one or more consecutive bridges
- sheet = one or more consecutive ladders with shared residues
- similar definition for anti-parallel sheets

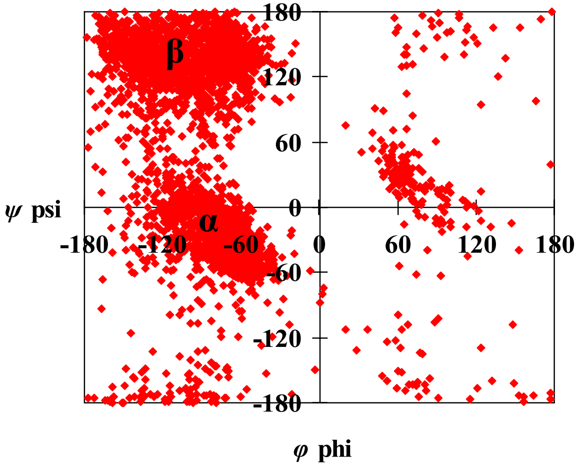


Are these problems real ?

Do thresholds matter ?

- do programs give the same answer ?
- if we use secondary structure for comparisons of proteins

- comparisons with experiment



Can we set perfect thresholds ?

- not all H-bonds are the same
- look at *ψ*-*φ* map, borders are not clear
  - mobility, finite energy
- coordinates have experimental error
- our programs generate worse coordinates (holes, distortions)

From secondary to higher levels
Classification

- Why classify proteins ?
- Why recognise similarities
  - function prediction
  - structure prediction
    - vague idea of structure for mutagenesis, applications
- Why might this be useful ?
  - how many structures are there ?

How Many Protein Structures Are There ?

- Protein Databank ≈ 5.5×10^4
- 90 % sequence similarity ≈ 1.7×10^4
- different shapes 2 to 5×10^3
- implications for structure prediction ?
    - how many possible structures can we think of ?
        - exponential
    - how big is the real search space ?
        - really 10^3 to 10^4

Why So Few Structures

- discretization of space (makes it look smaller)
- physical reasons
  - compactness, stability
  - advantages of H-bonded conformations
- history / evolution
  - imagine all proteins evolve from some original molecule
  - evidence
    - theoretical – geometric constructions
    - chemical – construction of artificial protein(s)

Before Classifying

- earlier description of structure
- primary (sequence)
- secondary (α-helices, β-sheets, …)
  - supersecondary ?
- tertiary
  - arrangement of helices / sheets or
  - where atoms are in space
- quaternary…

- we need idea of domains, then supersecondary structures

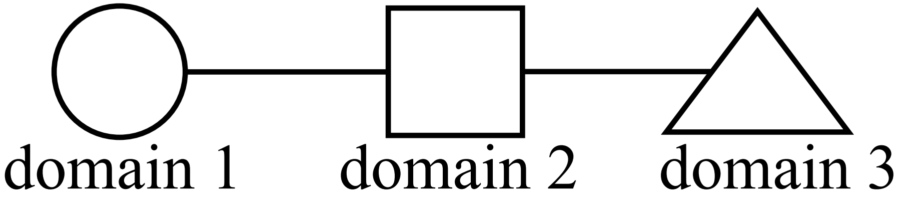
Domains in Biochemistry

History / biochemistry / no structures

- invented story
- we have a big protein
  - catalyses A → B
  - C regulates it
- cleave protein (break with enzyme) to two parts
  - 1 still converts A →B
  - 2 binds C
  - interpretation
    - catalytic domain
    - C binding domain
- more generally
  - different pieces of protein, responsible for different functions

Domain Concept Useful ?

- Many times a whole protein cannot be crystallised, solved by NMR
- attack protein with enzymes to break up
- look for activity in pieces, solve structures of pieces

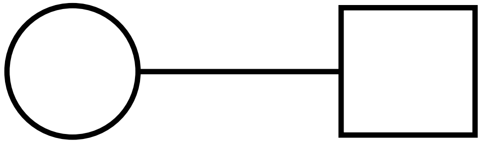


} each domain 50 to 250 residues

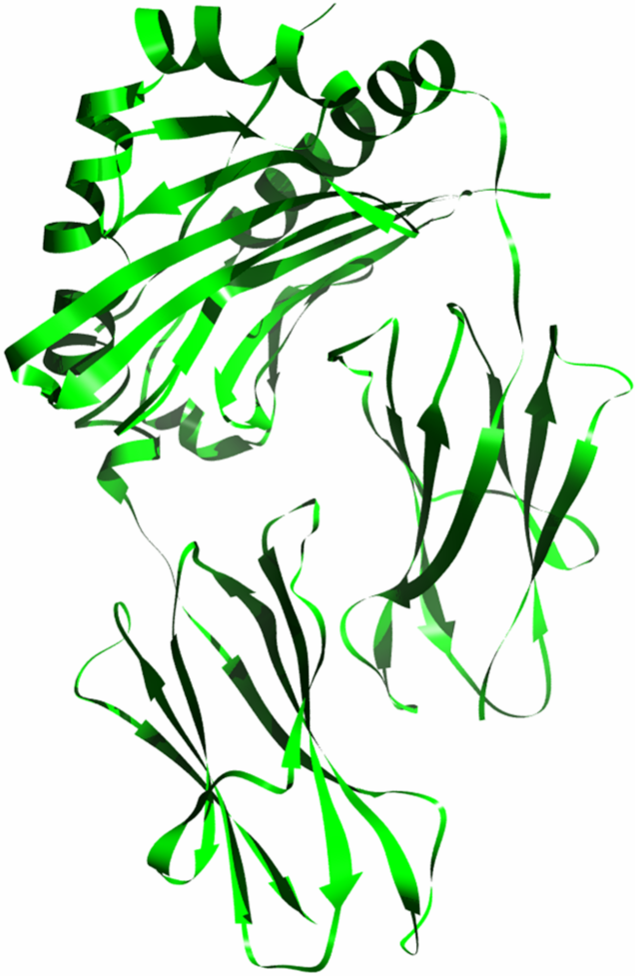
- literature / PDB full of "xxx domain of yyy"
- attractive ?
  - makes big proteins seem manageable
  - building block concept
    - attractive in evolutionary terms

Domains in Structures

- Many structures solved look like…



- histocompatibility module (1iak)
  - 3 domains + another protein



- are they always so clear ?
- porphobilinogen deaminase (1gtk)

Domain definition version 3

Three reasonable definitions

- biochemistry
- structures
- look for conserved units in sequence comparisons

Domains for today

- compact structural units

Domains for classification

- structural classifications often domain based

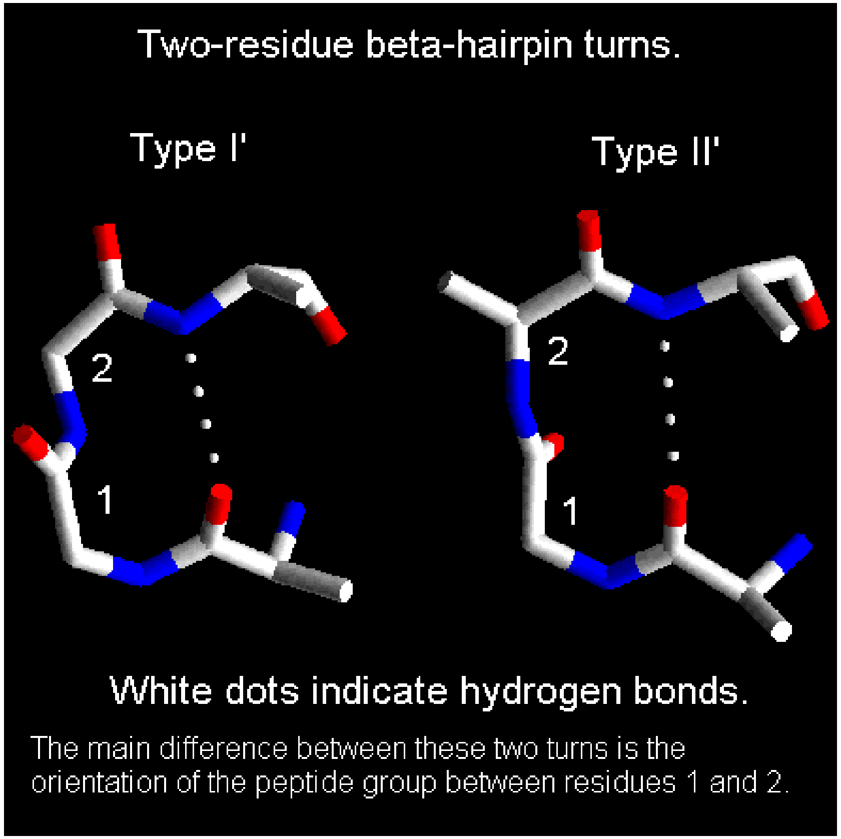
Classifications In General

- 1. secondary structure
  - we see collections of residues and classify into recognisable types
- 2. different types of domain
  - soon

- 1b. supersecondary elements ?
  - are there some common small arrangements of α-helices, β-sheets ?

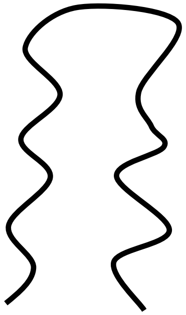
A Supersecondary Structure

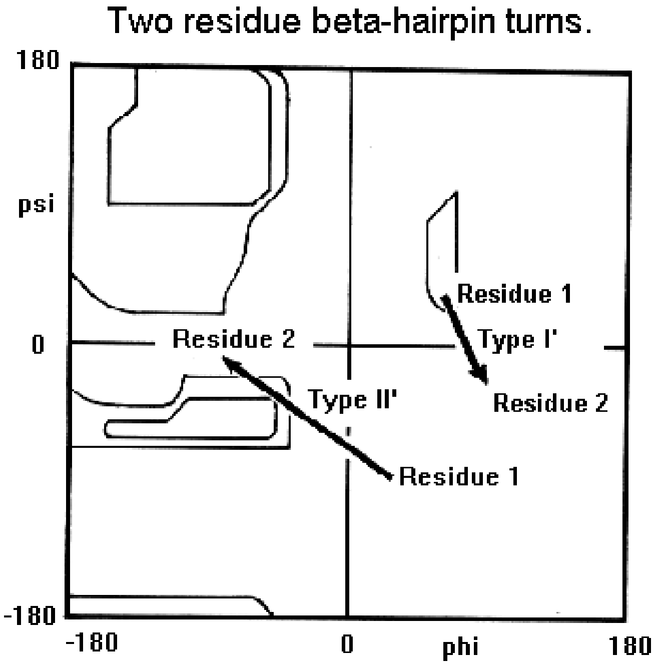
- β-hairpin (β-turn-β) fits idea of common motif
  - described as built on secondary structure + specific H-bonds


Two-residue beta-hairpin turns.

