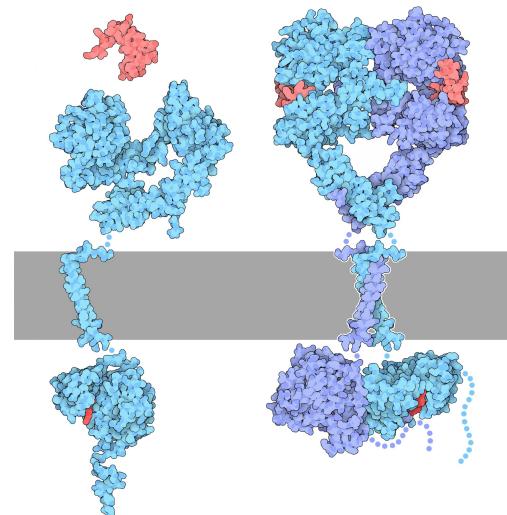


Structural Biology

The Epidermal Growth Factor Receptor

Analysing the extracellular domain of EGFR and its ligands
EGF, TGF- α , and EREG binding



Source: PDB-101 Molecule of the month. June 2010, David Goodsell

Daniel Martín López, Guillem Miró Almuni, and Gisela Rusiñol Gálvez

INTRODUCTION



1

Receptor Tyrosine Kinases

2

EGFR Family

3

Signaling Pathways

4

EGFR Family and Disease

5

EGFR Receptor (ErbB1)

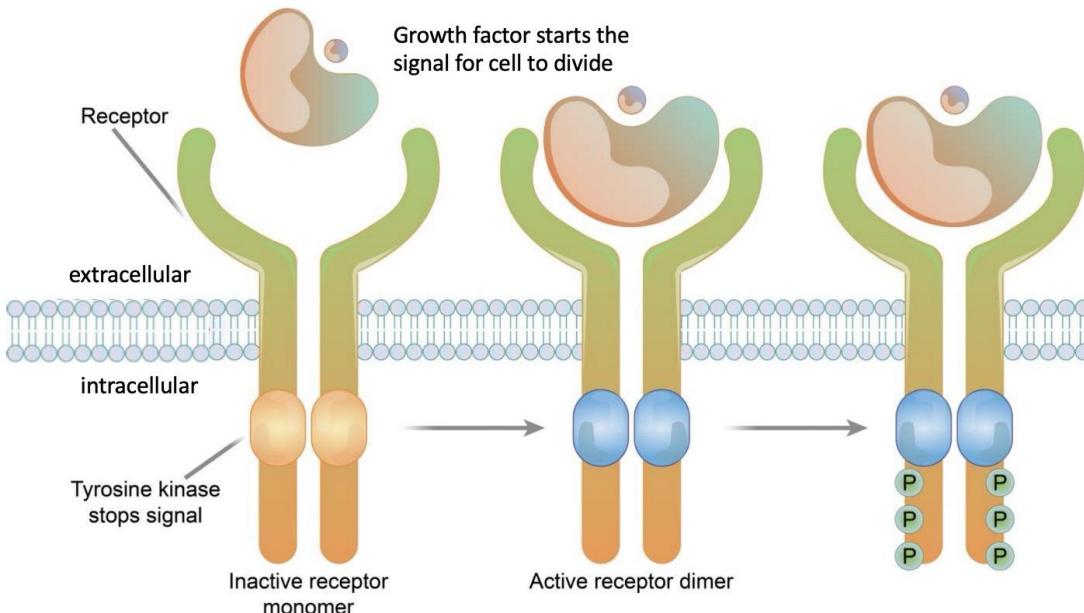
6

EGFR Extracellular Domain

1. RECEPTOR TYROSINE KINASES



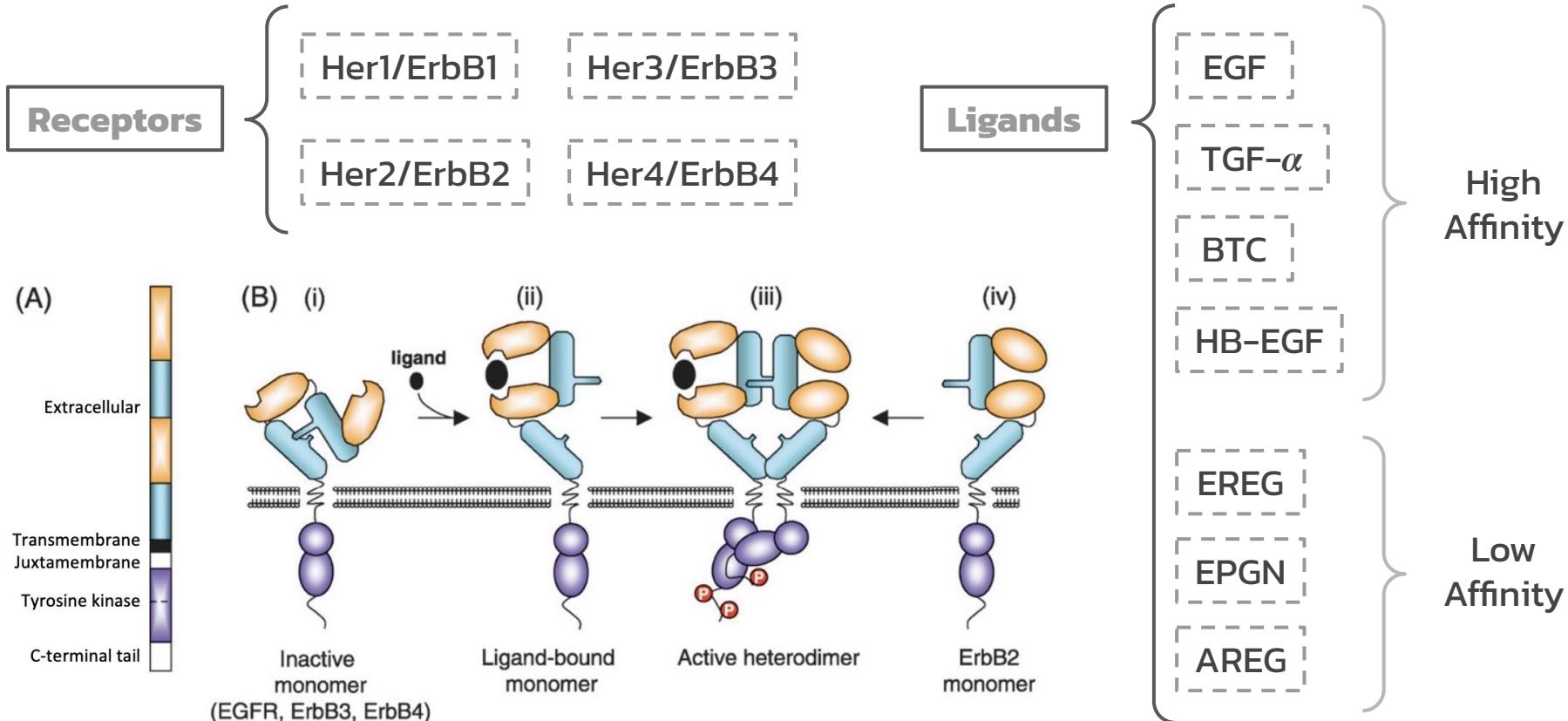
- Key regulators of **normal cellular processes**
- Critical role in the development of **cancer**



Adapted from Yeqing He *et al*, 2021

Class	Family
I	EGFR receptor
II	Insulin receptor
III	PDGF receptor
IV	VEGF receptor
V	FGF receptor
VI	CCK receptor
VII	NGF receptor
VIII	HGF receptor
...	

2. EGFR FAMILY



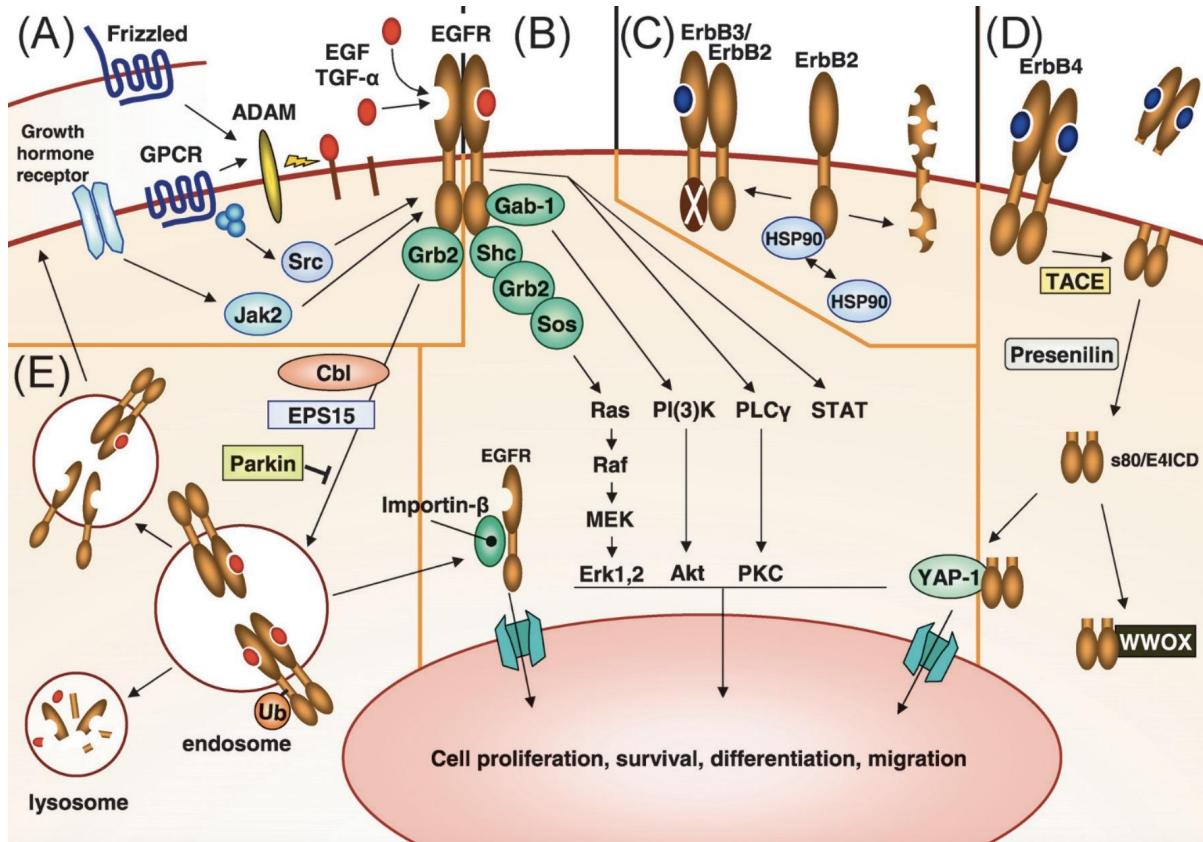
Adapted from Mark A. Lemmon *et al*, 2014

3. SIGNALING PATHWAYS



Direct activation or via adaptor proteins of:

- Ras/MAPK
- PI3K/Akt
- PLC γ 1/PKC
- STAT



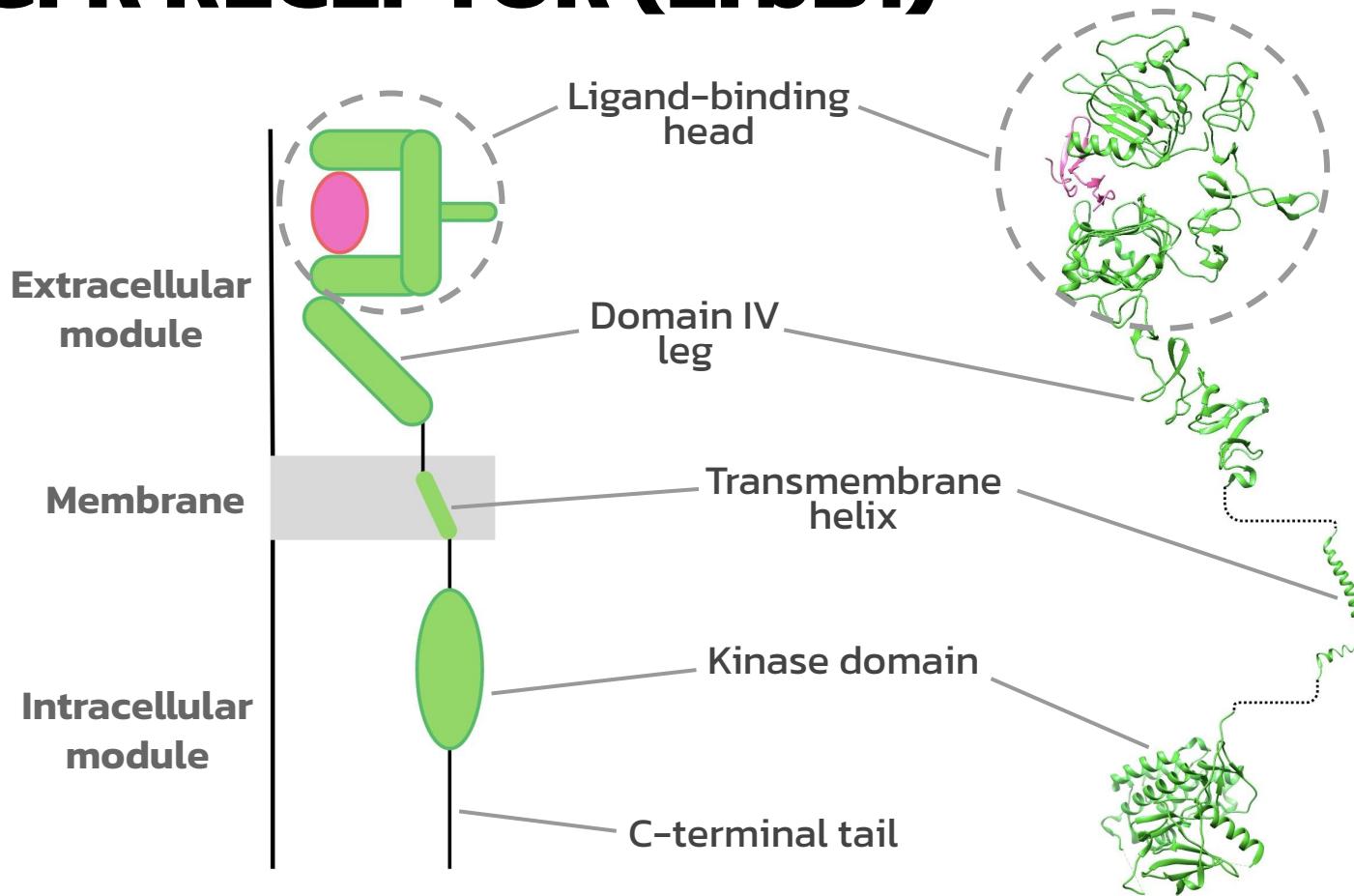
4. EGFR FAMILY AND DISEASE



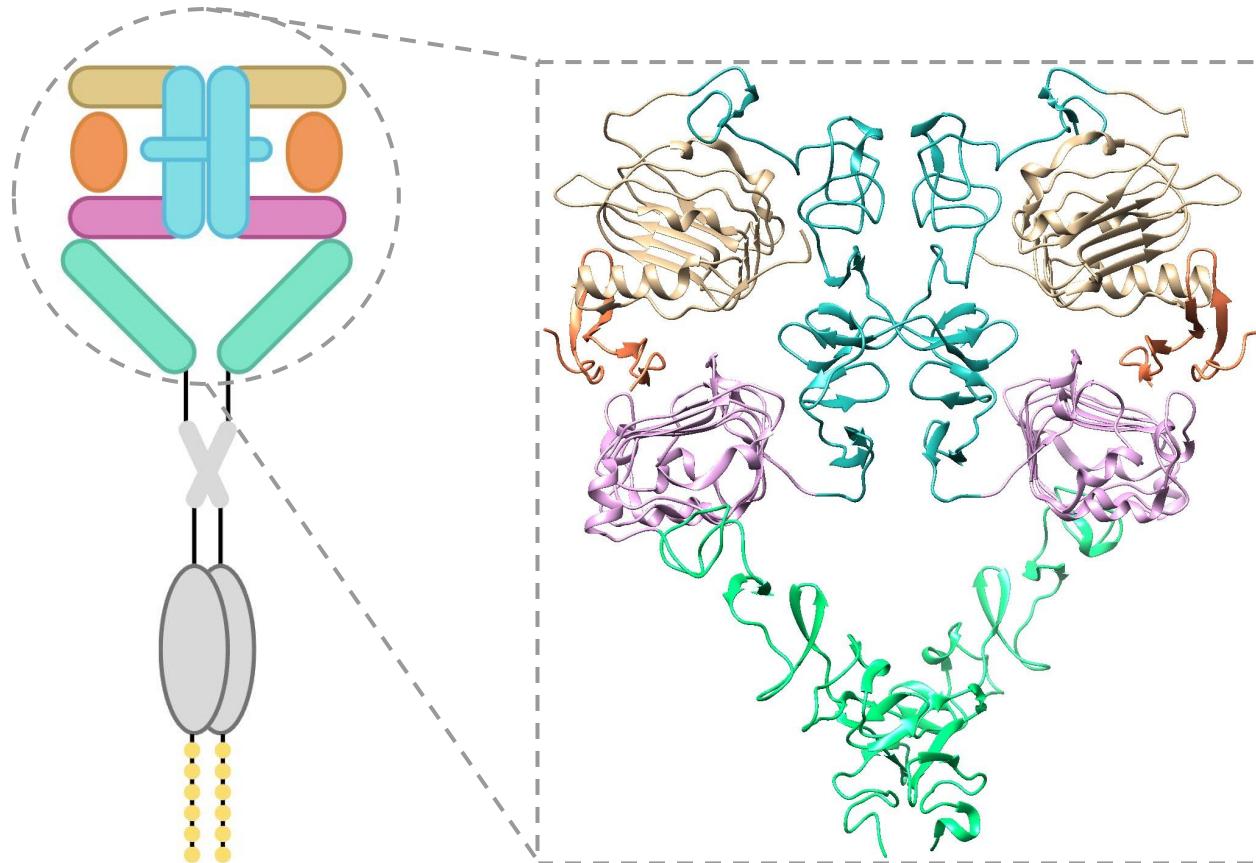
Aberrant signalling by ErbB receptors is associated with human disease, specially with different types of cancer:

Gene	Malignancy	Alteration
EGFR	Breast cancer	Gene amplification
	Glioblastoma multiforme	Extracellular domain deletions and mutations Gene amplification
	Non-small cell lung cancer	Kinase domain mutations
	Others	Gene amplification
Her2/ErbB2	Breast cancer	Gene amplification
	Non-small cell lung cancer	Kinase domain mutations
ErbB4	Multiple	Kinase domain mutations

5. EGFR RECEPTOR (ErbB1)



6. EGFR EXTRACELLULAR DOMAIN



ID	3NJP
Resolution	3.3 Å

Domain I

Domain II

Domain III

Domain IV

622 residues
68,699.3 Da

Ligand (EGF)

EGFR STRUCTURE



1

SCOP Classification

2

Domains I and III

3

Domains II and IV

4

MSA and Phylogeny

1. SCOP CLASSIFICATION



Domains I and III

Class	Alpha and beta proteins (a/b)
Fold	Leucine-rich repeat, LRR (right-handed beta-alpha superhelix)
Superfamily	L domain-like (less regular structure consisting of variable repeats)
Family	L domain
Protein	EGF receptor extracellular domain
Species	<i>Homo sapiens</i>

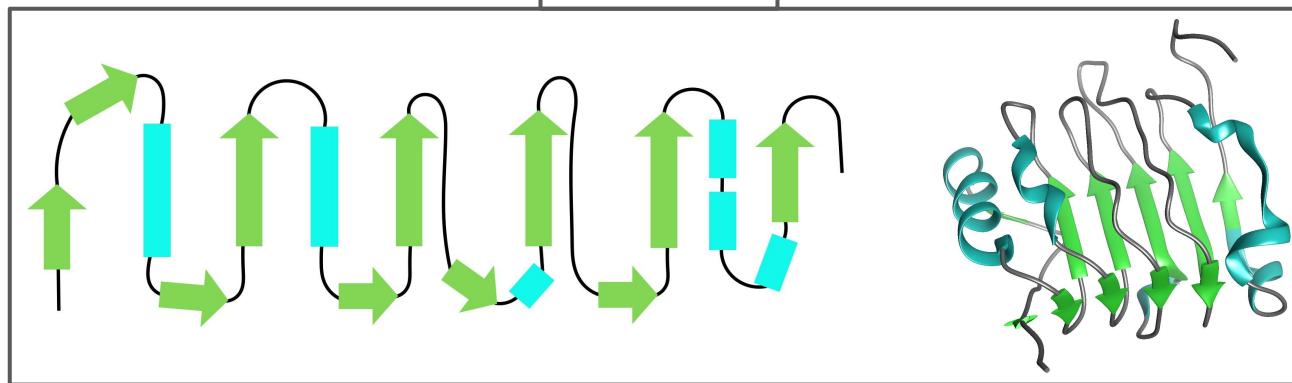
Domains II and IV

Class	Small proteins
Fold	Knottins (disulfide-bound fold, contains beta hairpin with two adjacent disulfides)
Superfamily	Growth factor receptor domain
Family	Growth factor receptor domain
Protein	EGF receptor Cys-rich domains
Species	<i>Homo sapiens</i>

