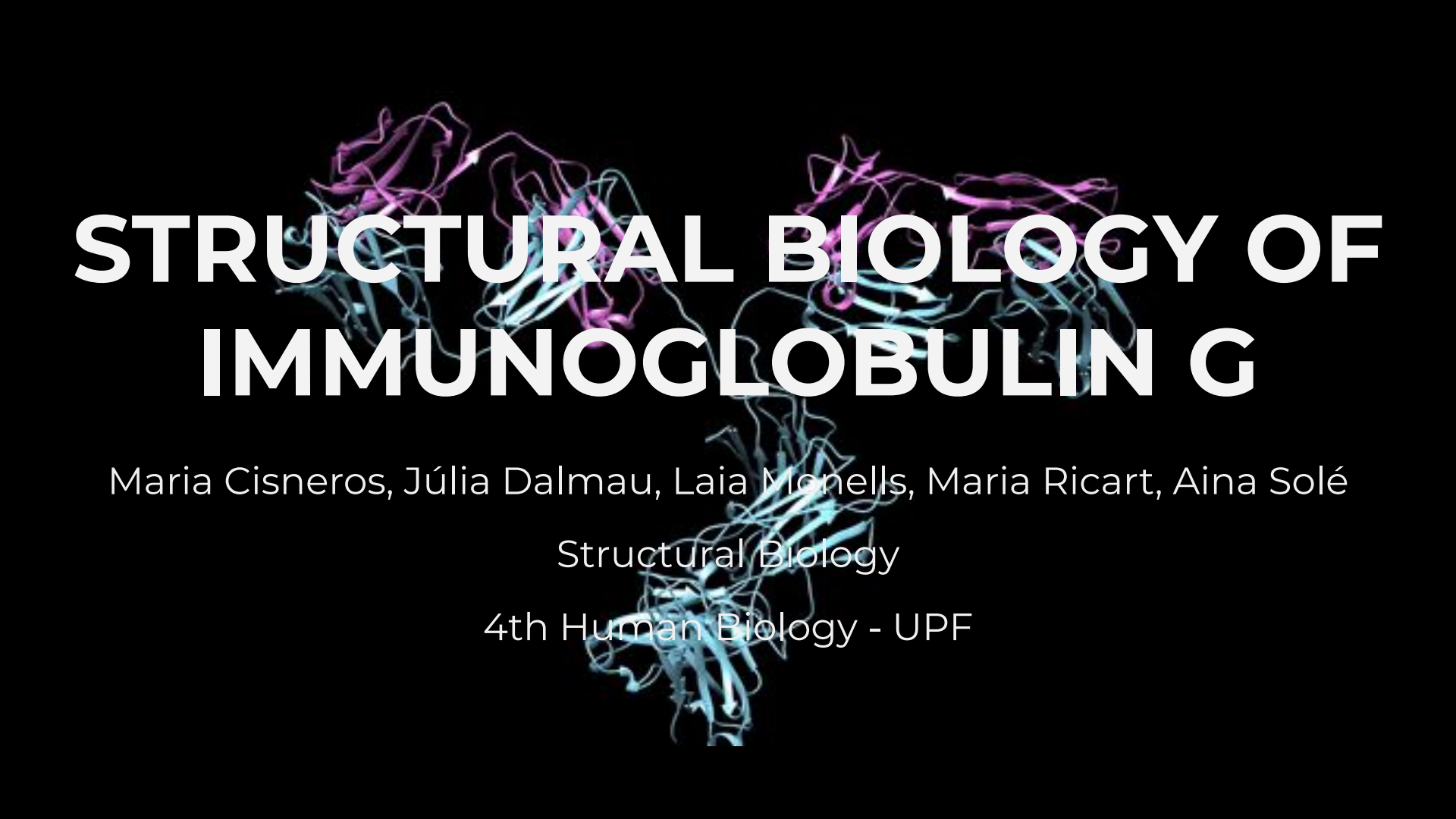


STRUCTURAL BIOLOGY OF IMMUNOGLOBULIN G

Maria Cisneros, Júlia Dalmau, Laia Monells, Maria Ricart, Aina Solé

Structural Biology

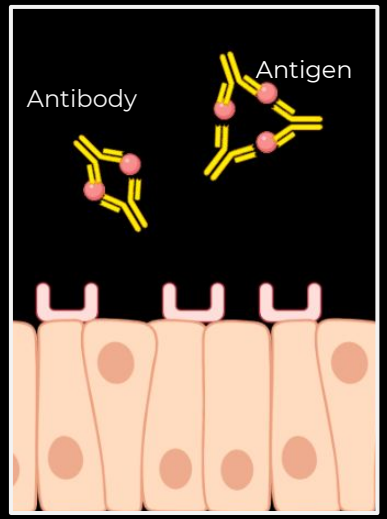
4th Human Biology - UPF

INDEX

BASIC CONCEPTS

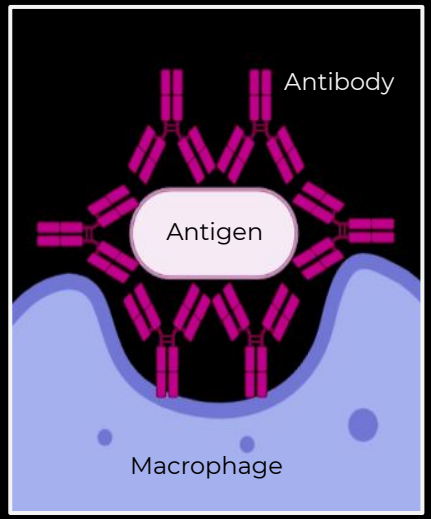
Definition and function

Neutralization



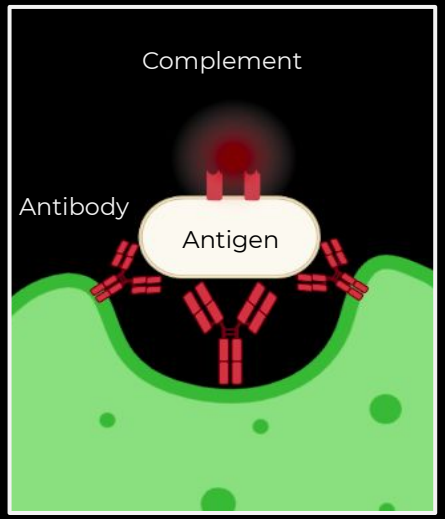
Antibody prevents bacterial adherence

Opsonization



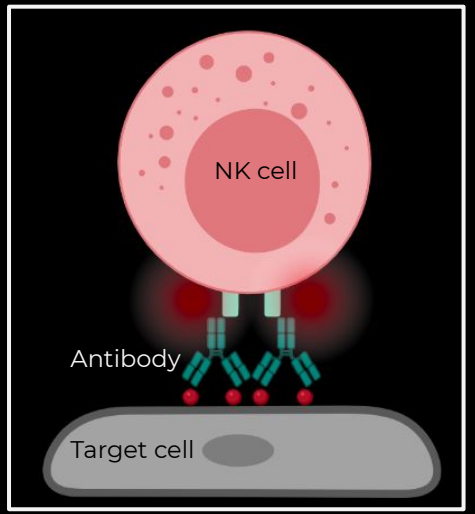
Antibody promotes phagocytosis

Complement activation



Antibody activates complement, which enhances opsonization

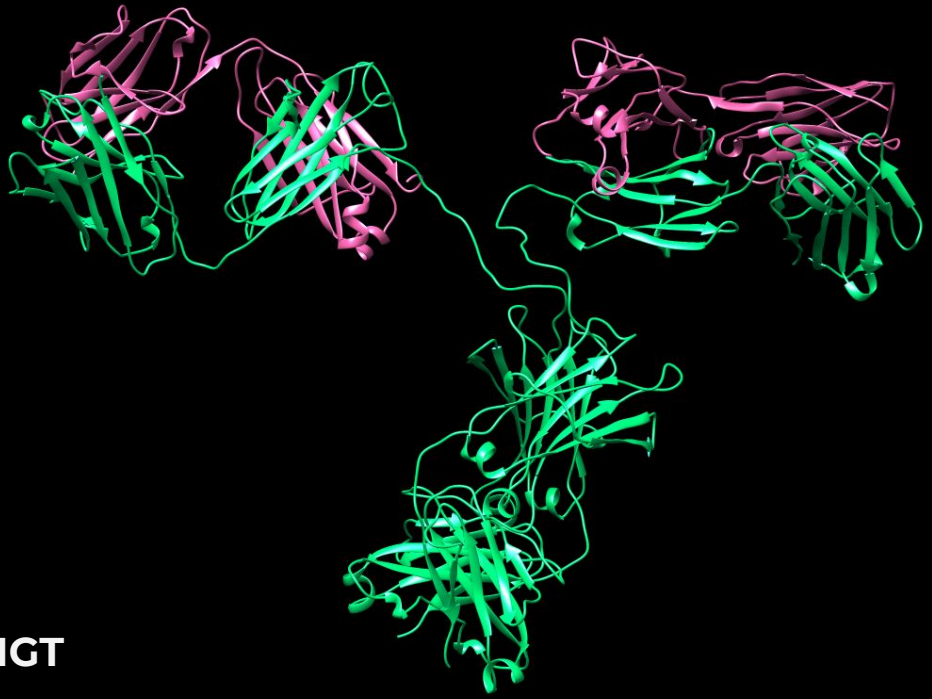
ADCC



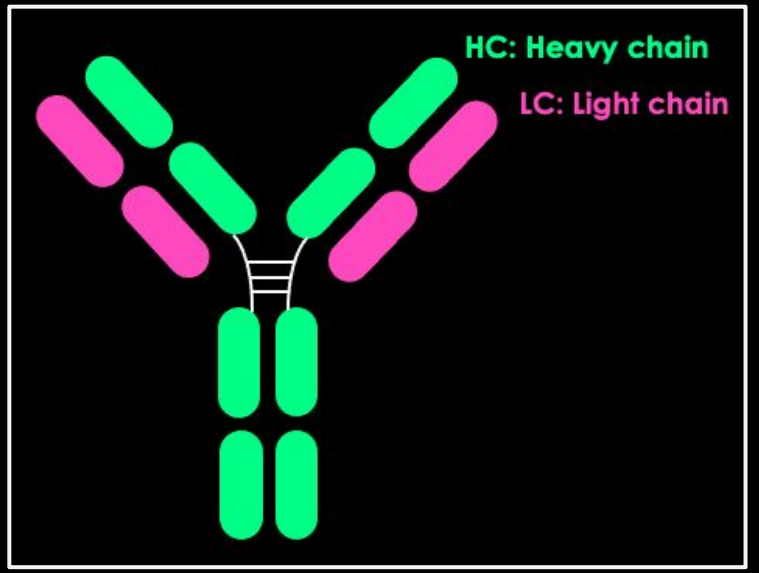
Antibody activates NK cells, which promotes the lysis

BASIC CONCEPTS

General structure: HC & LC

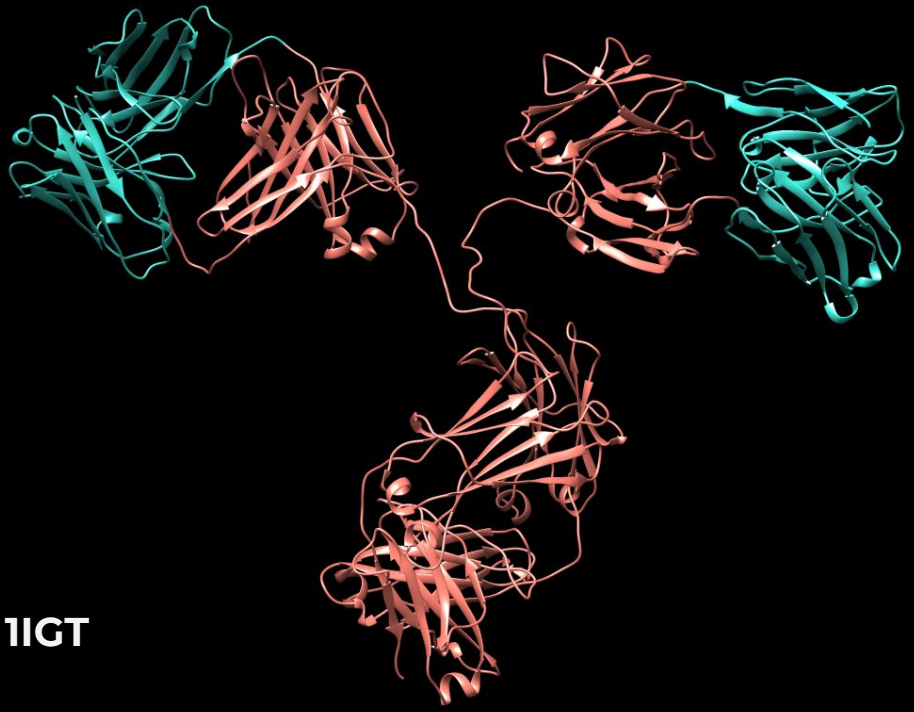


1IGT

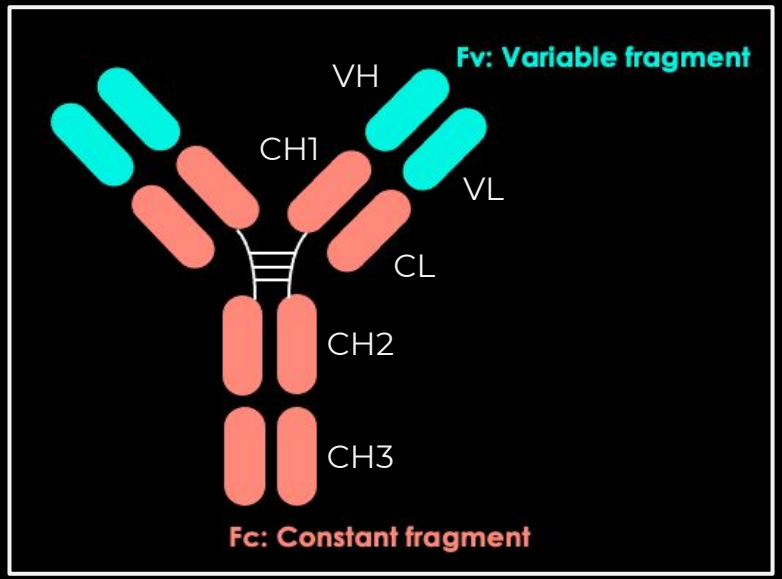


BASIC CONCEPTS

General structure: Fc & Fv

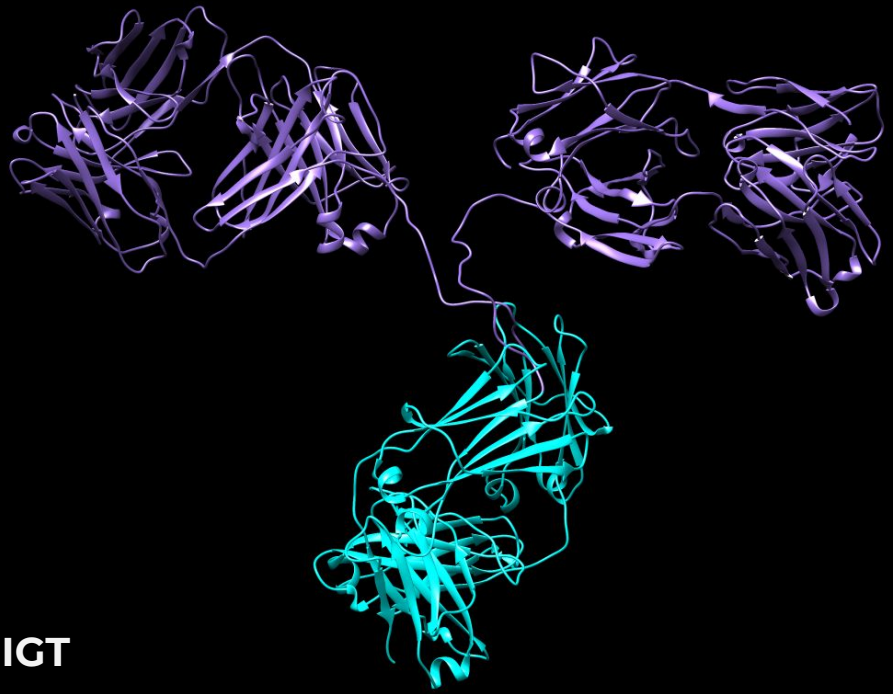


1IGT



BASIC CONCEPTS

General structure: Fab & Fc



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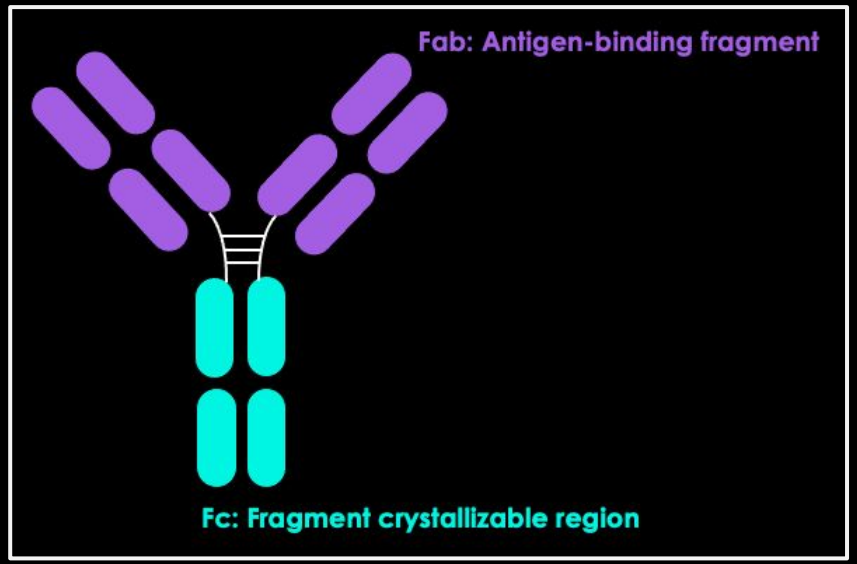
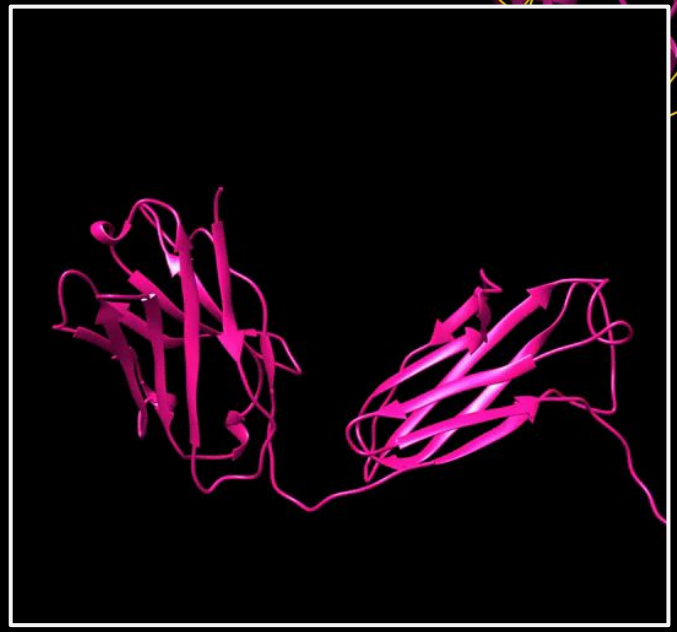