White dots indicate hydrogen bonds.

The main difference between these two turns is the orientation of the peptide group between residues 1 and 2.




Two residue beta-hairpin turns.

http://pps.life.nthu.edu.tw/PPS2/course/section9/sss/super1.html

More Supersecondary Structures

- helix-turn-helix (α-X-α)
  - DNA binding proteins
- helix-longer_loop-helix
  - Ca^{++} binding
- …

Who cares ?

- repeated patterns / motifs suggests there are smaller number of structural units to recognise
- modularity appeals
- functional association
- conforms to some ideas on protein folding (more next semester)

Why I Do Not Like Supersecondary Structure

- ideal picture…
  - primary structure arrangement →
  - secondary structure / arrangement →
  - supersecondary structure →
  - tertiary structure or domains
- implies supersecondary structure is useful hierarchical element
  - not really used !

Sequence vs Structural Similarity

Background

- in the real world we usually have sequence information first
- want to make guesses about protein structure

I have two aligned protein sequences

- are they structurally similar ?

Old rule

- > 25 % sequence similar – similar structures
- < 20 % cannot tell
- 20 % < x < 25 % - "twilight zone"

Is this universally valid ?

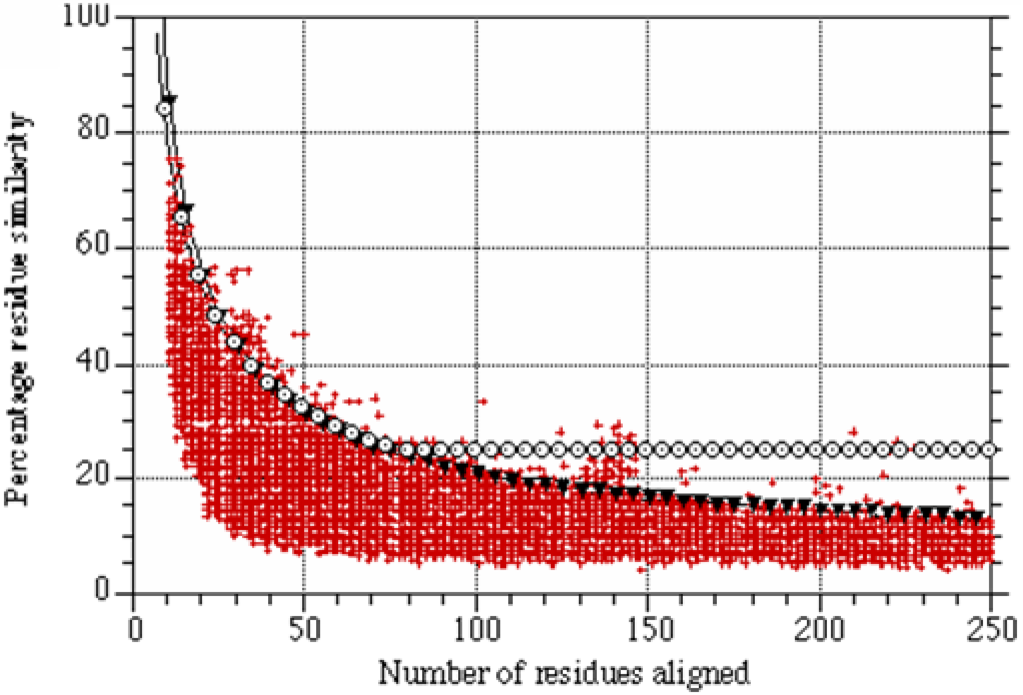
http://cubic.bioc.columbia.edu/papers/1999_twilight/paper.html

Sequence Similarity → Structure

Take a set of pairs of proteins

- find those which are not structurally similar
- look at sequence similarity

- old rule is not valid
- 50 residues
  - > 30 % seq
- 150 residues
  - > 20 %



- rule:
  - sequence similarity (length dependent) very good indicator of structural similarity

http://cubic.bioc.columbia.edu/papers/1999_twilight/paper.html

Using Sequence Similarity

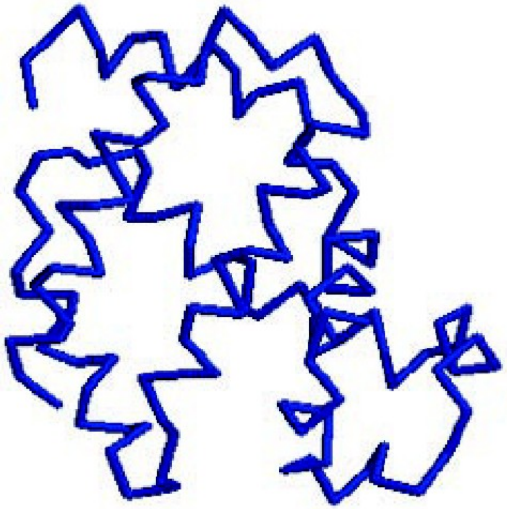
- consequence
  - I could try to categorise proteins based on sequence
- tools
  - any alignment program (blast, fasta, clustal, …)
- method
  - survey all proteins in the protein databank
  - collect all pairs > x % (or use more sophisticated threshold)
- result (jan 2009)

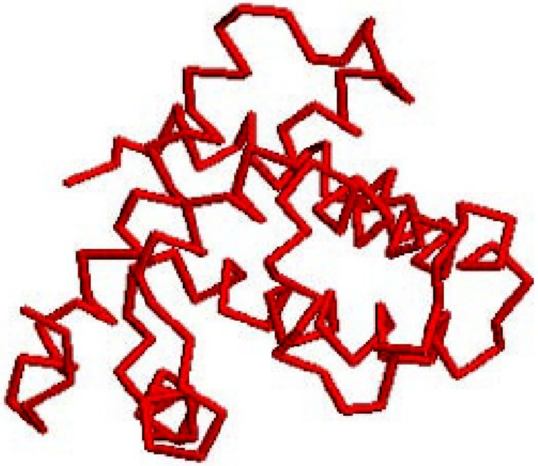
similarity	num clusters
90 %	20 002
70%	17 490
50%	14 906

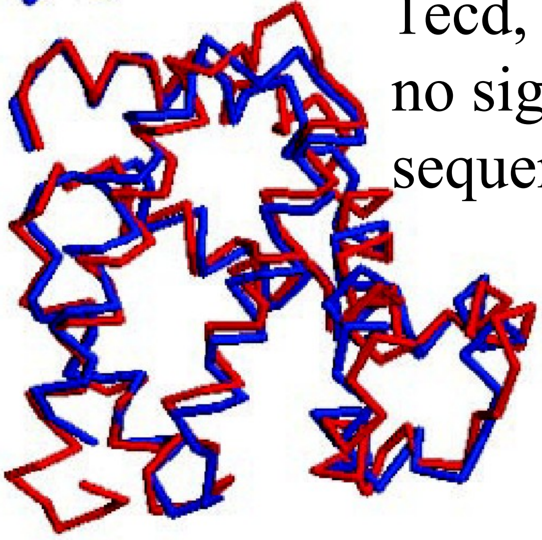
- much more than 2 to 5 × 10^3 ?
- maybe some of my classes are not really different

Sequences Not Similar

- Sequences similar ? .. similar structure
- Sequences different ?
  - ??
- Example
  - 100's examples







1ecd, 1mbd
no significant
sequence identity

Blankenbecler, Ohlsson, Peterson, Ringne PNAS (2003)100, 11936–11940

An Example Family

- example, neighbours of 1cun chain A
  - look at sequence identity (%id)
  - alignment length (lali = number of residues)
  - root mean square diff in Å

```
No Chain    %id lali rmsd  Description
 1 1cunA    100  213  0.0  ALPHA SPECTRIN
 2 1hciA     24  111  1.6  ALPHA-ACTININ 2
 3 1ek8A     12  106  4.4  RIBOSOME RECYCLING FACTOR
 4 1oxzA      9   91  2.5  ADP-RIBOSYLATION FACTOR BINDING PROTEIN GGA1
 5 1eh1A      8  102  4.6  RIBOSOME RECYCLING FACTOR
 6 1hx1B      5  105  3.1  HEAT SHOCK COGNATE 71 KDA
 7 1dd5A      8  103  4.7  RIBOSOME RECYCLING FACTOR
 8 1lvfA      9   98  2.6  SYNTAXIN 6
 9 1bg1A      9   99  2.3  STAT3B
10 1hg5A      5   98  3.0  CLATHRIN ASSEMBLY PROTEIN SHORT FORM
11 1hs7A     14   92  2.5  SYNTAXIN VAM3
12 1dn1B     10  101  2.7  SYNTAXIN BINDING PROTEIN 1
13 1ge9A      6  108  4.6  RIBOSOME RECYCLING FACTOR
14 1fewA      8  125  3.5  SECOND MITOCHONDRIA-DERIVED ACTIVATOR OF
15 1qsdA      4   90  2.4  BETA-TUBULIN BINDING POST-CHAPERONIN COFACTOR
16 1e2aA      6   95  2.8  ENZYME IIA
17 1i1iP      7   95  3.3  NEUROLYSIN
18 1fioA      8  100  2.6  SSO1 PROTEIN
19 1m62A      8   81  2.8  BAG-FAMILY MOLECULAR CHAPERONE REGULATOR-4
20 1k4tA      6  147 25.8  DNA TOPOISOMERASE I
```

http://ekhidna.biocenter.helsinki.fi/dali/start

DIVERSION Sequence Space

- convenient way to explain ideas of sequence similarity
- conventional spaces
  - 1D (x), 2D (x, y), 3D (x, y, z), 4D (x, y, z, w), …
  - let us estimate how big a space or problem is
  - how many variables do I have ? (a, b, c, …)
  - how many values can each variable have ?
    - a 3 values, b 4 values, c 5
    - number of points in space = 3 × 4 × 5
- protein sequences
  - each position can have 1 of 20 values
  - total number of sequences = 20 × 20 ×…= 20^{Nres}
  - like a space of N_{res} dimensions