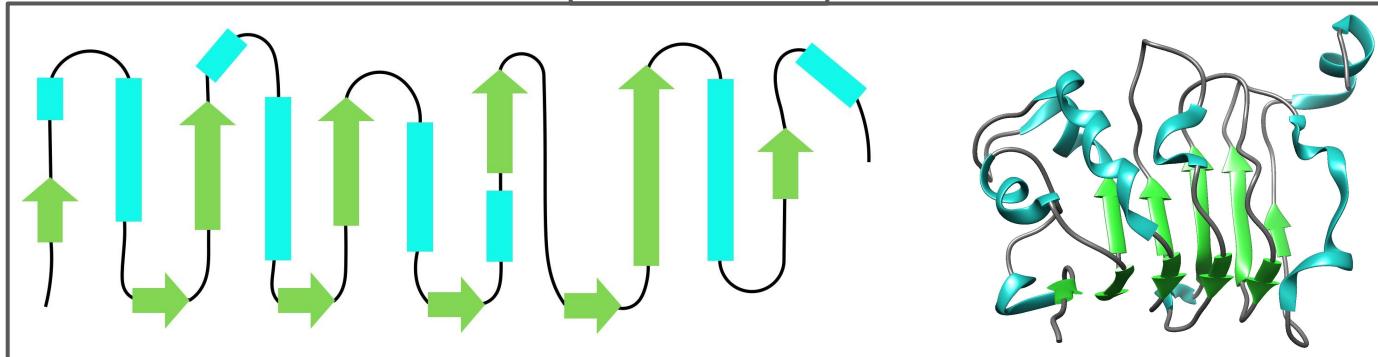
2. DOMAINS I and III



Domain I



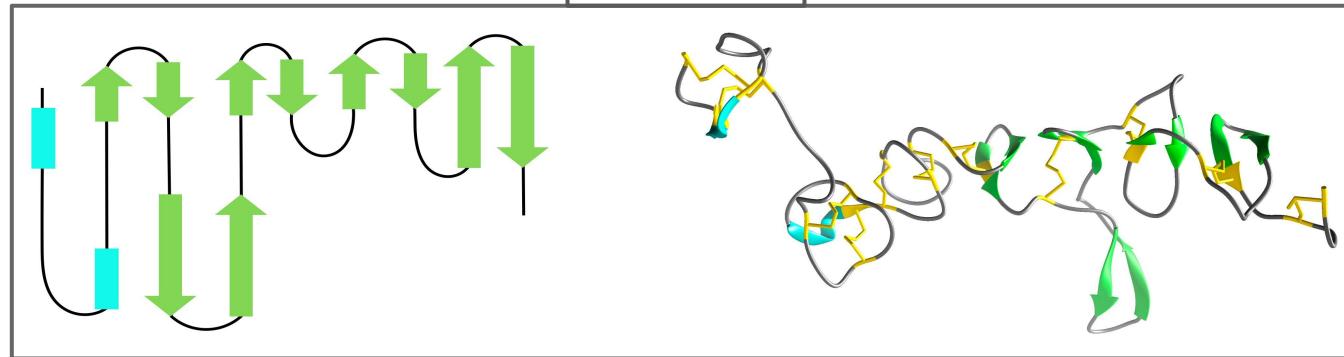
Domain III



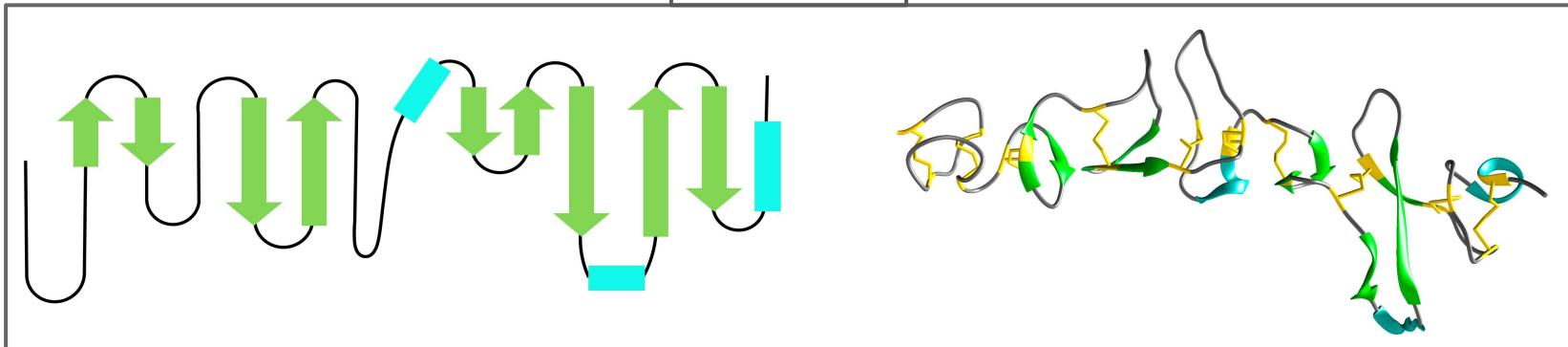
3. DOMAINS II and IV



Domain II



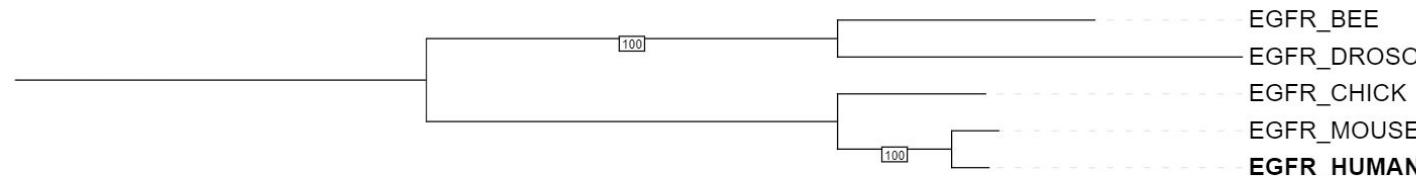
Domain IV



4. MSA and PHYLOGENY



	351	361	371	381	391
Consensus	q m d v N P E G K Y	s f G A T C V k k C	P - n y v v t D h G	s C V R s C p p D k	y e v e e e g - r k
Conservation					
EGFR_HUMAN	Q M D V N P E G K Y	S F G A T C V K K C	P R N Y V V T D H G	S C V R A C G A D S	Y E M E E D G V R K
EGFR_MOUSE	Q M D V N P E G K Y	S F G A T C V K K C	P R N Y V V T D H G	S C V R A C G P D Y	Y E V E E D G I R K
EGFR_CHICK	Q M D V N P E G K Y	S F G A T C V R E C	P H N Y V V T D H G	S C V R S C N T D T	Y E V E E N G V R K
EGFR_DROSO	V L E T N P E G K Y	A Y G A T C V K E C	P - G H L L R D N G	A C V R S C P Q D K	M D K G G E - - - -
EGFR_BEE	S W E P N P D G K Y	A Y G A T C V R R C	P - E H L L K D N G	A C V R S C P P K K	K A L N G E - - - -



LIGANDS STRUCTURE



- 1 SCOPE + EGF structure
- 2 EGF MSA and phylogeny
- 3 TGF- α structure
- 4 TGFA- α MSA and phylogeny
- 5 EREG structure
- 6 EREG MSA and phylogeny
- 7 Ligands superimposition

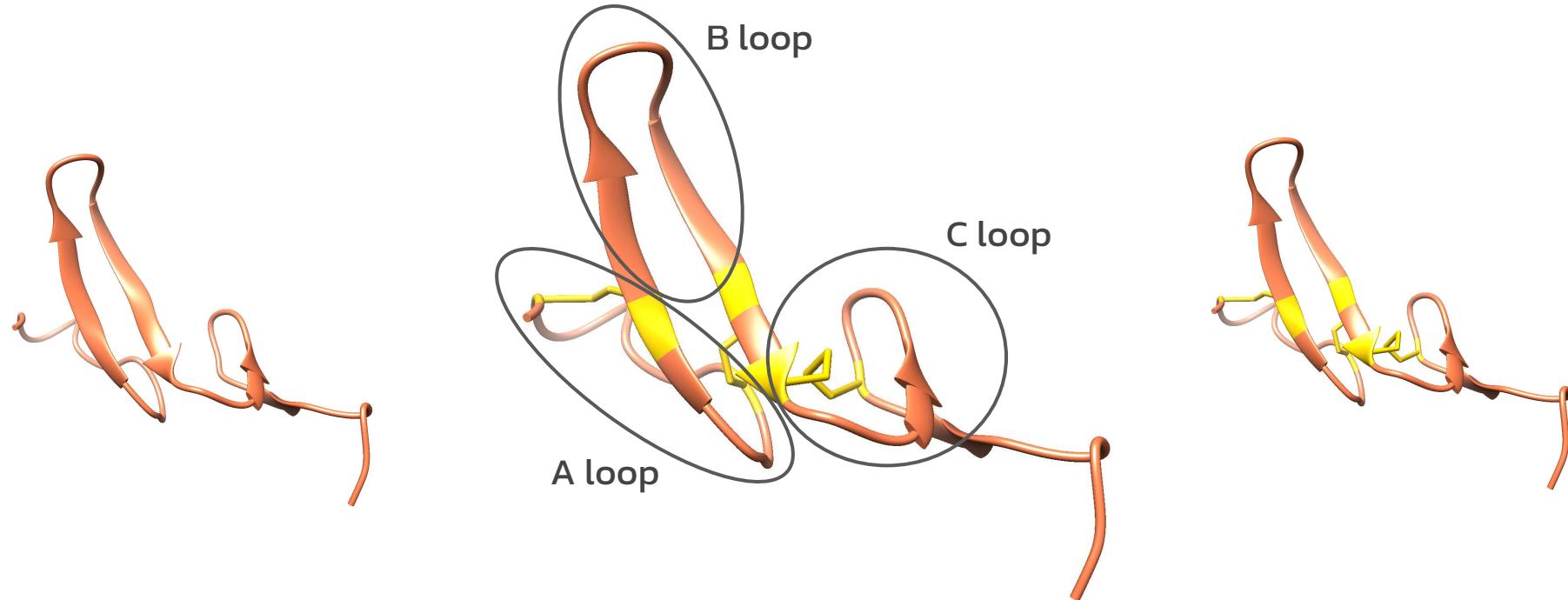
1.1 SCOP CLASSIFICATION



Epidermal growth factor (EGF)

Class	Small proteins
Fold	Knottins (disulfide-bound fold, contains beta hairpin with two adjacent disulfides)
Superfamily	EGF/laminin
Family	EGF type module
Protein	Epidermal growth factor (EGF)
Species	<i>Homo sapiens</i>

1.2 EGF STRUCTURE



EGF: ORANGE

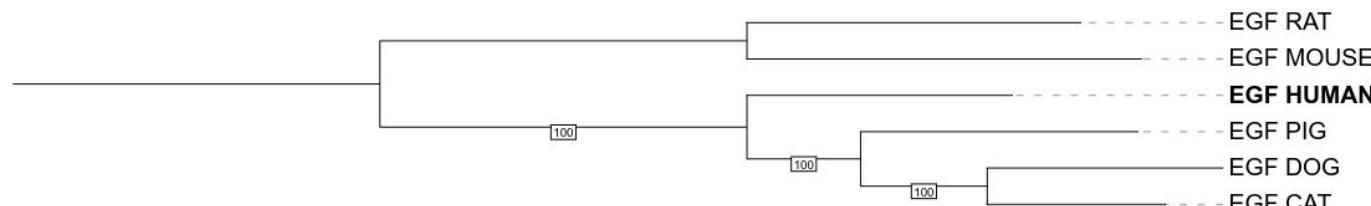
Cysteines and disulfide bonds

PDB: 3NJP

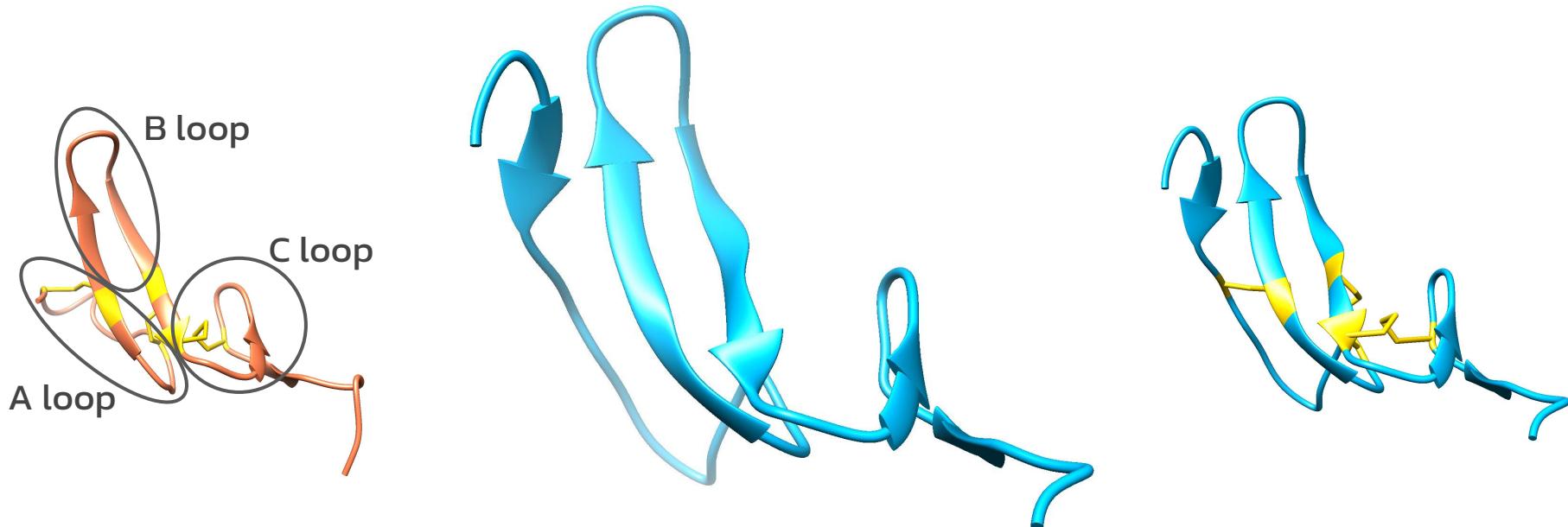
2. EGF MSA AND PHYLOGENY



	51	61	71	81	91
Consensus	GP a PFLIFSh	Gn SIFRIDle	GTN he QLVA D	AG v SV i IMDFH	Y n k E R i YWVD
Conservation					
EGF_CAT	GPE PFLIFSh	GG SIFRIDLD	GT NY EQLVAD	AG VSV IMDFH	YY K ERL YWVD
EGF_DOG	GPAP PFLIFSh	G I SIFRIDLE	GT NH EQLVAD	AG VSV IMDFH	YN K ERI YWVD
EGF_PIG	GPAP PFLIFSh	GNS IFRIDLE	GT NH EQLVAD	AG ISV LMDFH	YNE E R I YWVD
EGF_HUMAN	GPAP PFLIFSh	GNS IFRIDTE	GT NY EQLVVD	AG VSV IMDFH	YNE K R I YWVD
EGF_MOUSE	GPAP PFLVFSQ	GKS ISR IDPD	GT NH HQQLVVD	AG ISADMDI H	YKK ERL YWVD
EGF_RAT	GPP RFLIFLQ	GNS IFRINTD	GT NH HQQLVVD	AG VSV VMDFH	YKE ERL YWVD



3. TGF- α STRUCTURE



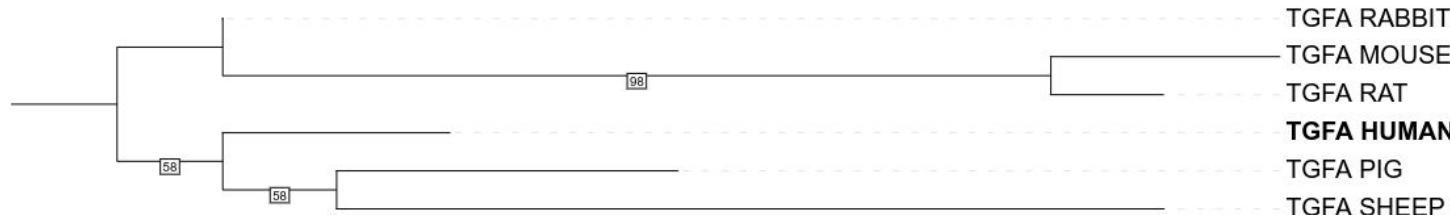
Cysteines and disulfide bonds

PDB: 1MOX

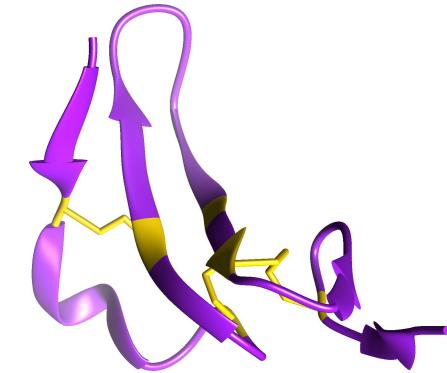
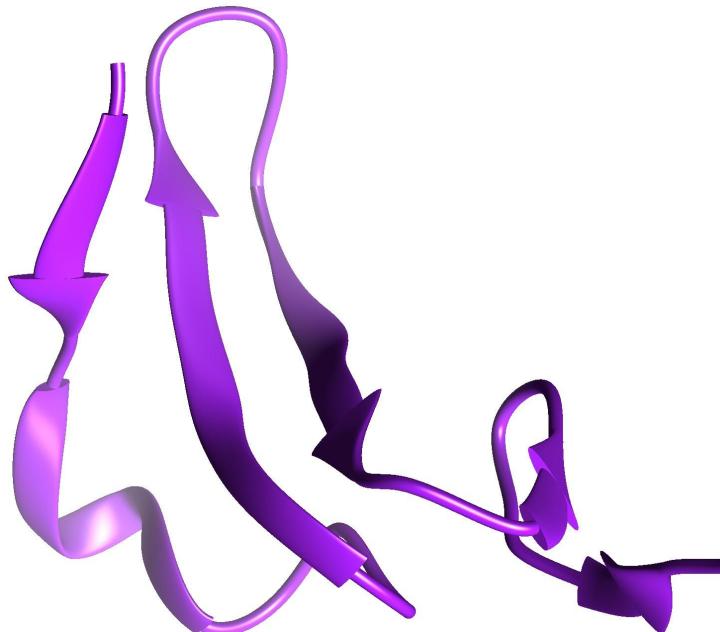
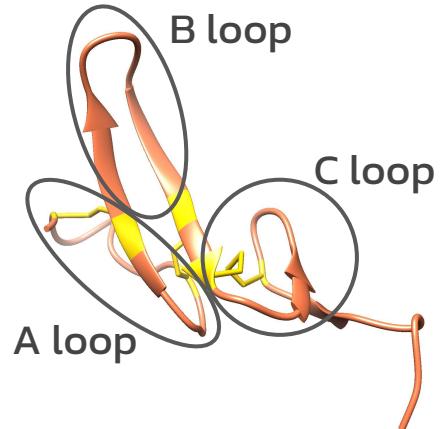
4. TGF- α MSA AND PHYLOGENY



Consensus Conservation	51	61	71	81	91
TGFA_RAT	HTQYCFHGT	RFLVQEEKPA	CVCHSGYVG	RCEHADLLA	VAA\$QKKQA
TGFA_MOUSE	HTQYCFHGT	RFLVQEEKPA	CVCHSGYVG	RCEHADLLA	VAA\$QKKQA
TGFA_PIG	HSQFCFHGT	RFLVQEDKPA	CVCHSGYVG	RCEHADLLA	VAA\$QKKQA
TGFA_HUMAN	HTQFCFHGT	RFLVQEDKPA	CVCHSGYVG	RCEHADLLA	VAA\$QKKQA
TGFA_SHEEP	HTQFCFHGT	RFLLQEEKPA	CVCHSGYVG	RCEHADLLA	VAA\$QKKQA
TGFA_RABBIT	HTQFCFHGT	RFLVQEDKPA	CVCHSGYVG	RCEHADLLA	VAA\$QKKQA