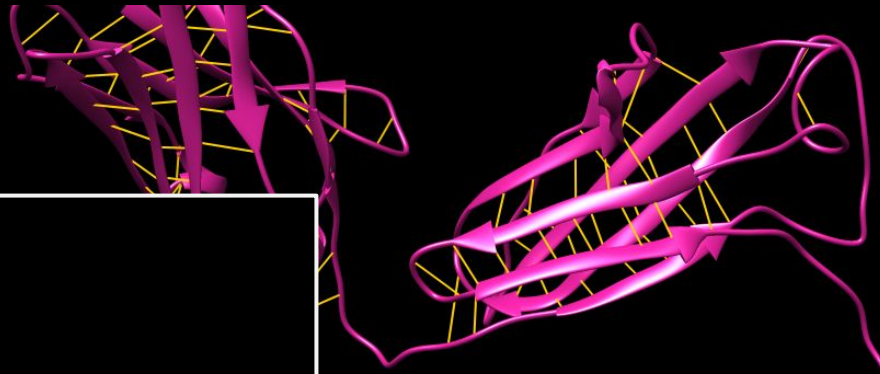
BASIC CONCEPTS

General structure: Stabilization

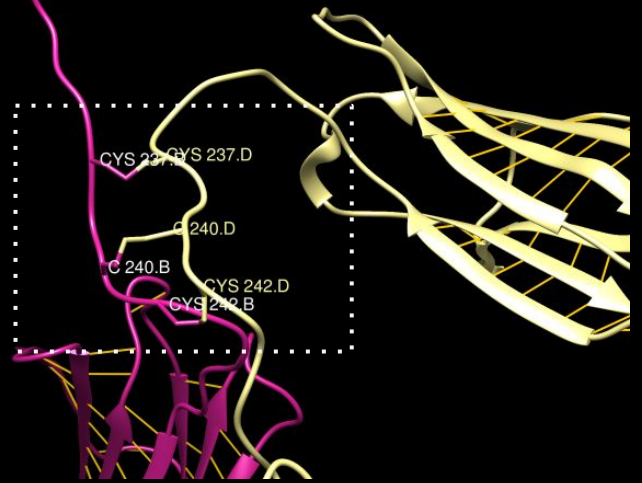
1IGT

Hydrogen bonds



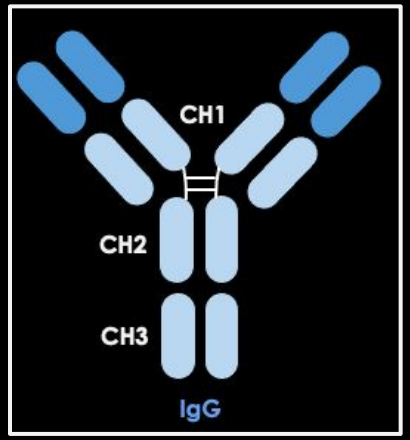


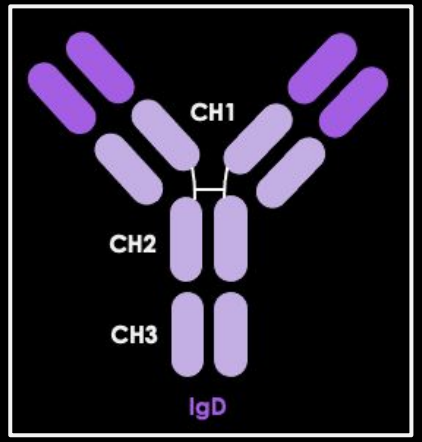
Disulfide bonds

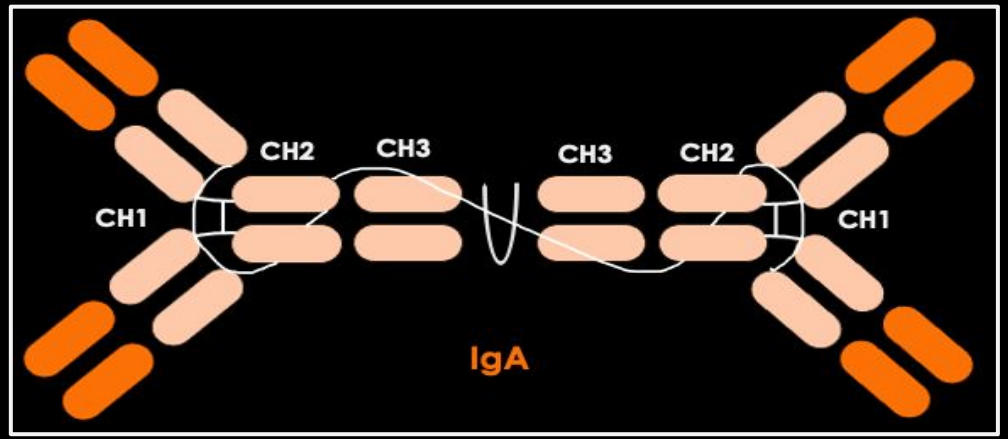


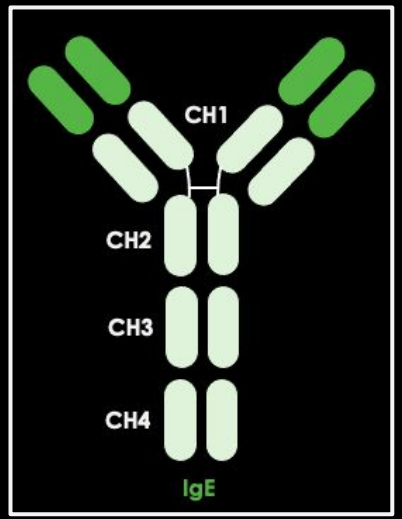
BASIC CONCEPTS

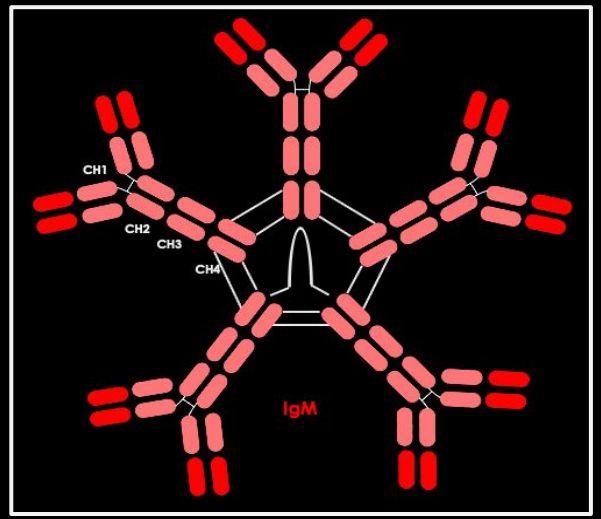
Isotypes









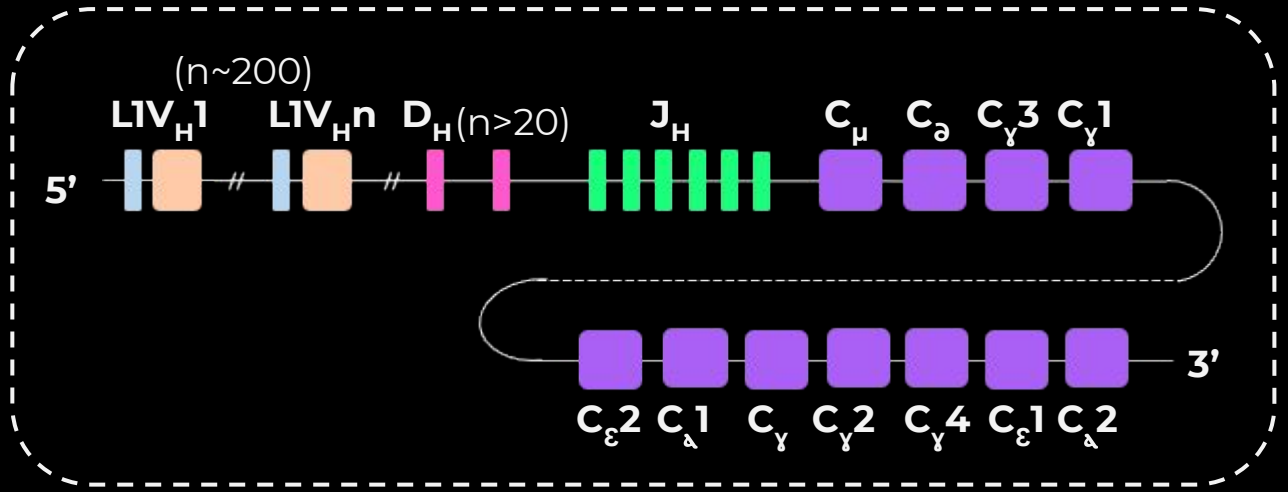


IgG, IgD and IgA have 3 constant domains

IgE and IgM have 4 constant domains

BASIC CONCEPTS

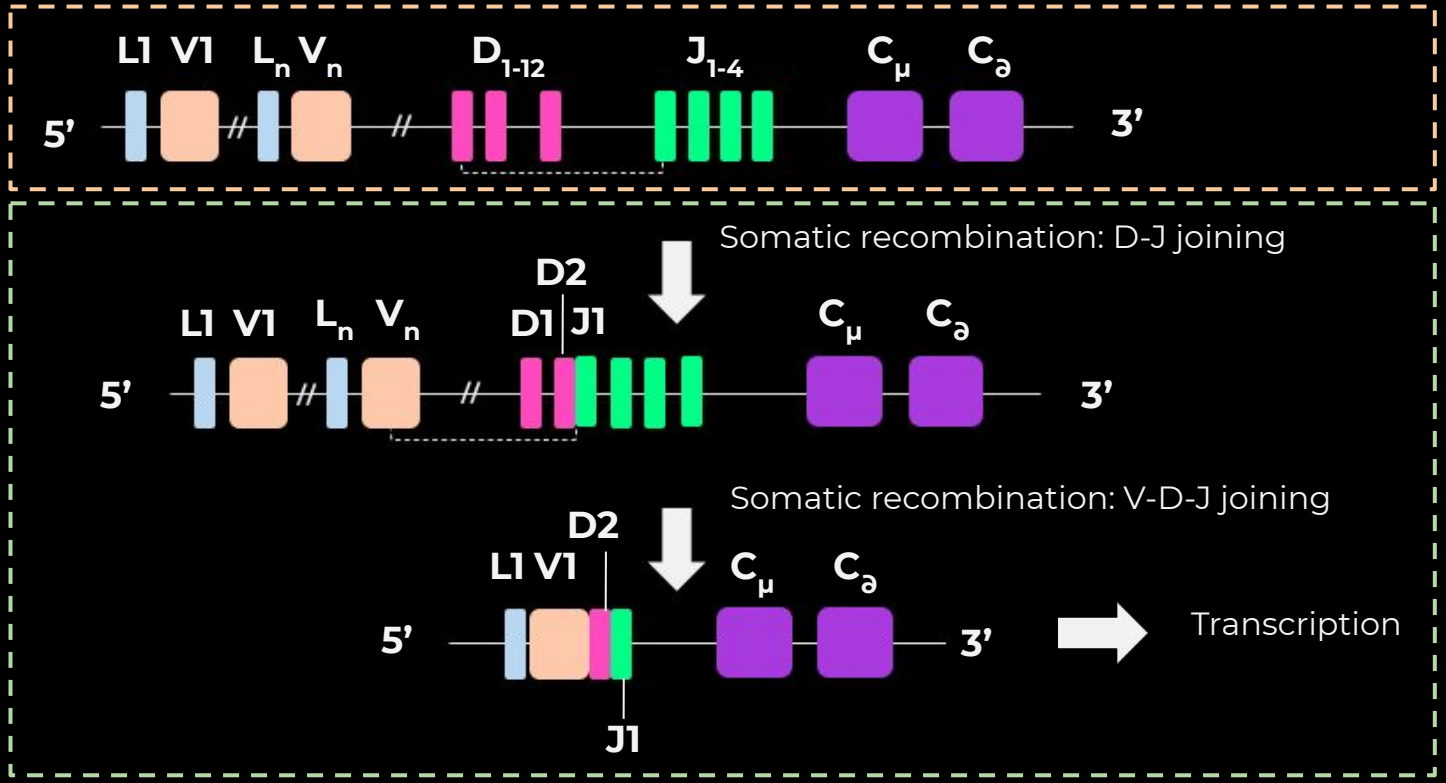
Rearrangement: Heavy Chain



H Chain locus (Chr 14)

BASIC CONCEPTS

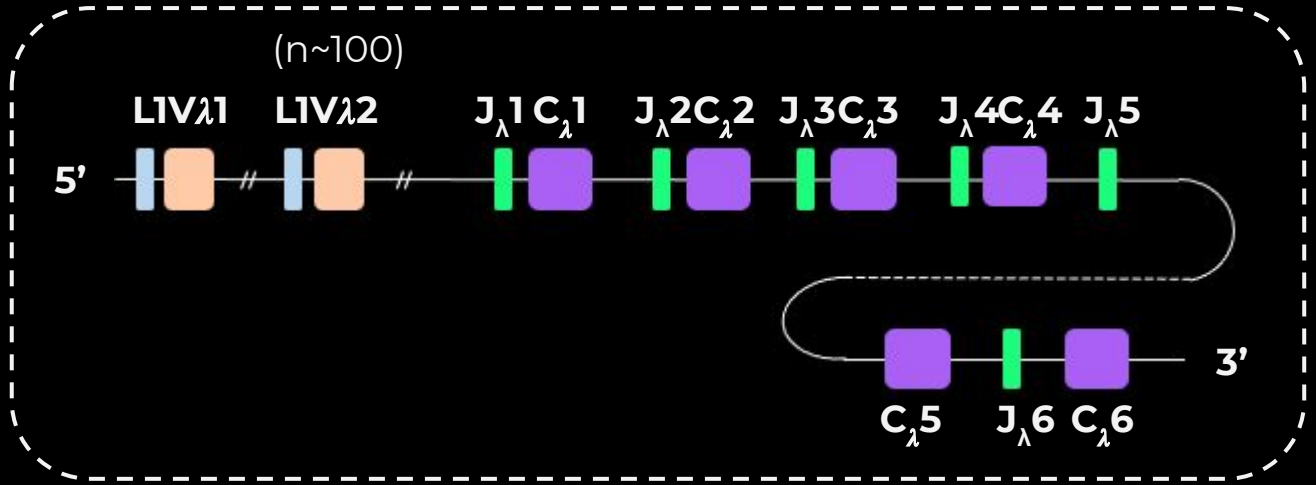
Rearrangement: Heavy Chain



BASIC CONCEPTS

Rearrangement: Light Chain

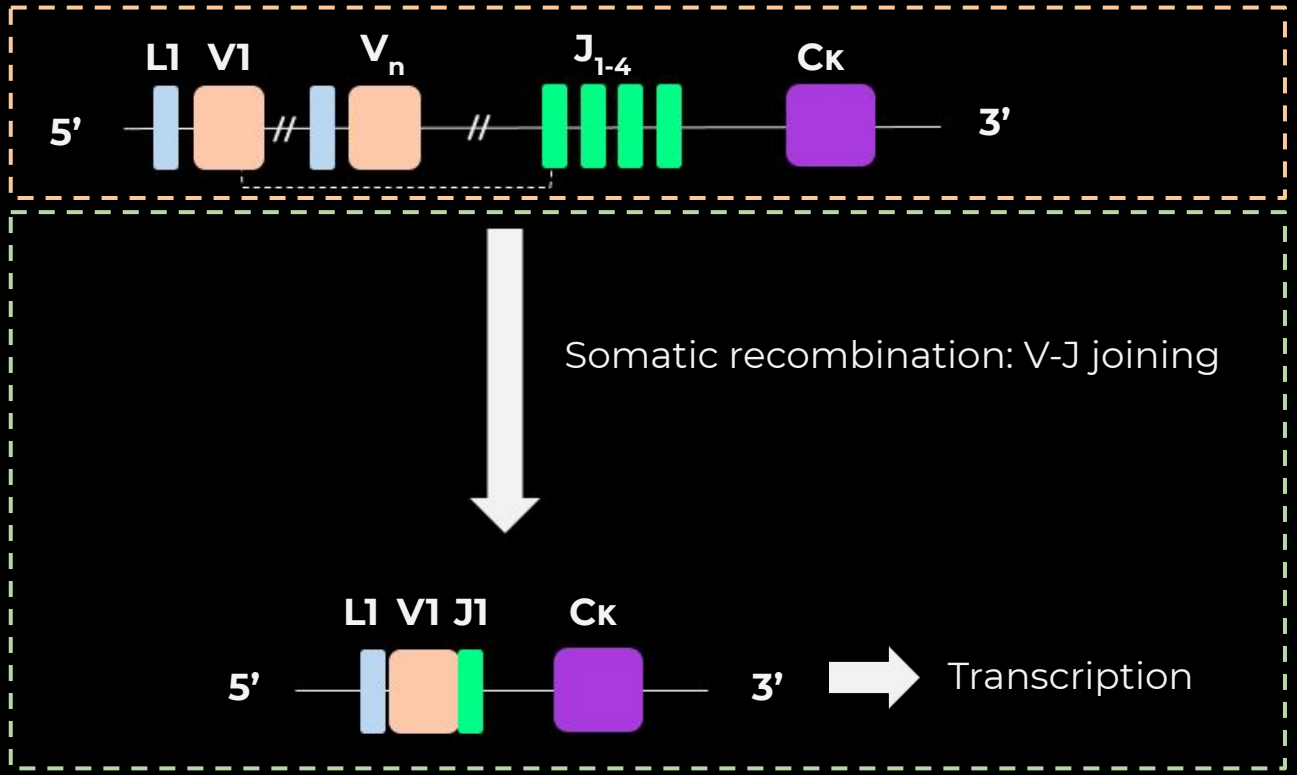
κ Chain locus (Chr 2)



λ Chain locus (Chr 22)

BASIC CONCEPTS

Rearrangement: Light chain



Germline DNA

Rearranged DNA

IgG

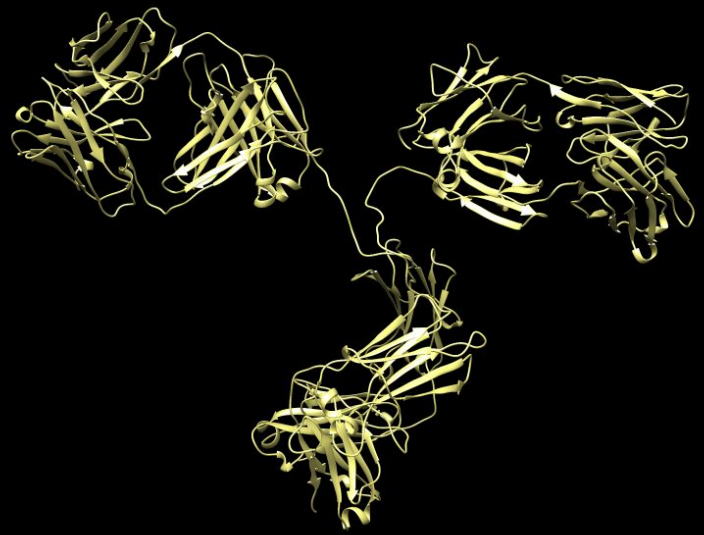
SCOP classification

Class b: All beta proteins

Fold b.1: Immunoglobulin-like beta-sandwich

Superfamily: Immunoglobulin

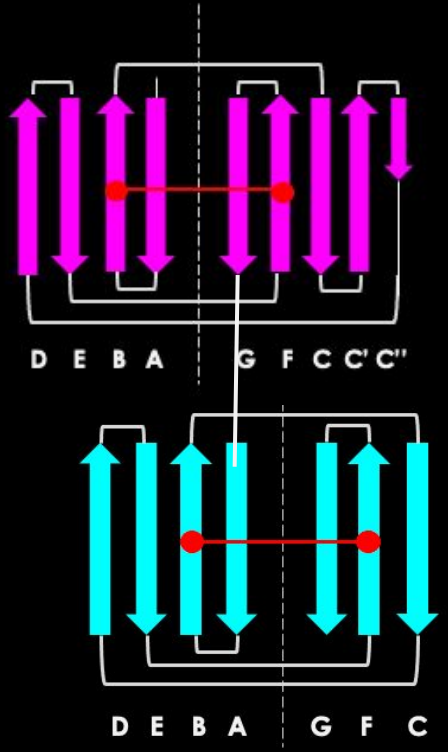
Family:
- C1 set domains (antibody constant domain-like)
- V set domains (antibody variable domain-like)