Representing a Sequence

- protein sequence and structural coordinates

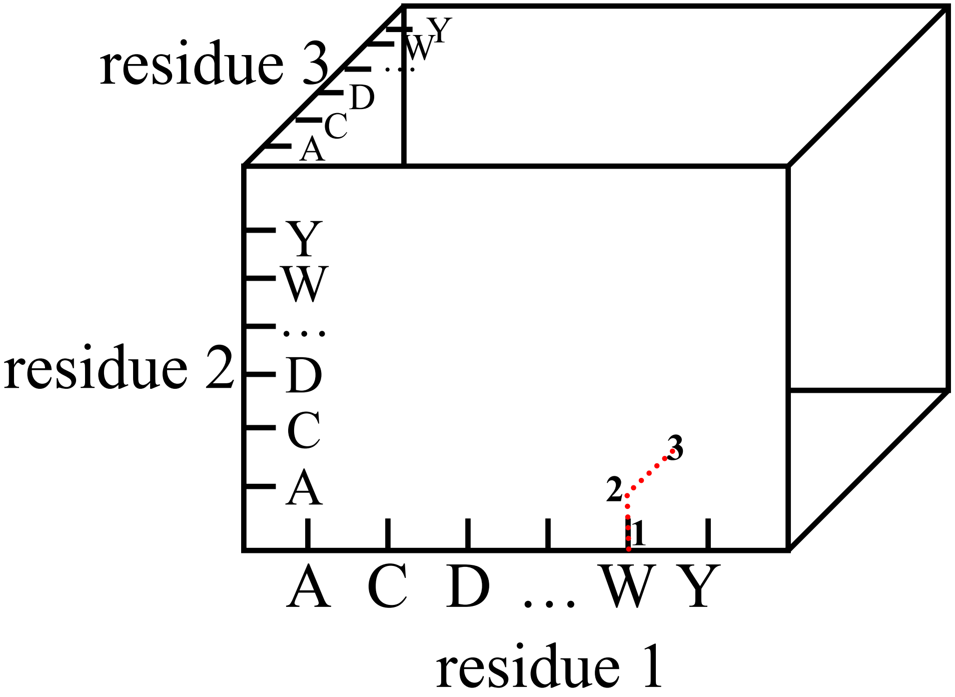
	1	2	3	4	5	6	7	…	N_{res}
x	1.2	2.3	…						10.3
y	2.4	3.5	…						11.1
z	1.7	2.9	…						15.5
seq	W	A	C	A	A	…			D

- consider the first three residues
  - WAC (for pictures only)

Finding a Sequence in This Space

- real diagram is a box of N_{res} dimensions
  - this one 3 dimensions

	1	2	3	4	5	6	7	…	N_{res}
x	1.2	2.3	…						10.3
y	2.4	3.5	…						11.1
z	1.7	2.9	…						15.5
seq	W	A	C	A	A	…			D

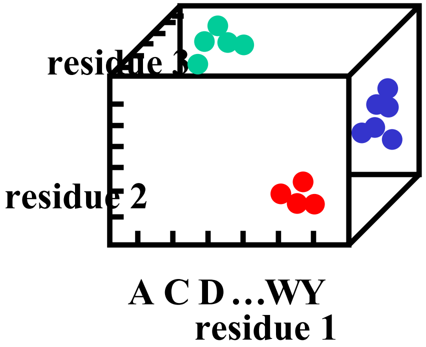


- looking for sequences…

Families in Sequence Space

- Similar sequences should land near each other

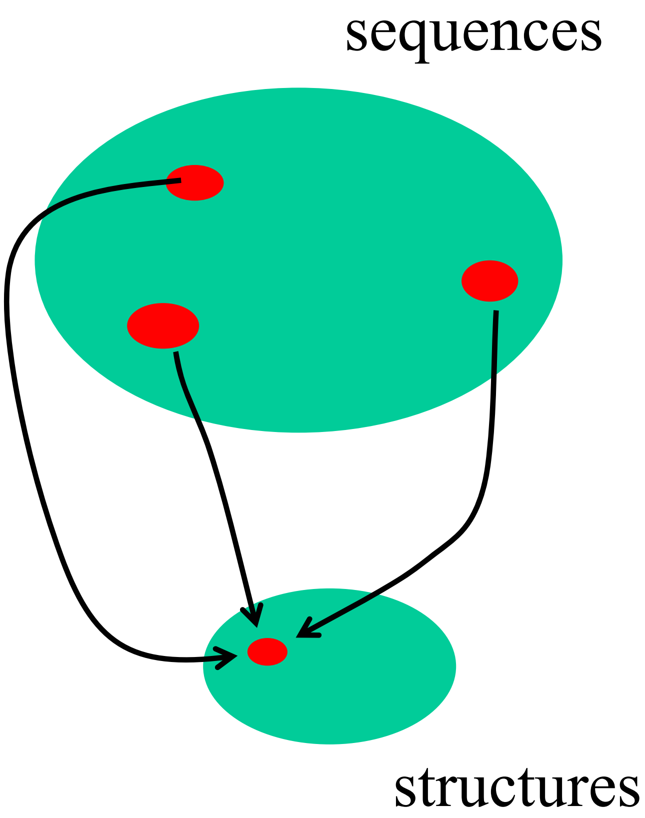
- How realistic ?
  - picture is a simplification
  - only works for $N_{seq1} = N_{seq2}$
  - very useful
    - distances between sequences



- Will return next semester

Structure vs Sequence

- there are 1000's of such families
- summarise
  - similar sequences
    - similar structures
  - very different sequences
    - similar or different structures
- why ?



Structures < Sequences… Why ?

Evolution 1

- many small changes
- if structure changes, function breaks, you die
- sequences change as much as possible within this constraint

Evolution 2

- maybe some cases of convergent evolution
- impossible to prove

Consequences of sequence based categorisation

- we will have different classes, but really same protein shape

Surprising ?

- consider near universal proteins
    - 100's millions years evolution, function largely preserved
- chemistry
    - sequence does determine structure, many sequences could fit structure

Back to Classification

Sequence classification

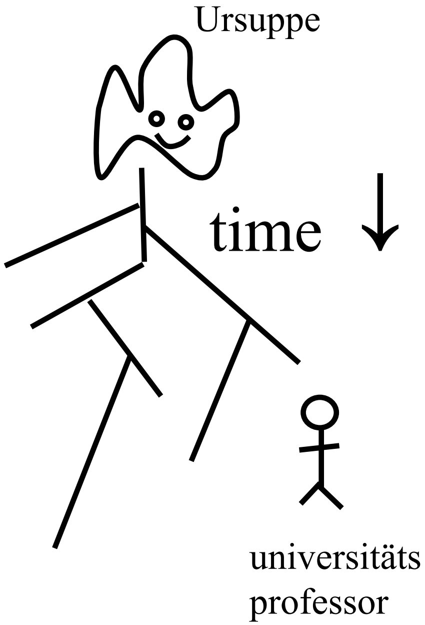
- good, reliable, similar class = similar structure
- not enough to find all similarities
- need for structure based methods