5. EREG STRUCTURE

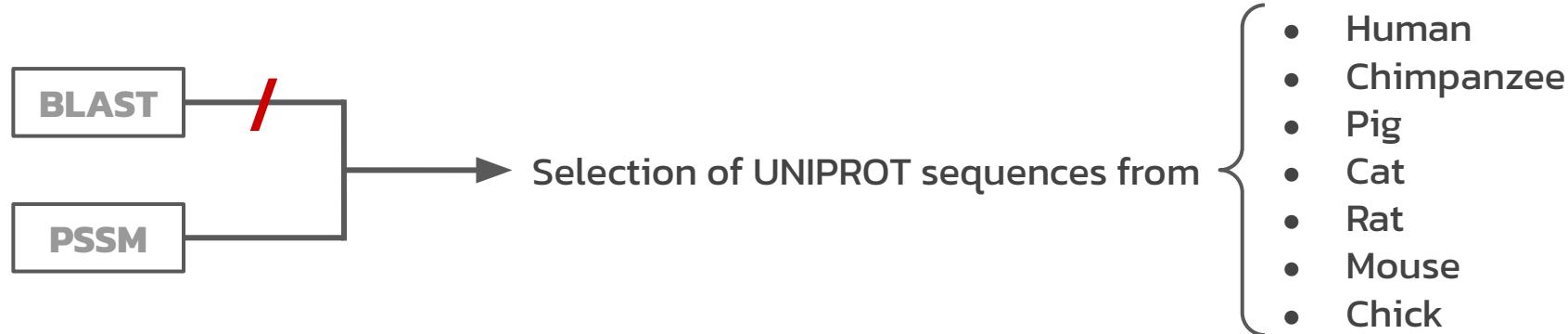


EGF: ORANGE
EREG: PURPLE

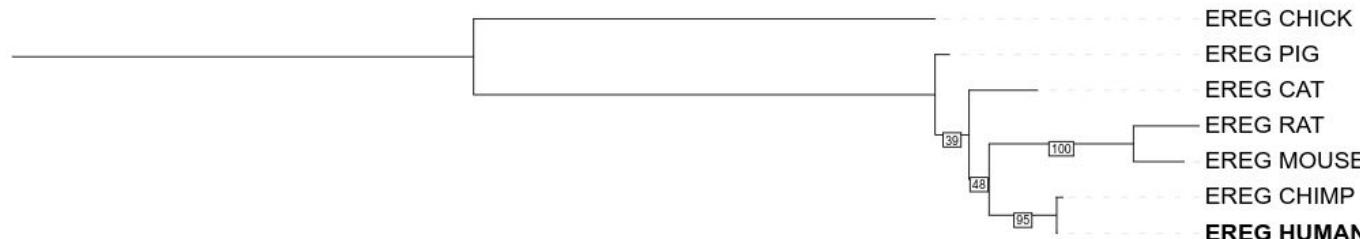
Cysteines and disulfide bonds

PDB: 5WB7

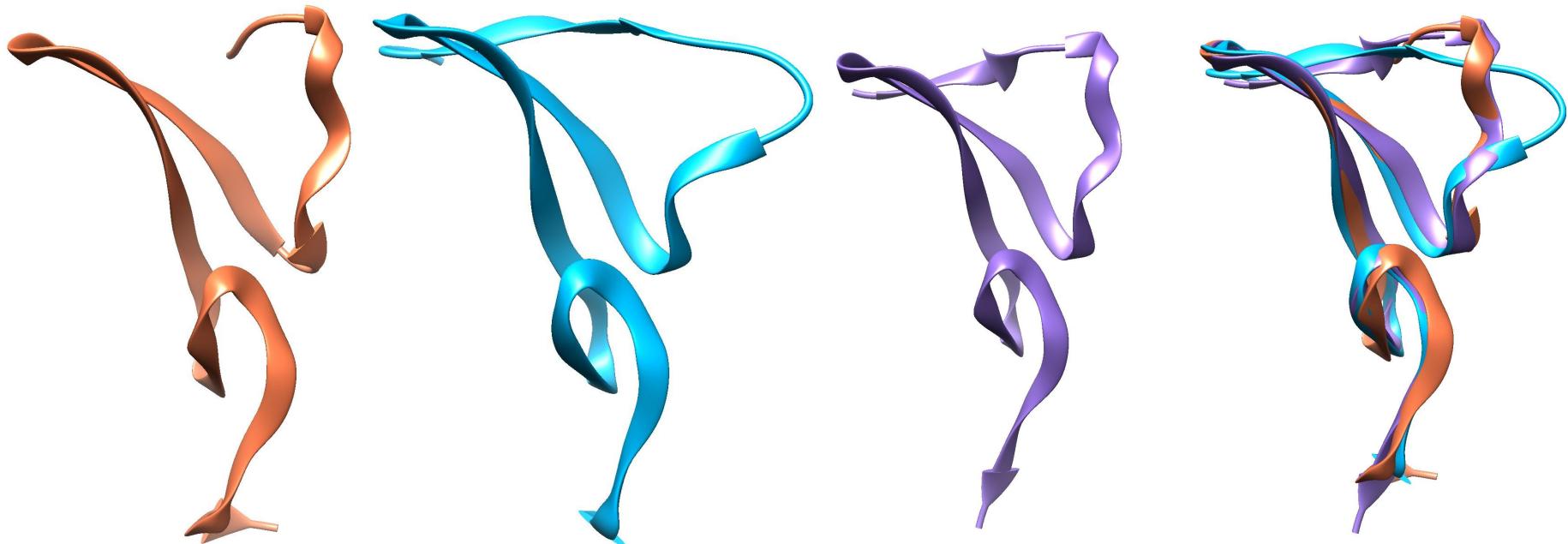
6. EREG MSA AND PHYLOGENY



	51	61	71	81	91
Consensus	LVQ t ED n PRV	A QV s I TK C s S	D M n G Y C L H G Q	C I Y L V D M s e n	Y C R C E V G Y T G
Conservation	L V Q M E D D P R V	A Q V Q I T K C S S	D M D G Y C L H G Q	C I Y L V D M R E K	F C R C E V G Y T G
EREG_MOUSE	LVQM EDDPRV	AQVQITKCSS	DMDGYCLHGQ	C I Y L V D M R E K	F C R C E V G Y T G
EREG_RAT	LVQM EDDPRV	AQVLITKCSS	DMDGYCLHGQH	C I Y L V D M S E K	Y C R C E V G Y T G
EREG_HUMAN	LVQTE DNP RV	A Q V S I T K C S S	D M N G Y C L H G Q	C I Y L V D M S Q N	Y C R C E V G Y T G
EREG_CHIMP	LVQTE DNP RV	A Q V S I T K C S S	D M N G Y C L H G Q	C I Y L V D M S Q N	Y C R C E V G Y T G
EREG_CAT	LVQTE DNP RV	A Q V S I T K C G S	D M N G F C L H G Q	C I Y L V D M S E N	Y C R C E V G Y T G
EREG_PIG	LVQTE DNP RV	A Q V S I T K C S S	D M N G Y C L H G Q	C I Y L V D M S E N	Y C R C E V G Y T G
EREG_CHICK	LVQTE N S P R V	A Q V G I T R C K P	E M K D Y C F H Q Q	C V Y I V D L D E H	Y C R C D V G F S G