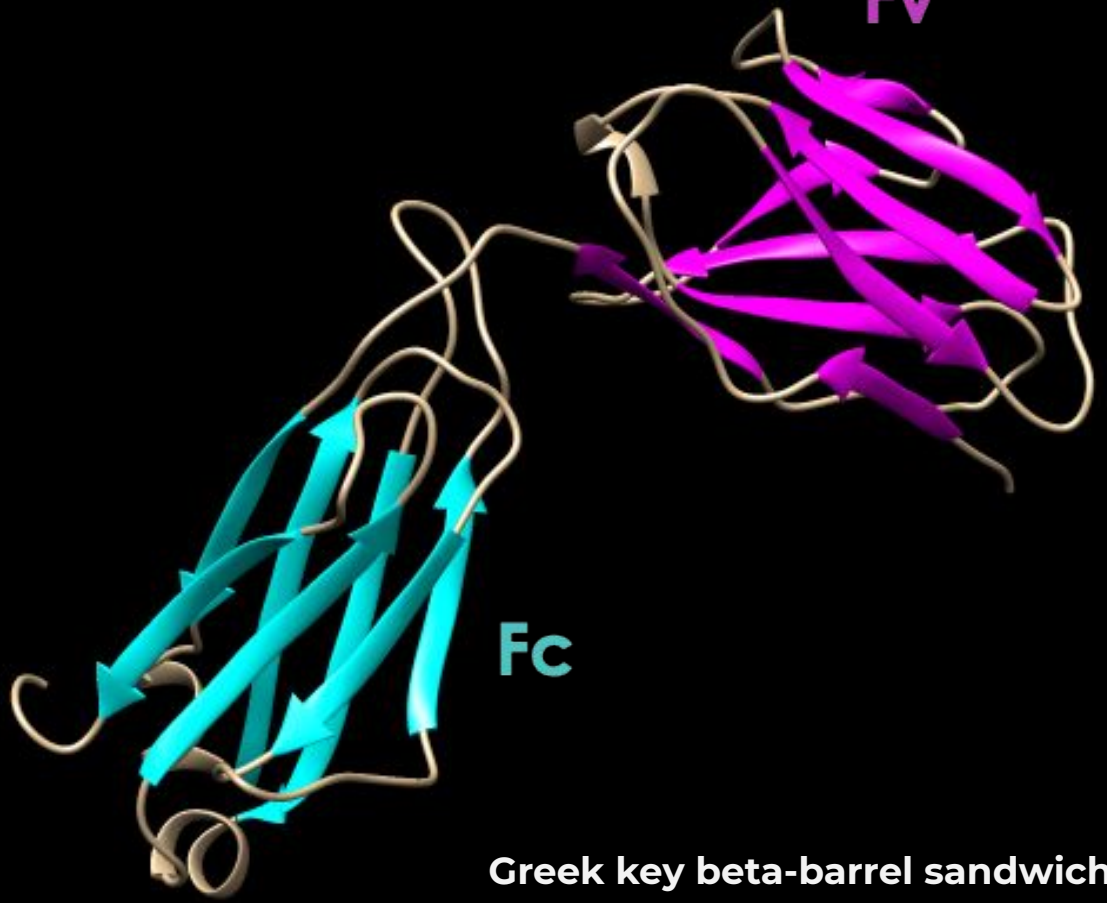


1IGT

IgG

Structure: Fold



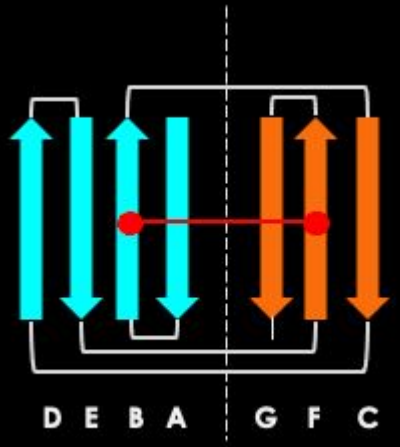


1IGT

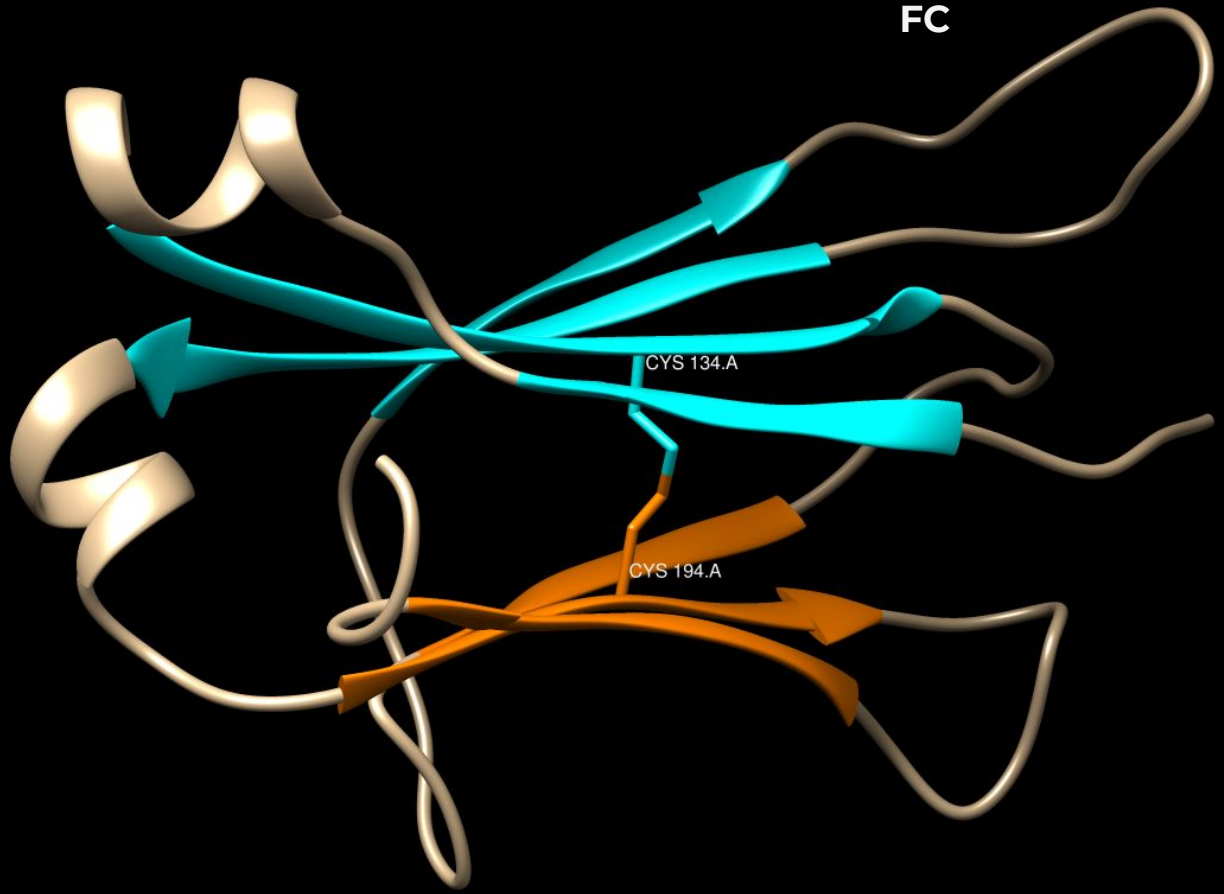
Greek key beta-barrel sandwich

IgG

Structure: Fold



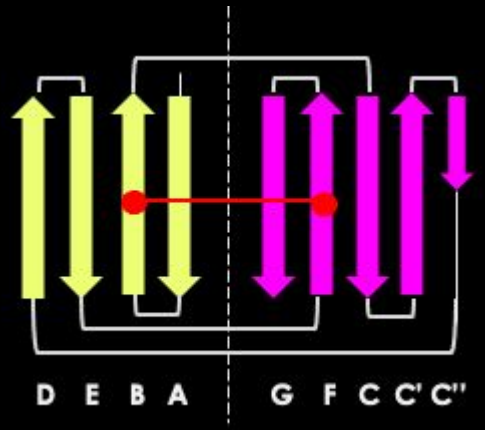
FC



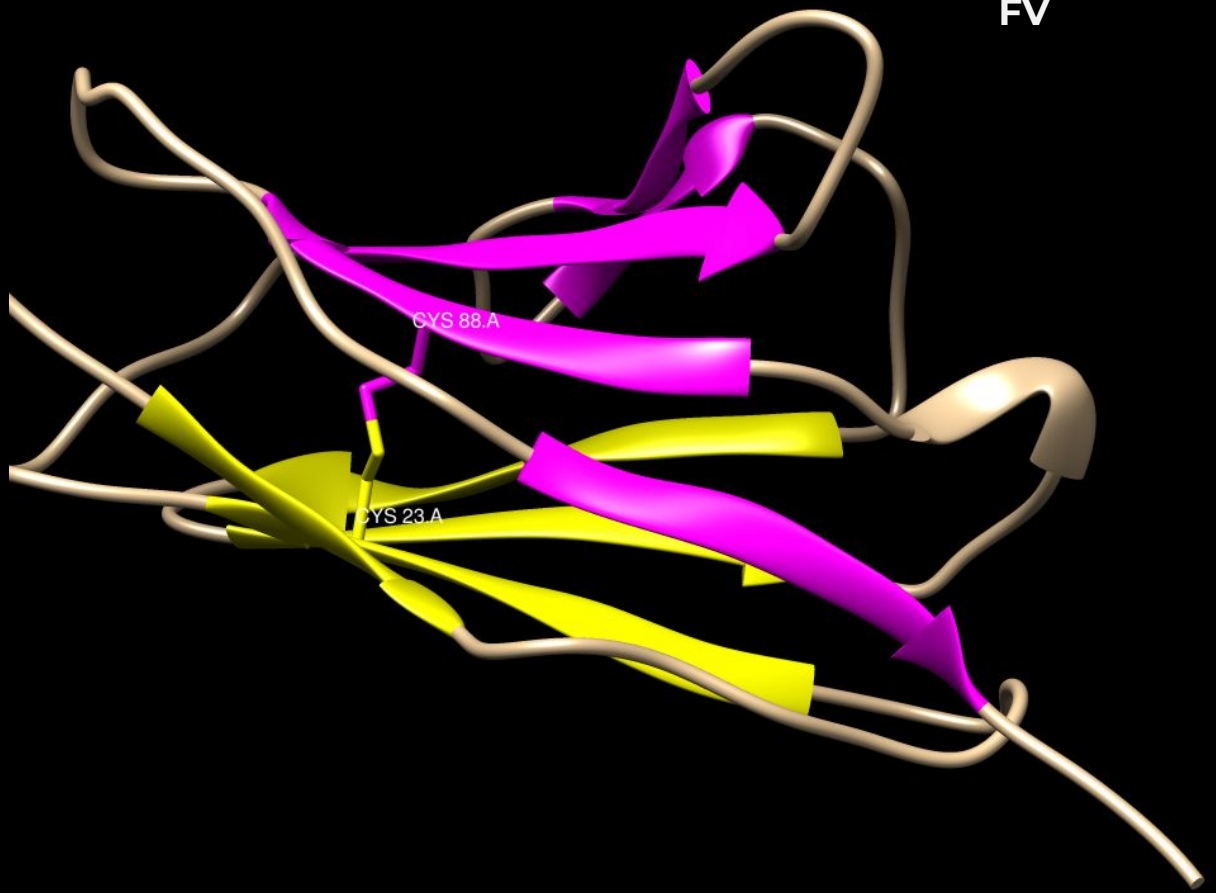
1IGT

IgG

Structure: Fold



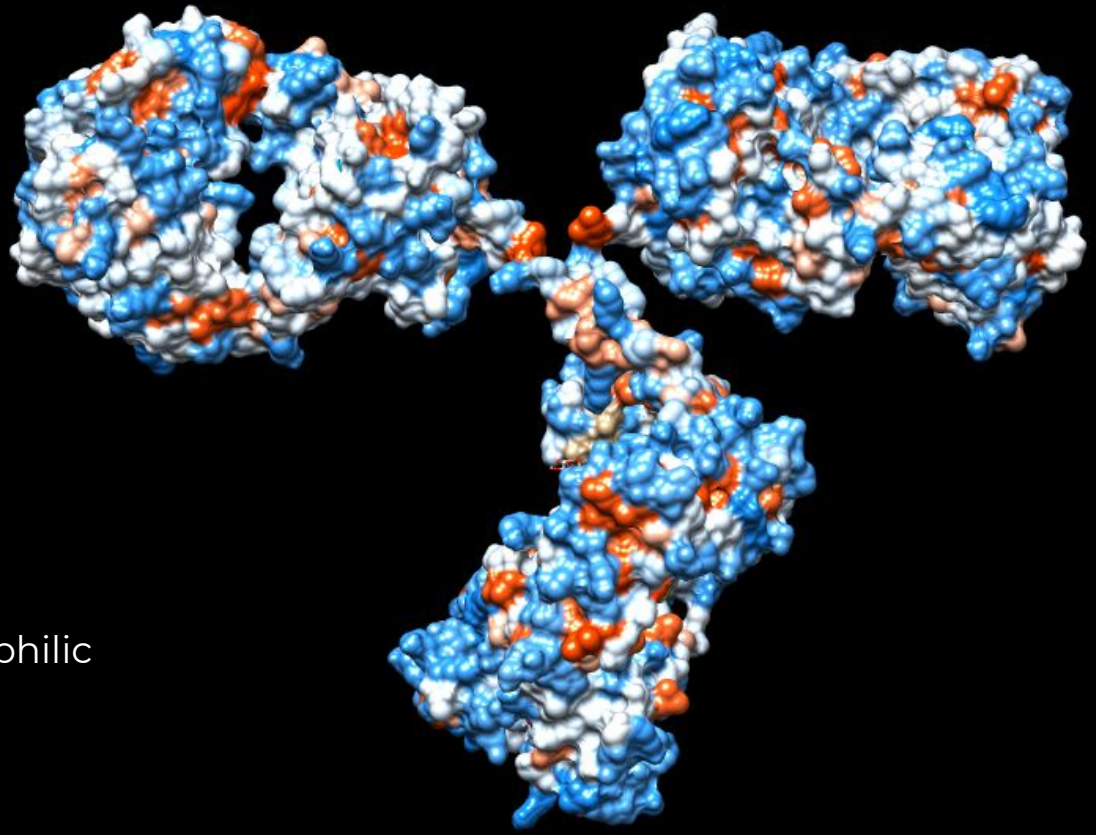
FV



1IGT

IgG

Structure: Architecture

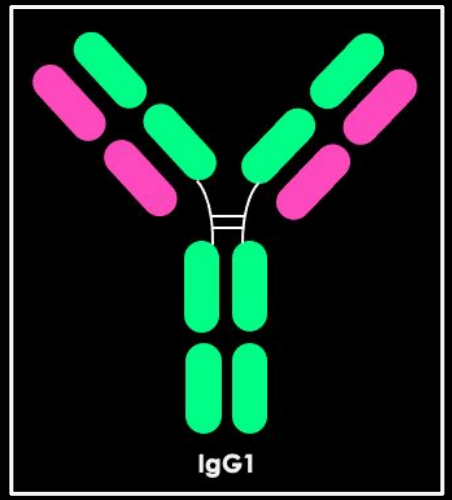


Hydrophobic Hydrophilic

1IGT

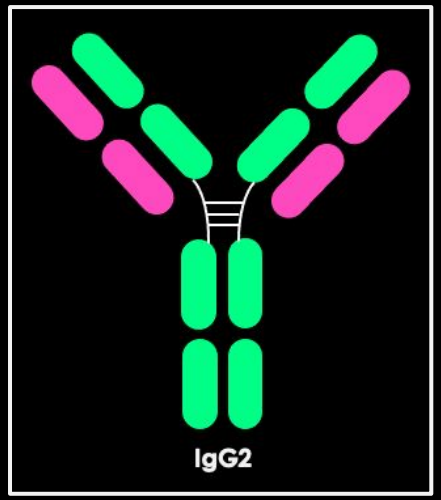
IgG

Hinge: Subtypes



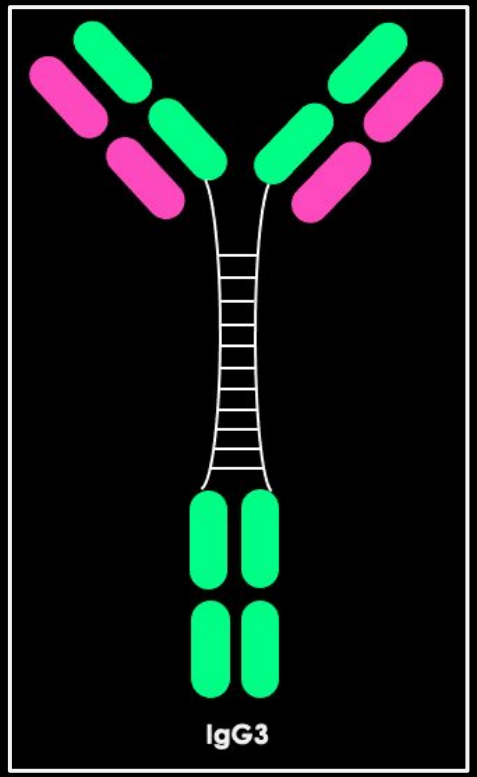
2 disulfide bonds

15 amino acids



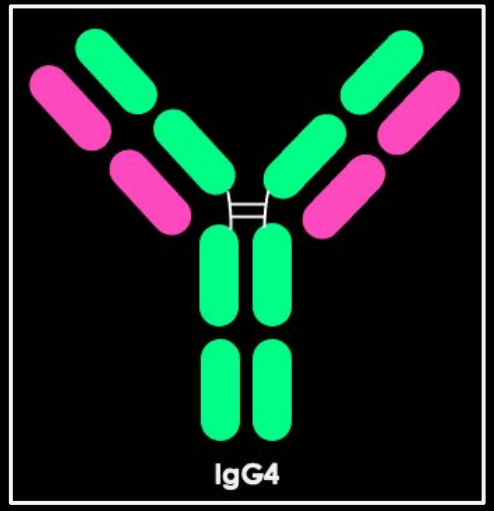
3 disulfide bonds

12 amino acids



11 disulfide bonds

62 amino acids



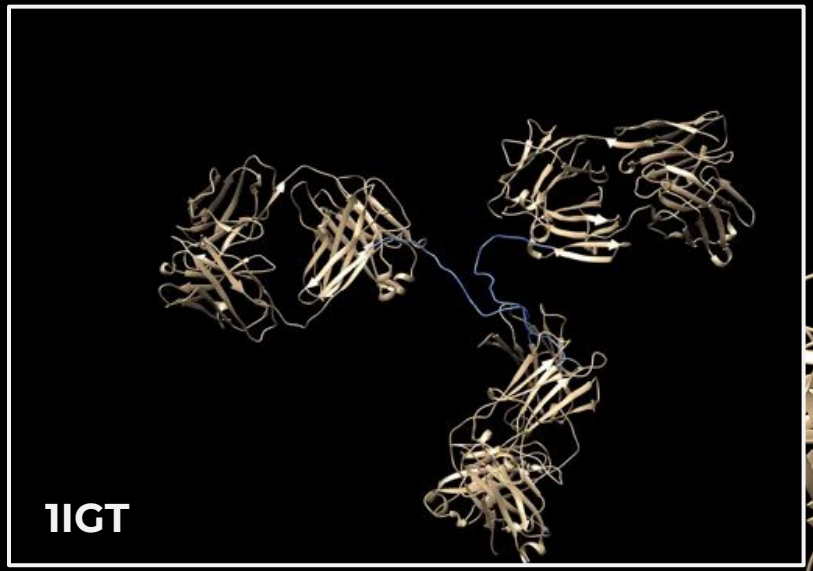
2 disulfide bonds

12 amino acids

IgG

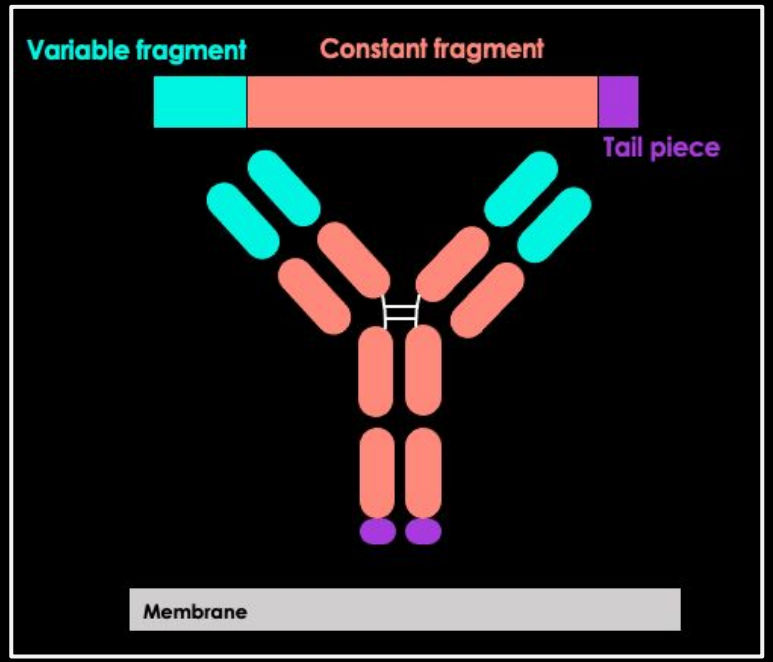
Hinge

Disulfide bonds

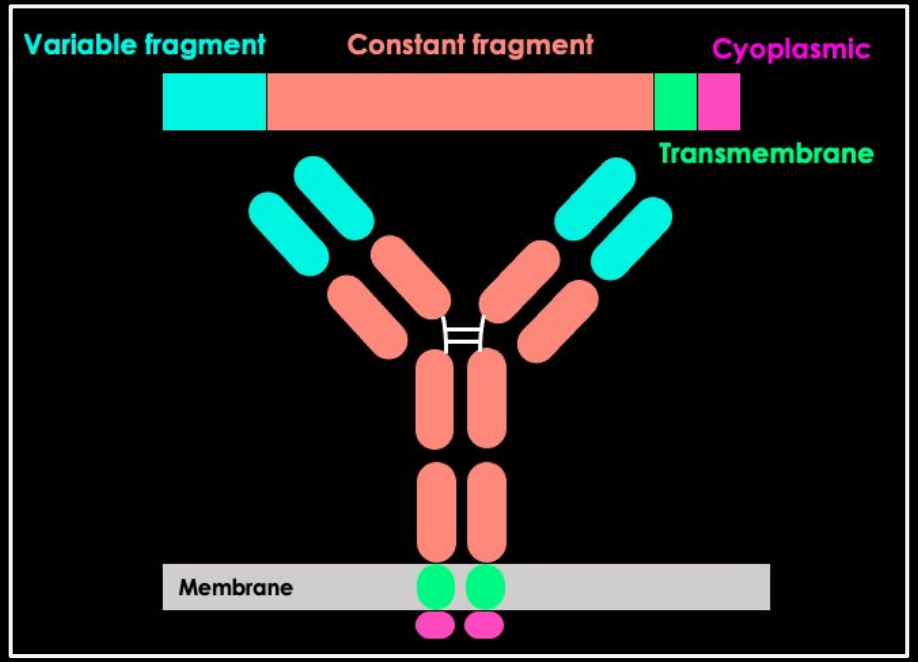


Fragments: FC

Types of Fc



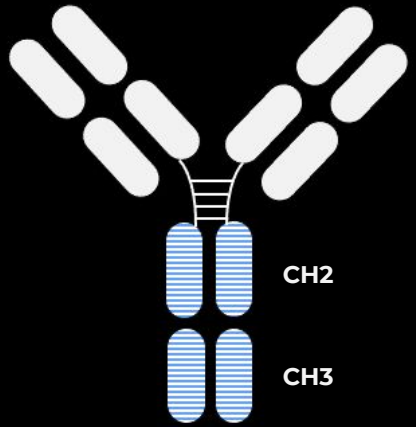
Soluble immunoglobulin



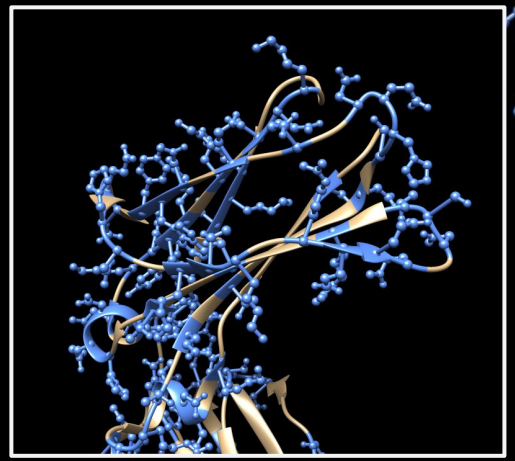
Membrane immunoglobulin

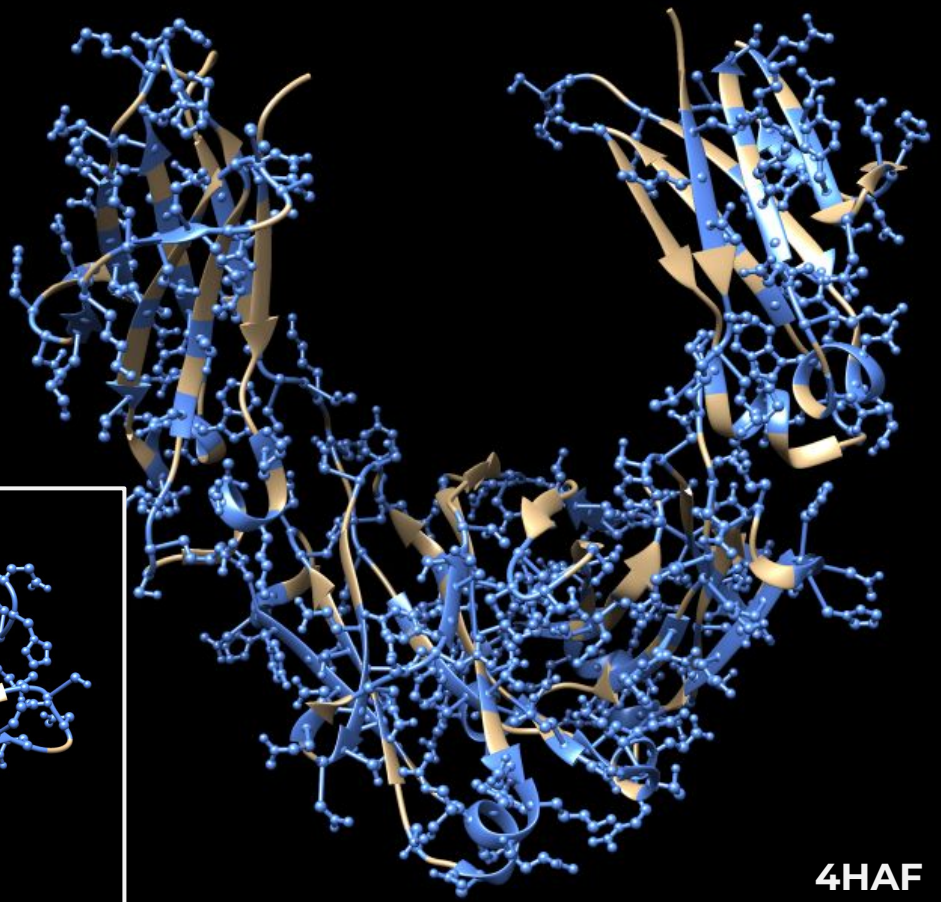
Fragments: FC

Soluble IgG



Polar residues



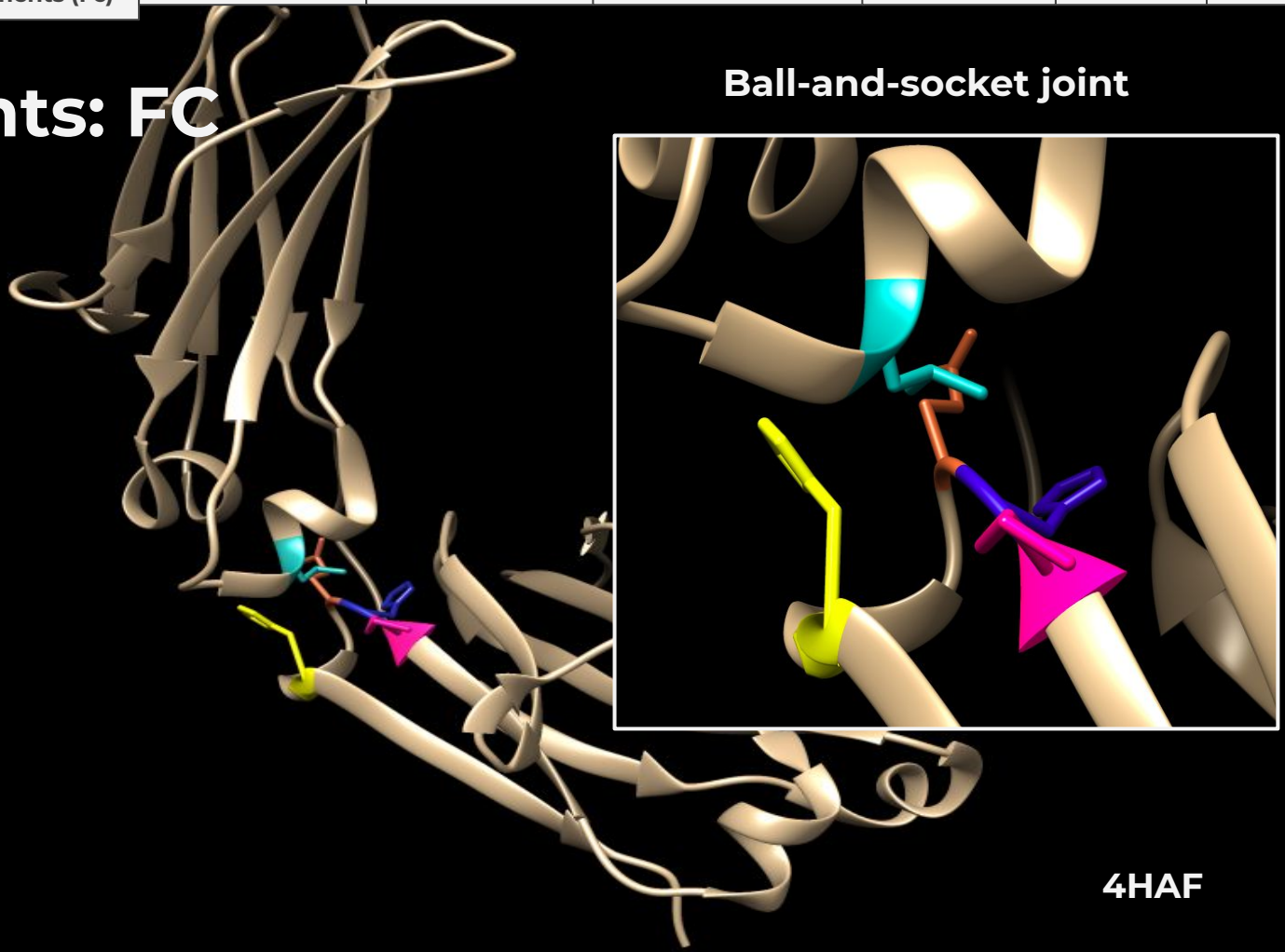


4HAF

Fragments: FC

Soluble IgG

Leu251
Met 428
His429
Glu430
His435



Ball-and-socket joint

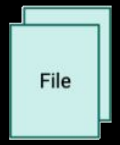
4HAF

Fragments: FC

Subtypes comparison

Downloads 4 fasta files

cat *fa



1 fasta file with the 4 seq.

Clustalw

MSA

Downloads 4 PDB files

Create file ".domains"

ROUGH STAMP

Structural alignment Superimposition

Transform → **pdb**

AconvertMod2 → **.aln**

Fragments: FC

Subtypes comparison

```
CLUSTAL 2.1 multiple sequence alignment


4HAF            VECPPCPAPPVAG-PSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVE
5W38            -TCPRCPAPELLGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFKWYVDGVE
3AVE            -TCPPCPAPELLGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVE
5LG1            ----------FLGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSQEDPEVQFNWYVDGVE
                          . *  ********************************:*****:*:*******

4HAF            VHNAKTKPREEQFNSTFRVVSVLTVVHQDWLNGKEYKCKVSNKGLPAPIEKTISKTKGQP
5W38            VHNAKTKPREEQFNSTFRVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTISKTKGQP
3AVE            VHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTISKAKGQP
5LG1            VHNAKTKPREEQFNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKGLPSSIEKTISKAKGQP
                ***********:***:********:****************.**:.*******:****

4HAF            REPQVYTLPPSREEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPMLDSDGS
5W38            REPQVYTLPPSREEMTKNQVSLTCLVKGFYPSDIAVEWESSGQPENNYKTTPPMLDSDGS
3AVE            REPQVYTLPPSRDELTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPVLDSDGS
5LG1            REPQVYTLPPSQEEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPVLDSDGS
                **********::*:************************.************:******

4HAF            FFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGK
5W38            FFLYSKLTVDKSRWQQGNIFSCSVMHEALHNRFTQKSLSLSPGK
3AVE            FFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGK
5LG1            FFLYSRLTVDKSRWQEGNVFSCSVMHEALHNHYTQKSLSLSLGK
                ****:********:**:***********::******** **
```

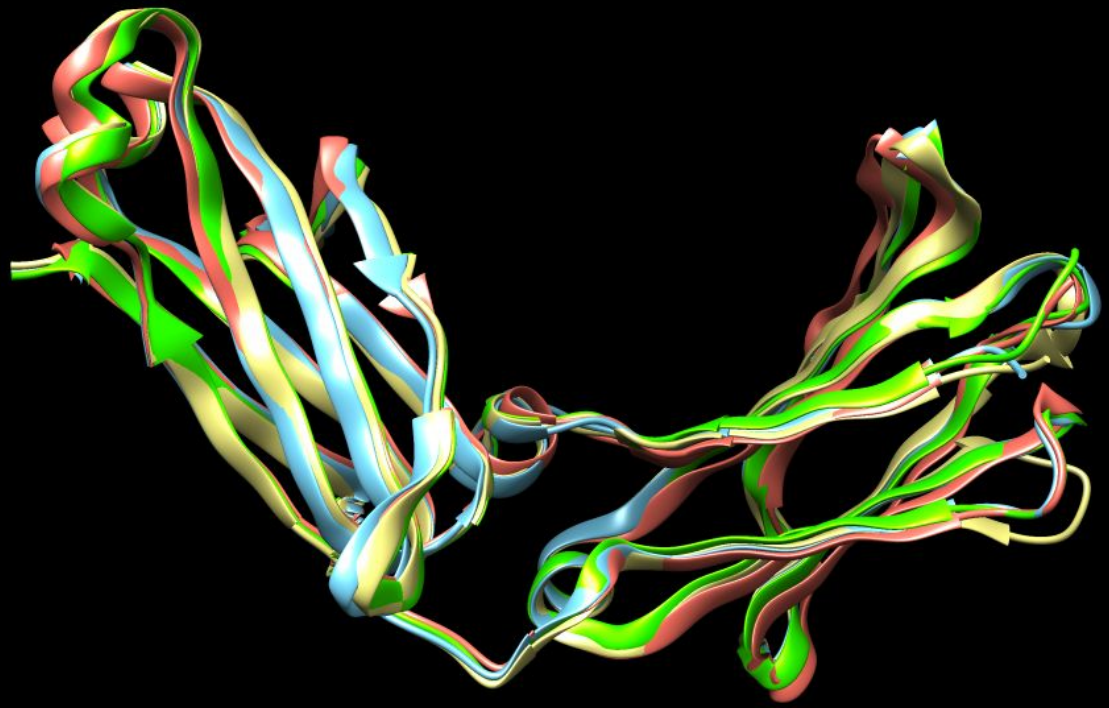
Sequence alignment

- `*` High conserved residues
- `:` Conserved residues
- `.` Low conserved residues, but same physico-chemical properties
- No conserved residues

Fragments: FC

Subtypes comparison

IgG1: 3ave
IgG2: 4haf
IgG3: 5w38
IgG4: 5lg1



RMSD: 0.80 Å
SC: 9.94

Fragments: FC

Subtypes comparison

IgG1: 3ave
IgG2: 4haf
IgG3: 5w38
IgG4: 5lg1

```
CLUSTAL W(1.60) multiple sequence alignment

5lg1        --GGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSQEDPEVQFNWYVDGVEVHNAKTKPRE
5w38        -LGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFKWYVDGVEVHNAKTKPRE
3ave        LLGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPRE
4haf        --AGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVEVHNAKTKPRE

5lg1        EQFNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKGL-PSSIEKTISKAKGQPREPQVYTLP
5w38        EQFNSTFRVVSVLTVLHQDWLNGKEYKCKVSN-KALPAPIEKTISKTKGQPREPQVYTLP
3ave        EQYNSTYRVVSVLTVLHQDWLNGKEYKCKVSN-KALPAPIEKTISKAKGQPREPQVYTLP
4haf        EQFNSTFRVVSVLTVVHQDWLNGKEYKCKVSN-KGLPAPIEKTISKTKGQPREPQVYTLP

5lg1        PSQEEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPVLDSDGSFFLYSRLTV
5w38        PSREEMTKNQVSLTCLVKGFYPSDIAVEWESSGQPENNYKTTPPMLDSDGSFFLYSKLTV
3ave        PSRDELTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPVLDSDGSFFLYSKLTV
4haf        PSREEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPMLDSDGSFFLYSKLTV

5lg1        DKSRWQEGNVFSCSVMHEALHNHYTQKSLSLS
5w38        DKSRWQQGNIFSCSVMHEALHNRFTQKSLSL-
3ave        DKSRWQQGNVFSCSVMHEALHNHYTQKSLSLS
4haf        DKSRWQQGNVFSCSVMHEALHNHYTQKSLSLS
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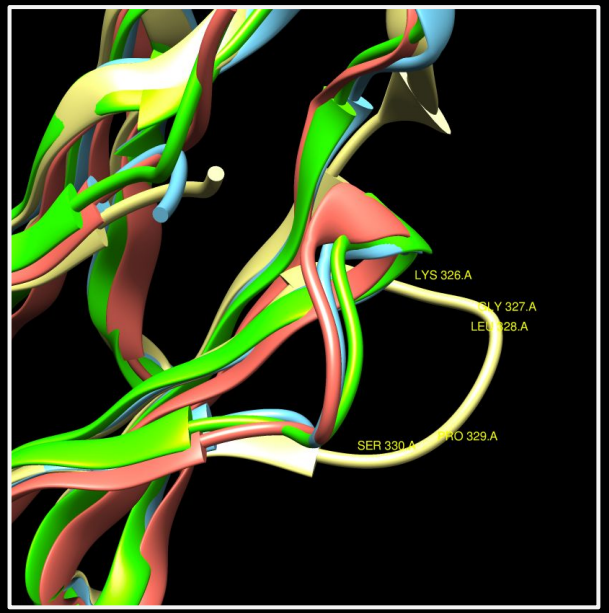


Less conserved region

Fragments: FC

Subtypes comparison

Leu251
Glu430 Met 428
His435 His429

IgG1: 3ave
IgG2: 4haf
IgG3: 5w38
IgG4: 5lg1