Philosophies

1. evolution
2. just classify proteins

Evolution

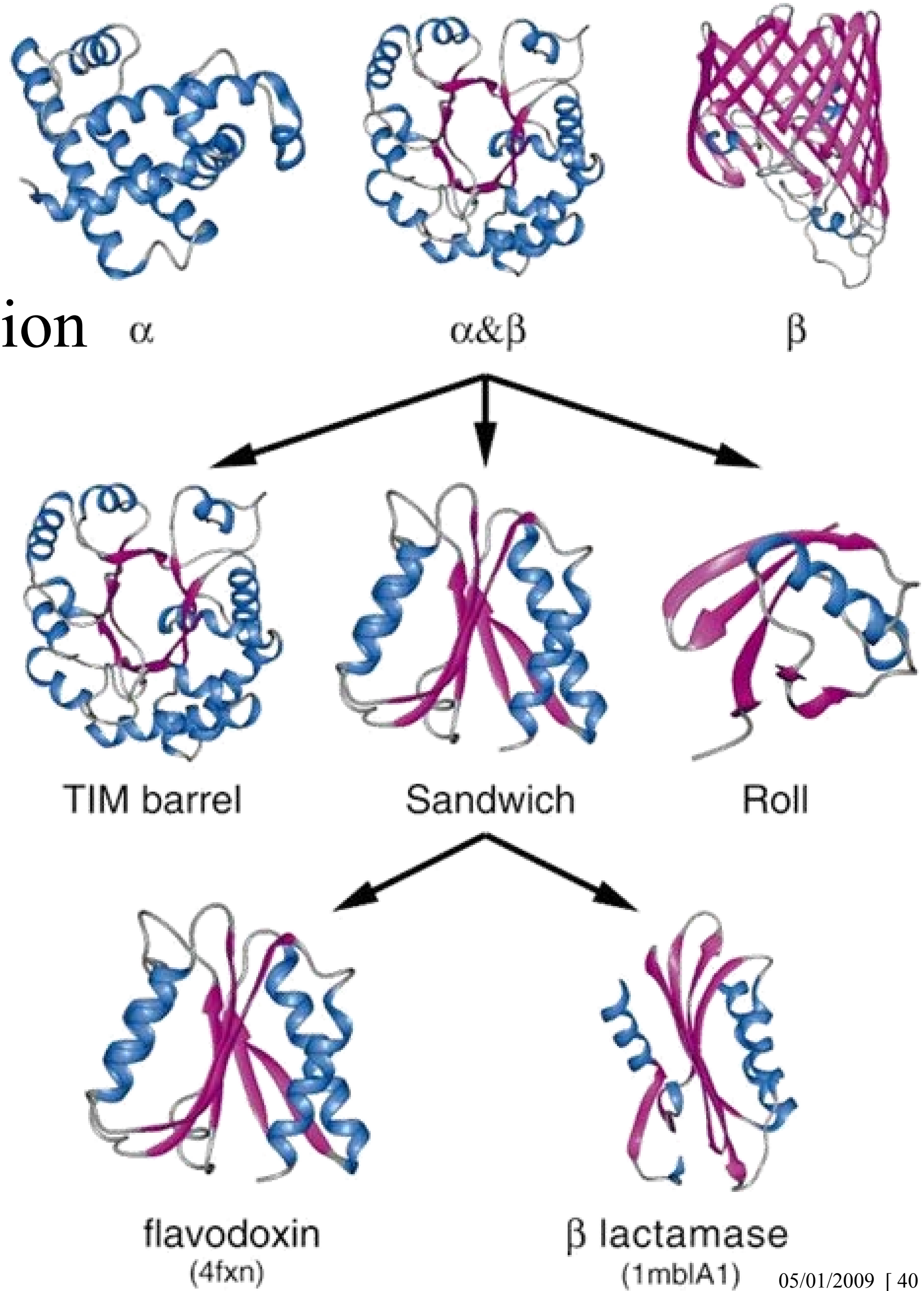
- diagram →
- we expect a hierarchy



Imposing a Hierarchy on Proteins

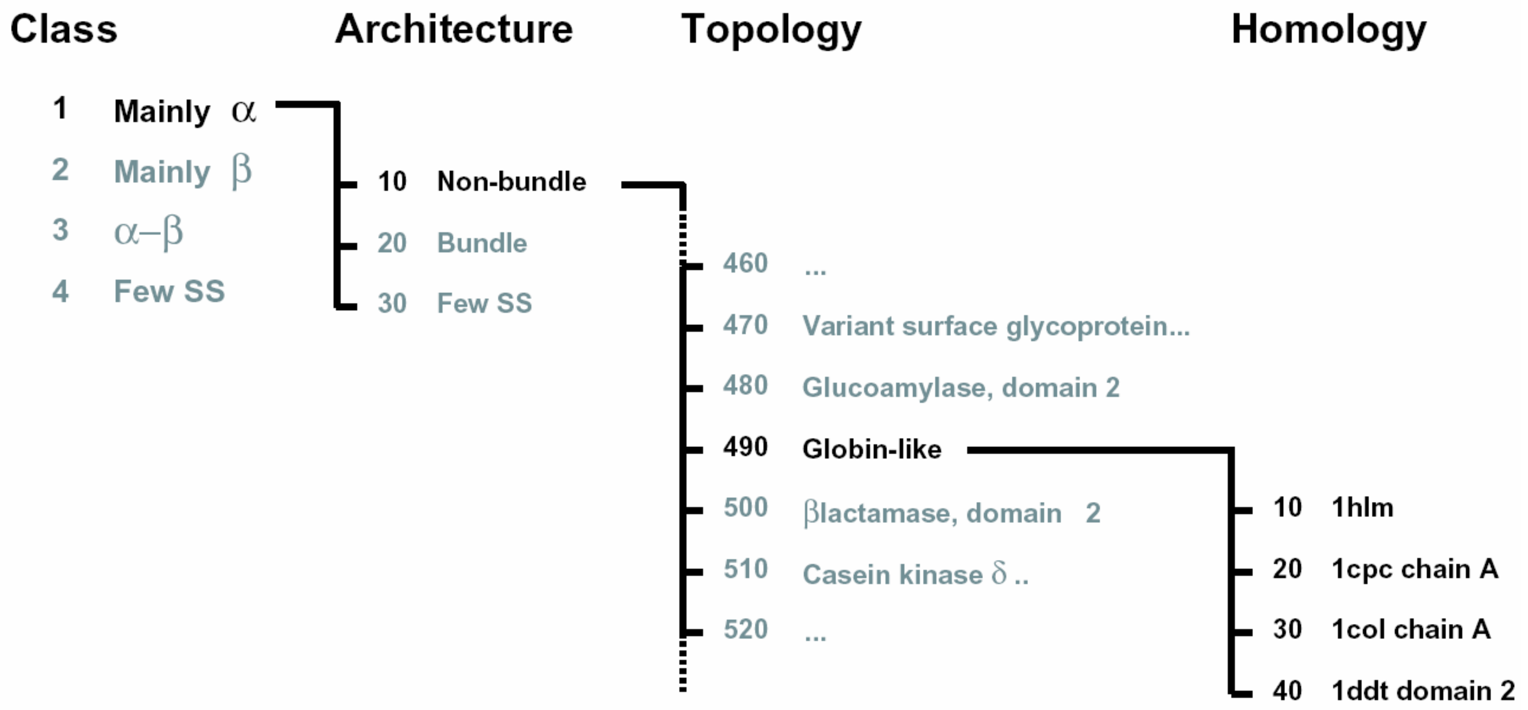
- parts may correspond to evolution
- top level ?

- How useful and applicable ?
  - examples



CA Orengo AD Michie, S Jones,DT Jones, MB Swindells,JM Thornton, Structure, 1997, 5,1093–1108

Example from "CATH"



Class

1 Mainly α

2 Mainly β

3 α–β

4 Few SS

Architecture

10 Non-bundle

20 Bundle

30 Few SS

Topology

460 ...

470 Variant surface glycoprotein...

480 Glucoamylase, domain 2

490 Globin-like

500 βlactamase, domain 2

510 Casein kinase δ ..

520 ...

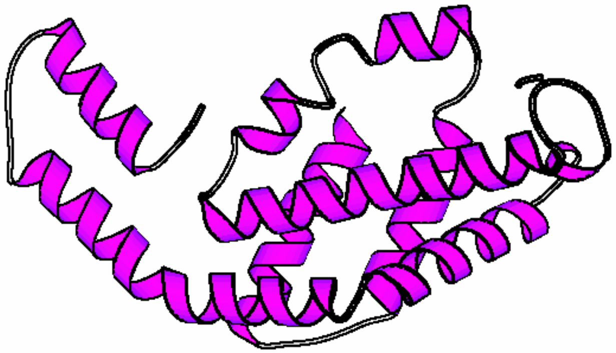
Homology

10 1hlm

20 1cpc chain A

30 1col chain A

40 1ddt domain 2



1.10.490.20

Mainly α.Non-bundle.Globin-like.1cpc chain A

CA Orengo AD Michie, S Jones,DT Jones, MB Swindells,JM Thornton, Structure, 1997, 5,1093–1108

Lots of families

α-helix bundles ?

- ≈226 domains, 3 % surveyed structures

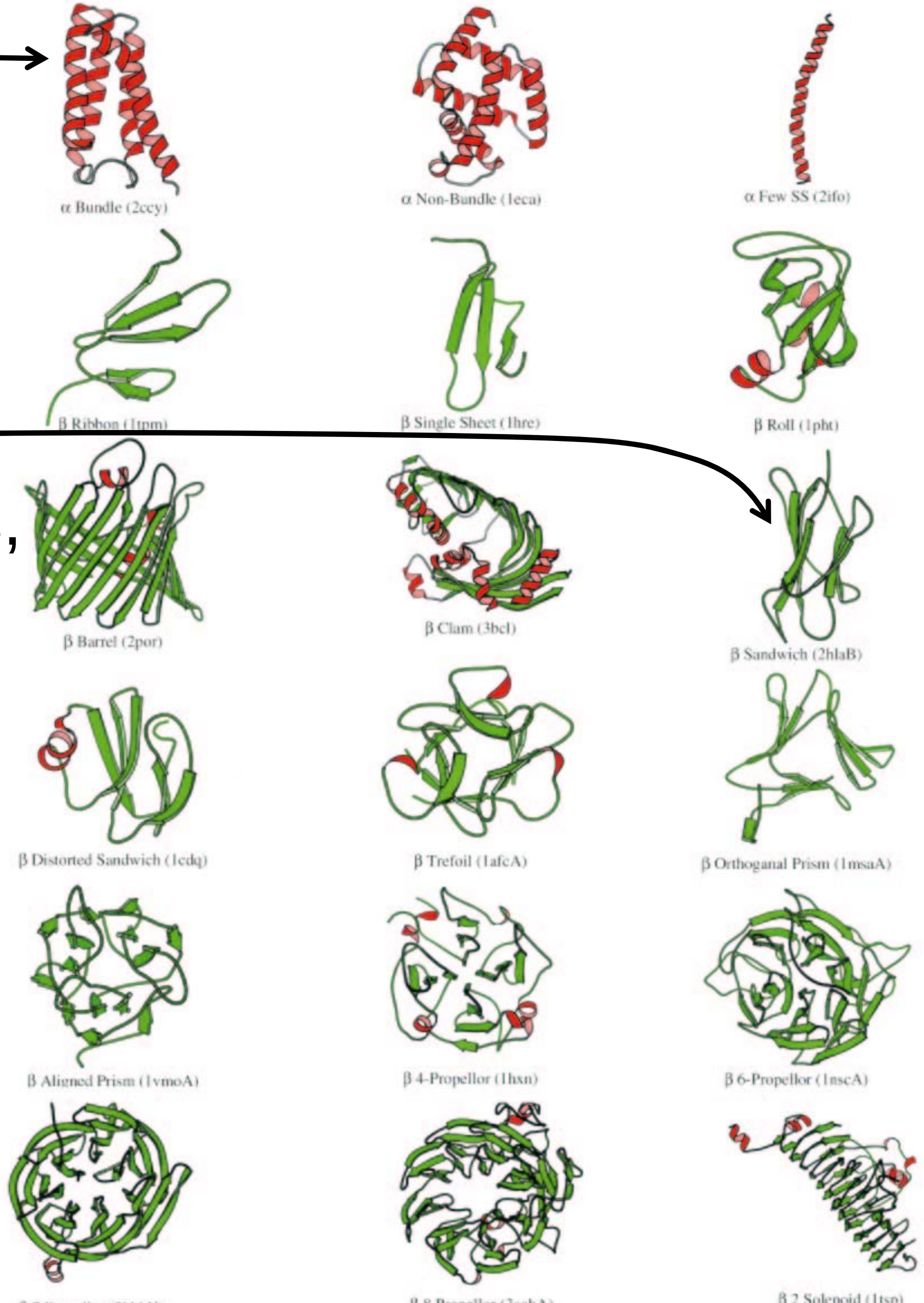
β-sandwich ≈1236 domains, 15 %

some families ?

- < 0.01 %

Interesting…

- some families very popular, some not



α Bundle (2ccy)

α Non-Bundle (1eca)

α Few SS (2ifo)

β Ribbon (1tpm)

β Single Sheet (1hre)

β Roll (1pht)

β Barrel (2por)

β Clam (3bcl)

β Sandwich (2hlaB)

β Distorted Sandwich (1cdq)

β Trefoil (1afcA)

β Orthoganal Prism (1msaA)

β Aligned Prism (1vmoA)

β 4-Propellor (1hxn)

β 6-Propellor (1nscA)

β 7 Propellor (2bbkH)

β 8 Propellor (3aahA)

β 2 Solenoid (1tsp)

CA Orengo AD Michie, S Jones,DT Jones, MB Swindells,JM Thornton, Structure, 1997, 5,1093–1108

Why are some families populated more than others ?