7. LIGANDS SUPERIMPOSITION



EGF

+

TGF- α

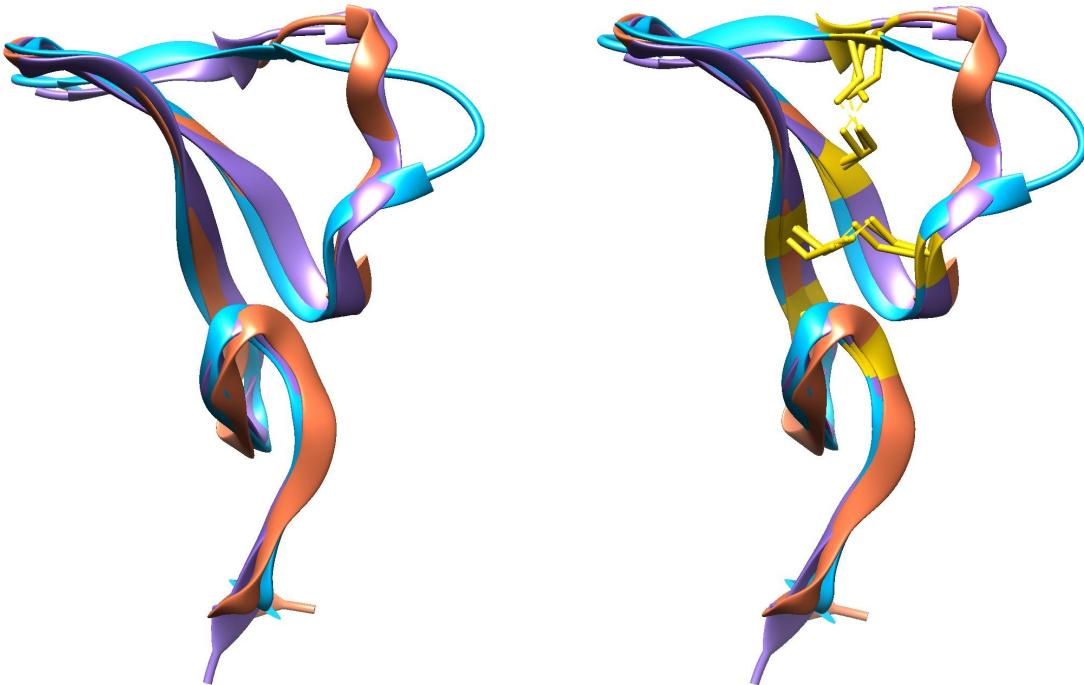
+

Epiregulin

\Rightarrow

Superimposition

7. LIGANDS SUPERIMPOSITION

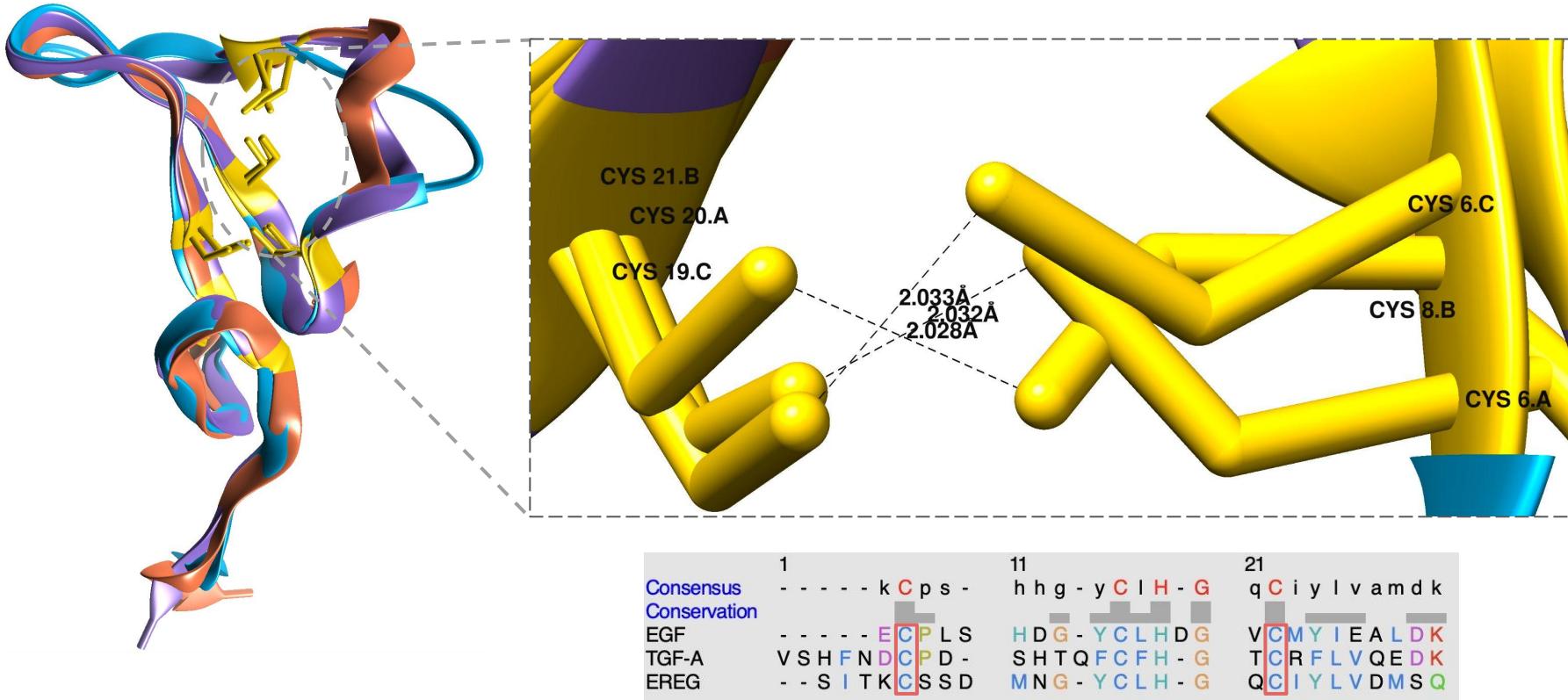


Score	8.34
RMSD	1.27
Len	48
nfit	42

7. LIGANDS SUPERIMPOSITION



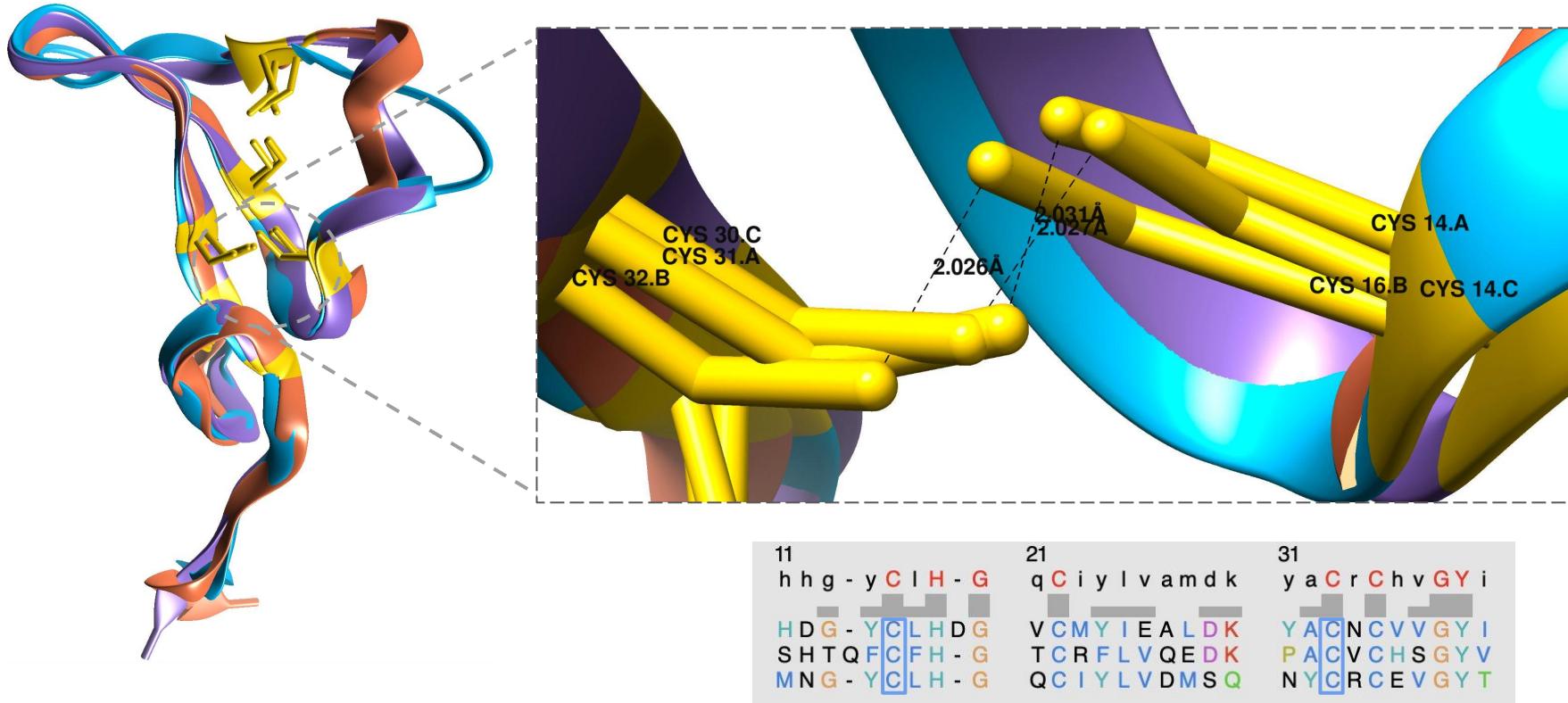
Disulphide bonds



7. LIGANDS SUPERIMPOSITION



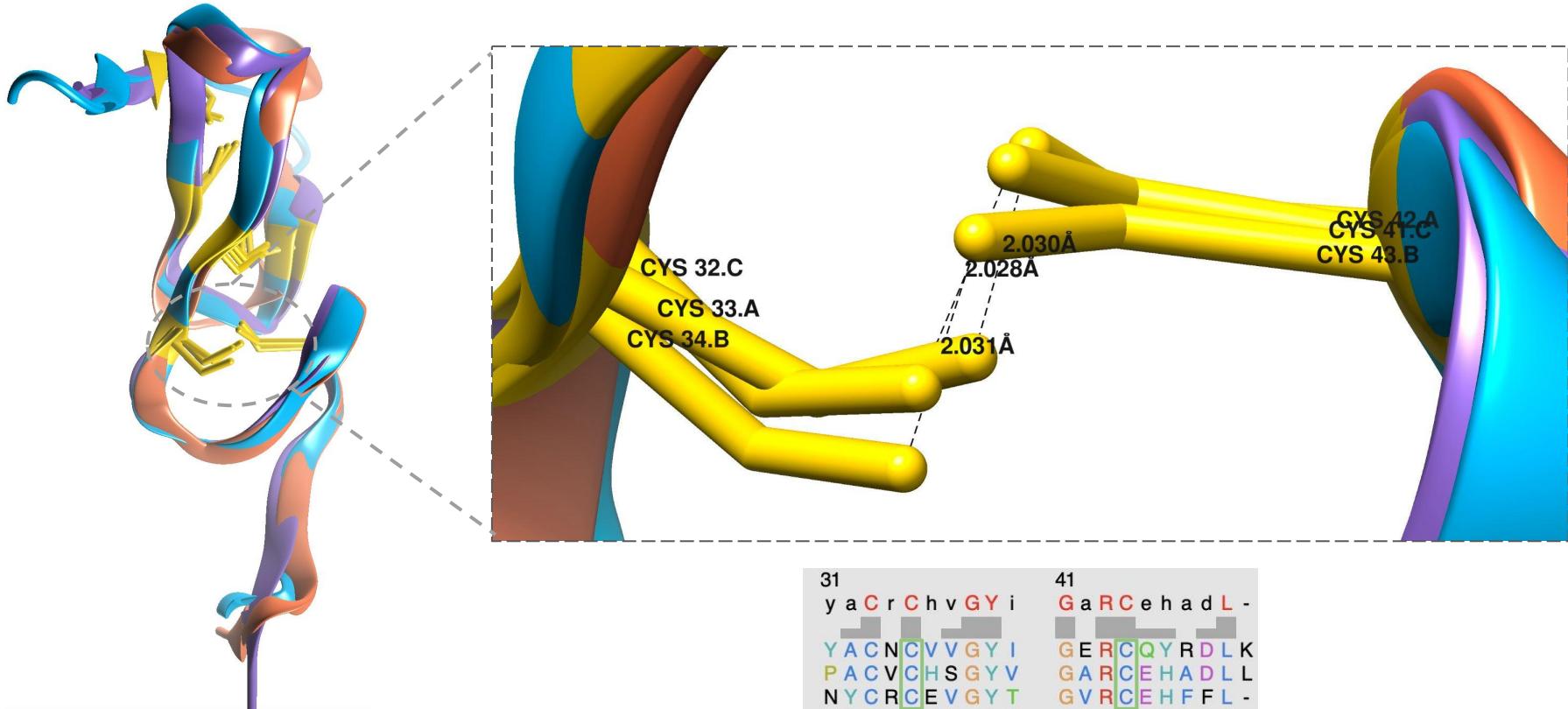
Disulphide bonds



7. LIGANDS SUPERIMPOSITION



Disulphide bonds



DIMERIZATION AND INTERACTIONS



1 Autoinhibited monomeric conformation

2 Dimerization

3 Symmetrical vs Asymmetrical dimerization

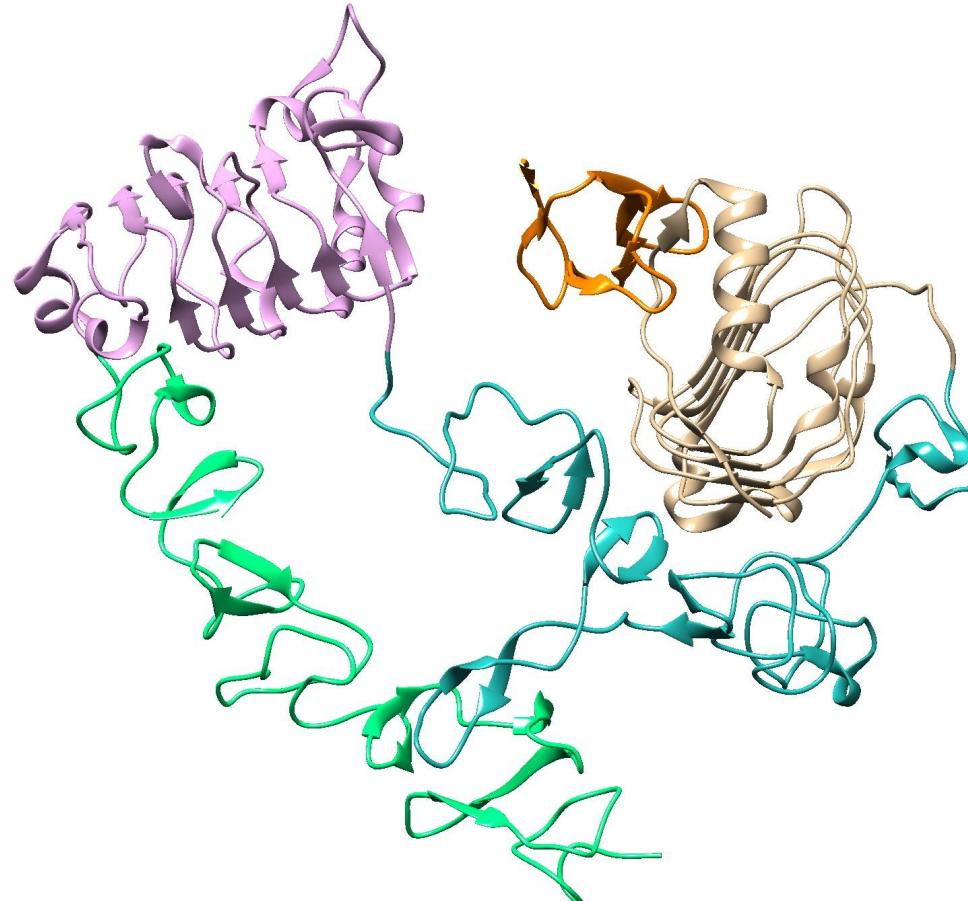
3.1 Symmetrical

3.2 Asymmetrical

1. AUTOINHIBITED MONOMERIC CONFORMATION



EGFR-EGF



*EGB-binding at
low pH conditions*

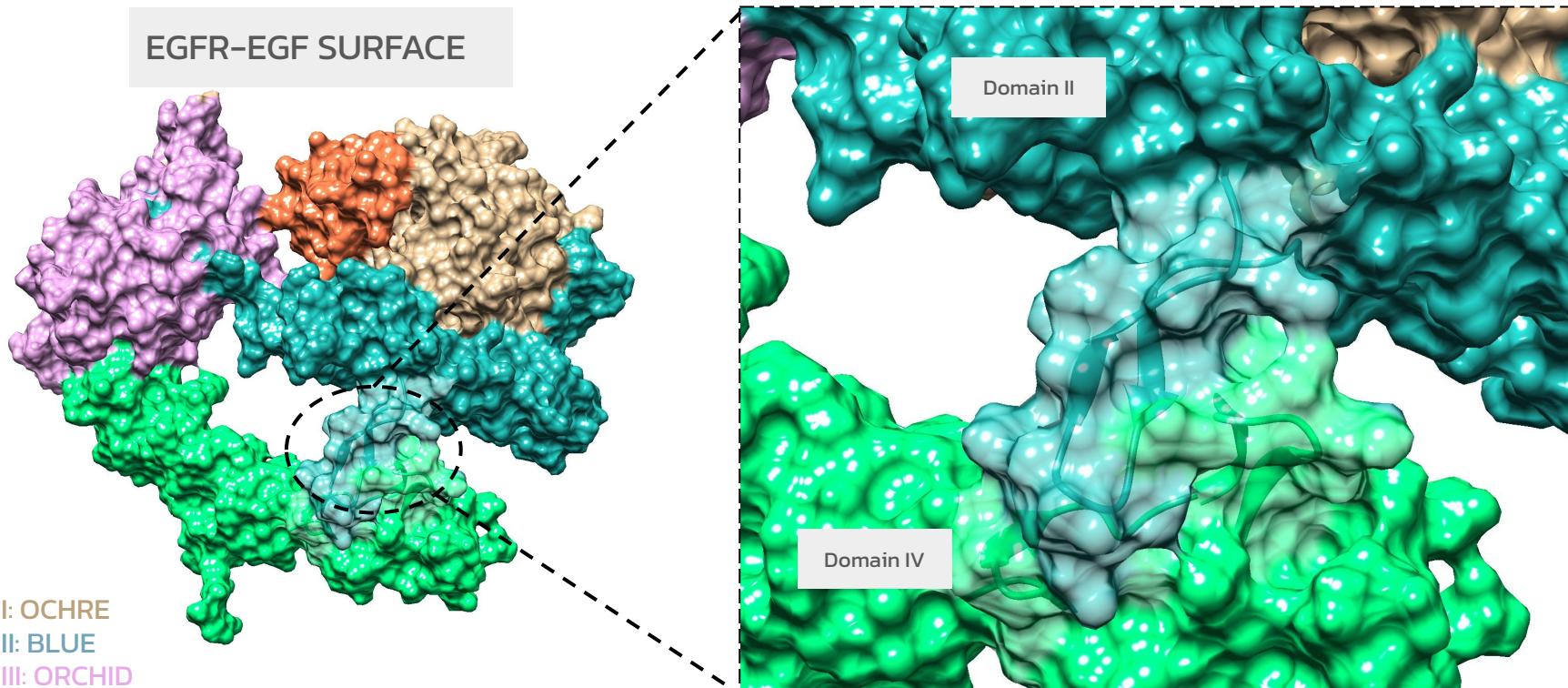
EGFR DI: OCHRE
EGFR DII: BLUE
EGFR DIII: ORCHID
EGFR DIV: GREEN
EGF: ORANGE

PDB: 1NQL

1. AUTOINHIBITED MONOMERIC CONFORMATION



Tethered dimerization arm

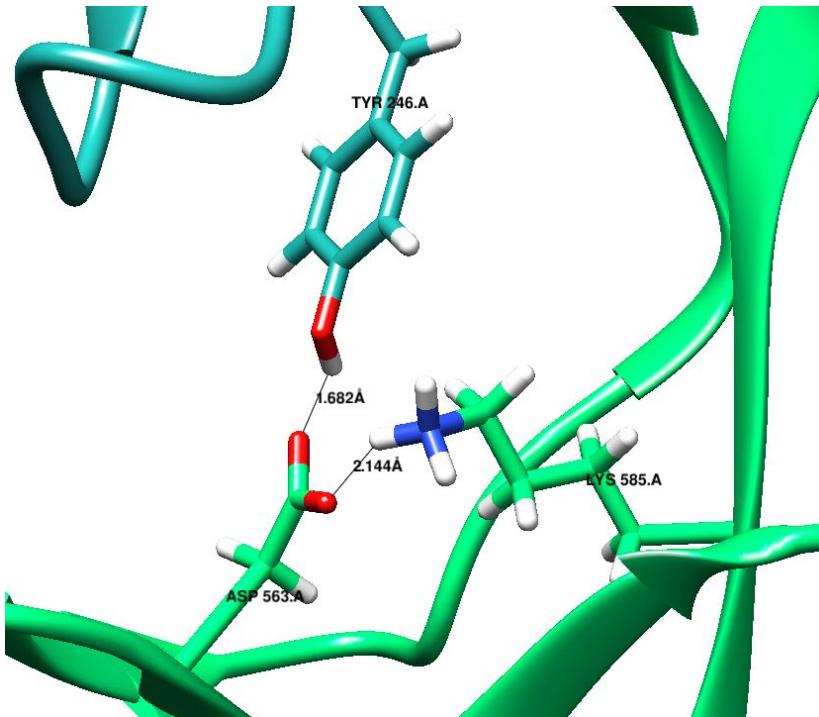


PDB: 1NQL

1. AUTOINHIBITED MONOMERIC CONFORMATION



Tethered dimerization arm



Consensus
Conservation
EGFR_HUMAN
EGFR_MOUSE
EGFR_CHICK
EGFR_DROSO
EGFR_BEE

341
P I m I Y N P T T Y
P L M L Y N P T T Y
P L M L Y N P T T Y
P L V L Y N P T T Y
P M R K Y N P T T Y
P M Q K Y N P T T Y

Consensus
Conservation
EGFR_HUMAN
EGFR_MOUSE
EGFR_CHICK
EGFR_DROSO
EGFR_BEE

671
H y i D G P H C V k
H Y I D G P H C V K
H Y I D G P H C V K
H F I D G P H C V K
H V R D G Q H C V S
H A R D G P F C V P

Consensus
Conservation
EGFR_HUMAN
EGFR_MOUSE
EGFR_CHICK
EGFR_DROSO
EGFR_BEE

741
- d t l v W k Y a d
- N T L V W K Y A D
- N T L V W K Y A D
- D T L V W K Y A D
P D G Y F W E Y V H
P D G Y Y Y E W V S

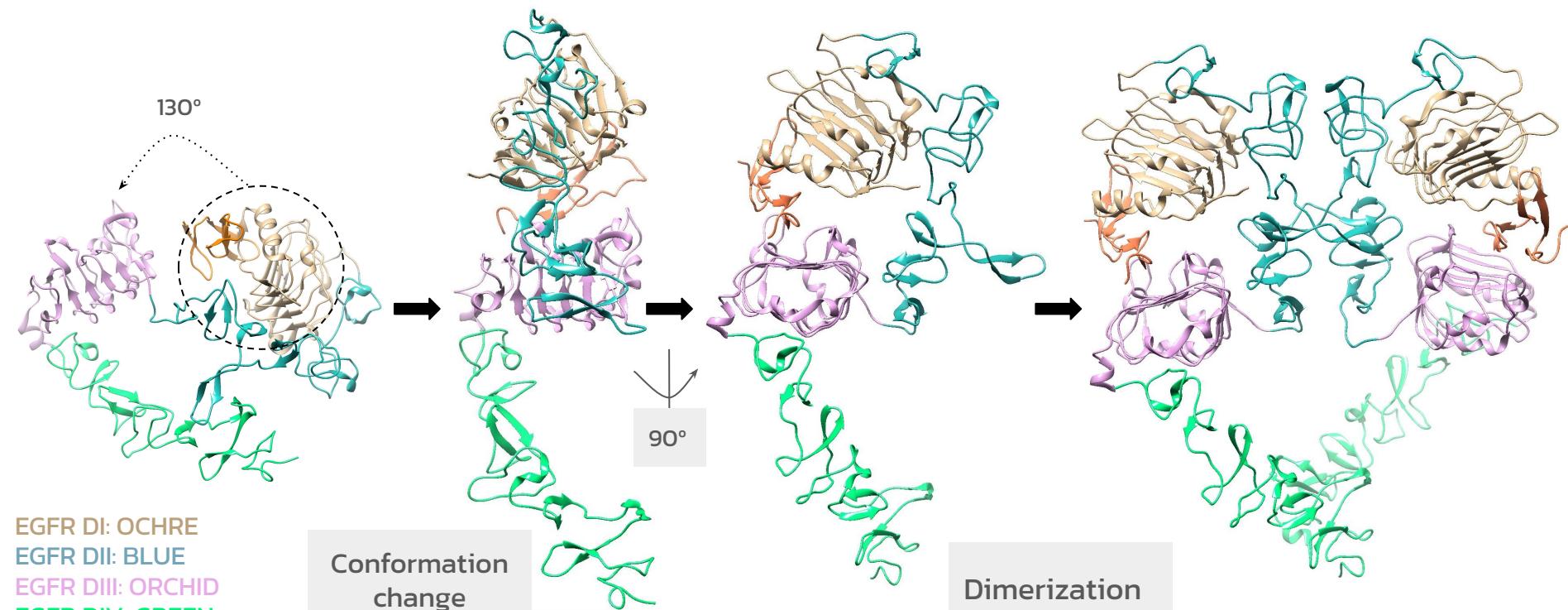
2. DIMERIZATION



Tethered
dimerization arm

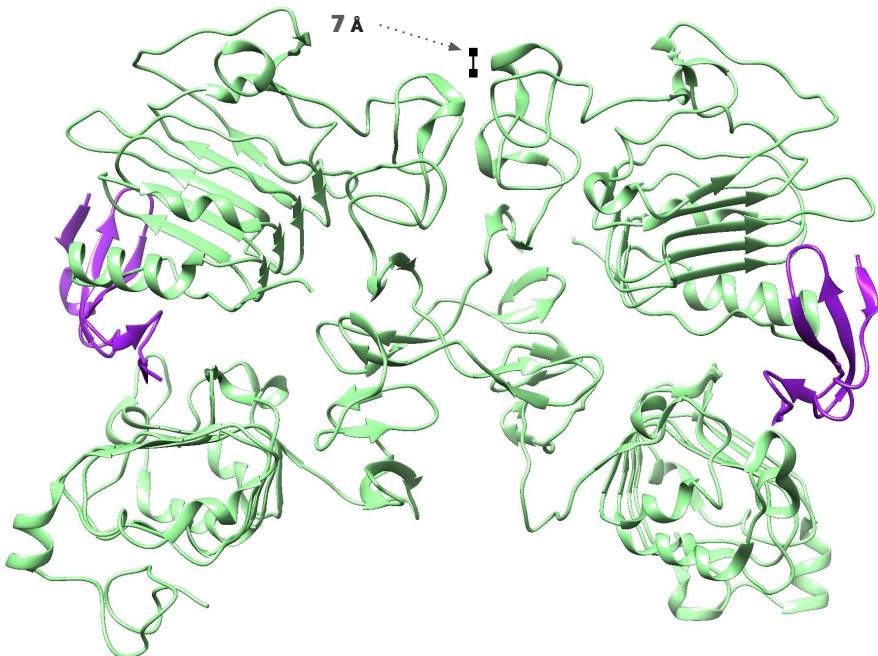
Monomeric
conformation

Dimeric
conformation



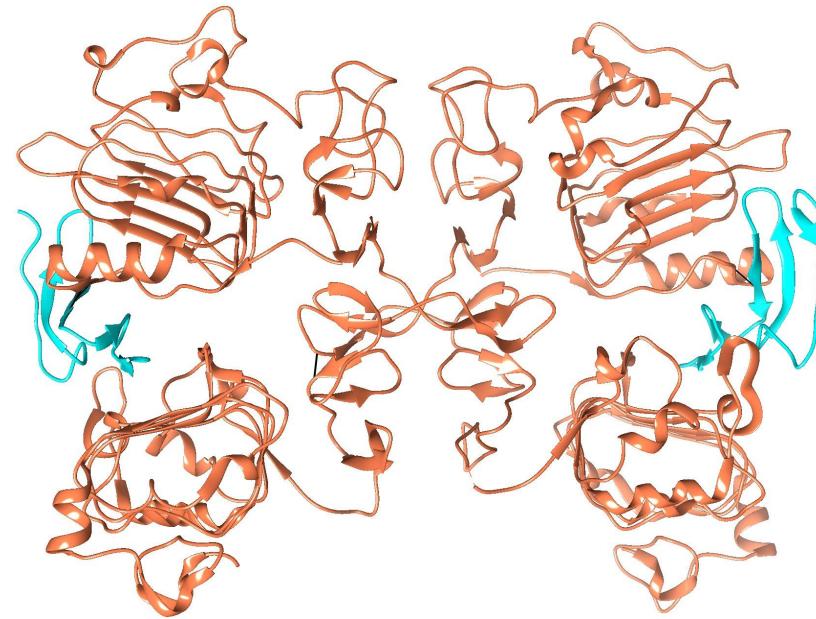
PDB: 1NQL, 3NJP

3. SYMMETRICAL VS ASYMMETRICAL DIMERIZATION



EGFR-EREG
asymmetrical

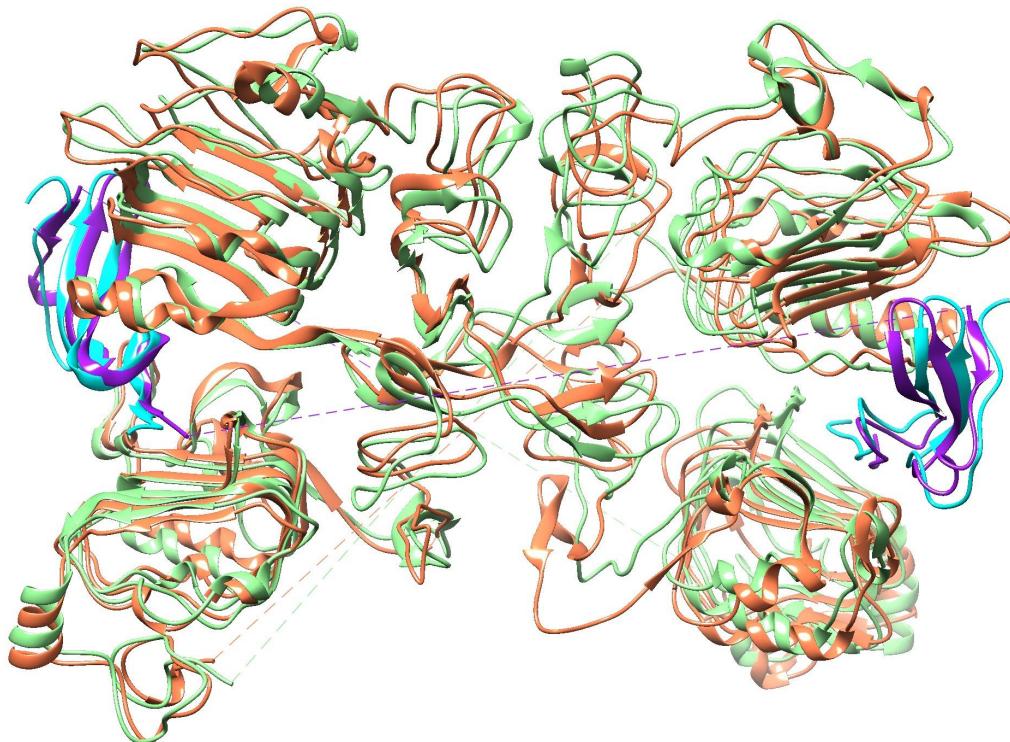
PDB: 5WB7



EGFR-TGF alfa
symmetrical

PDB: 1MOX

3. SYMMETRICAL VS ASYMMETRICAL DIMERIZATION



EGFR-EREG
asymmetrical

PDB: 5WB7

EGFR-TGF alfa
symmetrical

PDB: 1MOX

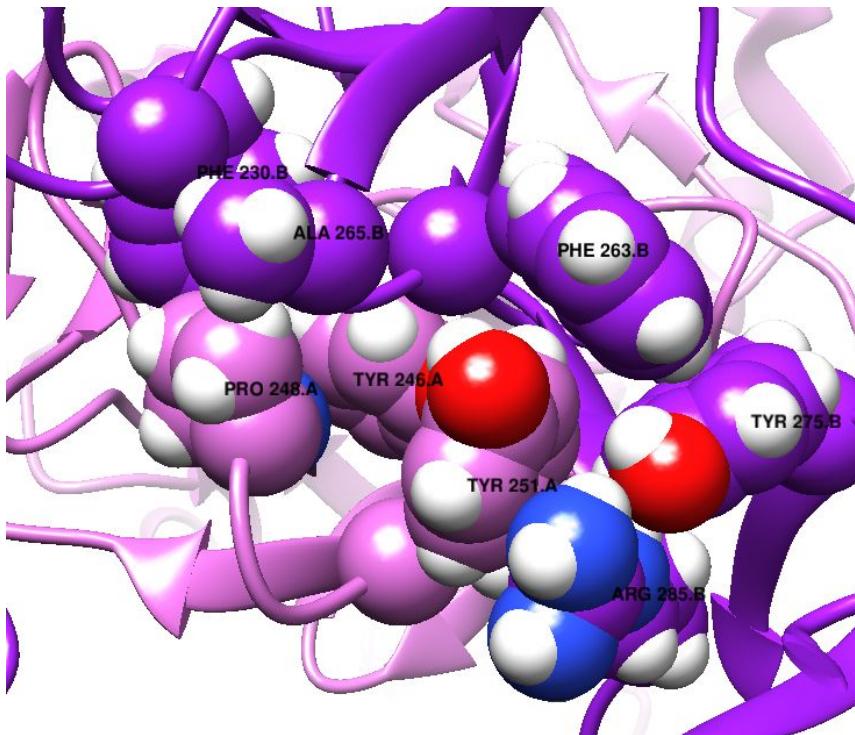
Score	6.69
RMSD	2.51
Len	1121
nfit	990

Data: STAMP

3.1. SYMMETRICAL DIMERIZATION



Receptor-Receptor interactions: DOMAIN II – DOMAIN II



Consensus
Conservation
EGFR_HUMAN
EGFR_MOUSE
EGFR_CHICK
EGFR_DROSO
EGFR_BEE

321
S D C I a C k k F r
S D C L V C R K F R
S D C L V C Q K F Q
S D C L A C R K F R
K D C I A C K N F F
S D C I A C K N F F

331
D e A t C K d t C P
D E A T C K D T C P
D E A T C K D T C P
D D A T C K D T C P
D E A V S K E E C P
D D G V C T Q E C P

341
P I m l Y N P T T Y
P L M L Y N P T T Y
P L V L Y N P T T Y
P M R K Y N P T T Y
P M Q K Y N P T T Y

Consensus
Conservation
EGFR_HUMAN
EGFR_MOUSE
EGFR_CHICK
EGFR_DROSO
EGFR_BEE

361
s f G A T C V k k C
S F G A T C V K K C
S F G A T C V K K C
S F G A T C V R E C
A Y G A T C V K E C
A Y G A T C V R R C

371
P - n y v v t D h G
P R N Y V V T D H G
P R N Y V V T D H G
P H N Y V V T D H G
P - G H L L R D N G
P - E H L L K D N G

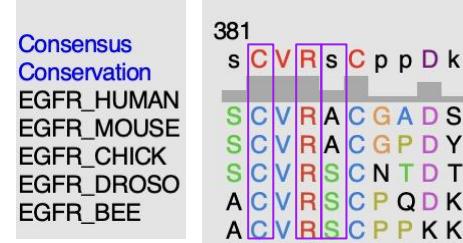
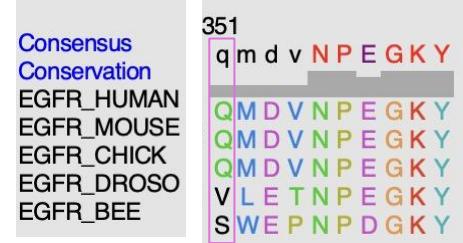
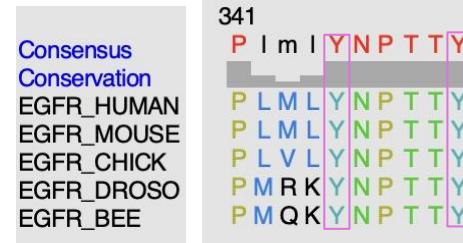
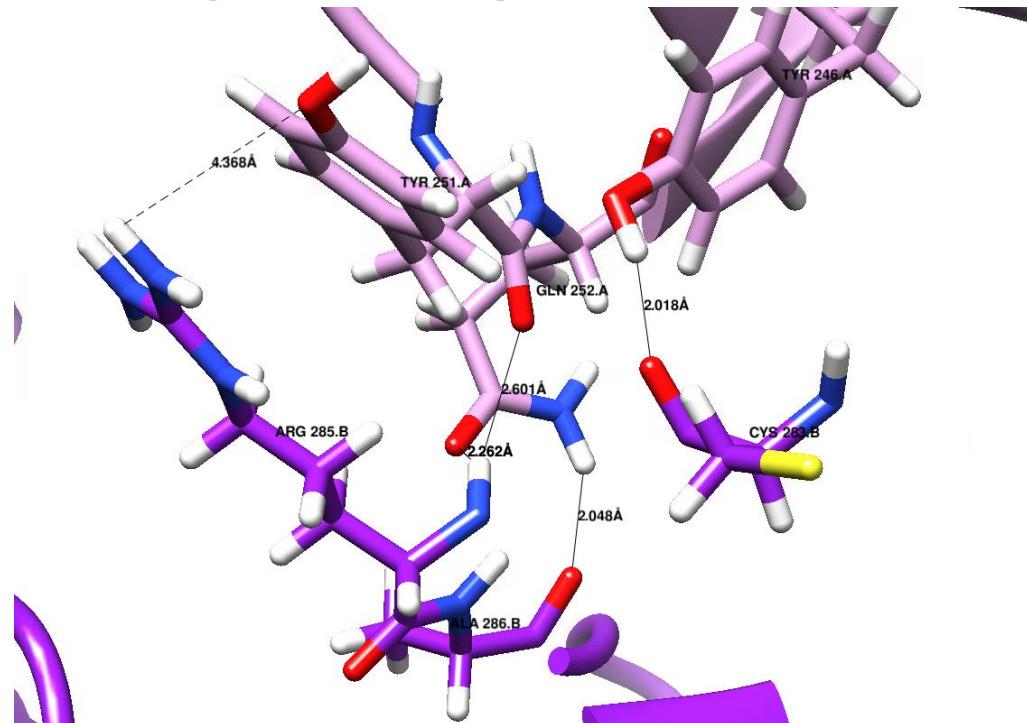
381
s C V R s C p p D k
S C V R A C G A D S
S C V R A C G P D Y
S C V R S C N T D T
A C V R S C P Q D K
A C V R S C P P K K