```
CLUSTAL W(1.60) multiple sequence alignment

5lg1        --GGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSQEDPEVQFNWYVDGVEVHNAKTKPRE
5w38        -LGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFKWYVDGVEVHNAKTKPRE
3ave        LLGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPRE
4haf        --AGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVEVHNAKTKPRE

5lg1        EQFNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKGL-PSSIEKTISKAKGQPREPQVYTLP
5w38        EQFNSTFRVVSVLTVLHQDWLNGKEYKCKVSN-KALPAPIEKTISKTKGQPREPQVYTLP
3ave        EQYNSTYRVVSVLTVLHQDWLNGKEYKCKVSN-KALPAPIEKTISKAKGQPREPQVYTLP
4haf        EQFNSTFRVVSVLTVVHQDWLNGKEYKCKVSN-KGLPAPIEKTISKTKGQPREPQVYTLP

5lg1        PSQEEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPVLDSDGSFFLYSRLTV
5w38        PSREEMTKNQVSLTCLVKGFYPSDIAVEWESSGQPENNYKTTPPMLDSDGSFFLYSKLTV
3ave        PSRDELTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPVLDSDGSFFLYSKLTV
4haf        PSREEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPMLDSDGSFFLYSKLTV

5lg1        DKSRWQEGNVFSCSVMHEALHNHYTQKSLSLS
5w38        DKSRWQQGNIFSCSVMHEALHNRFTQKSLSL-
3ave        DKSRWQQGNVFSCSVMHEALHNHYTQKSLSLS
4haf        DKSRWQQGNVFSCSVMHEALHNHYTQKSLSLS
```

Conserved residues: Ball-and-socket joint

Fragments: FC

Evolutive study

PSI BLAST → File

pdb & fasta files

Create file ".domains"

cat *fa → File

1 fasta file with the 5 seq.

Clustalw →

MSA

ROUGH STAMP →

Structural alignment Superimposition

Transform → **pdb**

AconvertMod2 → **.aln**

Fragments: FC

Evolutive study

Superimposition

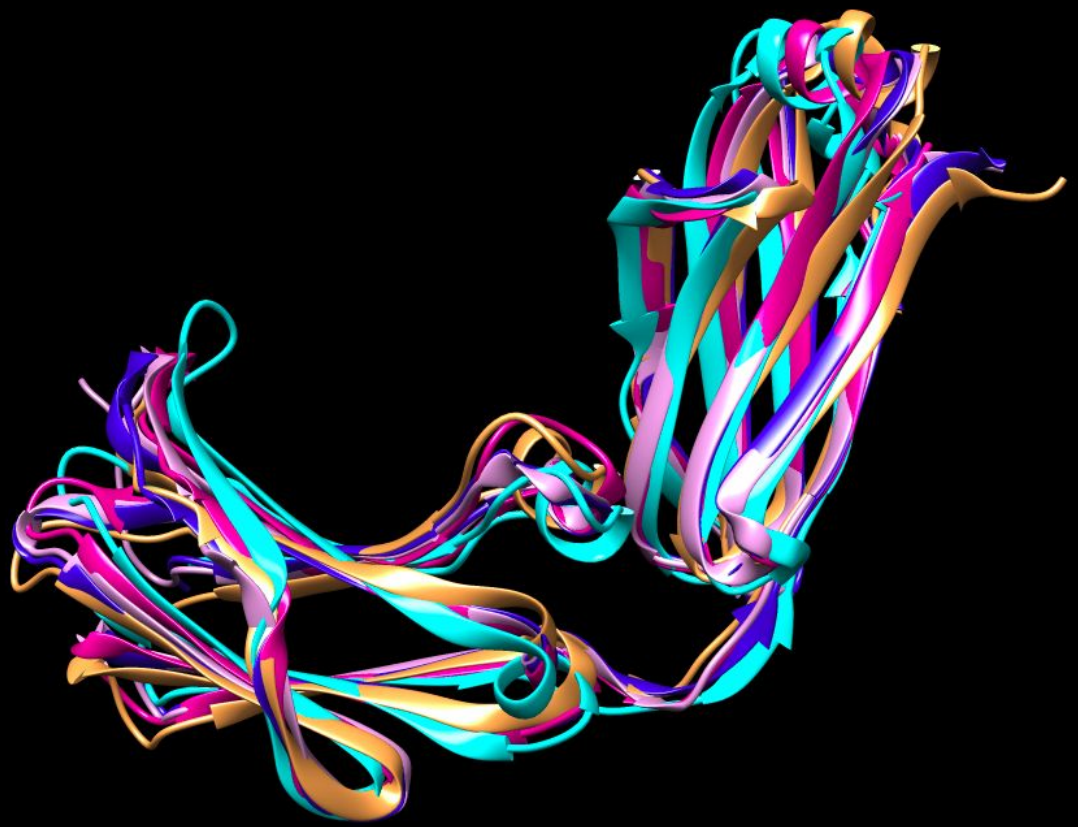
1l1c: Rattus norvegicus

2rgs: Mus musculus

4haf: Homo sapiens

2vuo: Oryctolagus cuniculus

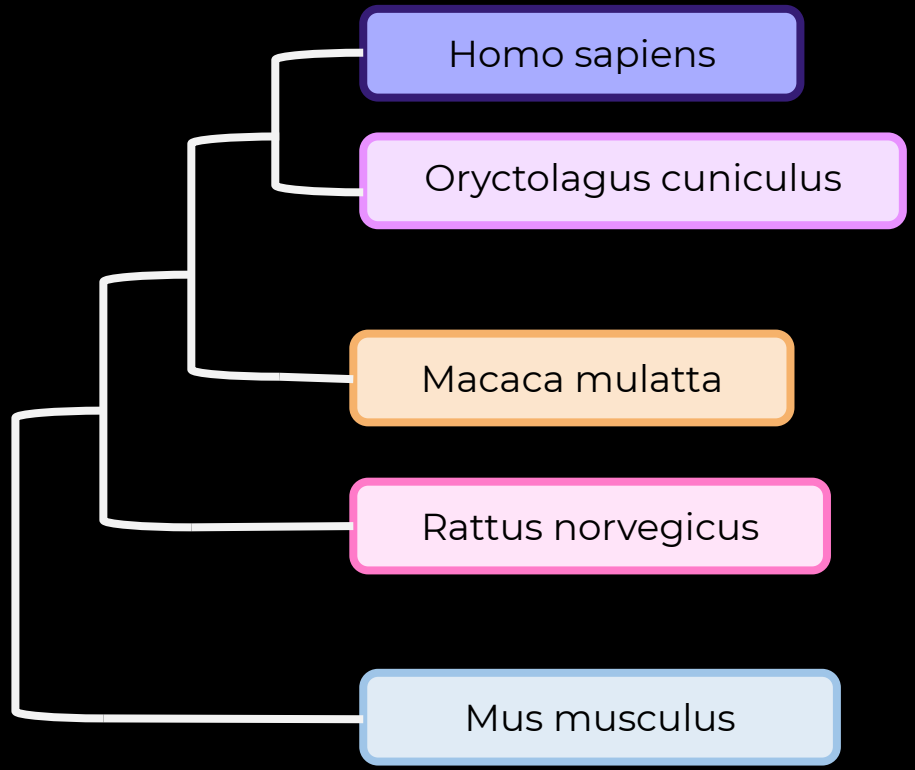
6d4i: Macaca mulatta



RMSD: 2.03 Å
SC: 9.26

Fragments: FC

Evolutive study



Fragments: FC

Evolutive study

1I1c: Rattus norvegicus
2rgs: Mus musculus
4haf: Homo sapiens
2vuo: Oryctolagus cuniculus
6d4i: Macaca mulatta

```
2RGS       -------GPSVFIFPPNIKDVLMISLTPKVTCVVVDVSEDDPDVQISWFVNNVEVHTAQT
1I1C       ---------SVFIFPPKTKDVLGGGLTPKVTCVVVDISQNDPEVRFSWFIDDVEVHTAQT
6D4I       -------GPSVFLFPPKPKDTLMISRTPEVTCVVVDVSQEEPDVKFNWYVDGVEVHNAQT
4HAF       ------AGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVEVHNAKT
2VUO       PPPELLGGPSVFIFPPKPKDTLMISRTPEVTCVVVDVSQDDPEVQFTWYINNEQVRTARP

2RGS       Q-THREDYNSTIRVVSTLPIQHQDWMSGKEFKCKVNNKDLPSPIERTISKIKGLVRAPQV
1I1C       HAPEKQ-SNSTLRSVSELPIVERDWLNGKTFKCKVNSGAFPAPIEKSISKPEGTPRGPQV
6D4I       KPREEQ-FNSTYRVVSVLTVTHQDWLNGKEYTCKVSNKALPAPRQKTVSKTKGQPREPQV
4HAF       KPREEQ-FNSTFRVVSVLTVVHQDWLNGKEYKCKVSNKGLPAPIEKTISKTKGQPREPQV
2VUO       PLREQQ-FNSTIRVVSTLPIAHQDWLRGKEFKCKVHNKALPAPIEKTISKARGQPLEPKV

2RGS       YILPPPAEQLSRKDVSLTCLVVGFNPGDISVEWTSNGHTEENYKDTAPVLDSDGSYFIYS
1I1C       YTMAPPKEEMTQSQVSITCMVKGFYPPDIYTEWKMNGQPQENYKNTPPTMDTDGSYFLYS
6D4I       YTLPPPREELTKNQVSLTCLVKGFYPSDIVVEWASNGQPENTYKTTPPVLDSDGSYFLYS
4HAF       YTLPPSREEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPMLDSDGSFFLYS
2VUO       YTMGPPREELSSRSVSLTCMINGFYPSDISVEWEKNGKAEDNYKTTPAVLDSDGSYFLYS

2RGS       KLNMKTSKWEKTDSFSCNVRHEGLKNYYLKKTIS---
1I1C       KLNVKKETWQQGNTFTCSVLHEGLENEHTEKSLSH--
6D4I       KLTVDKSRWQQGNTFSCSVMHEALHNHYTQKSLSLSP
4HAF       KLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLS-
2VUO       KLSVPTSEWQRGDVFTCSVMHEALHNHYTQKSISRS-
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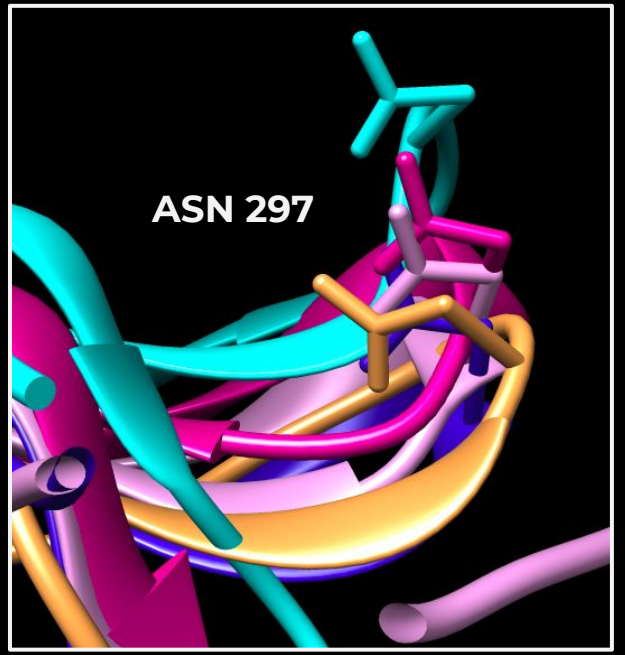
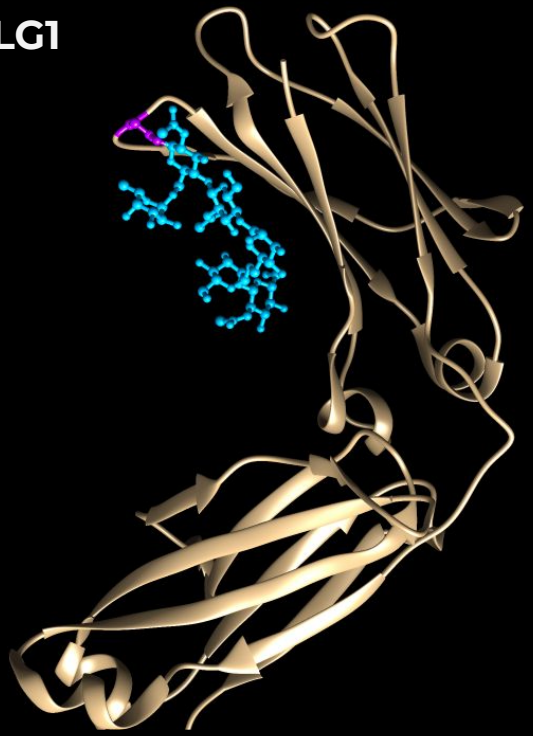
Conserved residue for N-Glycosylation



Fragments: FC

N-Glycosylation

5LG1



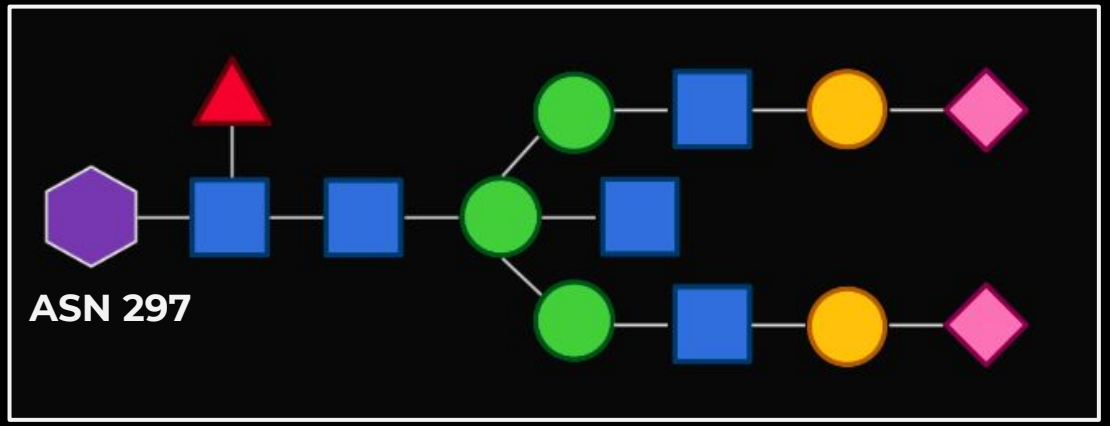
NAG: N-acetyl glucosamine

MAN: Mannose

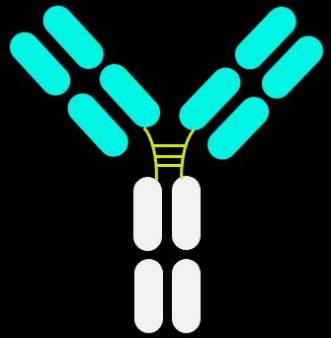
GAL: Galactose

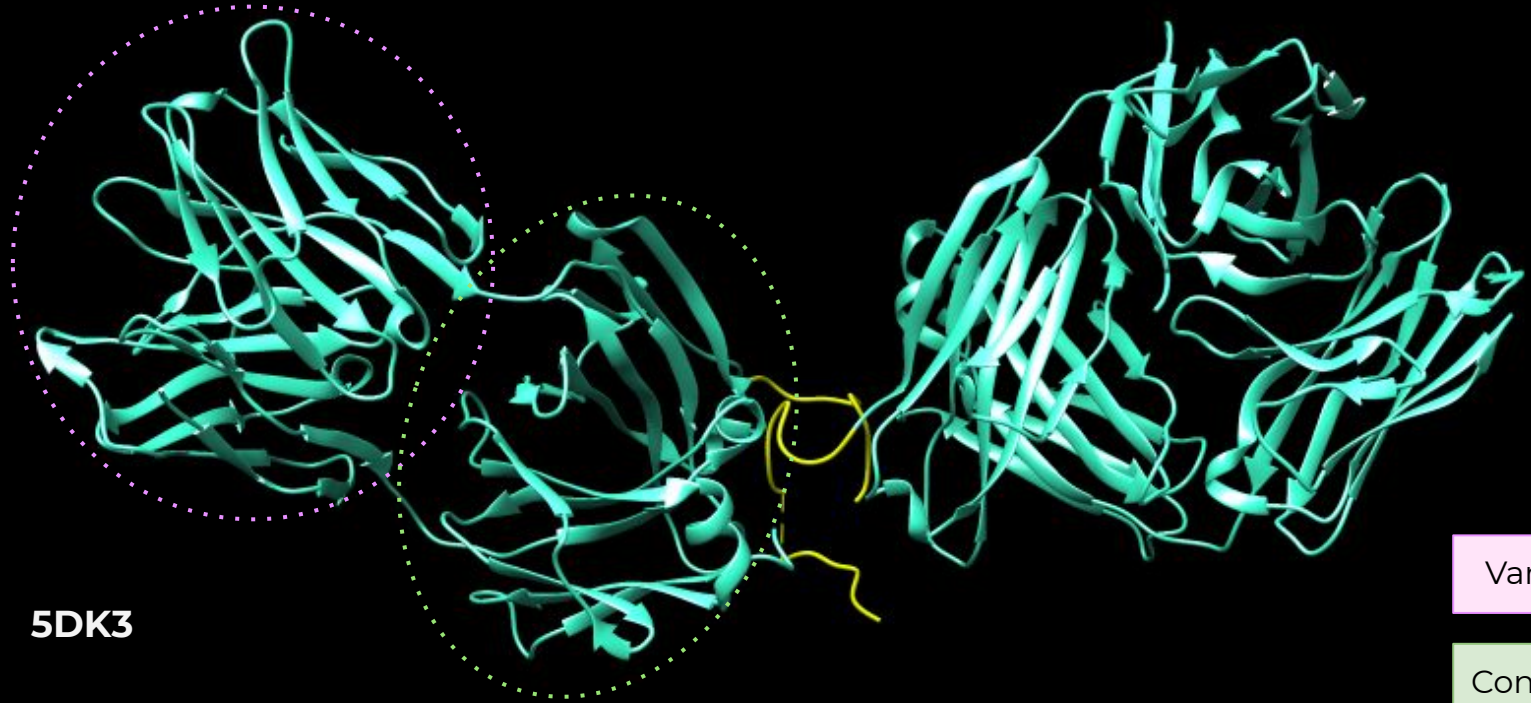
FUC: Fucose

SA: Sialic acid



Fragments: Fab



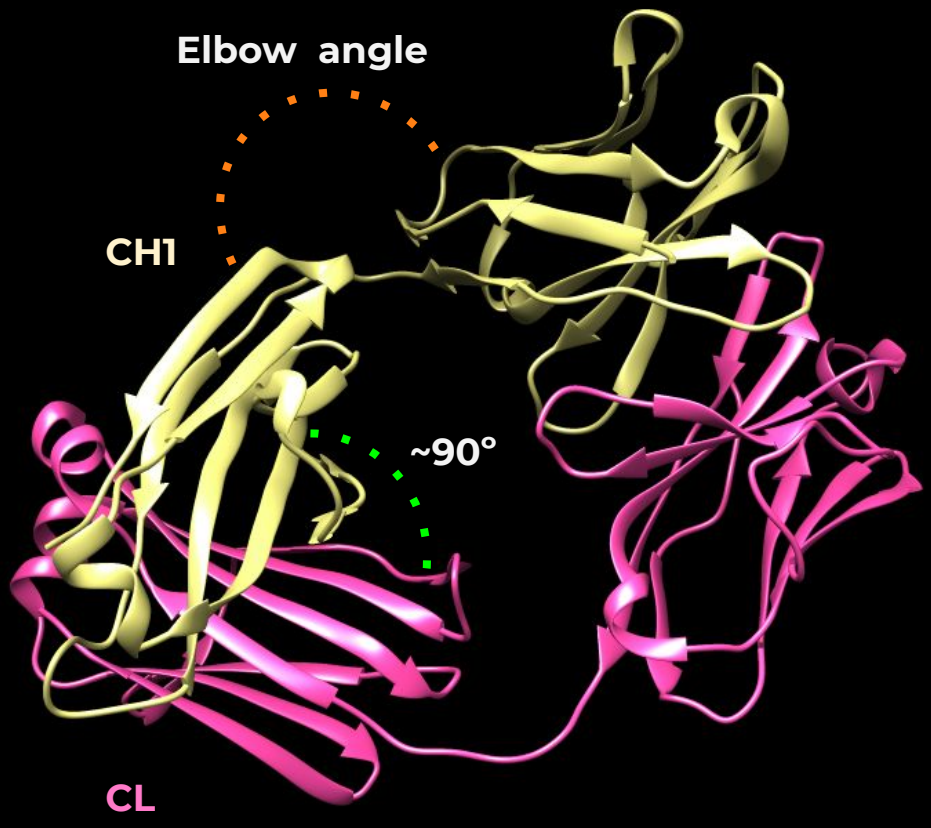


5DK3

Variable domain

Constant domain

Fragments: Fab

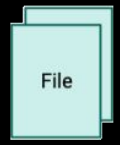


5DK3

Fragments: Fab

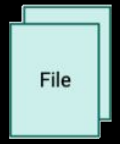
Evolutive study

PSI BLAST



pdb & fasta files

cat *fa



1 fasta file with the 4 seq.

Clustalw

MSA

Create file ".domains"