- more next semester
- are some structures more stable ?
- are some older in evolutionary terms ?
- can some "accommodate" more sequences / tolerate more mutations

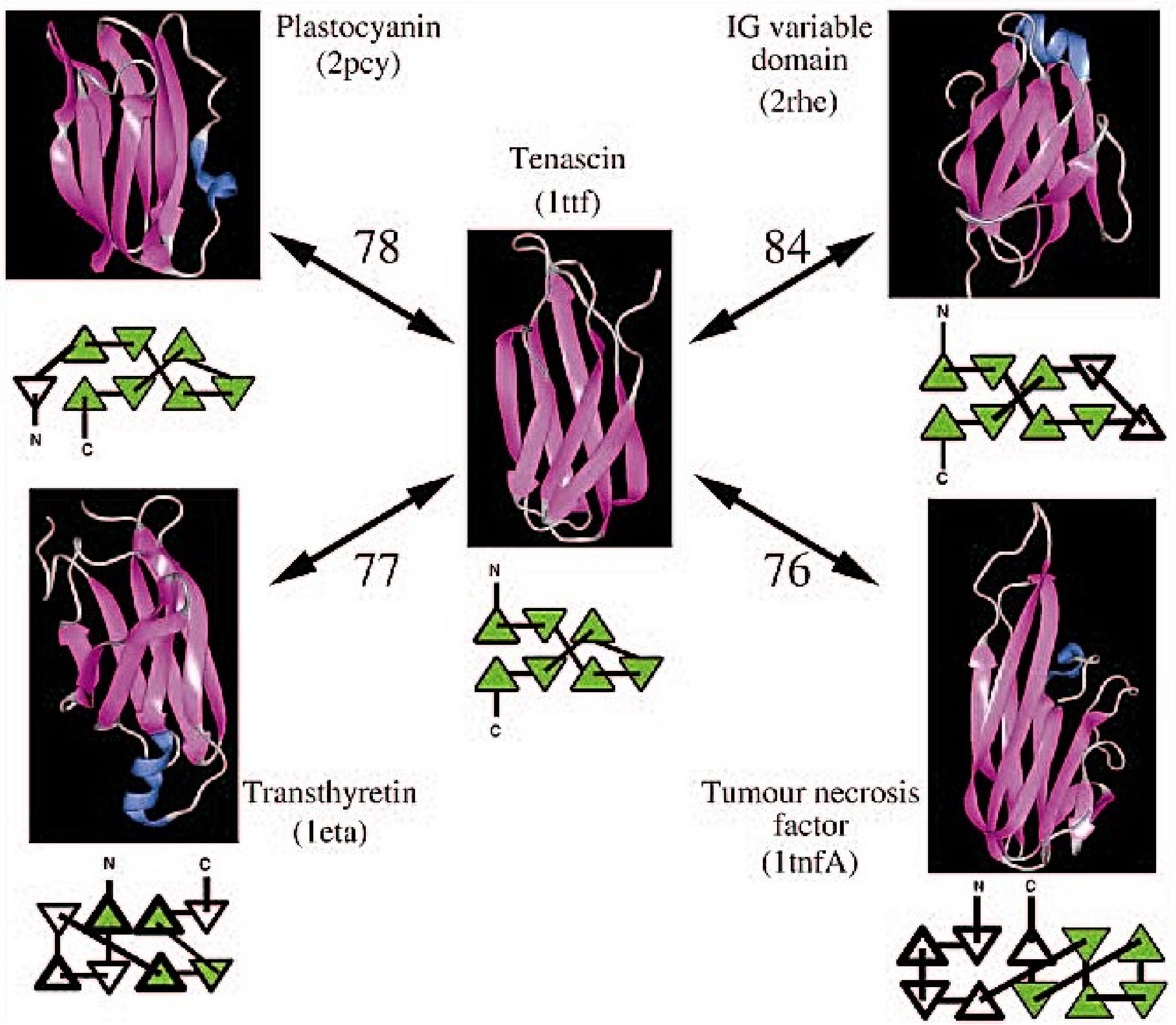
- is this a reflection of physics ?
- no – PDB is very biased
    - mainly soluble, globular proteins which crystallised
    - very few membrane-bound proteins

Supersecondary Motifs

- members of a given family probably have common supersecondary motifs.
  - helpful ?
  - not all proteins can be generated as a collection of motifs
- can we interpret in terms of evolution ?
  - sometimes

Evolution and Classification

- for very similar proteins, easy
- more remote ?
  - maybe

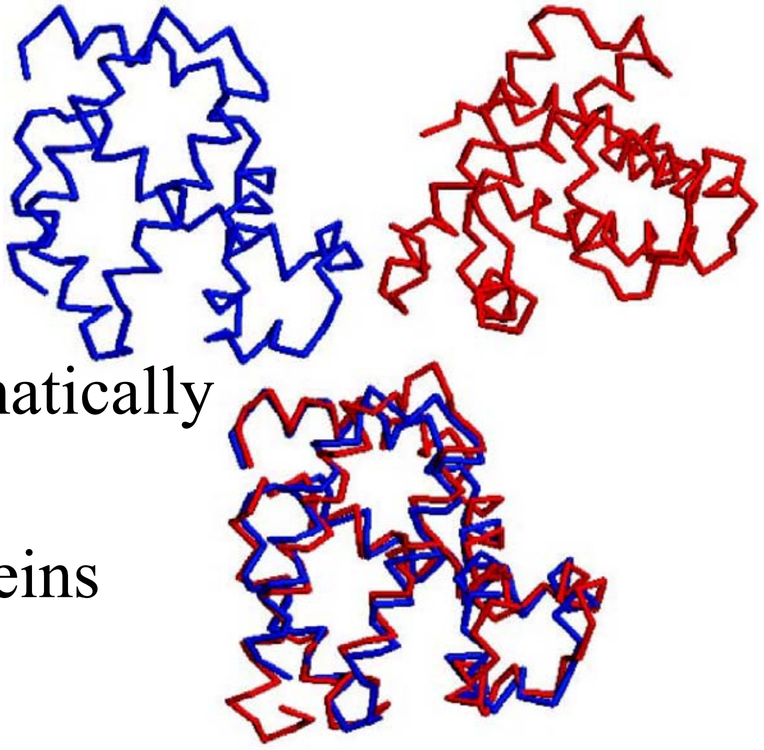


CA Orengo AD Michie, S Jones,DT Jones, MB Swindells,JM Thornton, Structure, 1997, 5,1093–1108

Forget Evolution

- Is the hierarchy really justified ?
  - at low levels maybe
  - at higher levels ? (α,α/β, ..)

- better to discover relationships automatically



- Imagine I can compare arbitrary proteins
- have some measure of similarity
- use this to classify

- Huge problem
  - proteins are different sizes and shapes
  - how to compare ?

Blankenbecler, Ohlsson, Peterson†, Ringne PNAS (2003)100, 11936–11940

Protein Structure Comparison / Numerical

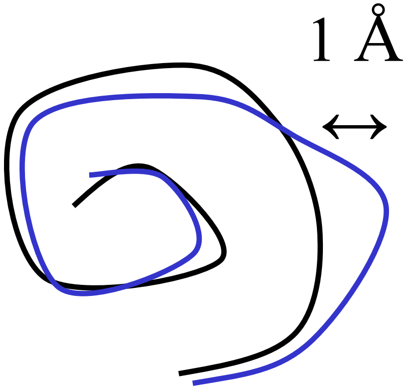
Most common protein structural question

- how much has my protein moved over a simulation ?
- how similar are these NMR models for a structure ?
- how close is my model to the correct answer ?

- more difficult
  - how similar is rat to human haemoglobin ?

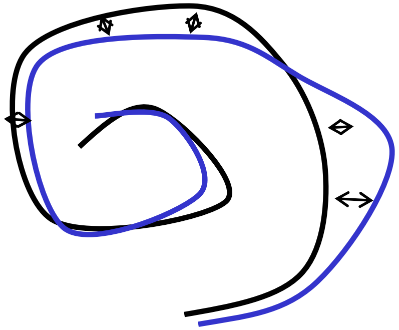
- two cases
  1. same protein, same number of atoms
  2. different proteins
- first
  - measures for easy cases

Numerical Comparison of Structures - Easy



- what units would we like ?
  - scale of similarity (0 to 1.0) ?
  - comparison of angles
  - distance / Å ? most common / easy to interpret

- looks a bit like the average difference between coordinates
- consider analogy with standard deviation / variance



From Standard Deviation to RMSD

Analogy with comparing a set of numbers

- get average (mean)

$$\bar{x} = N^{-1}\sum_{i=1}^{N} x_i$$

$$\sigma^2 = N^{-1}\sum_{i=1}^{N}\left(x_i - \bar{x}\right)^2$$

- variance and standard deviation, σ
- apply this to coordinates of *r* and *r′*

$$\sigma = \left(N^{-1}\sum_{i=1}^{N}\left(x_i - \bar{x}\right)^2 \right)^{1/2}$$

$$RMSD = \left(N^{-1}\sum_{i=1}^{N}\left|\vec{r}_i - \vec{r}_i{}'\right|^2 \right)^{1/2}$$

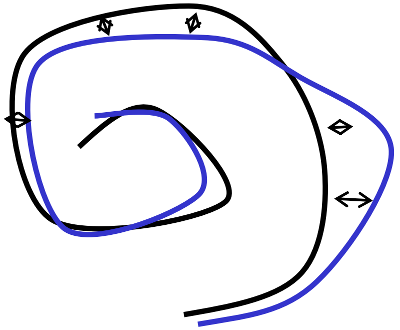
Vital

- formula above, names below
- rms = rmsd = RMSD = root mean square difference

Applying this…

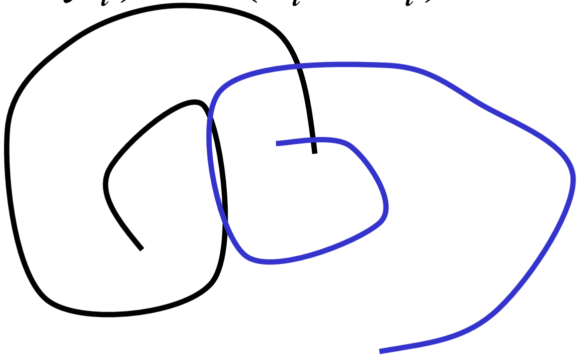
Calculating rmsd

$$RMSD = \left(N^{-1} \sum_{i=1}^{N} \left| \vec{r}_i - \vec{r}_i' \right|^2 \right)^{1/2}$$