EGFR DII_R: ORCHID
EGFR DII_L: PURPLE

3.1. SYMMETRICAL DIMERIZATION



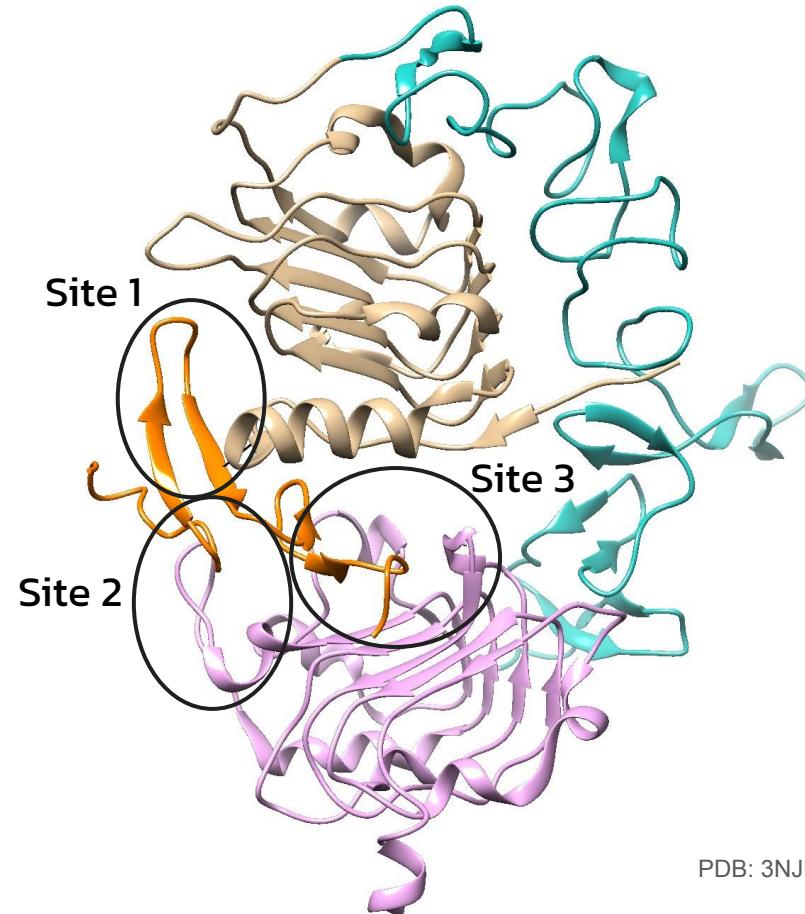
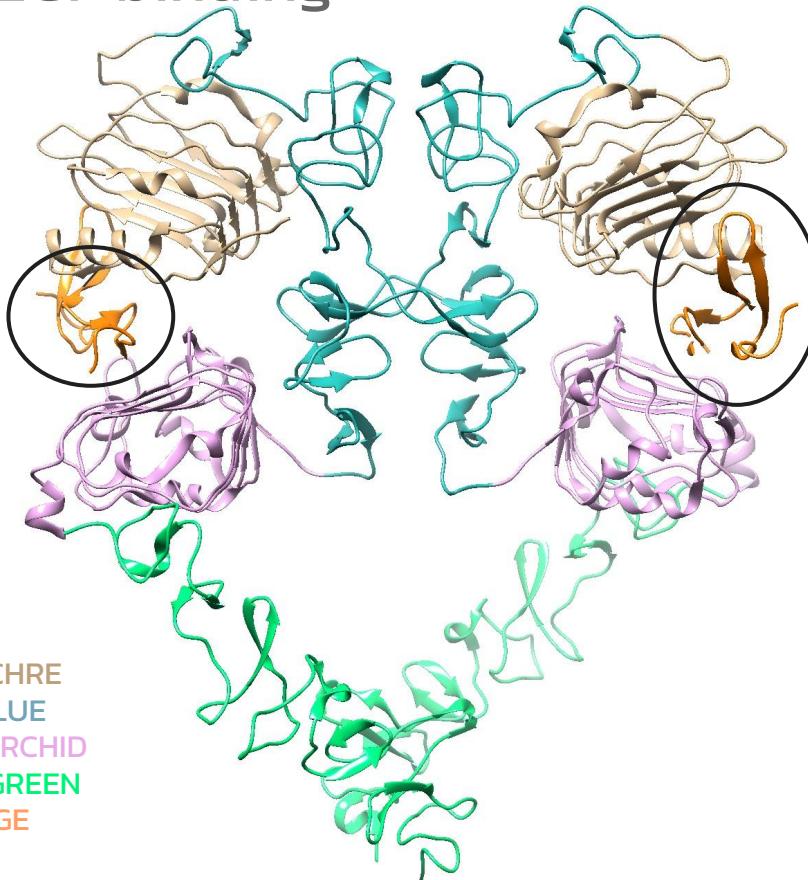
Receptor-Receptor interactions: DOMAIN II – DOMAIN II



3.1. SYMMETRICAL DIMERIZATION



EGF binding

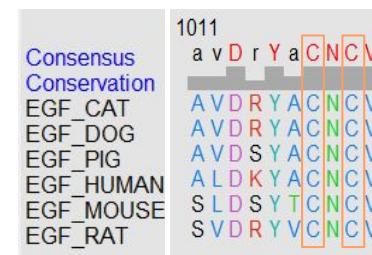
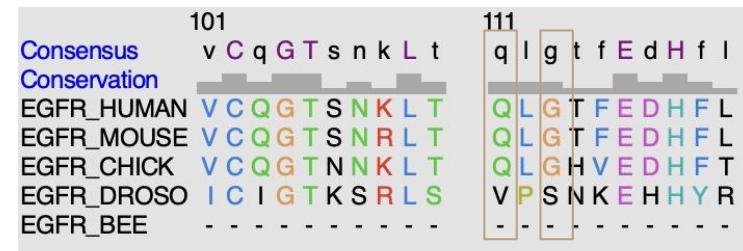
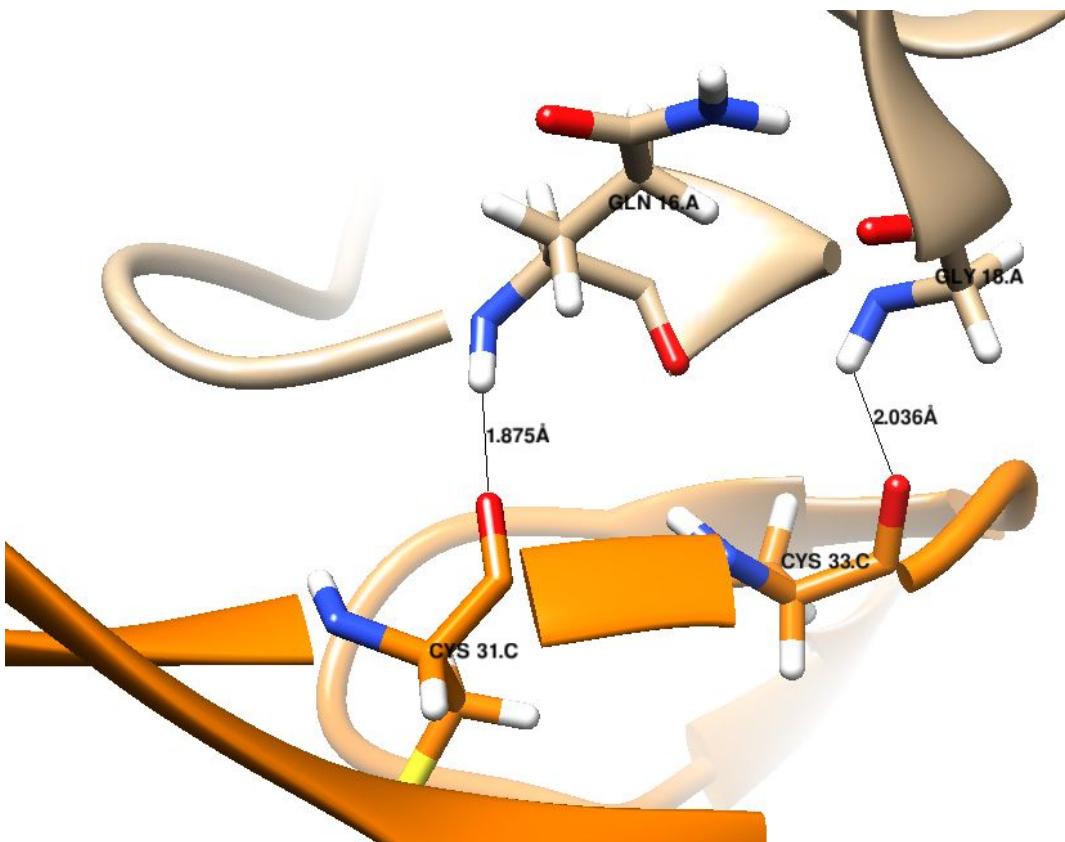


PDB: 3NJP

3.1. SYMMETRICAL DIMERIZATION



EGF binding: S1 Hydrogen bonds – DOMAIN I

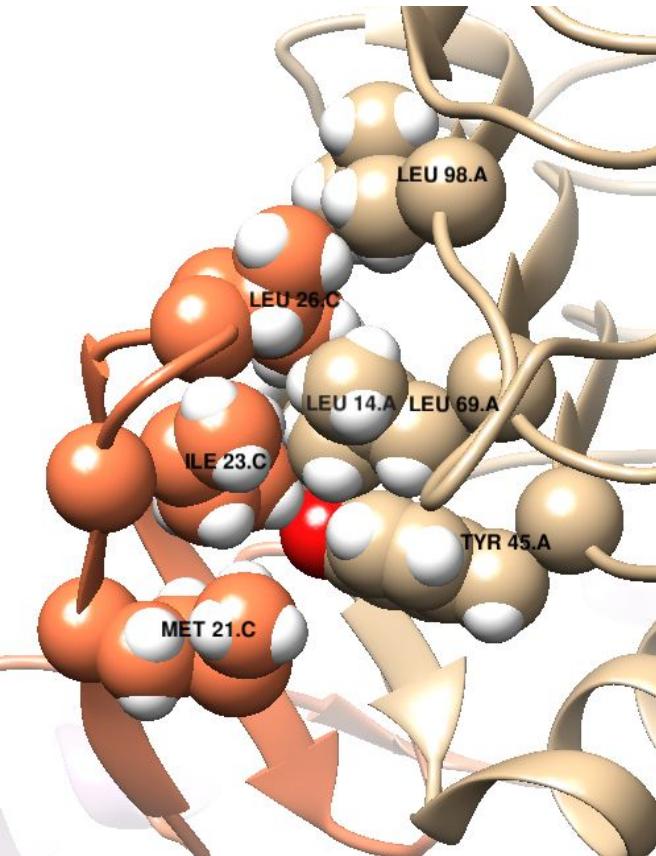


EGFR DI: OCHRE
EGF: ORANGE

3.1. SYMMETRICAL DIMERIZATION



EGF binding S1: Hydrophobic contacts – DOMAIN I



Consensus	101	111	121	131	141
Conservation	v C q G T s n k L t	q l g t f E d H f l	s L q r m y n N C e	v V l g N L e I t Y	v Q R - N y D L S F
EGFR_HUMAN	VCQGTSNKLT	QLGTFEDHFL	SLQRMFNNCE	VVLGNLEITY	VQR-NYDLSSF
EGFR_MOUSE	VCQGTSNKLT	QLGTFEDHFL	SLQRMYNNCE	VVLGNLEITY	VQR-NYDLSSF
EGFR_CHICK	VCQGTTNNKL	QLGHVEDHFT	SLQRMYNNCE	VVLSNLEITY	VEH-NRDLTF
EGFR_DROSO	ICIGTKSRLS	VPSNKEHYR	NLRDRYTNCT	YVDGNLKL	LPNENLDLSF
EGFR_BEE					
Consensus	151	161	171	181	191
Conservation	L k t I q E V a G Y	v L I a l n t V e R	i p l e n L Q I I R	G n - l y e n s y a	l a v l s n y - t n
EGFR_HUMAN	LKTIQEVAGY	VLIALNTVER	IPLENLQIIR	GNMYYENSYA	LAVLSNYDAN
EGFR_MOUSE	LKTIQEVAGY	VLIALNTVER	IPLENLQIIR	GNALYENTYA	LAIALSNYGTN
EGFR_CHICK	LKTIQEVAGY	VLIALNMVDV	IPLENLQIIR	GNVLYDNSFA	LAIVLSNYHMN
EGFR_DROSO	LDNIREVTGY	ILLISHVDVKK	VVFPKLQIIR	GRTLFSLSV	EEKYALFVT
EGFR_BEE					
Consensus	1001	1011			
Conservation	L y g G V C M Y I E	a v D r Y a C N C V			
EGF_CAT	LYNGVCMYIE	AVDRYACNCV			
EGF_DOG	LYNGVCMYIE	AVDRYACNCV			
EGF_PIG	LHGGVCMYIE	AVDSYACNCV			
EGF_HUMAN	LHDGVCAMYIE	ALDKYACNCV			
EGF_MOUSE	LNGGVCMHIE	SLDSYTCNCV			
EGF_RAT	LNGGVCAMYIE	SVDRYVCNCV			

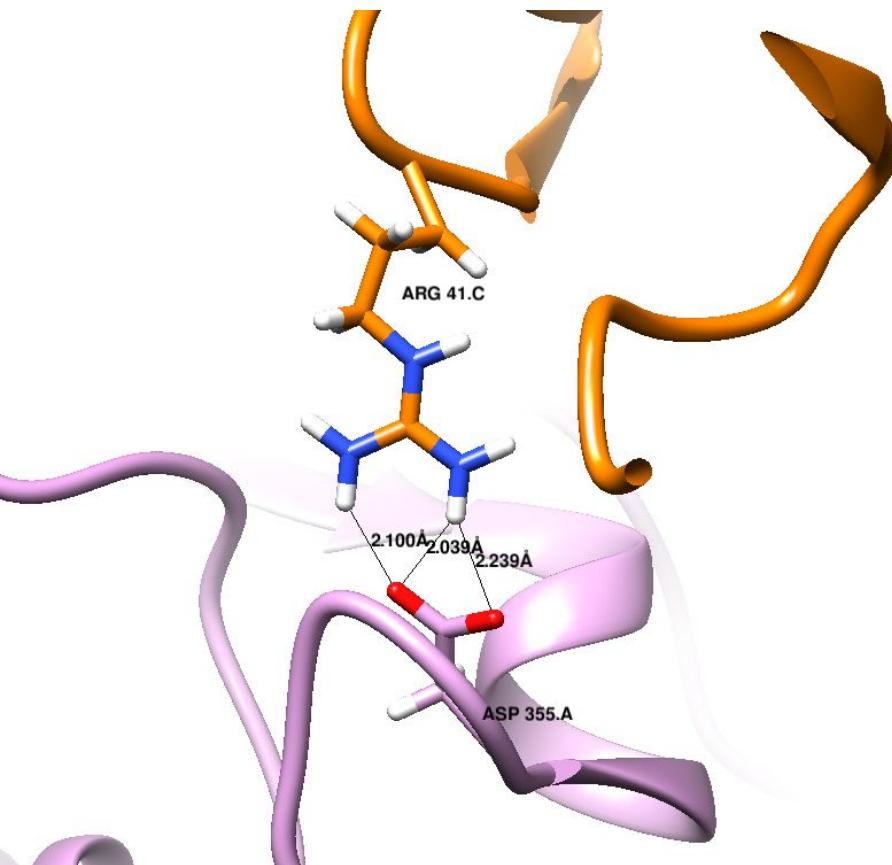
EGFR DI: OCHRE

EGF: ORANGE

3.1. SYMMETRICAL DIMERIZATION



EGF binding S2: Salt bridge – DOMAIN III



Consensus	F	s	G	d	q	f	t	y	t	-
Conservation	█	█	█	█	█	█	█	█	█	█
EGFR_HUMAN	F	R	G	D	S	F	T	H	T	-
EGFR_MOUSE	F	K	G	D	S	F	T	R	T	-
EGFR_CHICK	F	L	G	D	A	F	T	K	T	-
EGFR_DROSO	F	S	C	F	Q	D	V	Y	A	N
EGFR_BEE	F	Q	G	F	Q	H	V	Y	R	N

Consensus	f	G	Y	v	G	E	R	C	Q	h
Conservation	█	█	█	█	█	█	█	█	█	█
EGF_CAT	F	G	Y	V	G	E	R	C	Q	H
EGF_DOG	F	G	Y	V	G	E	R	C	Q	H
EGF_PIG	F	G	Y	V	G	E	R	C	Q	H
EGF_HUMAN	V	G	Y	I	G	E	R	C	Q	Y
EGF_MOUSE	I	G	Y	S	G	D	R	C	Q	T
EGF_RAT	I	G	Y	I	G	E	R	C	Q	H

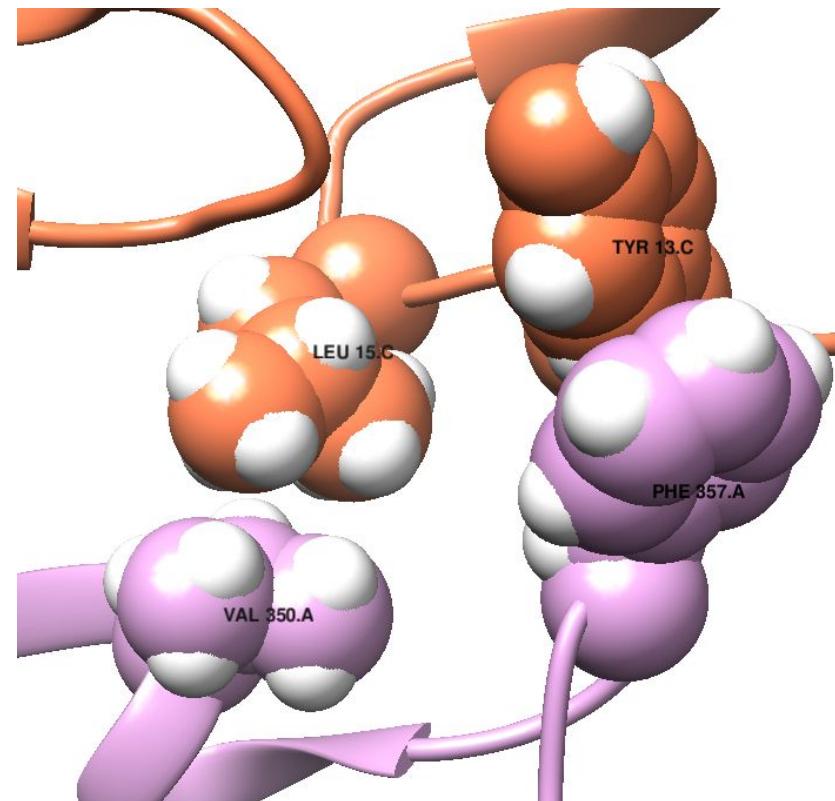
EGFR DIII: ORCHID

EGF: ORANGE

3.1. SYMMETRICAL DIMERIZATION



EGF binding S2: Hydrophobic contacts – DOMAIN III



Consensus	441	451
Conservation	s G d i h I L p v a F s G d c f t y t -	
EGFR_HUMAN	S G D L H I L P V A F R G D S F T H T -	
EGFR_MOUSE	S G D L H I L P V A F K G D S F T R T -	
EGFR_CHICK	N G D V S I L P V A F L G D A F T K T -	
EGFR_DROSO	D G N I R I L D Q T F S G F Q D V Y A N	
EGFR_BEE	E G S I T I L D Q S F Q G F Q H V Y R N	

Consensus	991	1001
Conservation	e C P p S y D G Y C	L y g G V C M Y I E
EGF_CAT	E C P P S Y D G Y C	L Y N G V C M Y I E
EGF_DOG	E C P S S Y D G Y C	L Y N G V C M Y I E
EGF_PIG	E C P P S H D G Y C	L H G G V C M Y I E
EGF_HUMAN	E C P L S H D G Y C	L H D G V C M Y I E
EGF_MOUSE	G C P S S Y D G Y C	L N G G V C M H I E
EGF_RAT	G C P P S Y D G Y C	L N G G V C M Y V E