ROUGH STAMP

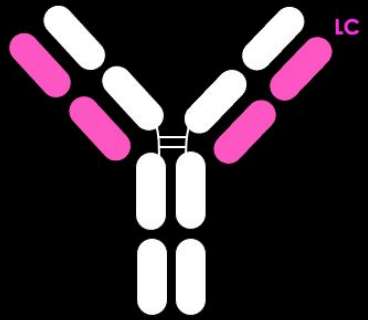
Structural alignment Superimposition

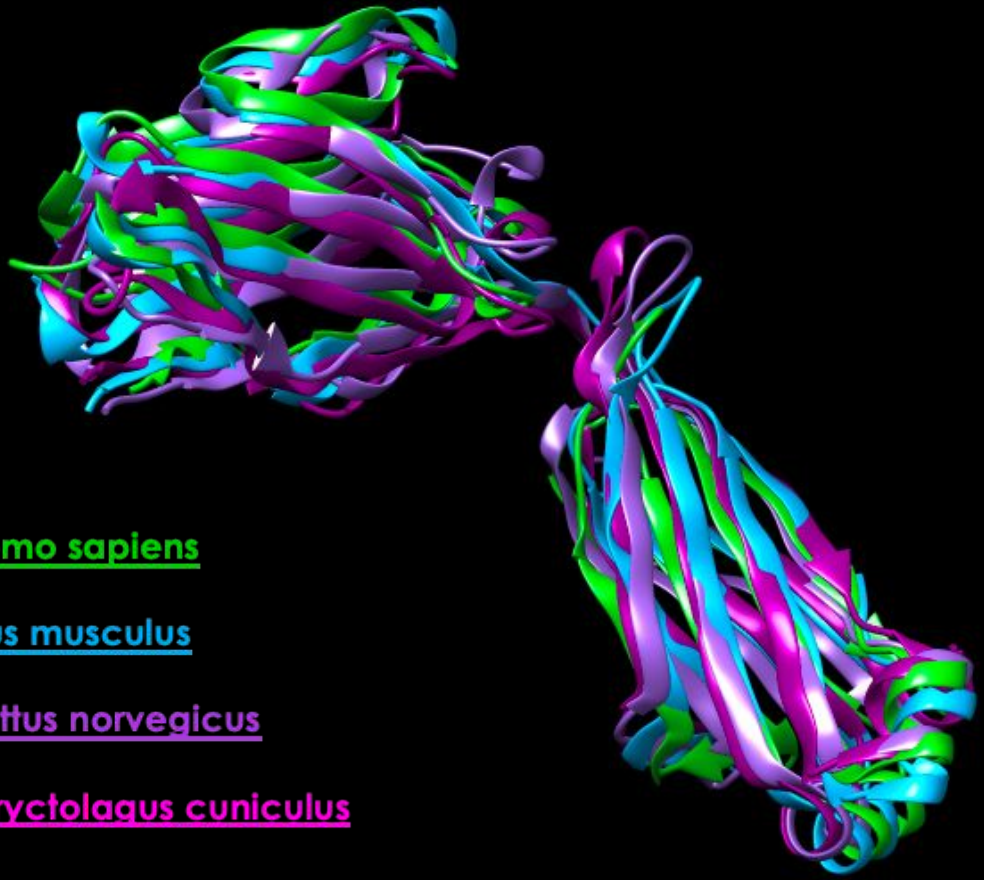
Transform → **pdb**

AconvertMod2 → **.aln**

Fragments: Fab

Evolutive study: LC





5whj: Homo sapiens

1gig: Mus musculus

1zan: Rattus norvegicus

4hbc: Oryctolagus cuniculus

RMSD: 2.16 Å
SC: 8.55

Fragments: Fab

Evolutive study: LC

5whj: Homo sapiens

1gig: Mus musculus

1zan: Rattus norvegicus

4hbc: Oryctolagus cuniculus

```
1gig        QAVVTQE-SALTTSPGETVTLTCRSSTG--AVTTSNYANWVQEKPDHLFTGLIGGTNNRA
5whj        -SALTQP-ASVSGSPGQSITISCTG-TGSDVGSYN-LVSWYQQHPGKAPKLMIYGDSQRP
1zan        DIQMTQSPASLSASLGETVTIECRASED---IYN--ALAWYQQKPGKSPQLLIYNTDTLH
4hbc        DVVMTQTPASVSEPVGGTVTIKCQASQS---ISS--YLAWYQQKPGQRPRLLIYETSTLA

1gig        PGVPARFSGSLIGDKAALTITGAQTEDEAIYFCALWY-S-NH-WVFGGGTKLTVLGQPKS
5whj        SGVSNRFSGSKSGNTASLTISGLQAEDEADYYCASYA-GSGI-YVFGTGTKVTVLGQPKA
1zan        TGVPSRFSGSGSGTQYSLKINSLQSEDVASYFCQHYF-G-YP-RTFGGGTKLELK-RADA
4hbc        SGVPSRFKGSGSGTDFTLTISDLECADAATYYCQSTYEN-PTYVSFGGGTEVGVK-GDPV

1gig        SPSVTLFPPSSEELETNKATLVCTITDFYPGVVTVDWKVDGTPVTQGMETTQPS-KQSN-
5whj        NPTVTLFPPSSEELQANKATLVCLISDFYPGAVTVAWKADGSPVKAGVETTKPS-KQSN-
1zan        APTVSIFPPSSEQLASGGASVVCLLNNFYPKDISVKWKIDGSERQNGVLDS-VTDQDSKD
4hbc        APTVLIFPPSADLVATGTVTIVCVANKYFP-DVTVTWEVDGTTQTTGIENS-KTPQNSAD

1gig        NKYMASSYLTLTARAWERHSSYSCQVTHE-GH-TVEKSLS----
5whj        NKYAASSYLSLTPEQWKSHRSYSCQVTHE-GS-TVEKTVAP---
1zan        STYSMSSTLTLTKAEYESHNSYTCEVTHKTSTSPVVKSFNRGE-
4hbc        CTYNLSSTLTLTSTEYNSHKEYTCKVTQG-TT-SVVQSFNRGDC
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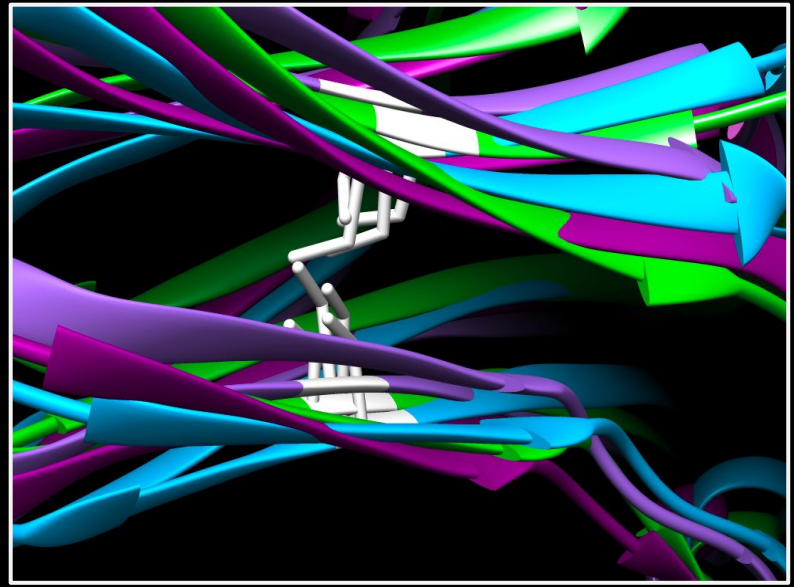
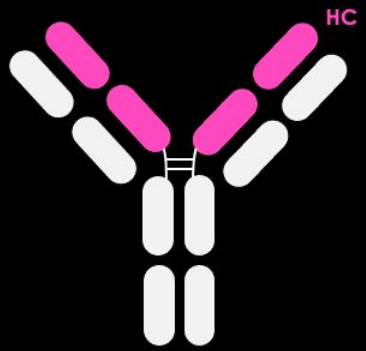
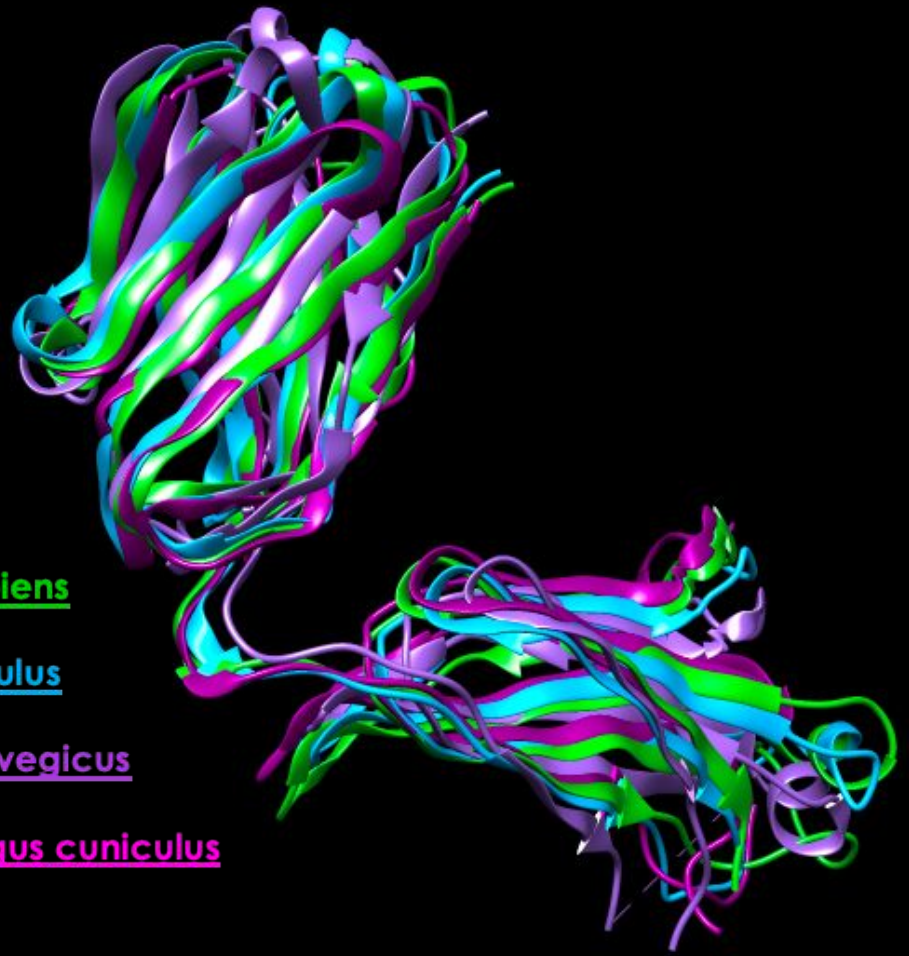


Conserved cysteines

Fragments: Fab

Evolutive study: HC





5whj: Homo sapiens

1gig: Mus musculus

1zan: Rattus norvegicus

4hbc: Oryctolagus cuniculus

RMSD: 2.93 Å
SC: 8.30

Fragments: Fab

Evolutive study: HC

5whj: Homo sapiens

1gig: Mus musculus

1zan: Rattus norvegicus

4hbc: Oryctolagus cuniculus

```
CLUSTAL W(1.60) multiple sequence alignment

1zan        QVQLKESGPGLVQPSQTLSLTCTVSGFSLTNNNVNWVRQATGRGLEWMGGVWAGG-ATDY
4hbc        -QSVEESGGRLVTPGTPLTLACTVSGFSLNTYSMFWVRQAPGKGLQWIGIISNFG-VIYY
1gig        QVQLKESGPGLVAPSQSLSITCTVSGFLLISNGVHWVRQPPGKGLEWLGVIWAGG-NTNY
5whj        EVQLLESGGGLVQPGGSLRLSCAASGFTFSEYAMGWVRQAPGKGLEWVSSIGSSGGQTKY

1zan        NSALKSRLTITRDTSKSQVFLKMHSLQSEDTATYYCARDGGYSSS-TLYAMD-AWGQGTT
4hbc        ATWAKGRFTISKT-S-TTVDLKITSPTTEDTATYFCVRKYG-----SEWGGD-LWGPGTL
1gig        NSALMSRVSISKDNSKSQVFLKMKSLQTDDTAMYYCARDFY-DYDVFYYAMD-YWGQGTS
5whj        ADSVKGRFTISRDNSKNTLYLQMNSLRAEDTAVYYCARLAI--------G-DSYWGQGTM

1zan        VTVSSASTTAPSVYPLAPG----S----MVTLGCLVKGYFPEPVTVTWNSGSLASGVHTF
4hbc        VTVSSGQPKAPSVFPLAPCCG--DT--PTVTLGCLVKGYLPEPVTVTWNSGTLTNGVRTF
1gig        VTVSSAKTTPPSVYPLAPGSA--AQTNSMVTLGCLVKGYFPEPVTVTWNSGSLSSGVHTF
5whj        VTVSSASTKGPSVFPLAPSSKSTSG--GTAALGCLVKDYFPEPVTVSWNSGALTSGVHTF

1zan        PAVLQ-SGLYTLSSSVTVPASPWASEAVTCNVAHPASSTKVDKKIV-PR-
4hbc        PSVRQSSGLYSLSSVVSVT---S---PVTCNVAHPATNTKVDKTVAPSTC
1gig        PAVLQ-SDLYTLSSSVTVPSSTWPSETVTCNVAHPASSTKVDKKIV-P--
5whj        PAVLQSSGLYSLSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKRVE-PK-
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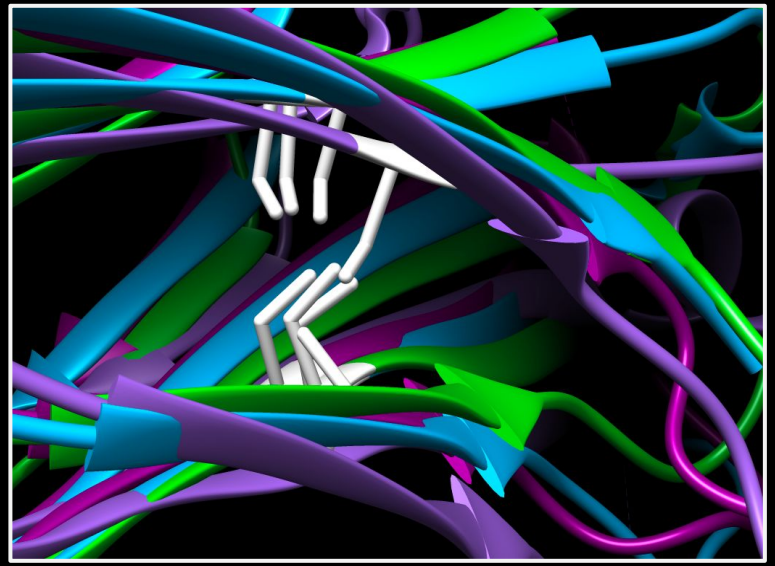
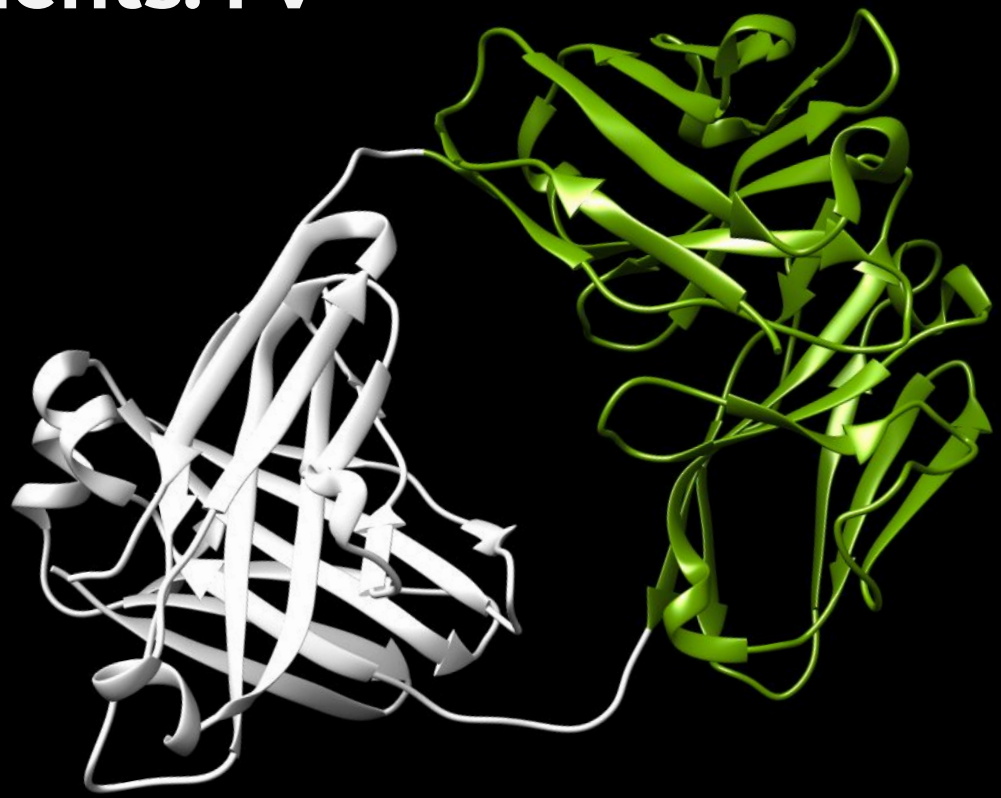
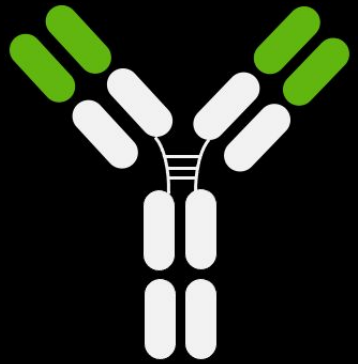


Conserved cysteines

Fragments: Fv



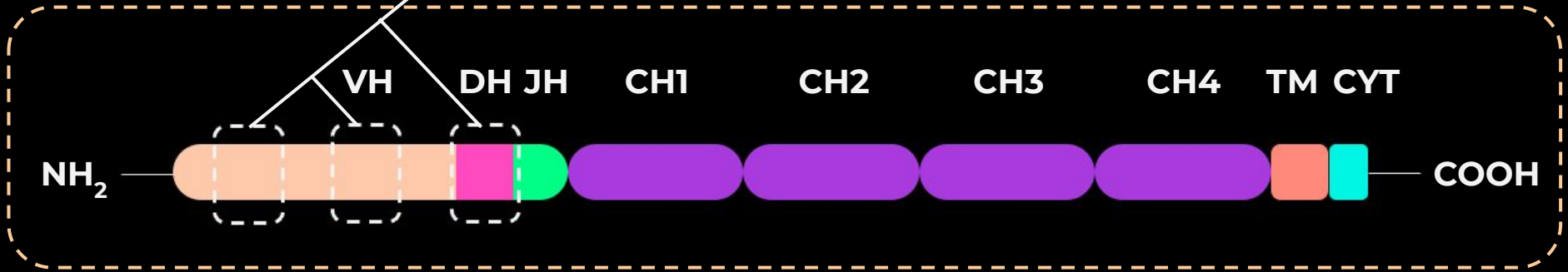


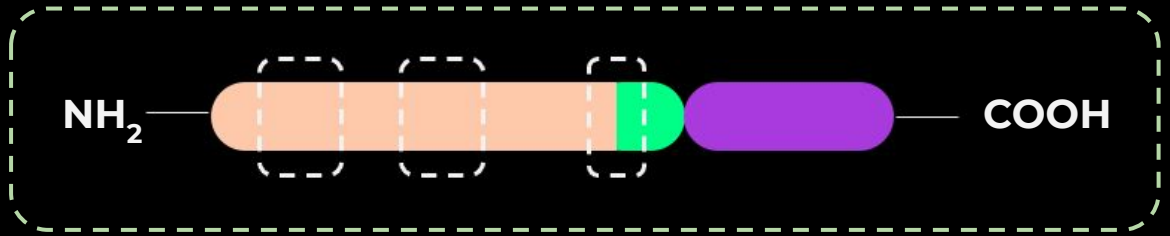
5WHJ

Fragments: Fv

CDR: Rearrangement

CDR: Hipervariable regions





Heavy chain (membrane form)

Light chain

Fragments: Fv

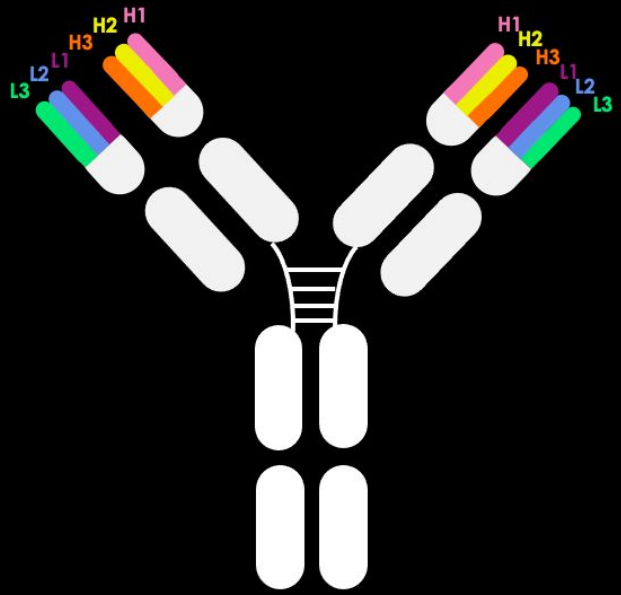
CDR's types

Light chain

	LC Kappa	LC Lambda
L1	1, **2a, 2b**, 3, 4, 5, 6	1, 2, 3a, 3b, 4
L2	**1**	
L3	**1, 2**, 3, 4, 5, 6	**1a, 1b, 1c, 2**