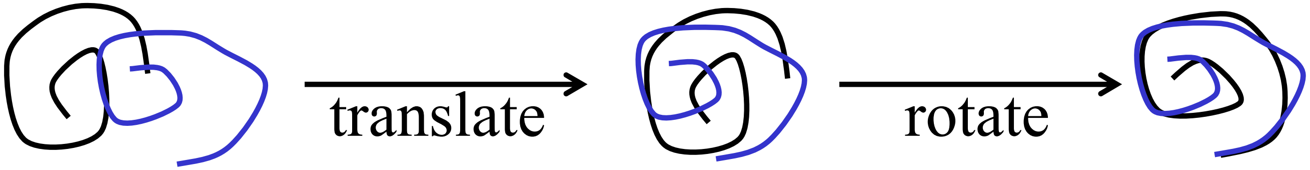


- start at one end
- difference between pairs of atoms

$$\left| \vec{r}_i - \vec{r}_i' \right|^2 = (x_i - x_i')^2 + (y_i - y_i')^2 + (z_i - z_i')^2$$



- huge problem..
  - coordinates are normally…
- what to do ?



Translation and Rotation

translation

- c.o.m. = centre of mass

$$\vec{r}^{\,c.o.m} = \left(\sum_{i=1}^{N} m_i \right)^{-1} \sum_{i=1}^{N} \vec{r}_i m_i$$

- subtract difference vector

$$\vec{r}_{diff} = \vec{r}^{\,c.o.m.} - \vec{r}'^{\,c.o.m.}$$

- rotation
  - messier..
  - find rotation matrix to minimise $RMSD = \left(N^{-1} \sum_{i=1}^{N} \left| \vec{r}_i - \vec{r}_i{}' \right|^2 \right)^{1/2}$

- summary
  - translate
  - rotate
  - apply formula
- still not finished

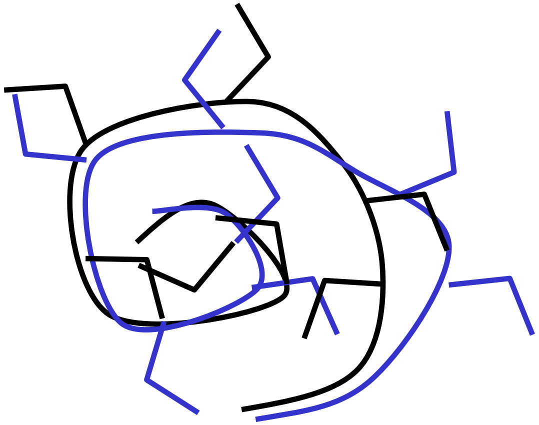
Which Atoms ?

What tells me the shape of a protein ?

- backbone trace

What happens if you include all atoms ?

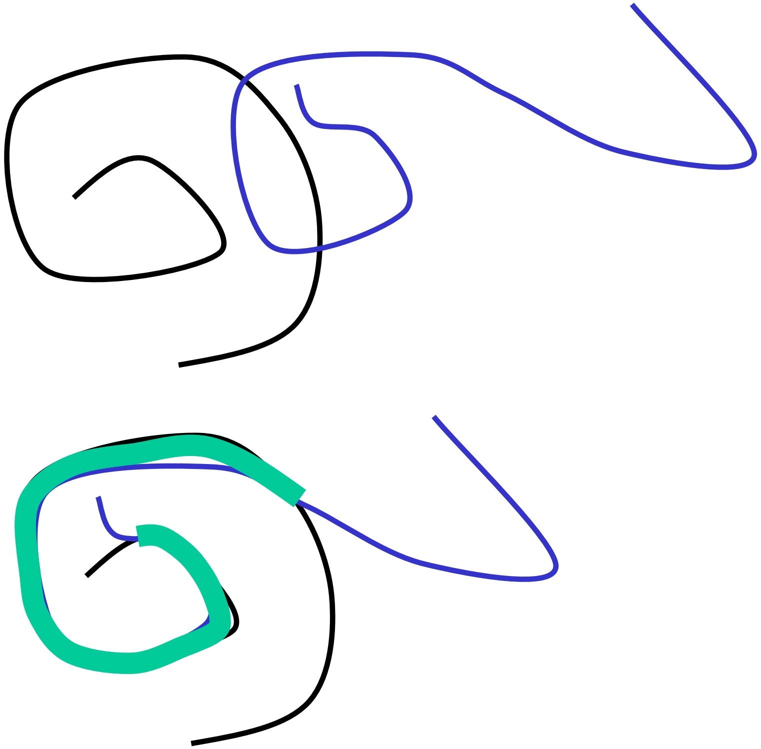
- bigger *rmsd*
- normal choice
  - C^{α}
- sometimes
  - N, C^{α}, C
- all atoms ?
  - when a model is very close



Still not finished with simple *rmsd*

Parts Of Proteins

- two models of a molecule
  - mostly very similar
  - is *rmsd* a good measure ?
- identify similar parts



define

```
superimpose ({r},{r'}, {d}) {
      translate ({r,},{r'}, {d})
      rotate ({r},{r'}, {d})
}
```

where **{d}** is some subset of sites

Selection of Interesting Atoms

- define a threshold like **thresh** =2 Å

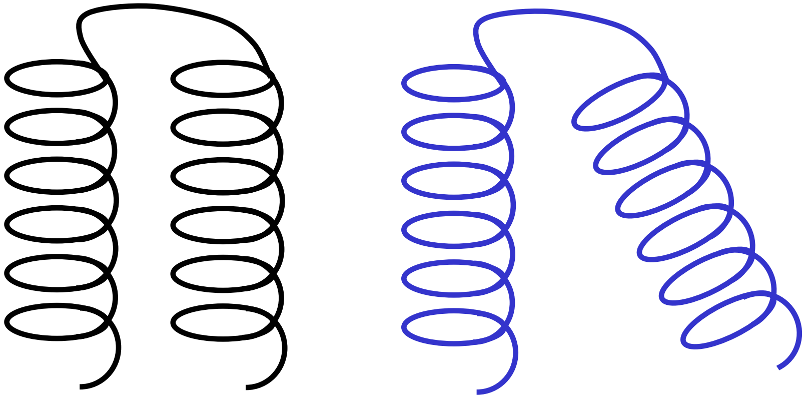
```
{d}={|$r_i$-$r'_i$|} i=1..N
sort {d}

diff= rmsd ({$r_i$},{$r_i'$})
while (diff > thresh) {
    remove largest d
    superimpose ({$r$},{$r'$}, {d})
    recalculate distances
    diff = rmsd ({$r$},{$r'$}, {d})
}
if (diff < thresh)
    return {d}, diff
else
    return broken
```

- result ? a subset of interesting atoms

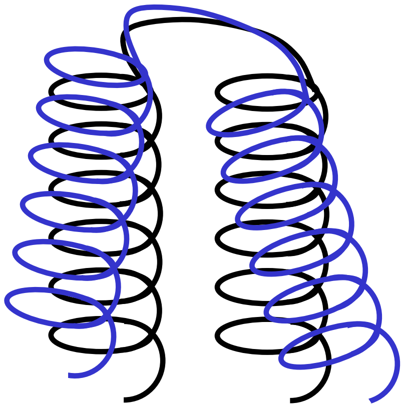
Subsets of Atoms

- Originally, quantify structural differences as Å *rmsd*
- Alternative quantity implied
  - number of residues used for *rmsd* below threshold
- implicit rule
  - as number of atoms ↓ calculated *rmsd* ↓

Why Not Use *rmsd*



- helices identical, fold identical *rmsd* ?



- superposition requires rotation, affects all atoms

- big *rmsd*, but structure has hardly changed
- do not see that helices are identical
- solutions
  - use angles (other problems)
  - distance matrices

Distance Matrices With Numbers

Another characteristic of structures

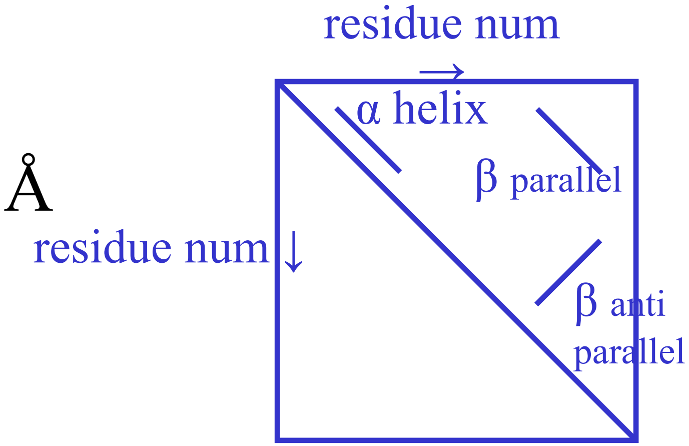
- C^{α} distance matrices
- simply measure the distance between C^{α} atoms

	1	2	3	4	5	6	7	…		N
1	0	3.8	6	7	…					
2		0	3.8	5	…					
3			0	3.8	4.5	…				
4				0	3.8					
5					0	3.8				
6						0	3.8			
7							0	3.8		
…								0	3.8	
									0	3.8
N										0