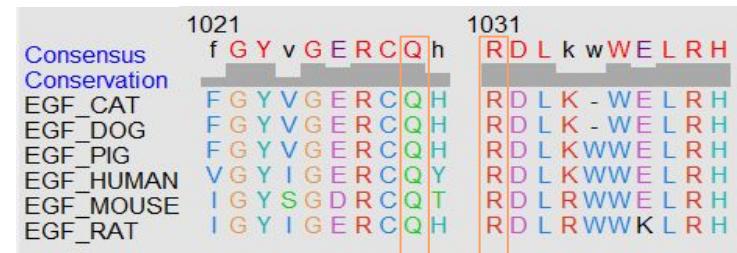
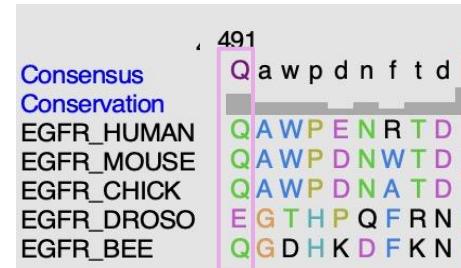
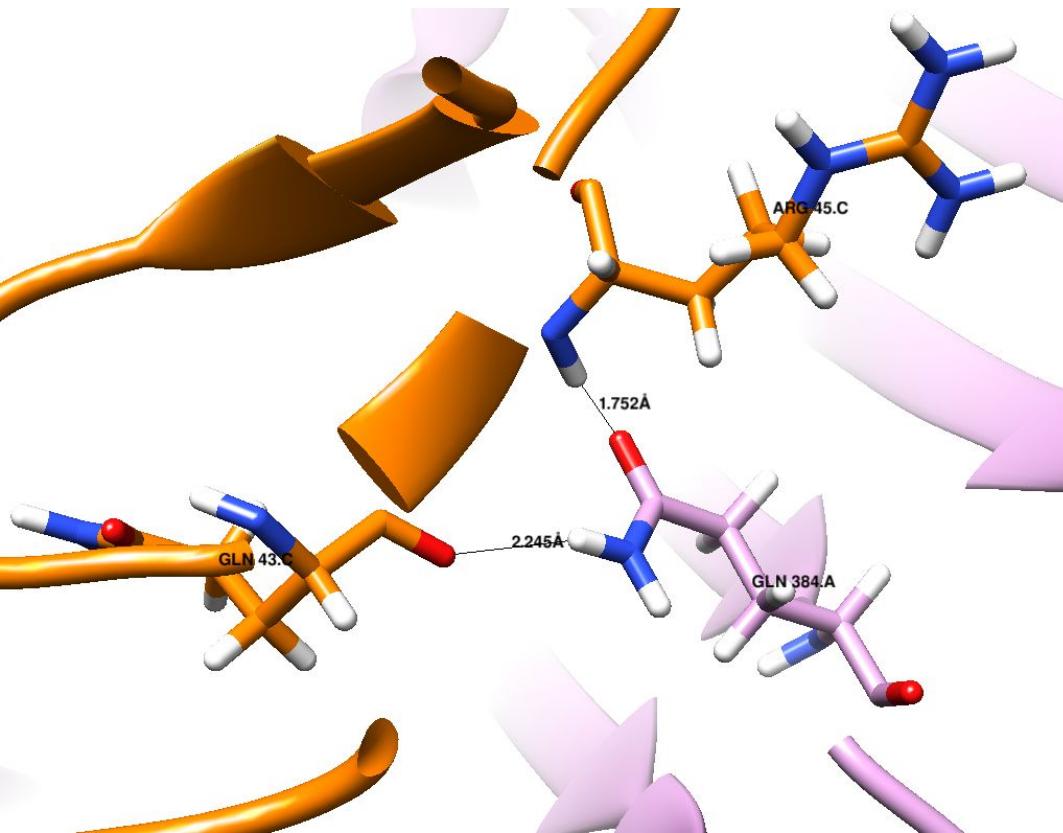
EGFR DIII: ORCHID
EGF: ORANGE

PDB: 3NJP

3.1. SYMMETRICAL DIMERIZATION



EGF binding S3: Hydrogen bonds – DOMAIN III

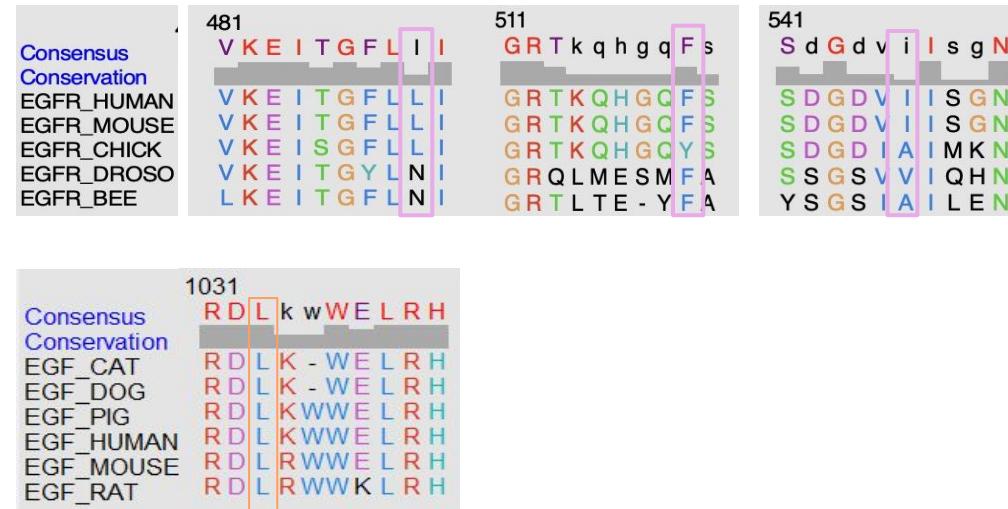
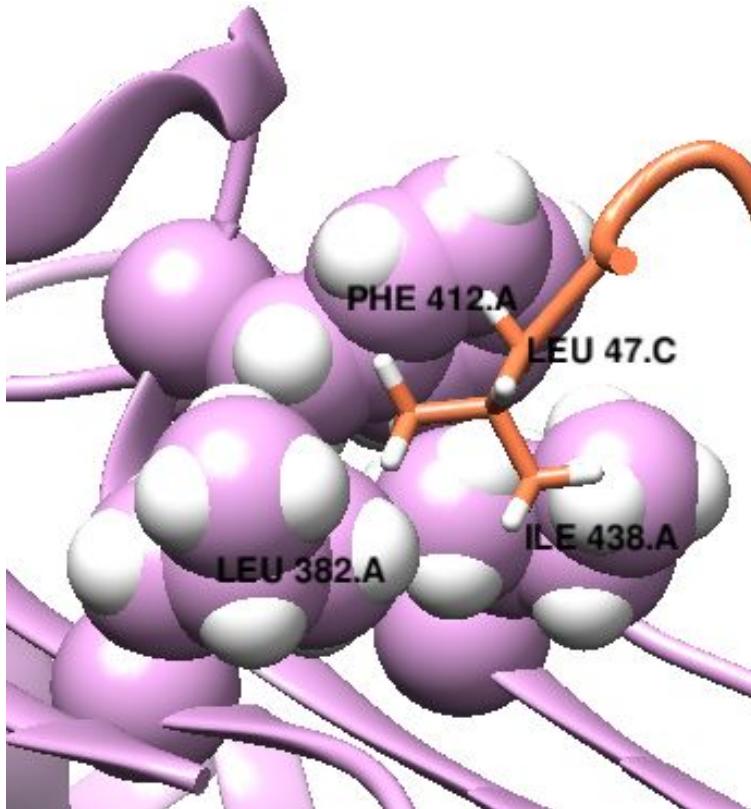


EGFR DIII: ORCHID
EGF: ORANGE

3.1. SYMMETRICAL DIMERIZATION



EGF binding S3: Hydrophobic contacts – DOMAIN III



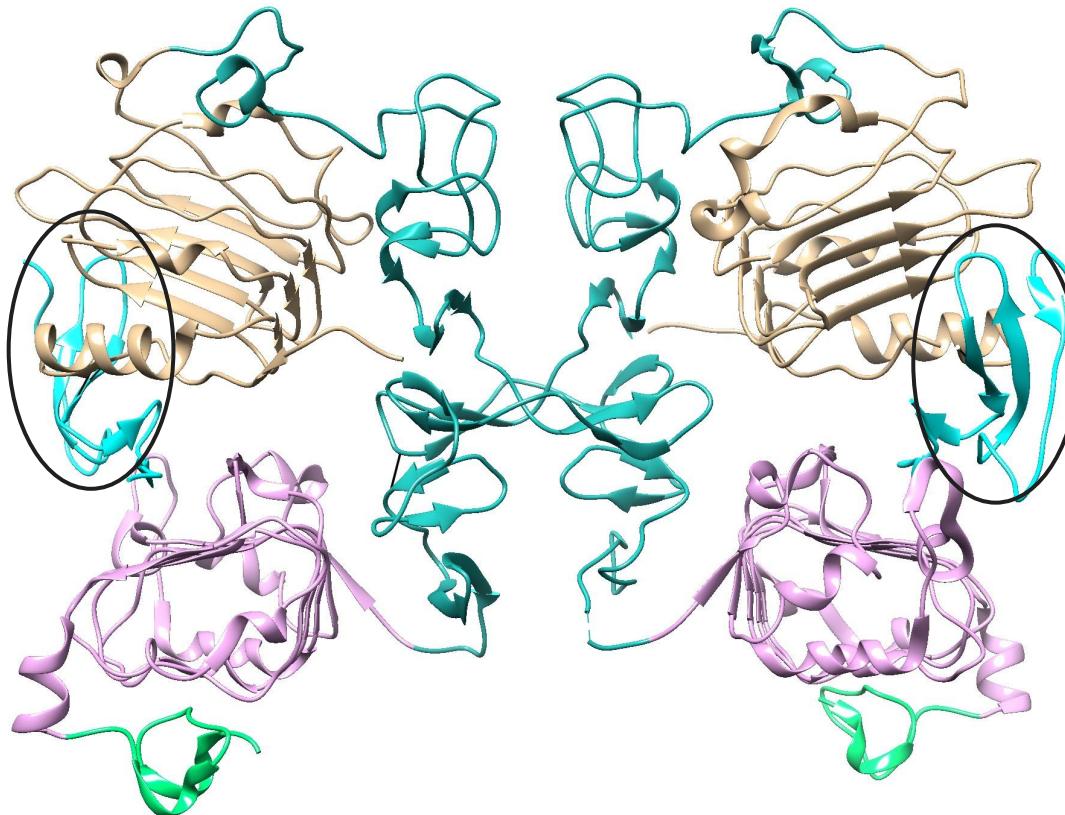
EGFR DIII: ORCHID

EGF: ORANGE

3.1. SYMMETRICAL DIMERIZATION



TGF- α binding

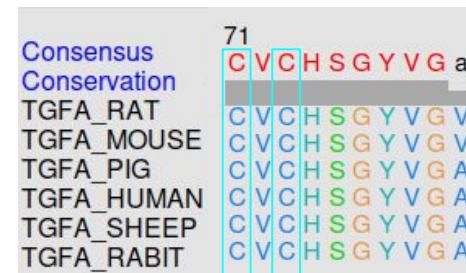
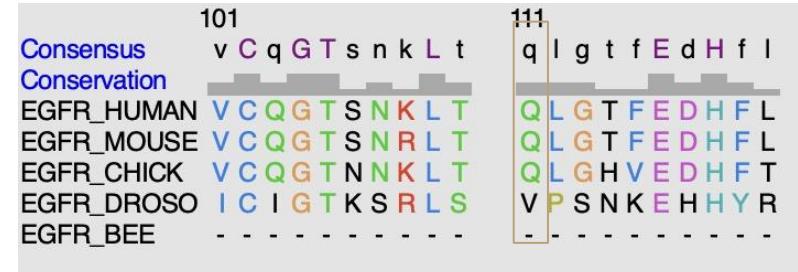
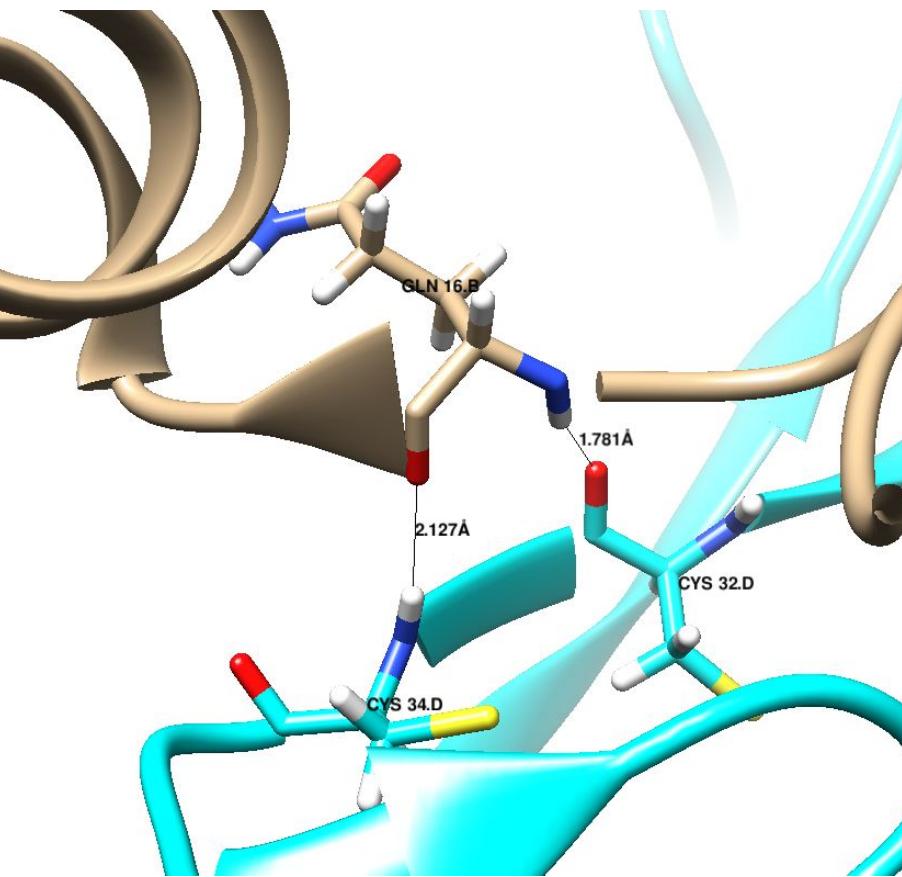


PDB: 1MOX

3.1. SYMMETRICAL DIMERIZATION



TGF- α binding: Hydrogen bonds – DOMAIN I



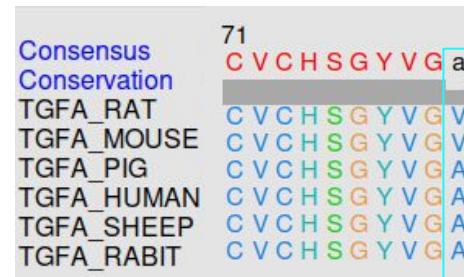
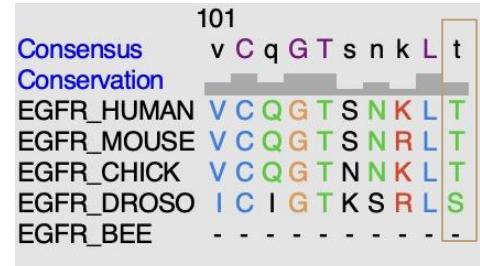
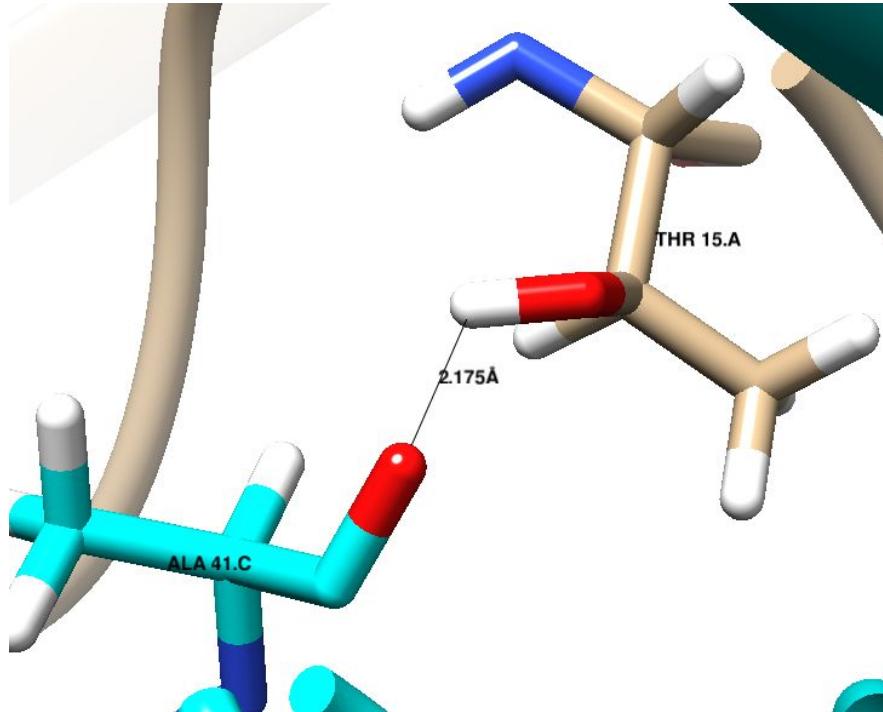
EGFR DI: OCHRE

TGF ALFA: CYAN

3.1. SYMMETRICAL DIMERIZATION



TGF- α binding: Hydrogen bonds – DOMAIN I

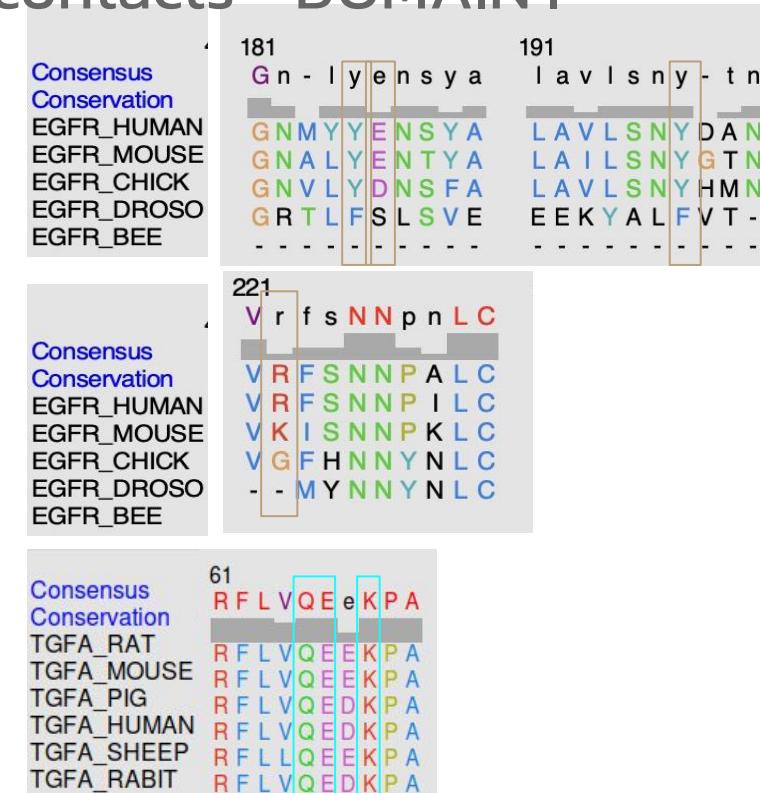
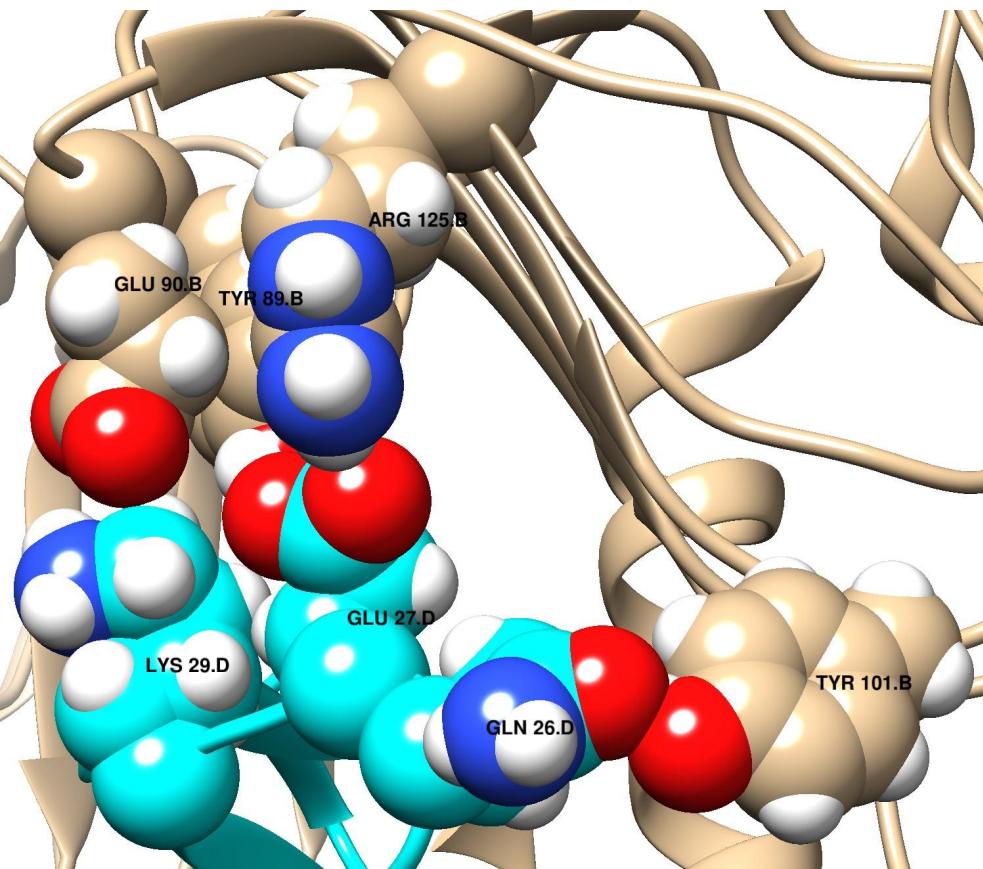