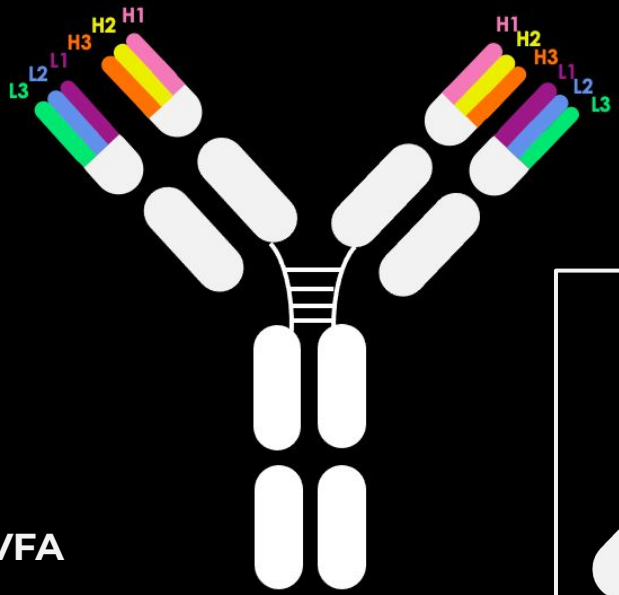
Heavy chain

	HC
H1	**1**, 2, 3
H2	1, **2a, 2b**, **3a, 3b, 3c**, 4
H3	No canonical structures

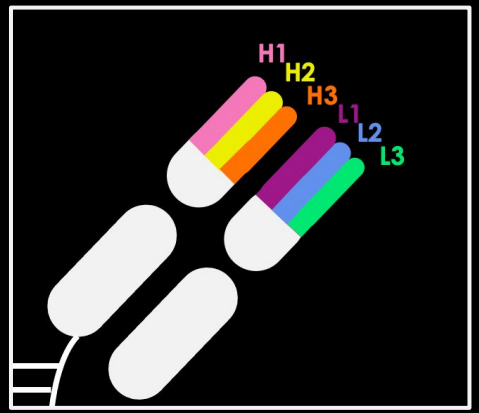


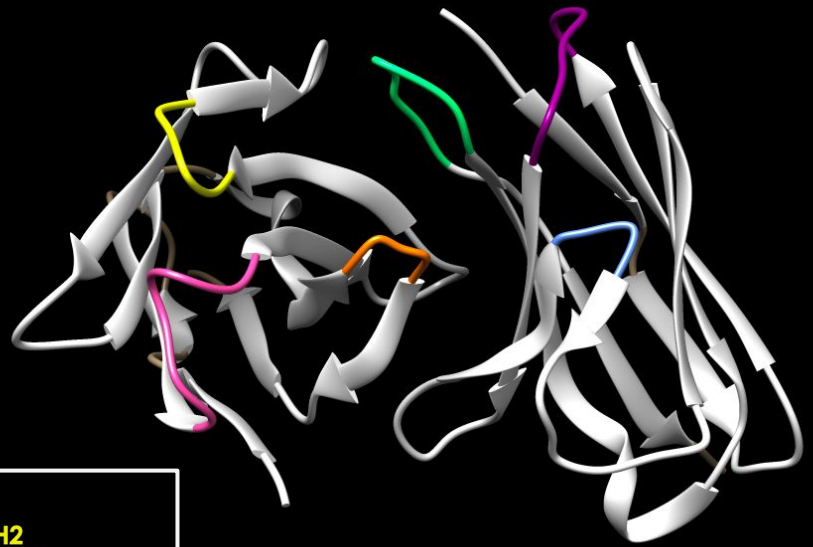
Fragments: Fv

CDR's types



1VFA





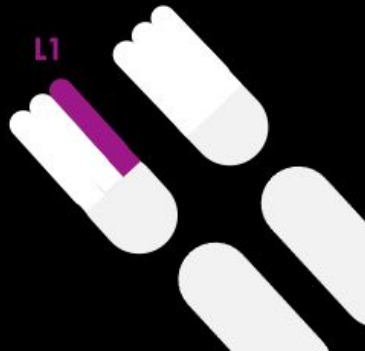
Fragments: Fv

CDR: L1-κ

2a

2a human: 1DFB

2a human: 1FVC

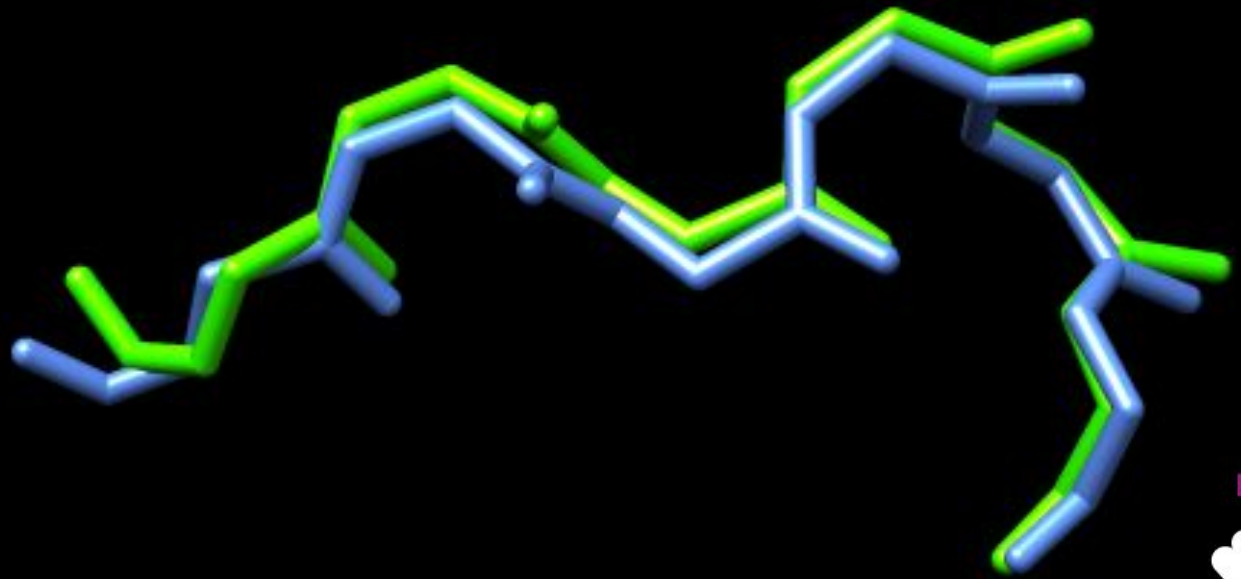


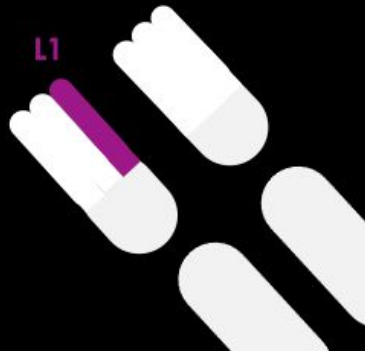
RMSD: 0.79 Å
SC: 4.59

Fragments: Fv

CDR: L1-κ

2b



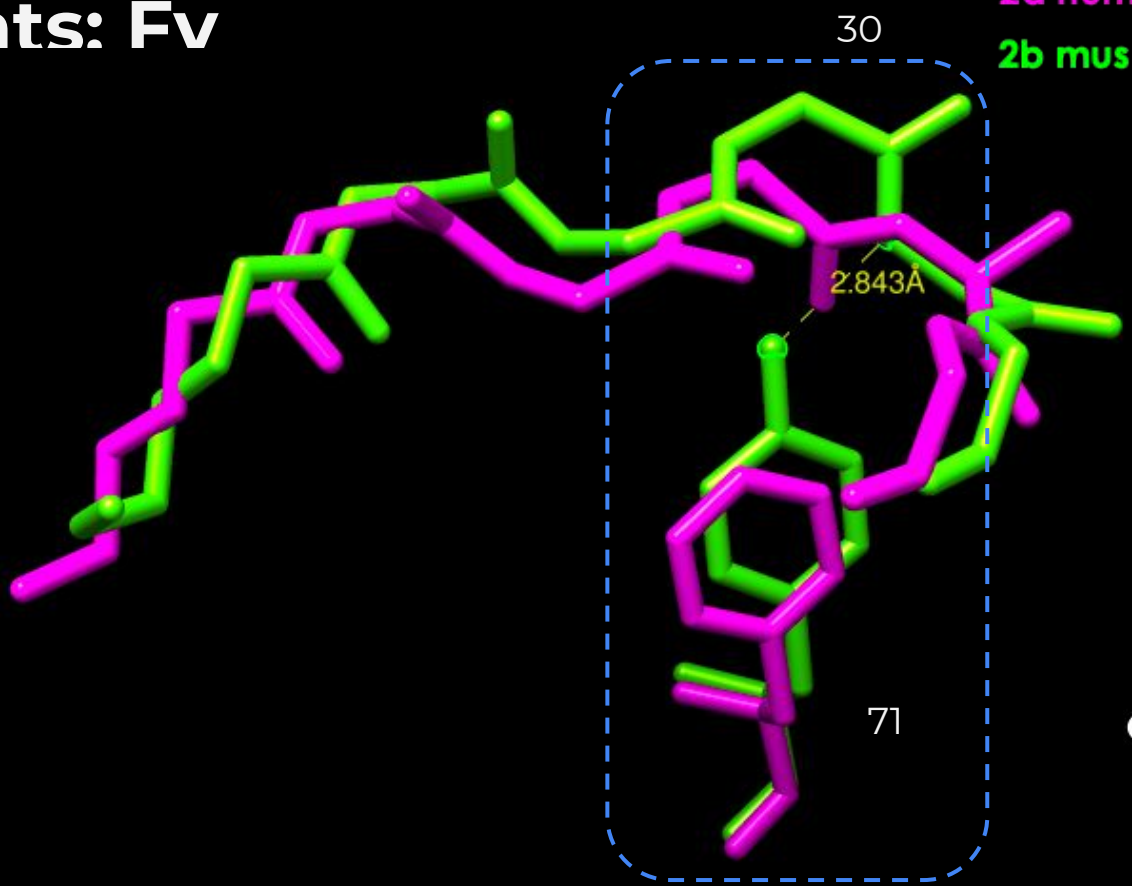


RMSD: 0.76 Å
SC: 4.74

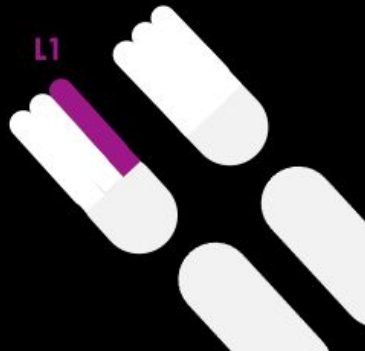
Fragments: Fv

CDR: L1-κ

2a vs 2b



RMSD: 1.32 Å
SC: 8.71

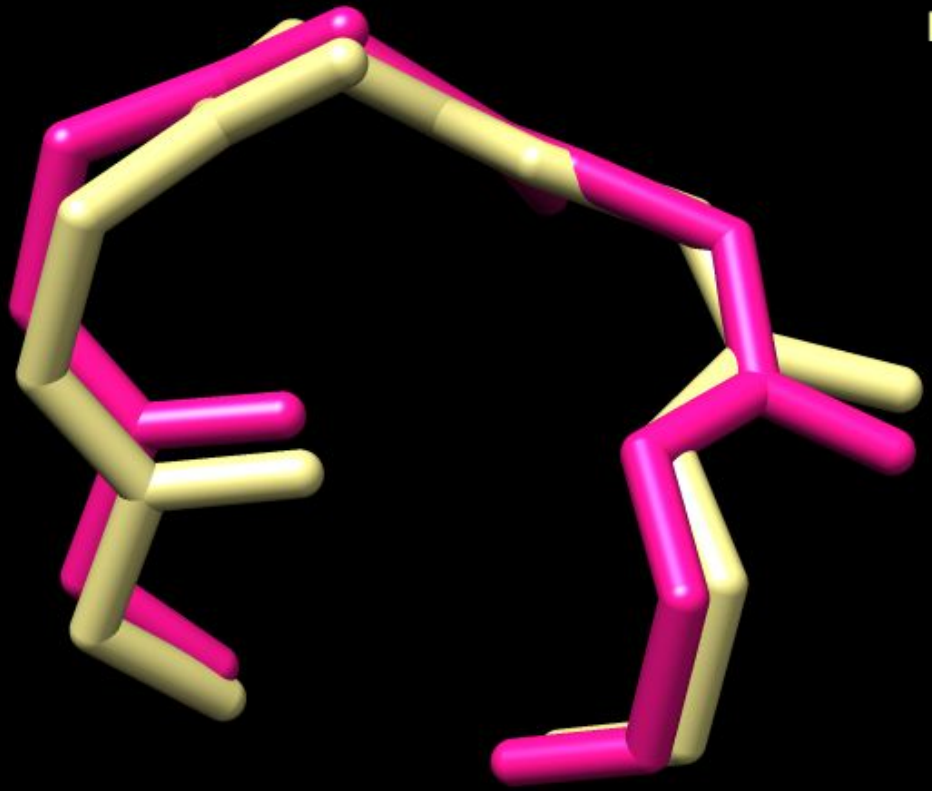


Fragments: Fv

CDR: L2

L2κ (mouse): 1FVG

L2 λ (human): 2RHE



RMSD: 1.14 Å

SC: 7.19

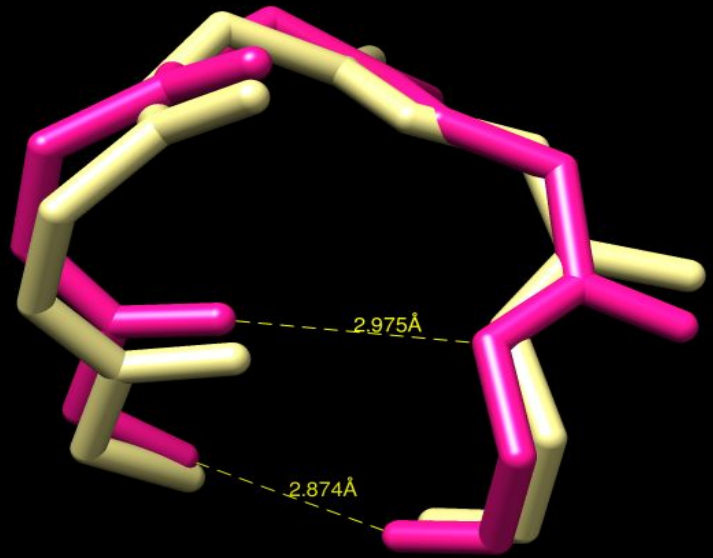
Fragments: Fv

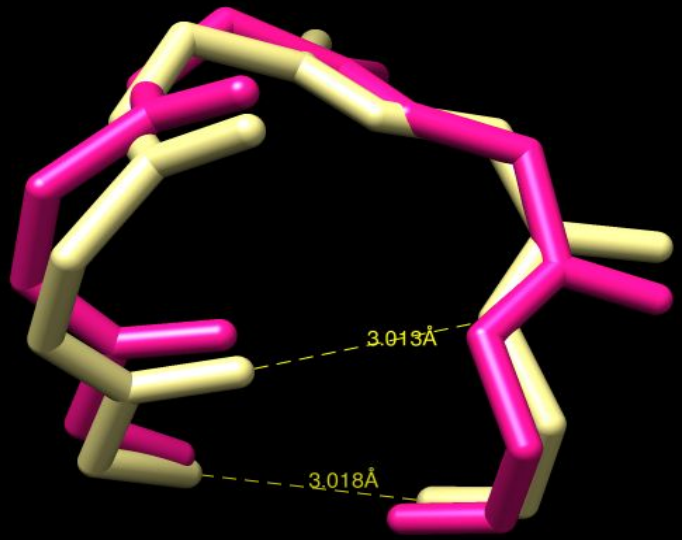
CDR: L2

Hydrogen bonds

49 N ---- O 53
49 O ---- N 53

L2κ (mouse): 1FVG
L2 λ (human): 2RHE



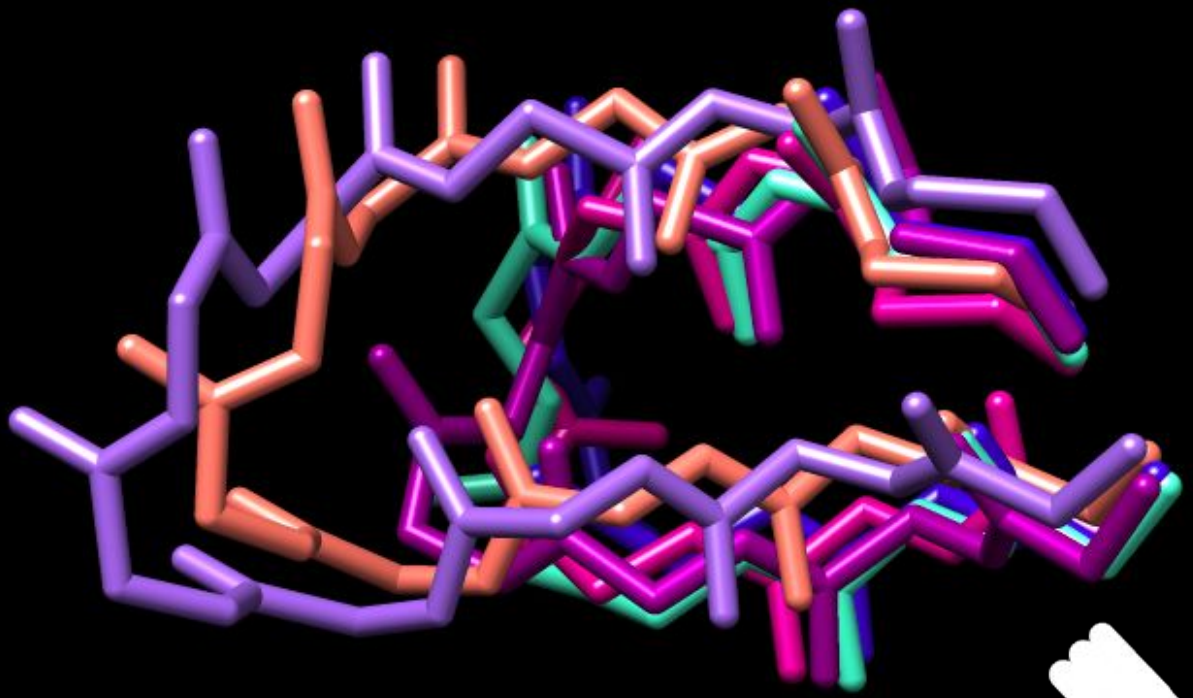


Fragments: F

CDR: L3-λ

L3-2λ: 2FBA
L3-2λ: 2RHE

L3-1λ: 1IND
L3-1λ:7FAB
L3-1λ:1GIG
L3-1λ:1MFA



RMSD: 0.96 Å
SC: 4.76

Fragments: Fv

CDR: L3-λ

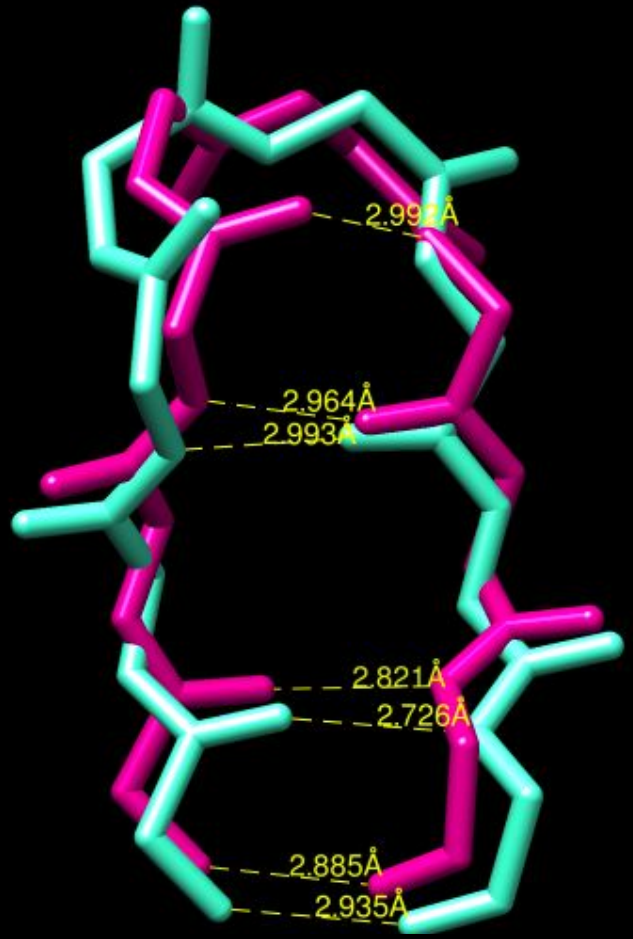
Hydrogen bonds

1λA & 1λB

90 O ---- N 97
90 N ---- O 97
92 N ---- O 95

1λB

92 O ---- N 95

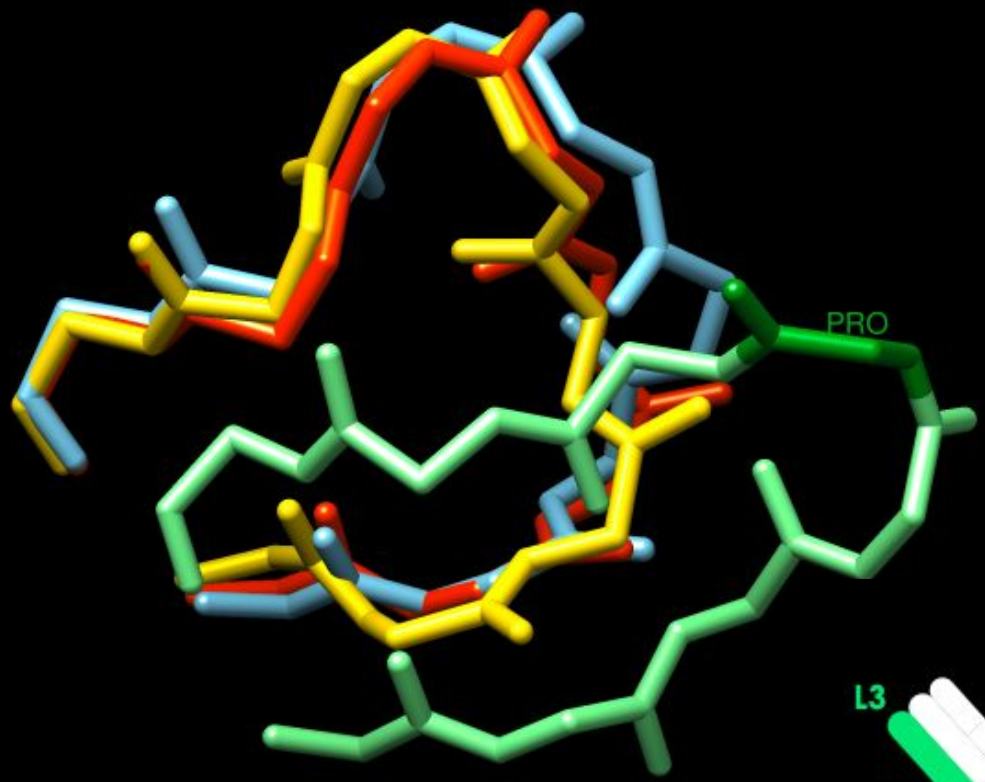


Fragments: Fv

CDR: L3-κ

L3-1κ : 2IMM
L3-1κ : 1FGV
L3-1κ : 1VFA

L3-2κ : 2FBJ



RMSD: 1.01 Å
SC: 6.34

Fragments: Fv

CDR: L3-κ

L3-1κ : 2IMM

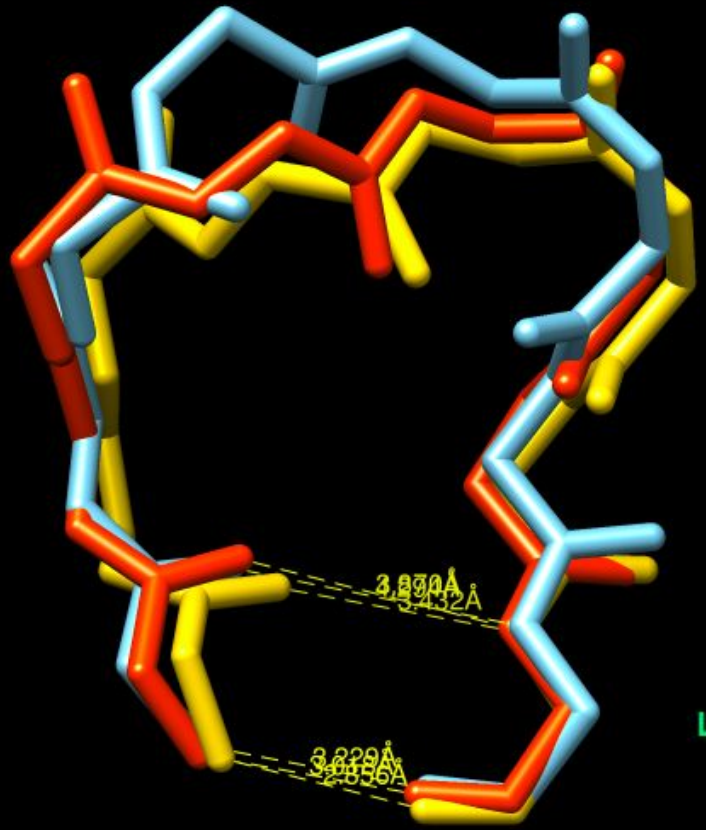
L3-1κ : 1FGV

L3-1κ : 1VFA

Hydrogen bonds

90 O ---- N 97

90 N ---- O 97



Fragments: Fv

CDR: L3-κ

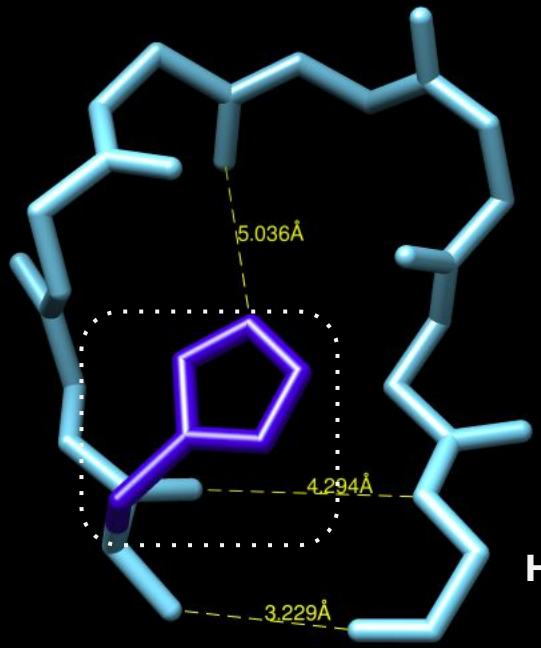
Hydrogen bonds in the side chain

L3-1κ : 2IMM

90 OE1 ---- N 92
90 NE1 ---- O 93
90 NE ---- O 95

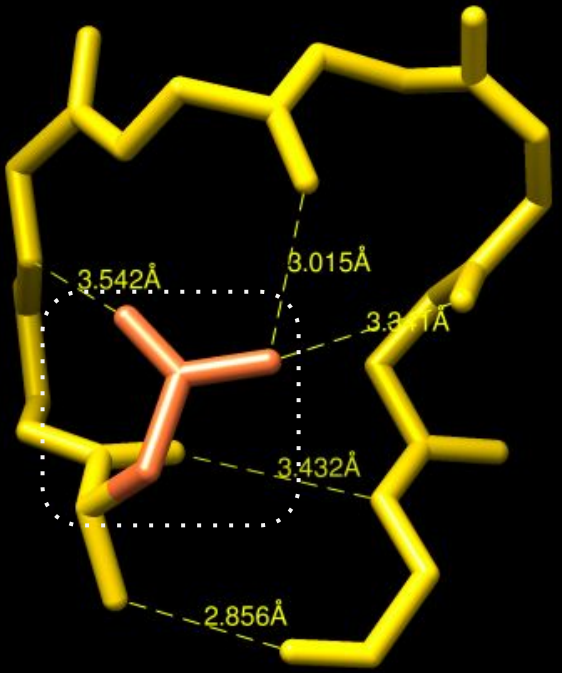
L3-1κ : 1VFA

90 NE ---- O 93
90 NE1 ---- O 97



Histidine 90

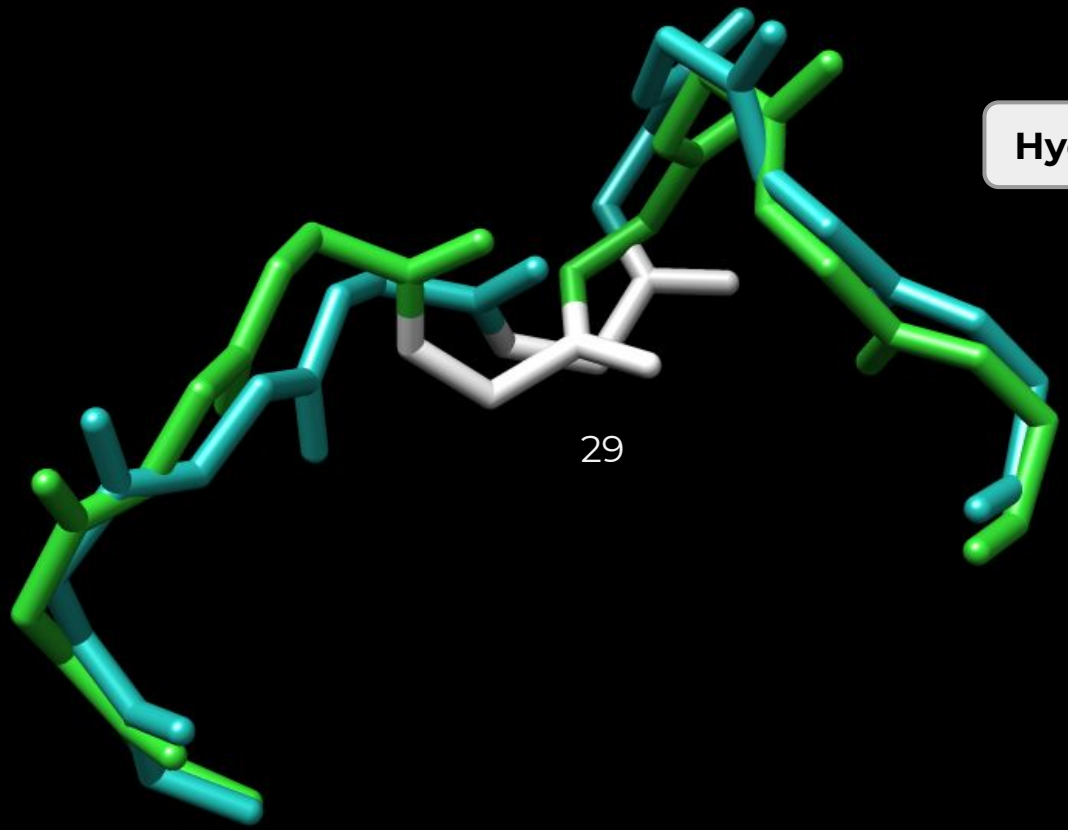
Asparagine 90



Fragments: Fv

CDR: H1

1



H1: 1VFA

H1: 2CGR

Hydrophobic residue

RMSD: 0.91 Å
SC: 4.8

Fragments: Fv

CDR: H1

Hydrogen bonds

28 O ---- N 31

29 N ---- O 32

H1: 1VFA

H1: 2CGR

Fragments: Fv

CDR: H2

3A VS 3B VS 3C

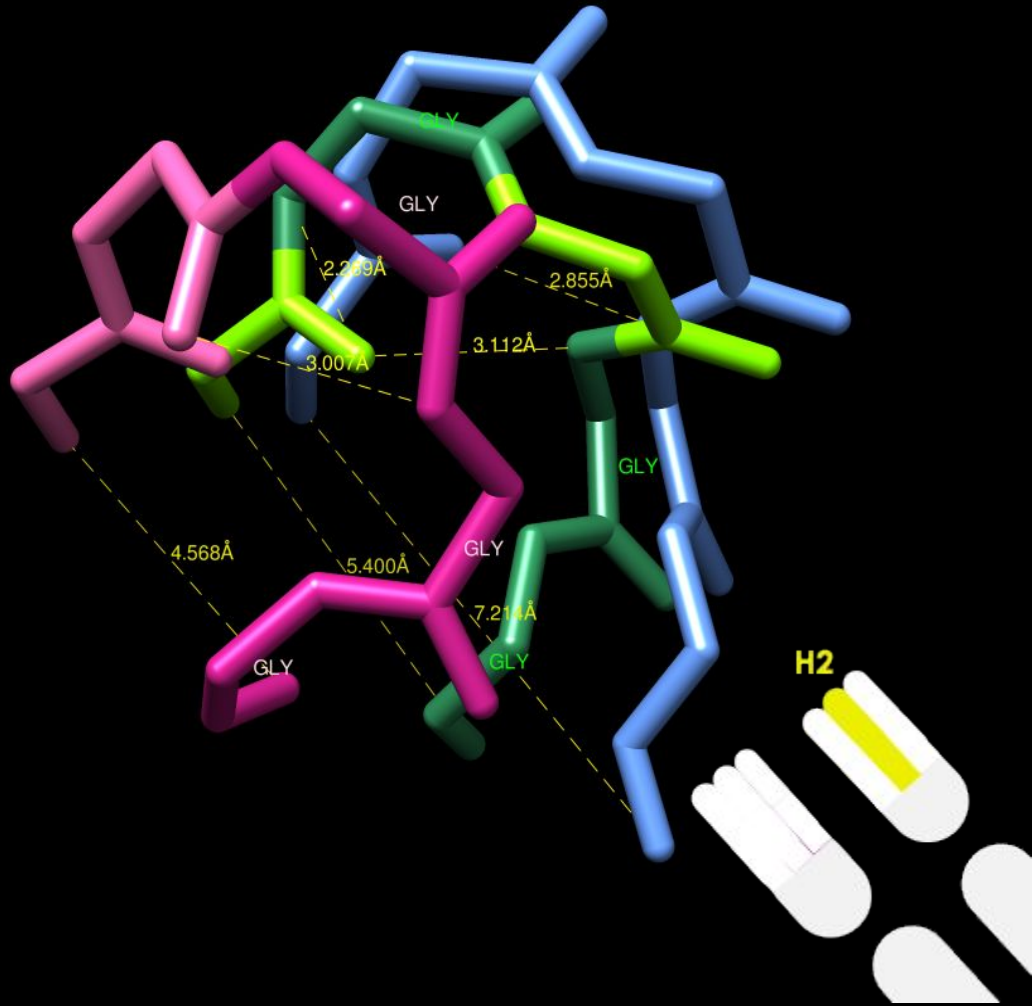
Hydrogen bonds

3a	3b	3c
52 N ---- O 56	52 N ---- O 56	52 N ---- O 56
52 O ---- N 54	53 O ---- N 55	52 O ---- N 53
		52 O ---- N 55

H3a: 2FB4

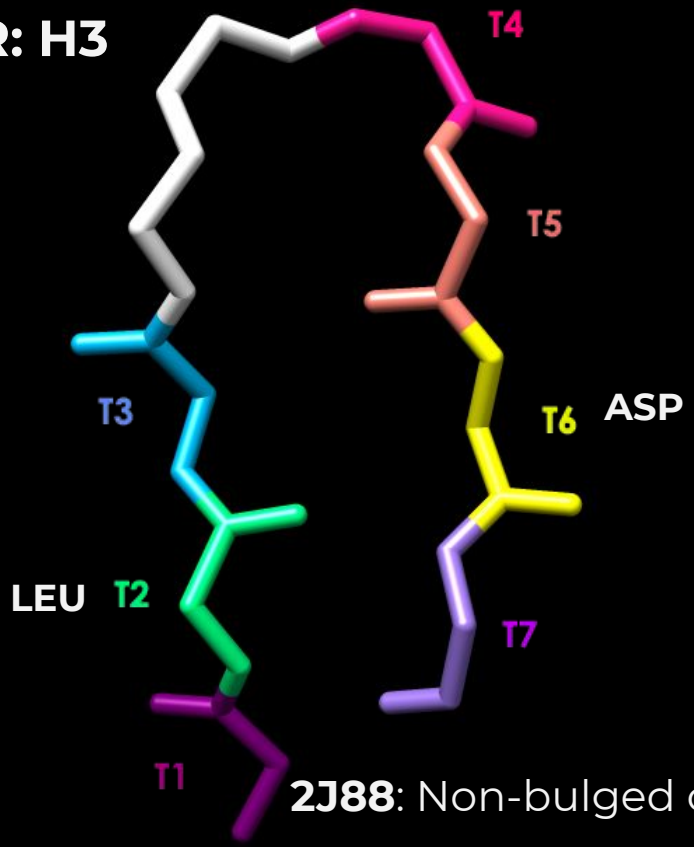
H3b: 1IND H3b Gly: residues 54, 55 and 56