Distance Matrix for Recognising Structure

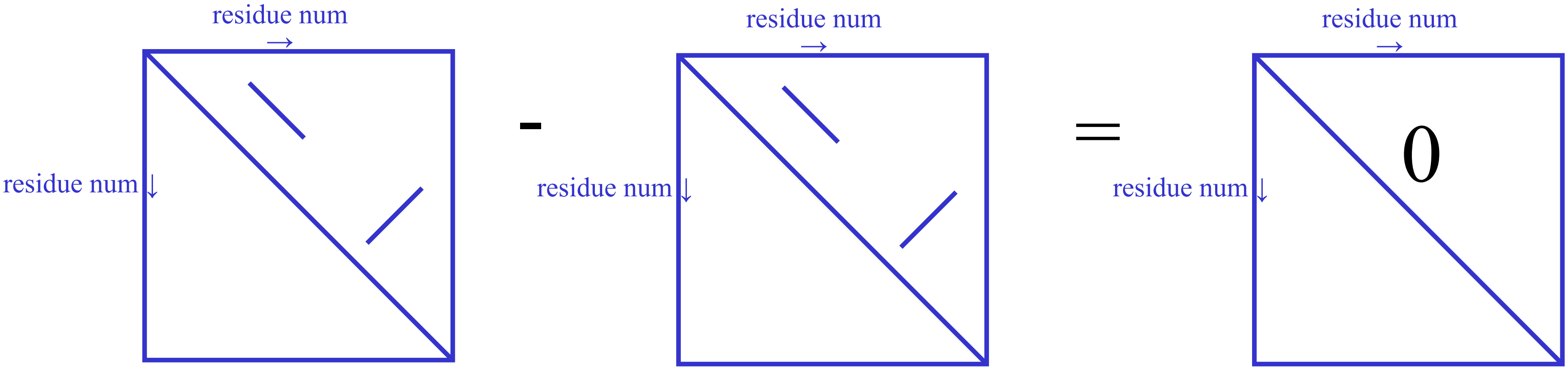
One way to summarise a structure

- plot C^{α} distance matrix, points below 4 Å
- can make α-helices and β-sheets clear



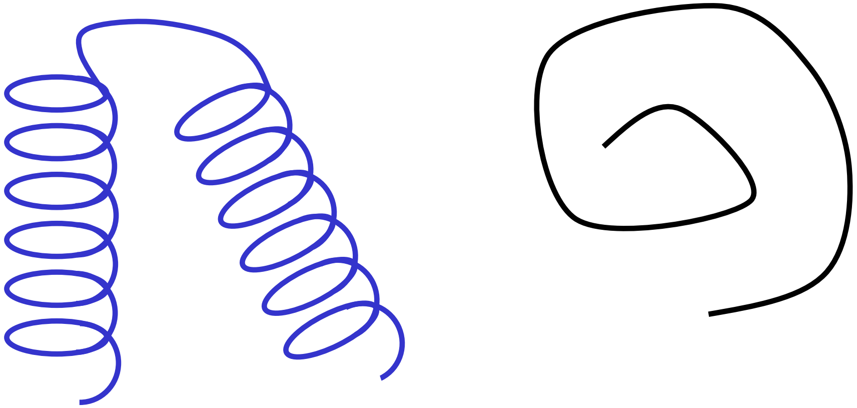
Distance matrix for comparing structures

- take two similar proteins
  - look at the difference of distance matrices

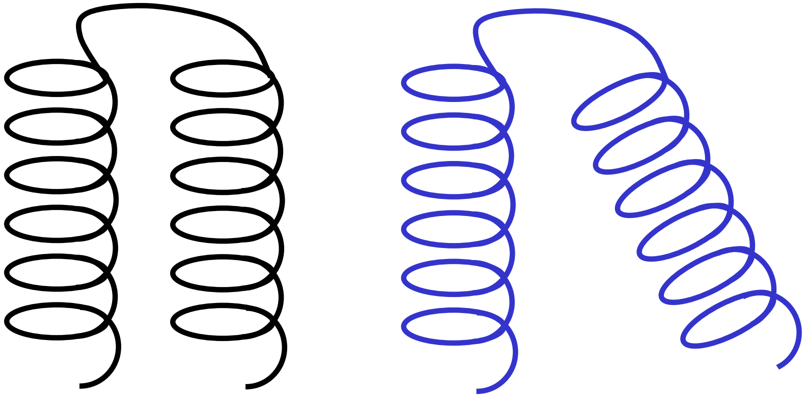


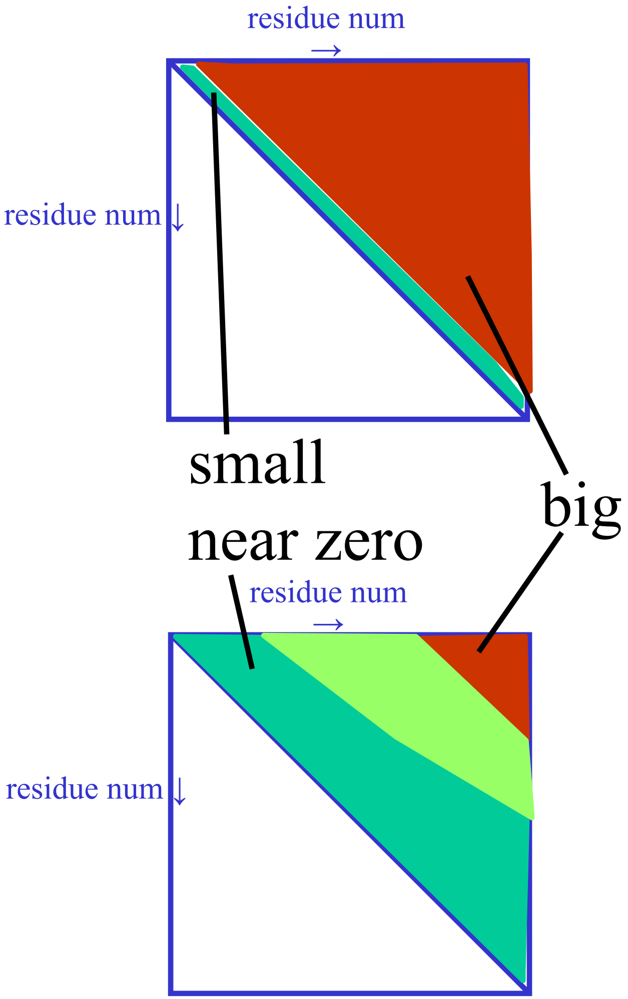
Comparing Distance Matrices

- consider two very different structures



- two related structures





- pictures are better than any single measure, but…

From Distance Matrices to Single Number

For lots of comparisons, single number is more convenient

- root mean square (*rms*) difference of distance matrices
  - define distance between C^{α} atoms *i* and *j*

$$d_{ij} = \left| \vec{r}_i - \vec{r}_j \right|$$

- *rms* of distance matrices measure is

$$rms = \left(\frac{2}{N(N-1)} \sum_{i=1}^{N} \sum_{j>i}^{N} \left(d'_{ij} - d_{ij} \right)^2 \right)^{1/2}$$

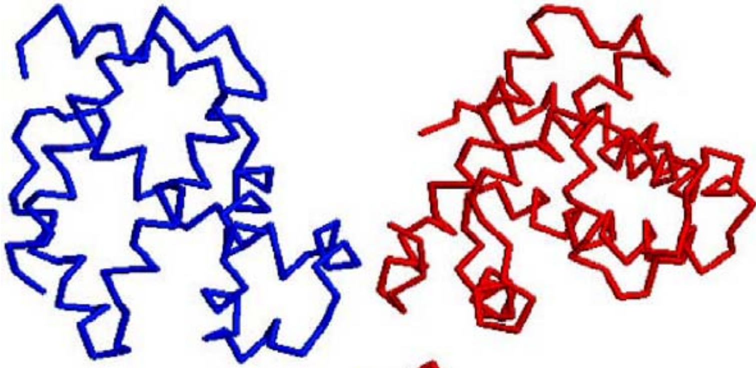
- just like all other *rms* quantities
  - normalised over top half of matrix

Summary – Comparing Models / Structures

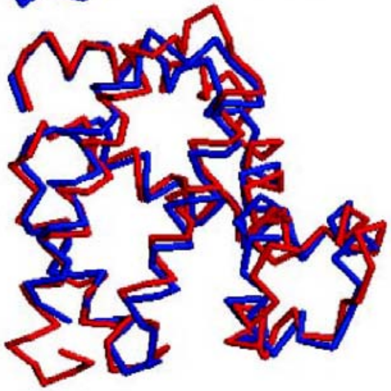
- *rmsd*
  - most popular
  - requires superposition (translate + rotate)
  - can be fooled by "hinge" movements
- to look at the shape of a molecule use C^{α} or backbone atoms
- numbers in Å have a physical meaning
- to look for the common core of a structure, find a subset of backbone
- other measures may be better than *rmsd*
- weakness of all measures
  - a single number can never capture all information

Comparing Different Proteins

- compare red and blue proteins
- if we know which residues match
  - easy (use any *rms* formula)
- which residues match ?
  - sequence alignment ?



protein 1	A	C	D	W	Y	T	R	P	K	L	H	G	F	D	S	A	C	V	N
protein 2	A	C	D	W	W	T	-	P	K	V	H	G	Y	D	S	A	C	V	N

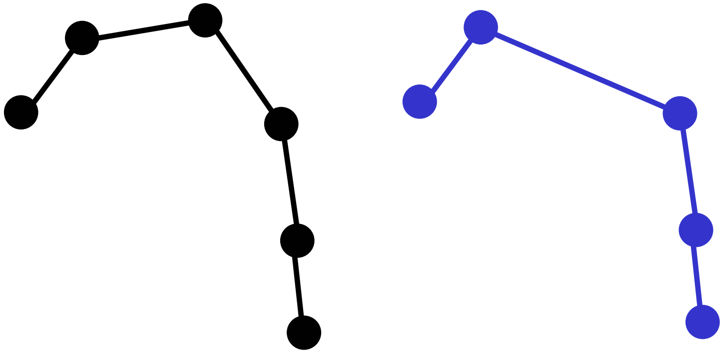


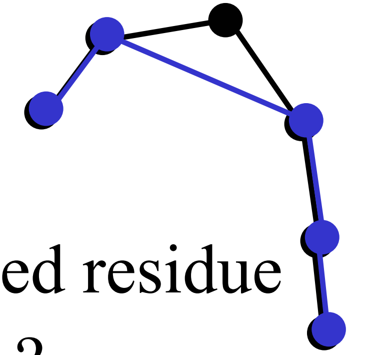
- green residues - backbone atoms
- pink residues – ignore
- is this useful for similar proteins ? very (rat vs human haemoglobin)
- for very different proteins ? no

picture from Blankenbecler, Ohlsson, Peterson†, Ringne PNAS (2003)100, 11936–11940