EGFR DI: OCHRE
TGF ALFA: CYAN

3.1. SYMMETRICAL DIMERIZATION



TGF- α binding: Hydrophobic contacts – DOMAIN I



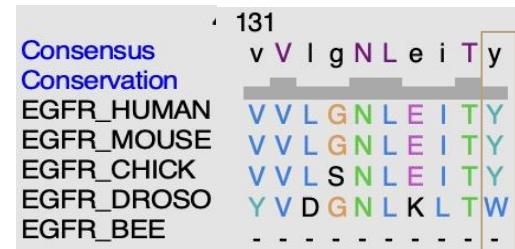
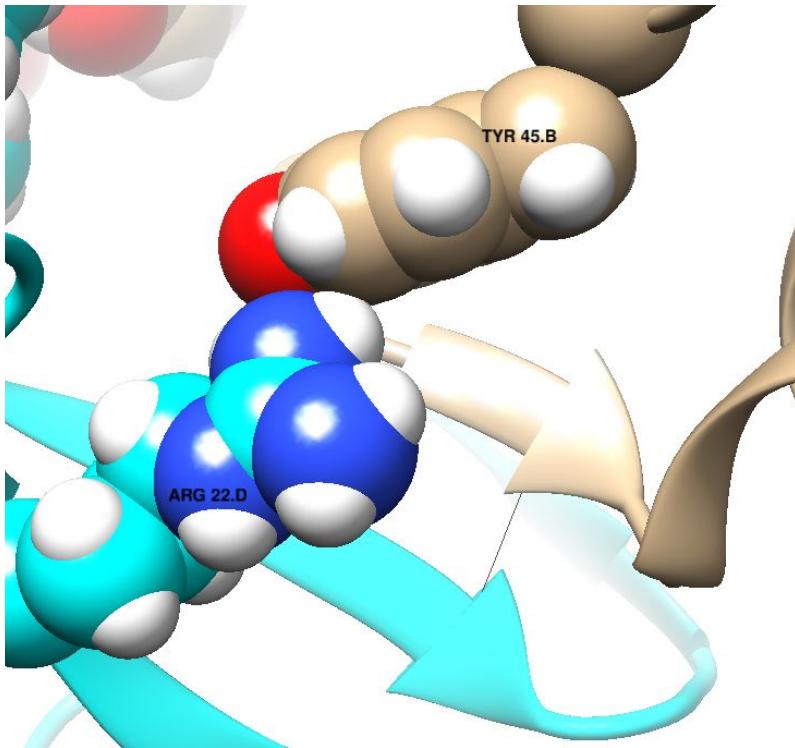
EGFR DI: OCHRE
TGF ALFA: CYAN

PDB: 1MOX

3.1. SYMMETRICAL DIMERIZATION



TGF- α binding: Hydrophobic contacts – DOMAIN I



EGFR DI: OCHRE

TGF ALFA: CYAN

3.1. SYMMETRICAL DIMERIZATION



TGF- α binding: Hydrophobic contacts – DOMAIN III



Consensus	F	s	G	d	q	f	t	y	t	-
Conservation	█	█	█	█	█	█	█	█	█	█
EGFR_HUMAN	F	R	G	D	S	F	T	H	T	-
EGFR_MOUSE	F	K	G	D	S	F	T	R	T	-
EGFR_CHICK	F	L	G	D	A	F	T	K	T	-
EGFR_DROSO	F	S	G	F	Q	D	V	Y	A	N
EGFR_BEE	F	Q	G	F	Q	H	V	Y	R	N

Consensus	H	T	Q	f	C	F	H	G	T	C
Conservation	█	█	█	█	█	█	█	█	█	█
TGFA_RAT	H	T	C	Y	C	F	H	G	T	C
TGFA_MOUSE	H	T	C	Y	C	F	H	G	T	C
TGFA_PIG	H	S	O	F	C	F	H	G	T	C
TGFA_HUMAN	H	T	O	F	C	F	H	G	T	C
TGFA_SHEEP	H	T	O	F	C	F	H	G	T	C
TGFA_RABBIT	H	T	O	F	C	F	H	G	T	C

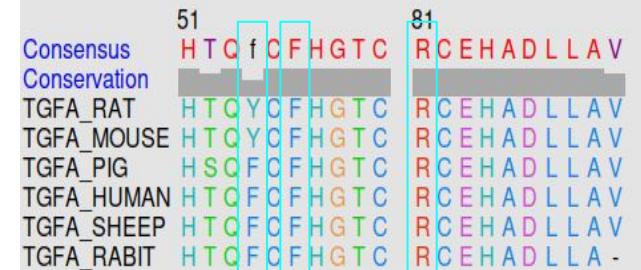
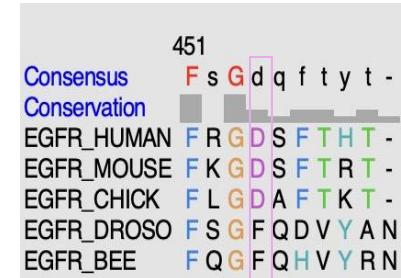
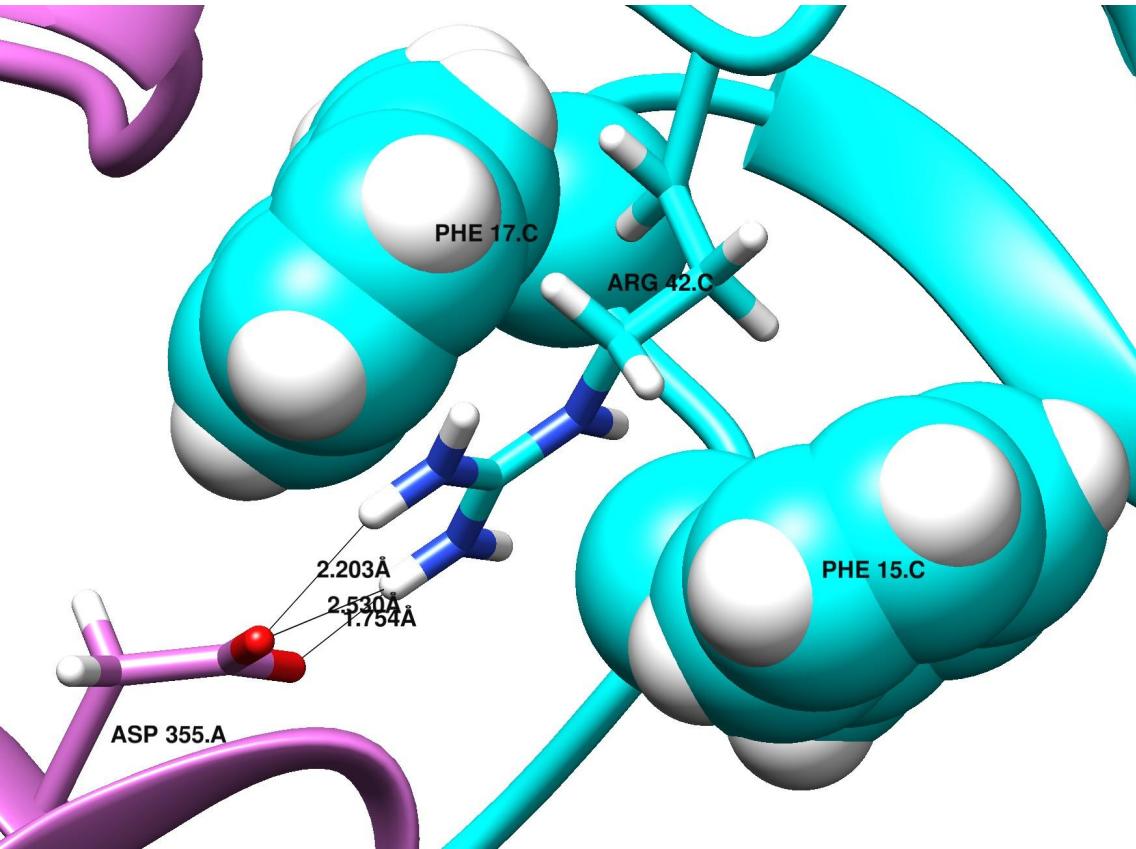
EGFR DIII: ORCHID

TGF ALFA: CYAN

3.1. SYMMETRICAL DIMERIZATION



TGF- α binding: Salt bridge – DOMAIN III

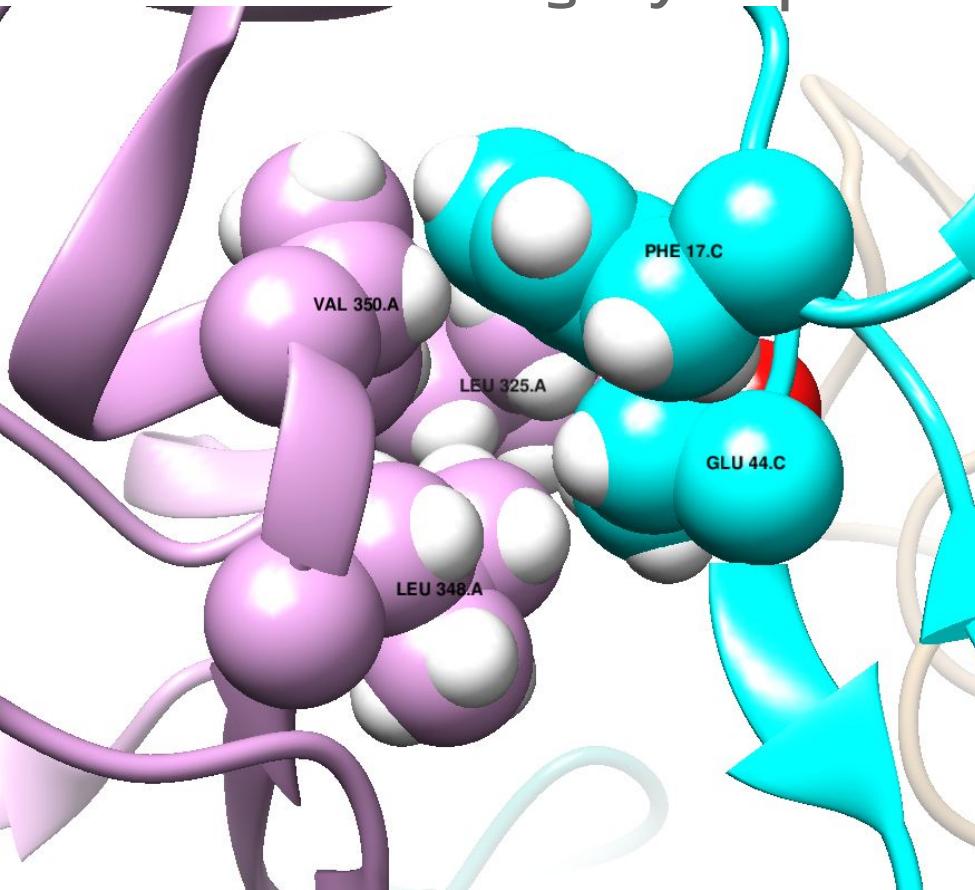


EGFR DIII: ORCHID
TGF ALFA: CYAN

3.1. SYMMETRICAL DIMERIZATION



TGF- α binding: Hydrophobic contacts – DOMAIN III



Consensus	421		441
Conservation			
EGFR_HUMAN	K D S	L S I N A T N	S G D L H I L P V A
EGFR_MOUSE	K D T	L S I N A T N	S G D L H I L P V A
EGFR_CHICK	K G I	L S I N A T N	N G D V S I L P V A
EGFR_DROSO	- - -	- L H A G N	D G N I R I L D Q T
EGFR_BEE	- - -	- V H S G N	E G S I T I L D Q S

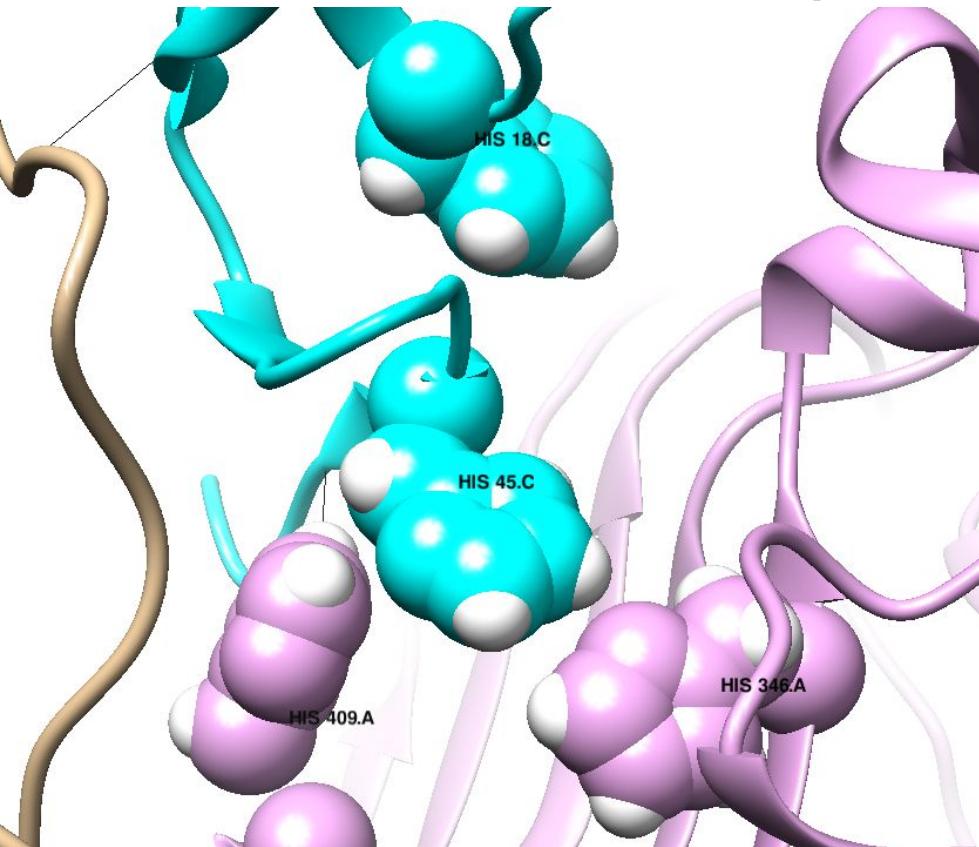
Consensus	51	81
Conservation		
TGFA_RAT	H T Q Y C F C F H G T C	R C E H A D L L A V
TGFA_MOUSE	H T Q Y C F C F H G T C	R C E H A D L L A V
TGFA_PIG	H S Q F C F C F H G T C	R C E H A D L L A V
TGFA_HUMAN	H T Q F C F C F H G T C	R C E H A D L L A V
TGFA_SHEEP	H T Q F C F C F H G T C	R C E H A D L L A V
TGFA_RABBIT	H T Q F C F C F H G T C	R C E H A D L L A -

EGFR DIII: ORCHID

TGF ALFA: CYAN

3.1. SYMMETRICAL DIMERIZATION

TGF- α binding: Hydrophilic region – DOMAIN III



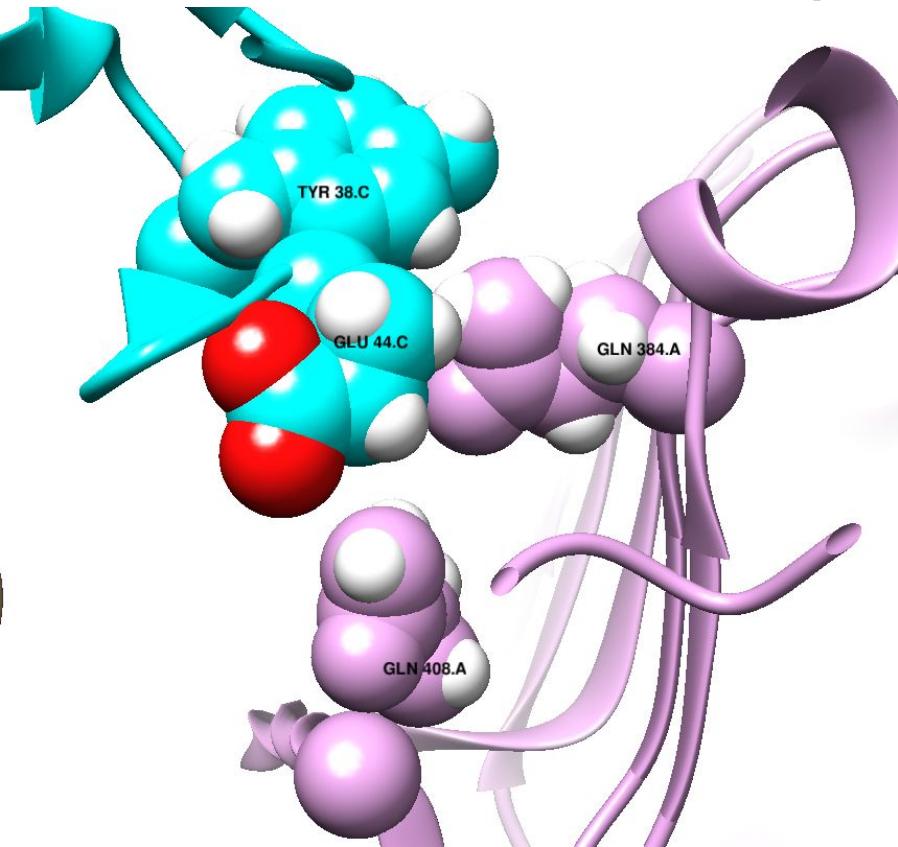
Consensus	441	s G d i h I L p v a <th>Conservation</th> <td>511</td> <td>G R T k q h g q f s</td>	Conservation	511	G R T k q h g q f s
Conservation			Conservation		
EGFR_HUMAN	S	G D L H I L P V A	EGFR_MOUSE	G R T K Q H G Q F S	
EGFR_MOUSE	S	G D L H I L P V A	EGFR_CHICK	G R T K Q H G Q F S	
EGFR_CHICK	N	G D V S I L P V A	EGFR_DROSO	G R T K Q H G Q Y S	
EGFR_DROSO	D	G N I R I L D Q T	EGFR_BEE	G R Q L M E S M F A	
EGFR_BEE	E	G S I T I L D Q S		G R T L T E - Y F A	

Consensus	51	H T Q f C F H G T C	Conservation	81	R C E H A D L L A V
Conservation			Conservation		
TGFA_RAT	H	T Q Y C F H G T C	TGFA_MOUSE	R C E H A D L L A V	
TGFA_MOUSE	H	T Q Y C F H G T C	TGFA_PIG	R C E H A D L L A V	
TGFA_PIG	H	S Q F C F H G T C	TGFA_HUMAN	R C E H A D L L A V	
TGFA_HUMAN	H	T Q F C F H G T C	TGFA_SHEEP	R C E H A D L L A V	
TGFA_SHEEP	H	T Q F C F H G T C	TGFA_RABBIT	R C E H A D L L A -	
TGFA_RABBIT	H	T Q F C F H G T C			

EGFR DIII: ORCHID
TGF ALFA: CYAN

3.1. SYMMETRICAL DIMERIZATION

TGF- α binding: Hydrophilic region – DOMAIN III



Consensus	491	Q a w p d n f t d l	511	G R T k q h g q F s
Conservation				
EGFR_HUMAN	Q	A W P E N R T D L	G R T K Q H G Q F S	
EGFR_MOUSE	Q	A W P D N W T D L	G R T K Q H G Q F S	
EGFR_CHICK	Q	A W P D N A T D L	G R T K Q H G Q Y S	
EGFR_DROSO	E	G T H P Q F R N L	G R Q L M E S M F A	
EGFR_BEE	Q	G D H K D F K N L	G R T L T E - Y F A	

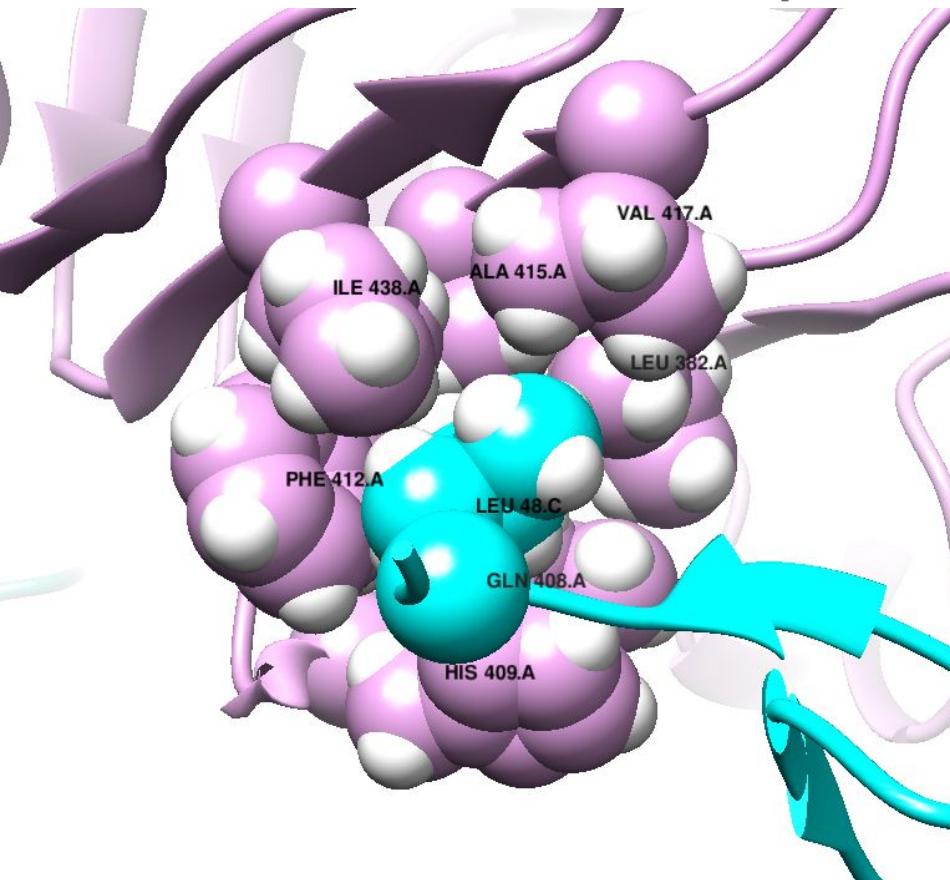
Consensus	71	C V C H S G Y V G a	81	R C E H A D L L A V
Conservation				
TGFA_RAT	C	C V C H S G Y V G V	R	C E H A D L L A V
TGFA_MOUSE	C	C V C H S G Y V G V	R	C E H A D L L A V
TGFA_PIG	C	C V C H S G Y V G A	R	C E H A D L L A V
TGFA_HUMAN	C	C V C H S G Y V G A	R	C E H A D L L A V
TGFA_SHEEP	C	C V C H S G Y V G A	R	C E H A D L L A V
TGFA_RABBIT	C	C V C H S G Y V G A	R	C E H A D L L A -