H3c: 1IGM H3c Gly: residues 53, 55 and 56



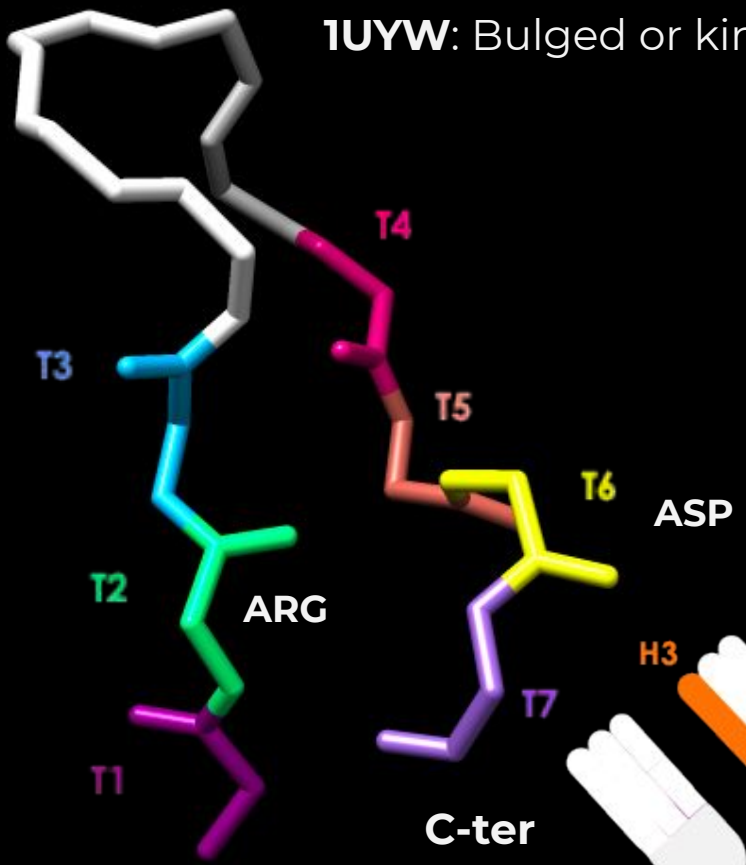
Fragments: Fv

CDR: H3



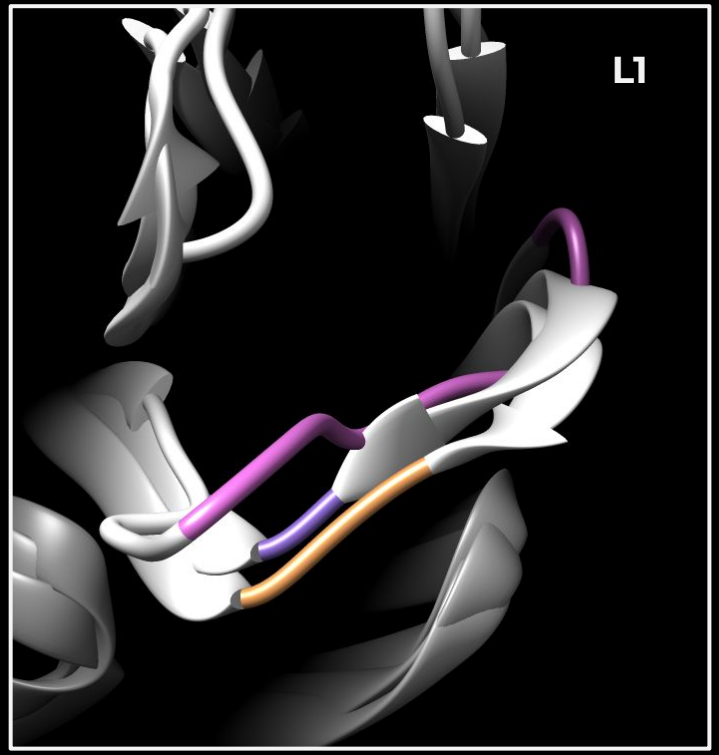
2J88: Non-bulged or Extended

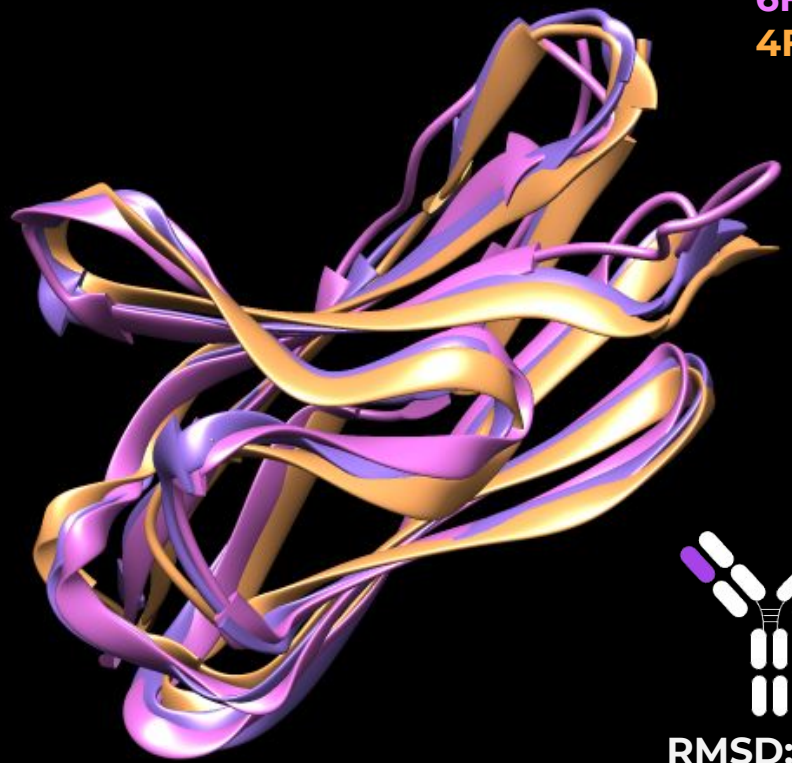
1UYW: Bulged or kinked



Antigen interaction

Introduction (LC-FV)

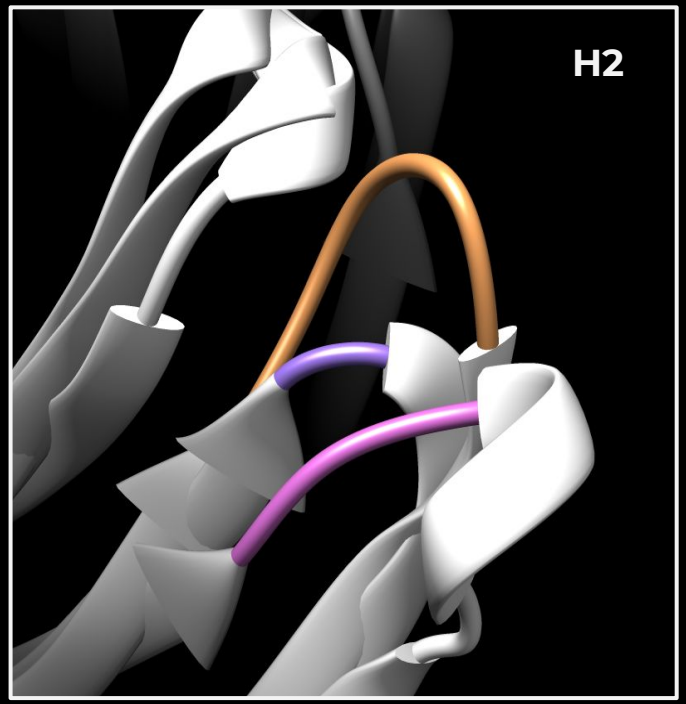


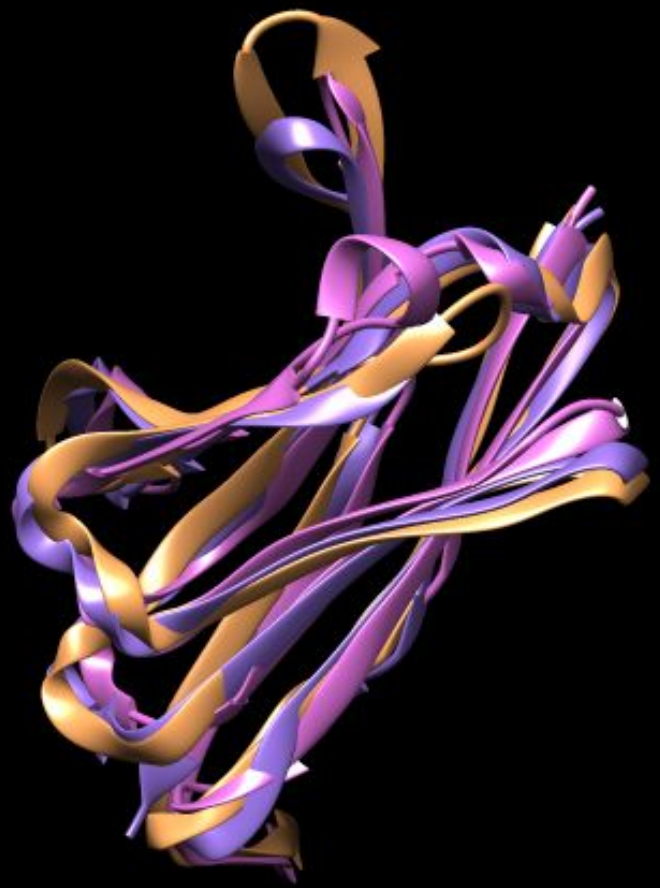


RMSD: 0.67 Å
SC: 6.71

Antigen interaction

Introduction (HC-FV)



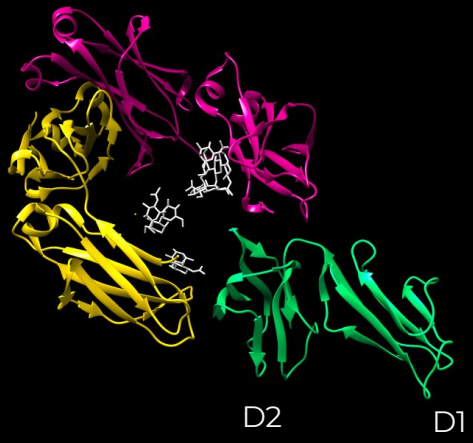


1VFA
6FAB
4FAB

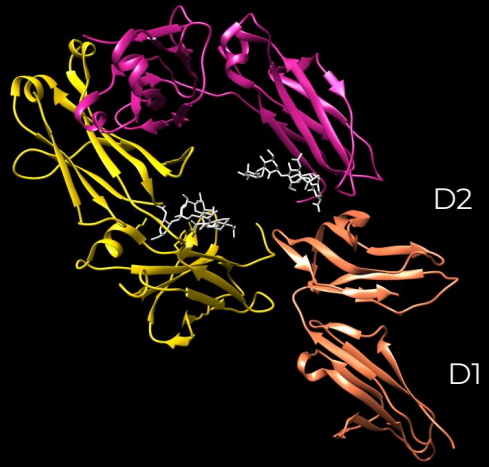
RMSD: 1.10 Å
SC: 6.71

Antigen interaction

Types of Fc-R

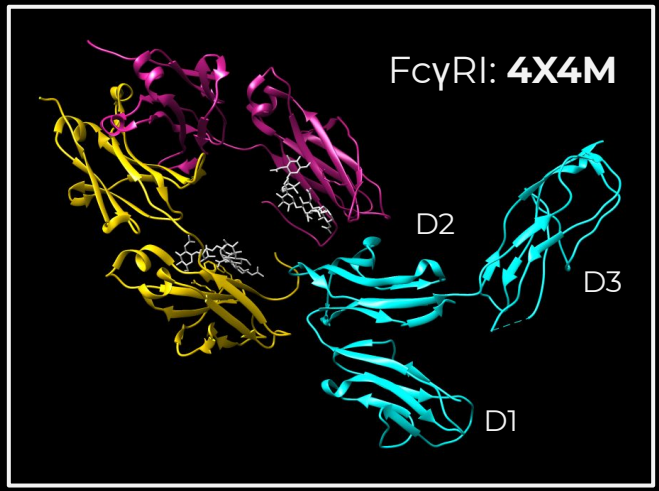


FcγRIIA/B: **3WJJ**



FcγRIII: **1T83**

FceRI: **1F6A**



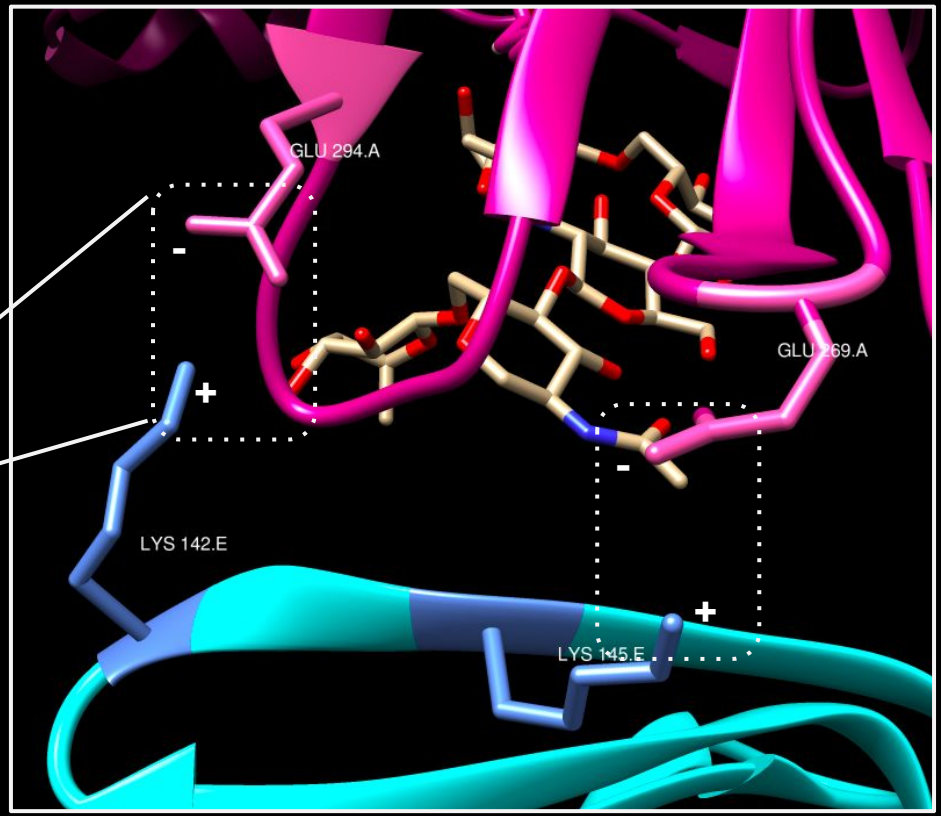
Antigen interaction

IgG/Fc-R: Region 1

IgG-FC

FcγRI

Salt bridges



4X4M

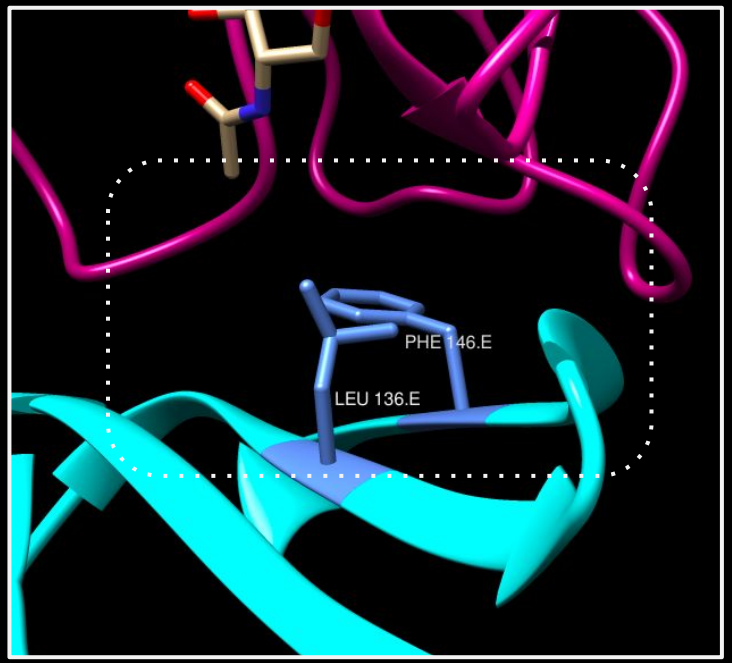
Antigen interaction

IgG-FC

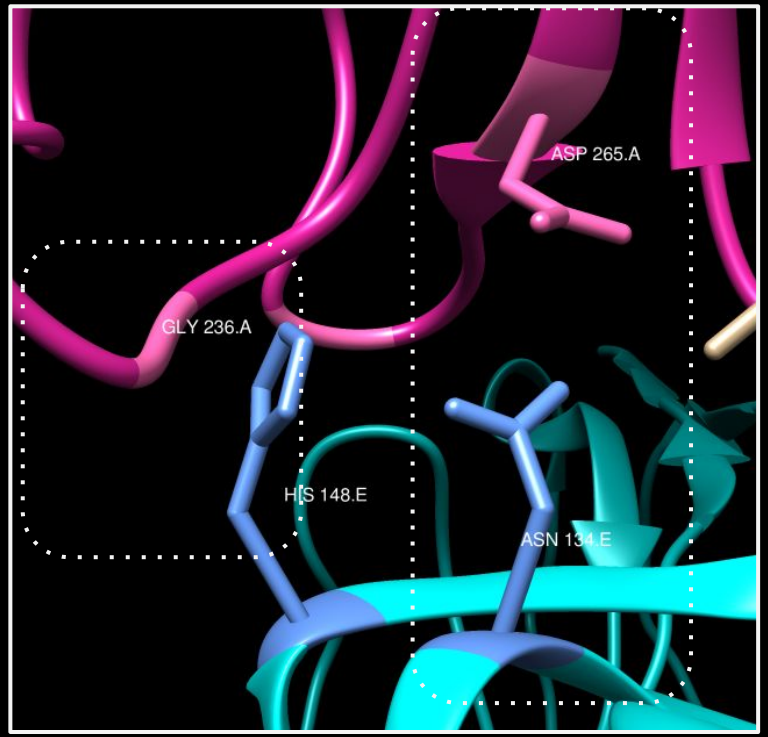
FcγRI

IgG/Fc-R: Region 1

VDW



Hydrogen bonds

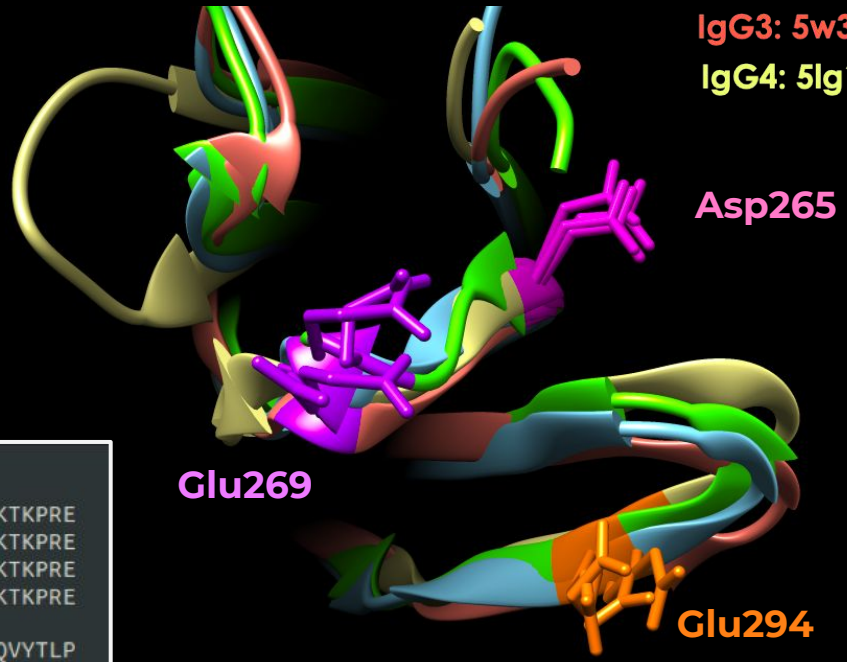


4X4M

Antigen interaction

IgG/Fc-R: Region 1

IgG1: 3ave
IgG2: 4haf
IgG3: 5w38
IgG4: 5lg1



Residues conserved

Conserved residues

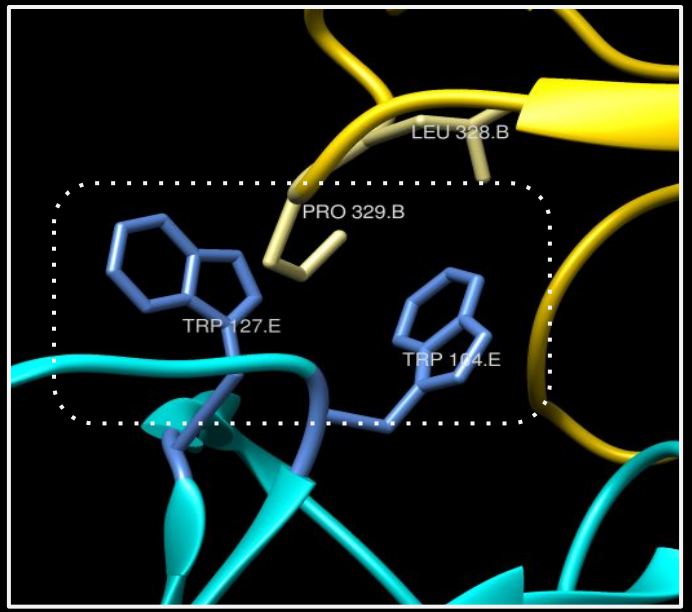
```
CLUSTAL W(1.60) multiple sequence alignment

5lg1        --GGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSQEDPEVQFNWYVDGVEVHNAKTKPRE
5w38        -LGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFKWYVDGVEVHNAKTKPRE
3ave        LLGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPRE
4haf        --AGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVEVHNAKTKPRE

5lg1        EQFNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKGL-PSSIEKTISKAKGQPREPQVYTLP
5w38        EQFNSTFRVVSVLTVLHQDWLNGKEYKCKVSN-KALPAPIEKTISKTKGQPREPQVYTLP
3ave        EQYNSTYRVVSVLTVLHQDWLNGKEYKCKVSN-KALPAPIEKTISKAKGQPREPQVYTLP
4haf        EQFNSTFRVVSVLTVVHQDWLNGKEYKCKVSN-KGLPAPIEKTISKTKGQPREPQVYTLP
```