Comparing Very Different Proteins

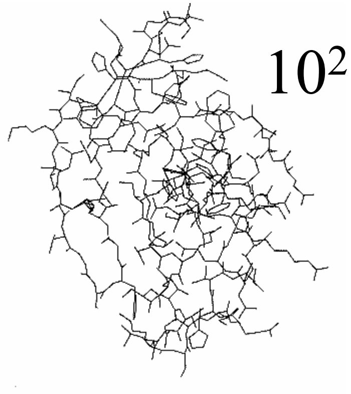
- sequence alignment vs identity
  - as identity ↓, errors ↑
- consequence
  - methods needed
    - operate on C^{α}
    - do not require sequence
- how difficult ?
  - superposition requires recognising the deleted residue
  - can we use standard dynamic programming ?
    - no
  - gap/insertion at any position, any length
    - combinatorial explosion



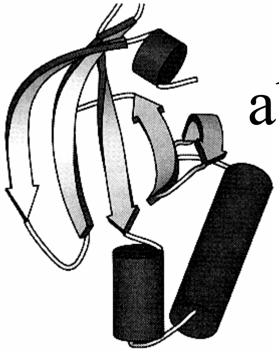


Strategies For Comparing Different Structures
1. use secondary structure

- Combinatorial explosion is the problem
  - reduce size of problem
  - use elements of secondary structure



10^2-10^3 atoms



about 8 units

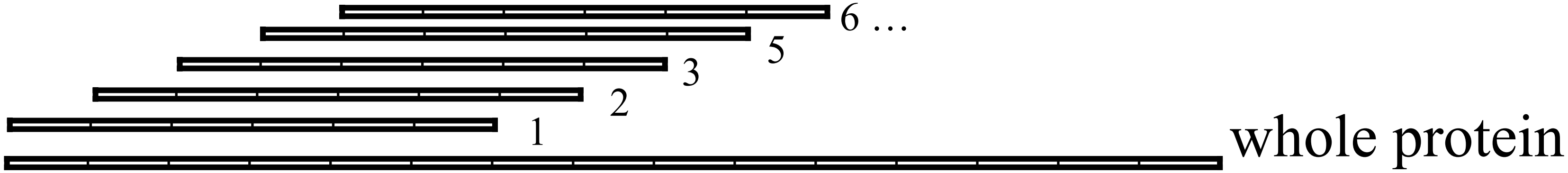
- define secondary structure
- search for superposition
- for each residue
  - find closest C^{α} in partner structure
  - use the set of matching residues to calculate *rmsd*

pictures from "Structural Bioinformatics", ed Bourne, PE and Weissig, H.

2. Peptide fragment strategy

- more general version of idea on previous page
- basis of most popular methods

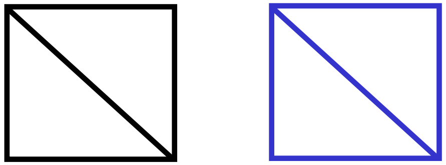
- Ingredients
    - break protein into overlapping fragments (length 6 or 8)
    - protein is no longer a string of residues nor a whole structure



- each fragment is a little distance matrix

Fragment Based Comparison

- any two distance matrices can be compared
- two proteins length *N* and *M* can now be compared…



protein 1 fragments →

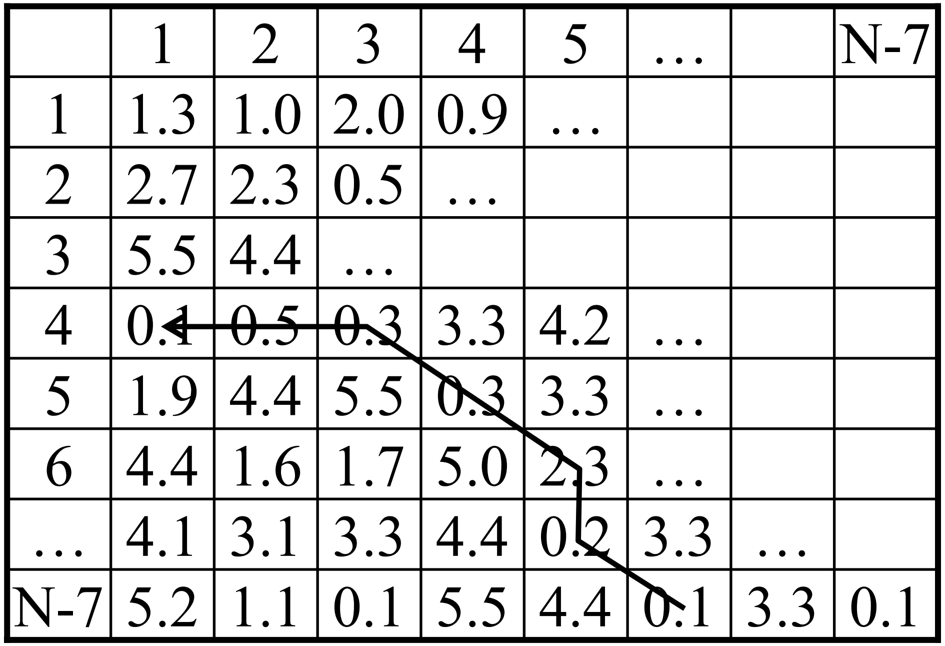
protein 2 fragments ↓

	1	2	3	4	5	…		*N*-7
1	1.3	1.0	2.0	0.9	…			
2	2.7	2.3	0.5	…				
3	5.5	4.4	…					
4	0.1	0.5	0.3	3.3	4.2	…		
5	1.9	4.4	5.5	0.3	3.3	…		
6	4.4	1.6	1.7	5.0	2.3	…		
…	4.1	3.1	3.3	4.4	0.2	3.3	…	
M-7	5.2	1.1	0.1	5.5	4.4	0.1	3.3	0.1

- imagine *rmsd*
- this is now like a sequence comparison problem

Finding Equivalent Fragments

- find optimal path through matrix
- classic dynamic programming method like sequence comparison



	1	2	3	4	5	…		N-7
1	1.3	1.0	2.0	0.9	…			
2	2.7	2.3	0.5	…				
3	5.5	4.4	…					
4	0.1	0.5	0.3	3.3	4.2	…		
5	1.9	4.4	5.5	0.3	3.3	…		
6	4.4	1.6	1.7	5.0	2.3	…		
…	4.1	3.1	3.3	4.4	0.2	3.3	…	
N-7	5.2	1.1	0.1	5.5	4.4	0.1	3.3	0.1

- like sequence comparison
  - find optimal path through matrix
  - classic dynamic programming method (N & W, S & W)
  - uses gap penalties

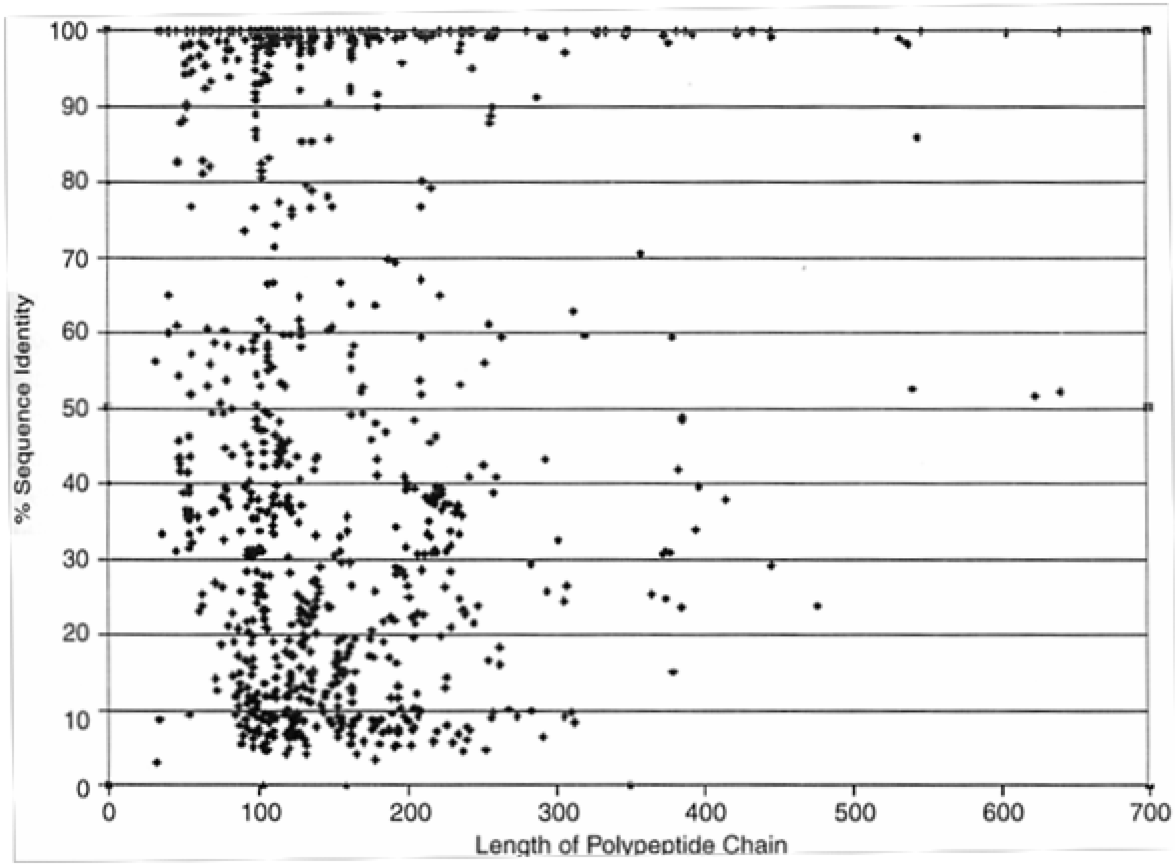
Comparing Different Size Protein Structures

- Break protein into overlapping fragments
- fragments can be compared to each other via distance matrices
- align like sequences
- from aligned fragments, get list of aligned residues
- using aligned residues, calculate *rmsd*, *rms* of overall distance matrices

How Important Are These Similarities ?

- survey 1000 proteins
- find structurally similar pairs
- plot sequence identity

may not be found by sequence methods {



pictures from "Structural Bioinformatics", ed Bourne, PE and Weissig, H.

Summary of All Protein Comparisons

Classification of proteins

- could be done by sequence, better by structure

Structure comparison

- for one protein
  - selection of atoms
- for different proteins
  - requires list of matching atoms
- for similar proteins
  - can use pairs from sequence alignment
- for often dissimilar proteins
  - pure structure based method