Antigen interaction

IgG-FC
FcγRI

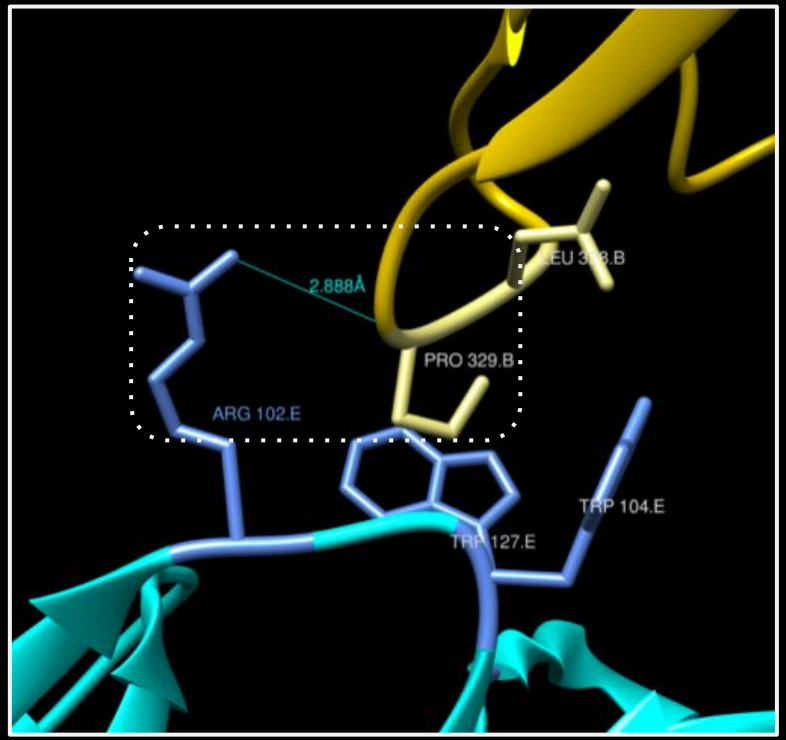
IgG/Fc-R: Region 2

Sandwich Trp-Pro-Trp: hydrophobic



4X4M

Hydrogen bonds

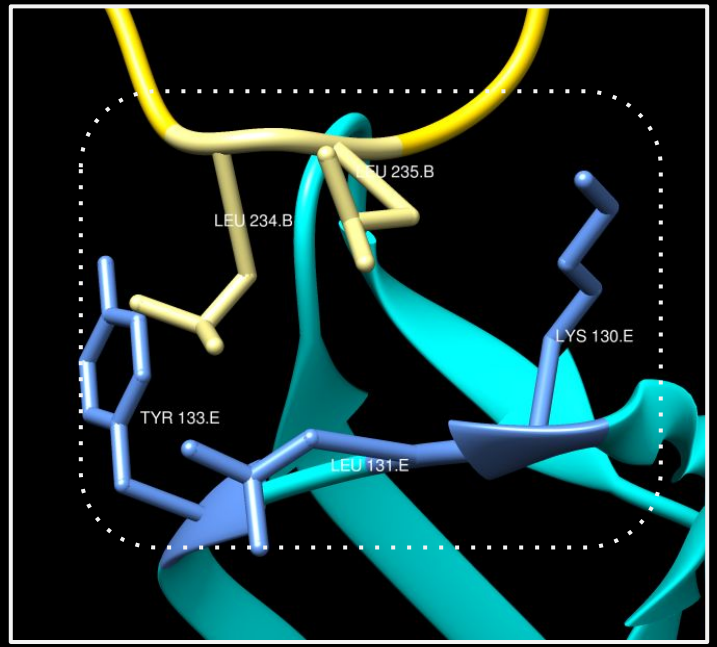


Antigen interaction

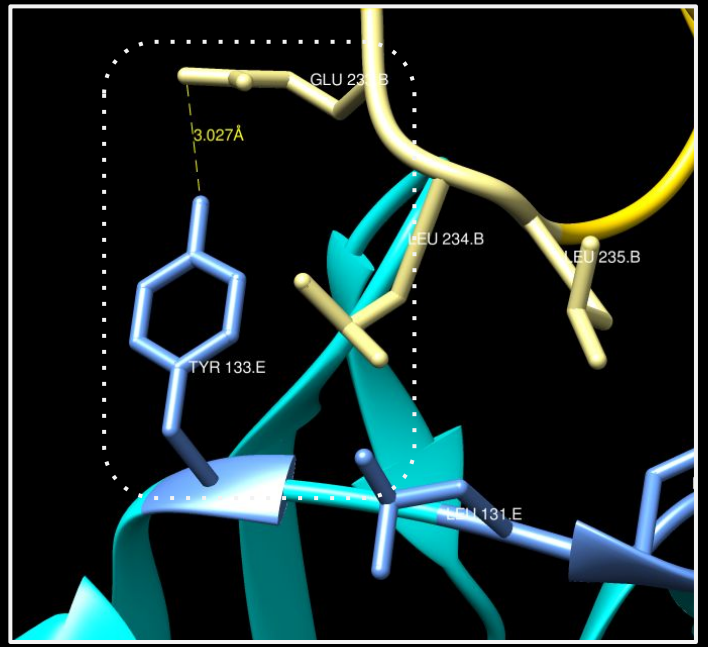
IgG-FC
FcγRI

IgG/Fc-R: Region 2

Hydrophobic cluster



Hydrogen bonds



4X4M

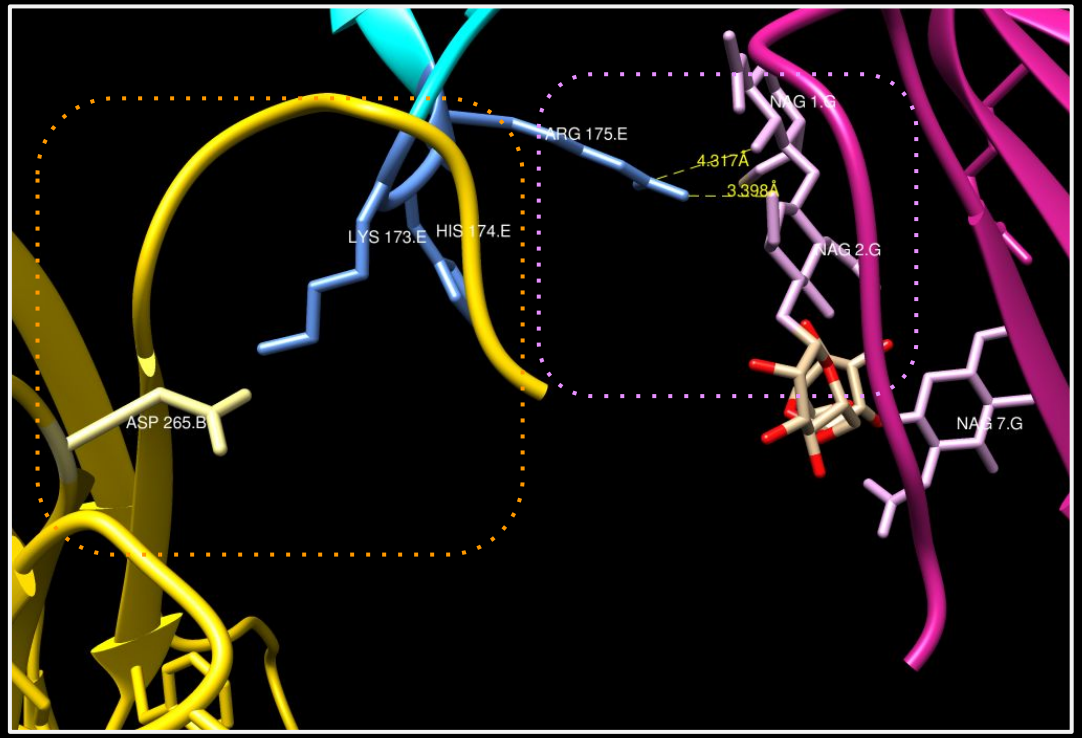
Antigen interaction

IgG/Fc-R: Region 3

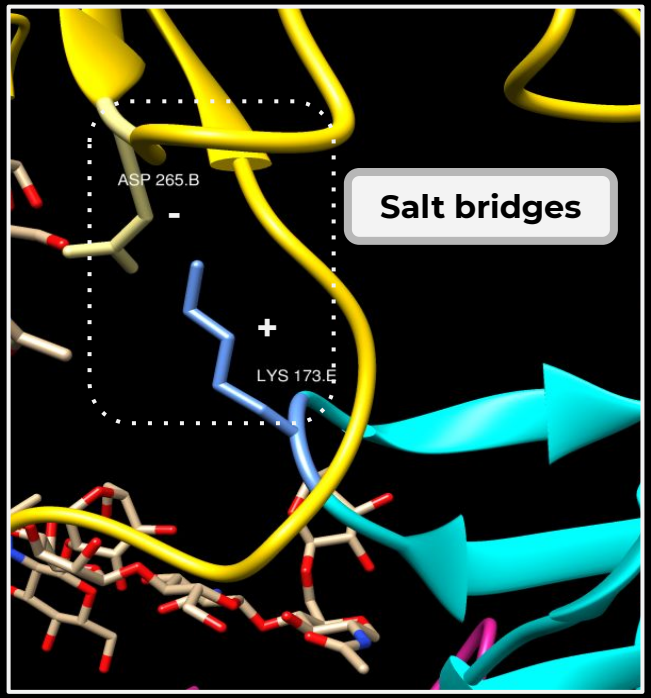
IgG-FC
IgG-FC
FcγRI

Interaction IgG-FcγRI

Interaction NAG-FcγRI



4X4M



Antigen interaction

IgG/SARS-CoV-2

IgG HC

IgG LC

RBD region: SARS-CoV-2

7C01

Interaction with HCDR1

Interaction with HCDR2

Antigen interaction

IgG/SARS-CoV-2

Interaction with HCDR3

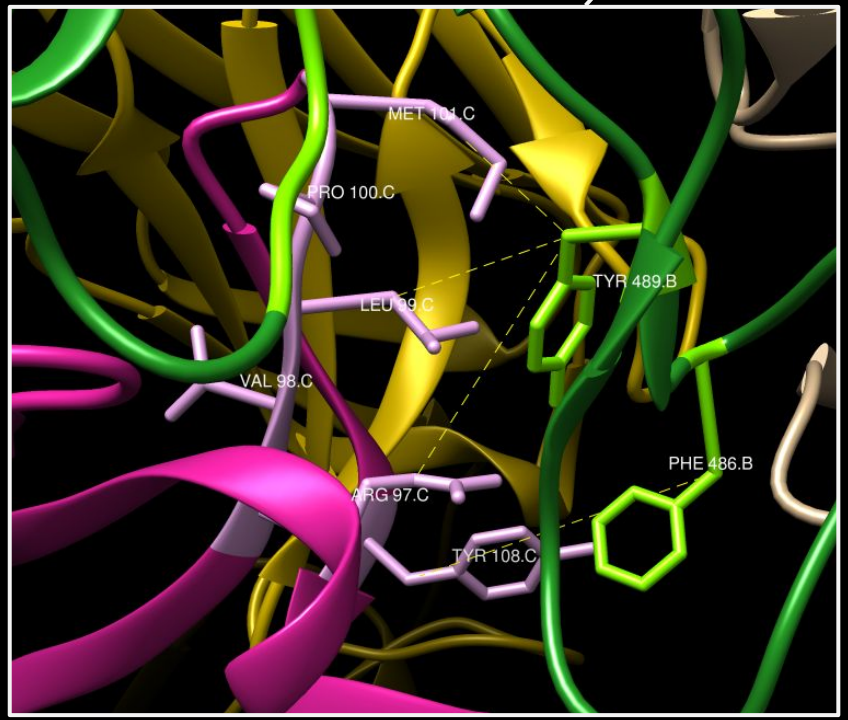
Interaction with LCDR1

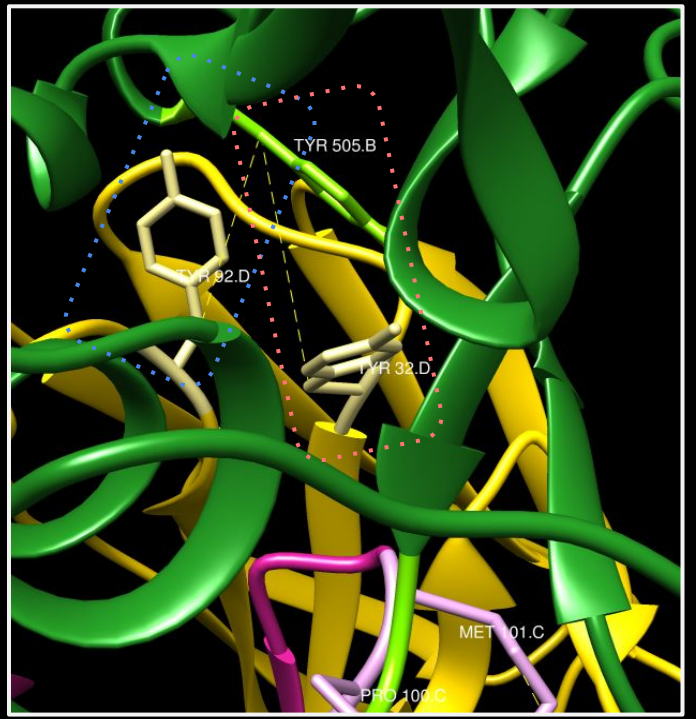
Interaction with LCDR3

IgG HC

IgG LC

RBD region: SARS-CoV-2





7C01

Antigen interaction

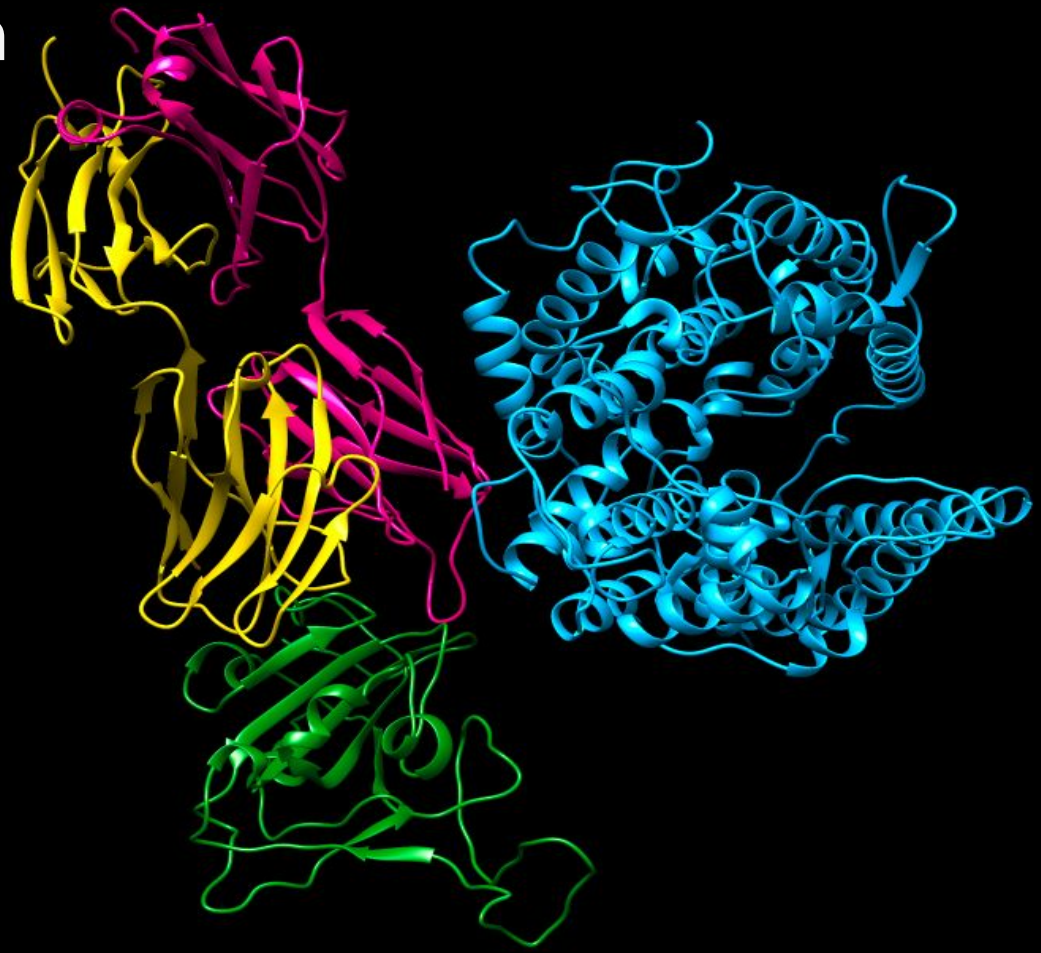
IgG/SARS-CoV-2

IgG HC

IgG LC

RBD region: SARS-CoV-2

ACE-2 Receptor



6W41 & 7C8D

Antigen interaction

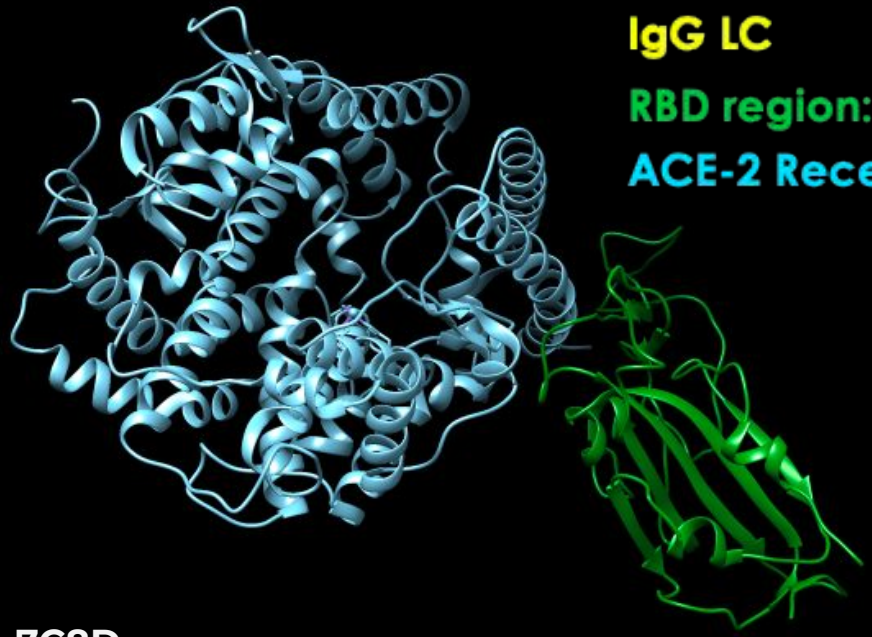
IgG/SARS-CoV-2

IgG HC

IgG LC

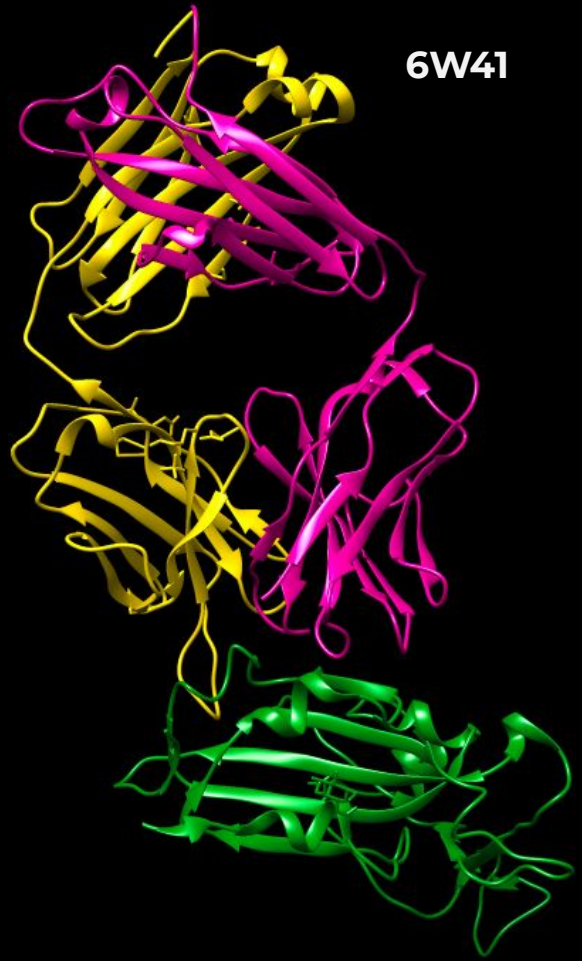
RBD region: SARS-CoV-2

ACE-2 Receptor



7C8D

6W41



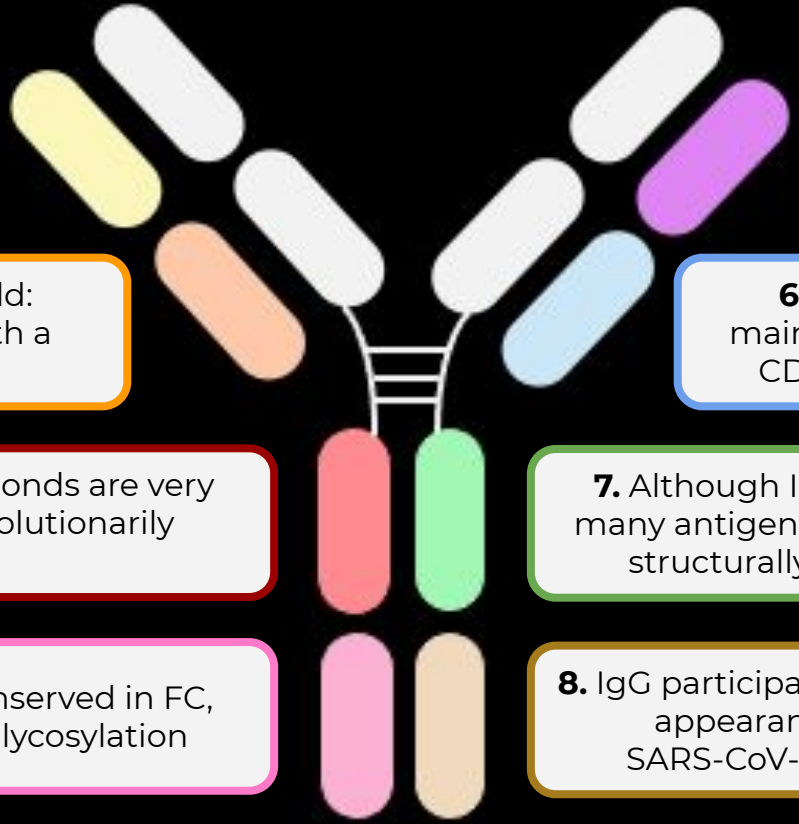
Conclusions

1. Immunoglobulins G are really well studied biostructurally

2. All present an Ig-like fold: antiparallel beta strand with a greek key topology

3. Hydrogen and disulfide bonds are very important, so they are evolutionarily conserved

4. The residue Asn297 is conserved in FC, which is necessary for N-glycosylation



5. CDR are hypervariable regions, which are found in the Fab region

6. Canonical structures are maintained in all CDRs, except in CDR3 (the most variable one)

7. Although IgG are variable to recognize many antigens, the parts whose function is structurally important are preserved

8. IgG participates in antigen interaction. The appearance of new antigens (like SARS-CoV-2) requires further studies

Questions

1. About basic concepts of the Immunoglobulins, select the correct answer:
   a. Structure stabilization is given by disulfide bonds and Van der Waals
   b. CH2 and CH3 domains form the Fab region
   c. There are two different types of heavy chain: kappa and lambda.
   d. The constant region of IgG is composed of four globular domains, 3 of them located in the heavy chain and one in the light chain.
   e. IgG and IgD have 4 constant domains, whereas IgA, IgM and IgE have 3 constant domains.
2. Regarding the structure of the IgG, select the correct answer:
   a. IgG belongs to the class of all beta proteins.
   b. These proteins contains two beta-sheets, that bend together in a greek key beta-barrel sandwich.
   c. Both A and B are correct
   d. The disulfide bond between beta sheets B and F is very important in the folding process of the protein.
   e. All of them are correct.

Questions

3. About the rearrangement:
   a. The light chain has an extra variable region.
   b. There isn't any alternative splicing process after the Ig's rearrangement.
   c. The rearrangement of the V-J segments define the most variable regions.
   d. The differences between the two light chains is functional.
   e. The lambda light chain is encoded by chromosome X.
4. The N-glycosylation:
   a. Occurs in the CH2 region.
   b. Carbohydrates are bound to the hydrophobic face.
   c. Both A and B are correct.
   d. The glycosylation takes place on the asparagine 297 (Asp297).
   e. All of them are correct.
5. The classes of canonical structures:
   a. Allow to divide immunoglobulins into several types of antigen recognition structures.
   b. Make a correlation between architecture of antigen-binding site and the antibody function.
   c. Divide the immunoglobulins in those that are anti-specific and those which are multi-specific.
   d. Show that there are no difference in H2 and L1 canonical structure types.
   e. Allow to conclude that there is no contribution of the geometry in the binding process.

Questions

6. About the antigen binding site:
   a. Contains 10 hypervariable regions close to each other.
   b. All the hypervariable regions have canonical structures.
   c. CDR-H3 is the only hypervariable region that don't have any canonical structure.
   d. The lambda and kappa chains have the same canonical structures
   e. All the canonical structures have the same length.
7. About the CDRL3 of the immunoglobulines, which of the following statements is true?
   a. There is a common L3 canonical structure for both Kappa and Lambda chains.
   b. L3 canonical structure 1 in Lambda chain can be divided in four: A, B, C and D.
   c. CDRL3 is stabilized by disulfide bonds.
   d. CDRL3 in Kappa chain has six canonical structures.
   e. The canonical 2 in Kappa chain is the most common.
8. Which of these sentences about CDR H3 is correct?
   a. CDR H3 has 4 canonical structures
   b. CDR H3 is divided in three different parts: feet, torso and head
   c. CDR H3 can adopt two different conformations in the torso region: kinked/bulged and extended/non-bulged
   d. CDR H3 is not very variable and this is the reason why it doesn't have canonical structures.
   e. CDR H3 is not involved in antigen interaction.

Questions

9. Which are the CDRs that participates, for sure, in the interaction of IgG and SARS-CoV-2?
   a. HCDR1, HCDR2 and HCDR3.
   b. LCDR1 and LCDR3.
   c. Both A and B are correct
   d. LCDR2
   e. All of them are correct
10. Which of these sentences is incorrect?
   a. Docking has been done with HEK program
   b. IgG binds to the Receptor Binding Domain Region of the S1 region of Spike (SARS-CoV-2)
   c. Receptor Binding Domain of S1 region of Spike (SARS-CoV-2) binds to ACE2
   d. ACE2 can bind the RDB domain of SARS-CoV-2 meanwhile IgG is bound to it
   e. If IgG is bound to RBD, SARS-CoV-v2 virus cannot get inside the cells.

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THANKS FOR YOUR ATTENTION