EGFR DIII: ORCHID
TGF ALFA: CYAN

3.1. SYMMETRICAL DIMERIZATION



TGF- α binding: Hydrophobic pocket – DOMAIN III



481
Consensus: VKE I T G F L I I
Conservation:
EGFR_HUMAN: VKE I T G F L I I
EGFR_MOUSE: VKE I T G F L I I
EGFR_CHICK: VKE I S G F L L I I
EGFR_DROSO: VKE I T G Y L N I I
EGFR_BEE: LKE I T G F L N I I

541
S d G d v i I s g N
S D G D V I I S G N
S D G D V I I S G N
S D G D I A I M K N
S S G S V V V Q H N
Y S G S I A I L E N

511
Consensus: G R T k q h g q f s
Conservation:
EGFR_HUMAN: G R T K Q H G Q F S
EGFR_MOUSE: G R T K Q H G Q F S
EGFR_CHICK: G R T K Q H G Q Y S
EGFR_DROSO: G R Q L M E S M F A
EGFR_BEE: G R T L T E - Y F A

521
- L A V V k l s i t
- L A V V S L N I T
- L A V V G L N I T
- L A V V N L K I Q
A L A I V K S S L Y
S L Y V V K T S L V

81
Consensus: R C E H A D L L A V
Conservation:
TGFA_RAT: R C E H A D L L A V
TGFA_MOUSE: R C E H A D L L A V
TGFA_PIG: R C E H A D L L A V
TGFA_HUMAN: R C E H A D L L A V
TGFA_SHEEP: R C E H A D L L A V
TGFA_RABBIT: R C E H A D L L A -

EGFR DIII: ORCHID

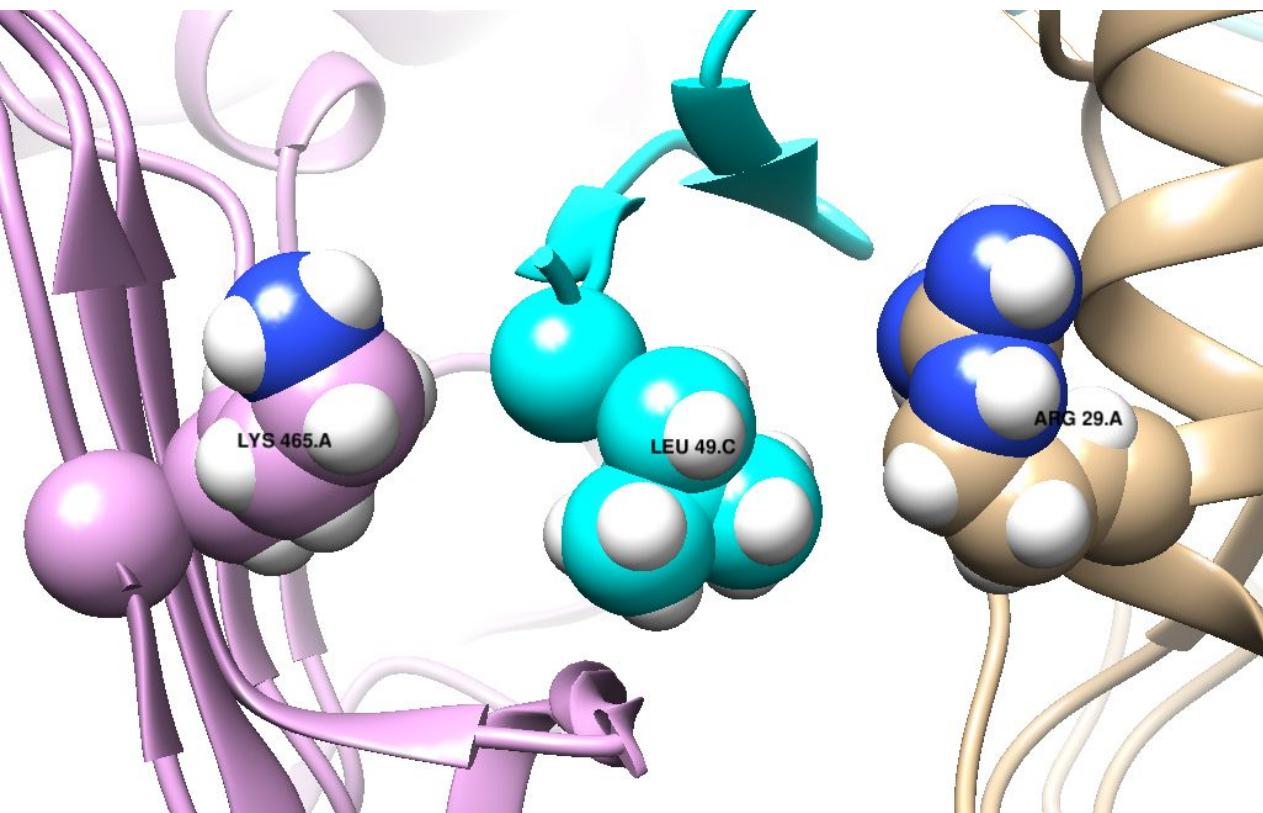
TGF ALFA: CYAN

PDB: 1MOX

3.1. SYMMETRICAL DIMERIZATION



TGF- α binding: C-terminal – DOMAIN I and III



121	s L q r m y n N C e
Consensus	
Conservation	
EGFR_HUMAN	S L Q R M F N N C E
EGFR_MOUSE	S L Q R M Y N N C E
EGFR_CHICK	S L Q R M Y M N N C E
EGFR_DROSO	N L R D R Y T N C T
EGFR_BEE	- - - - -
571	K t k i i n N R n e
Consensus	
Conservation	
EGFR_HUMAN	K T K I I S N R G E
EGFR_MOUSE	K T K I M N N R A E
EGFR_CHICK	K T K I I Q N R N K
EGFR_DROSO	K V W V N E N L R A
EGFR_BEE	E S L L S N N R N E
81	R C E H A D L L A V
Consensus	
Conservation	
TGFA_RAT	R C E H A D L L A V
TGFA_MOUSE	R C E H A D L L A V
TGFA_PIG	R C E H A D L L A V
TGFA_HUMAN	R C E H A D L L A V
TGFA_SHEEP	R C E H A D L L A V
TGFA_RABBIT	R C E H A D L L A -

EGFR DI: OCHRE

EGFR DIII: ORCHID

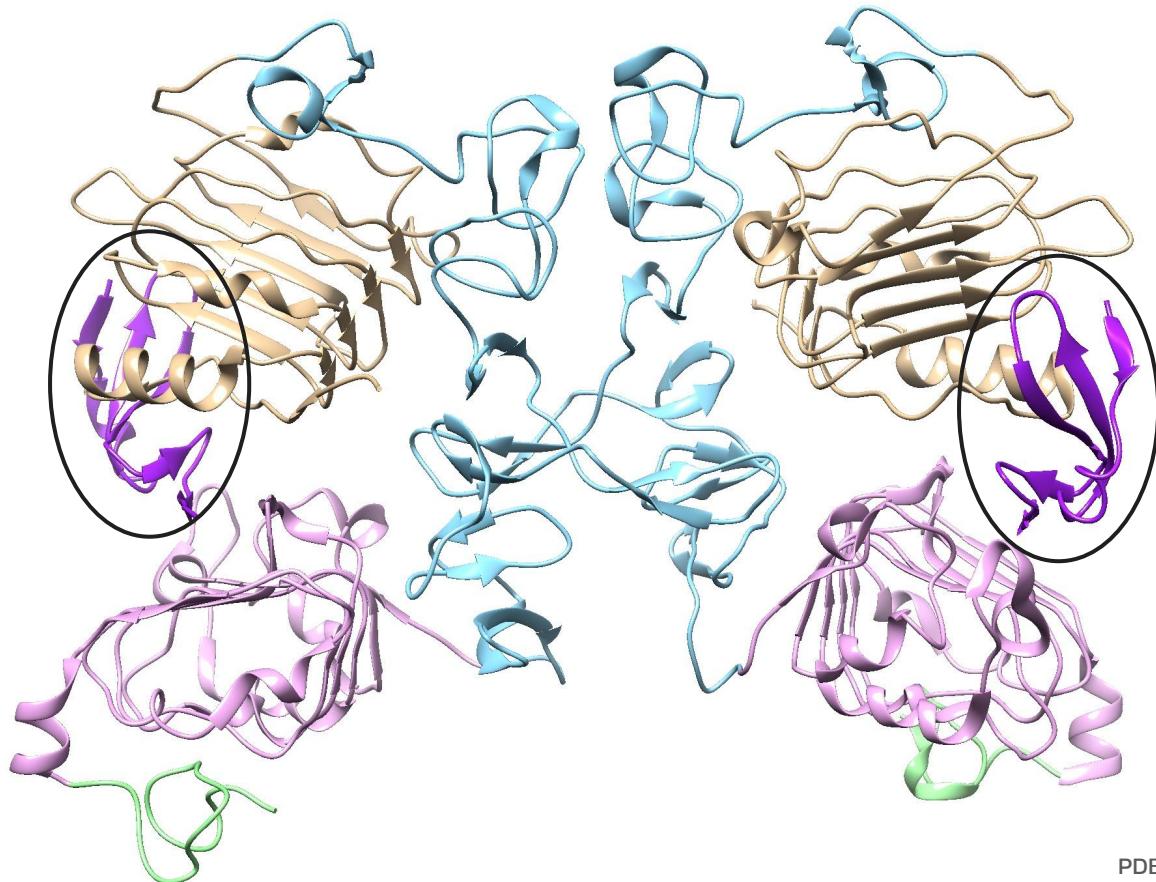
TGF ALFA: CYAN

PDB: 1MOX

3.2. ASYMMETRICAL DIMERIZATION



EREG Binding

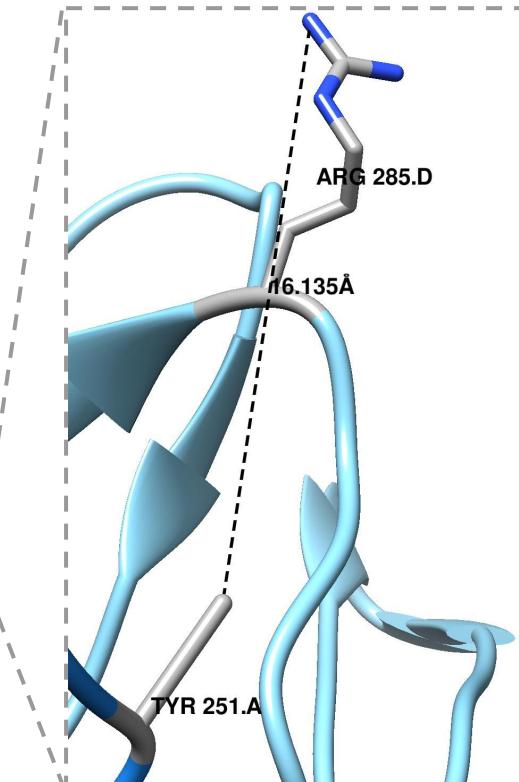
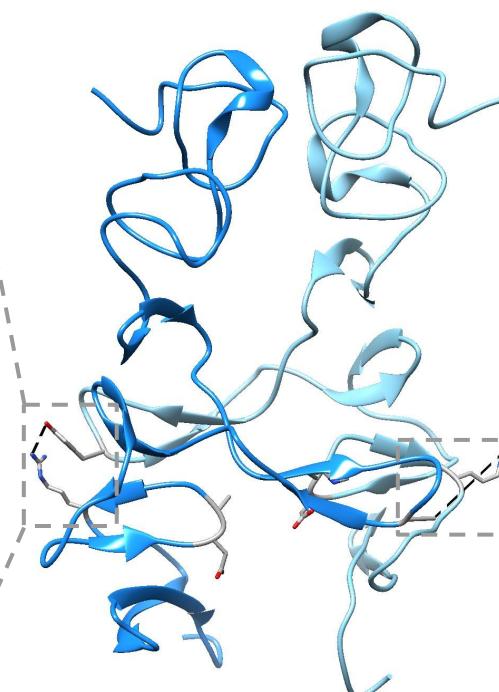
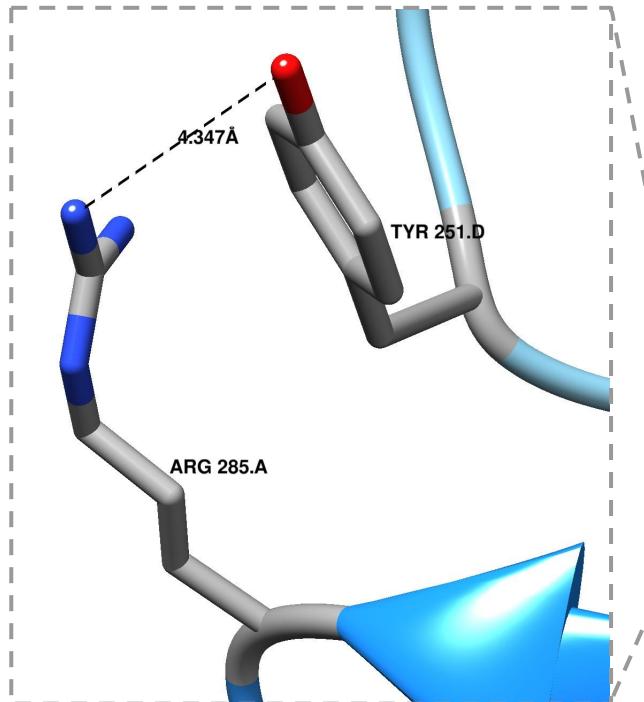


PDB: 5WB7

3.2. ASYMMETRICAL DIMERIZATION



EREG Binding: Receptor-Receptor interactions

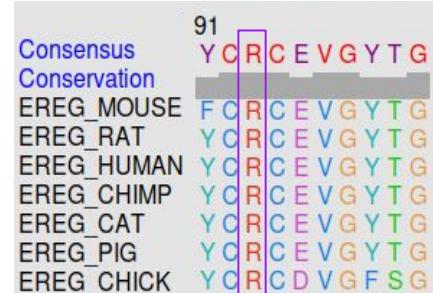
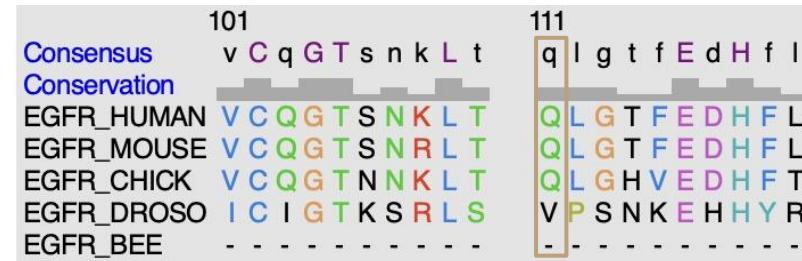
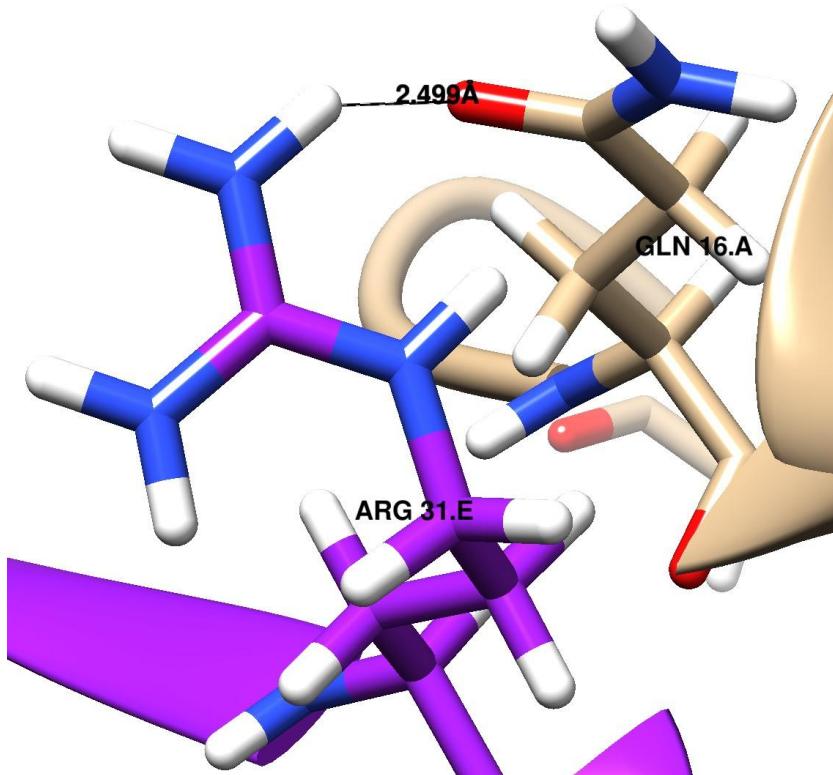


EGFR DII_L: BLUE
EGFR DII_R: SKY BLUE

3.2. ASYMMETRICAL DIMERIZATION



EREG Binding: Hydrogen Bonds – DOMAIN I (left)

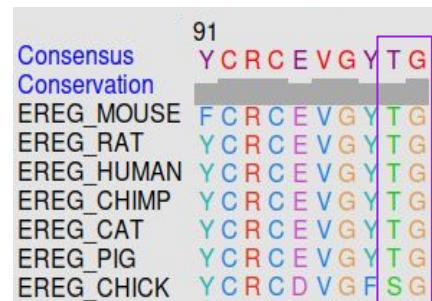
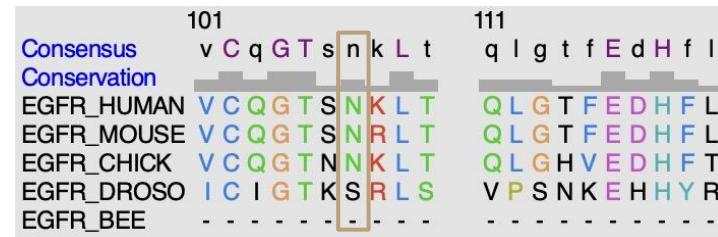
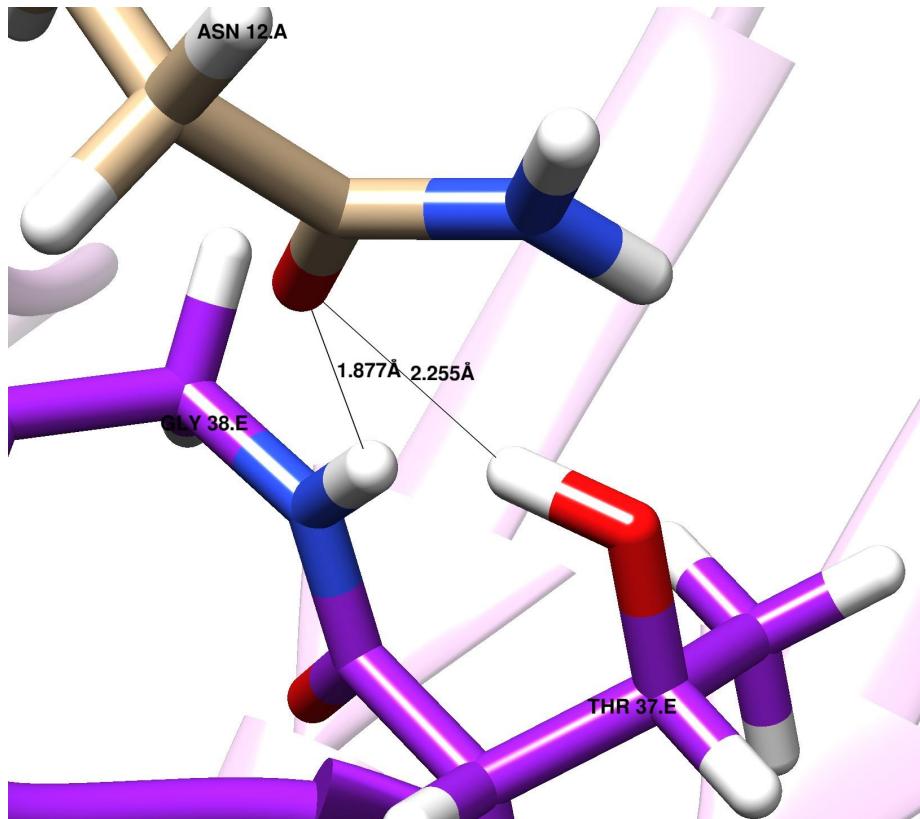


EGFR DI_L: OCHRE
EREG: PURPLE

3.2. ASYMMETRICAL DIMERIZATION



EREG Binding: Hydrogen Bonds – DOMAIN I (left)

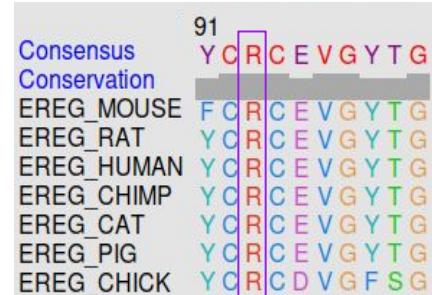
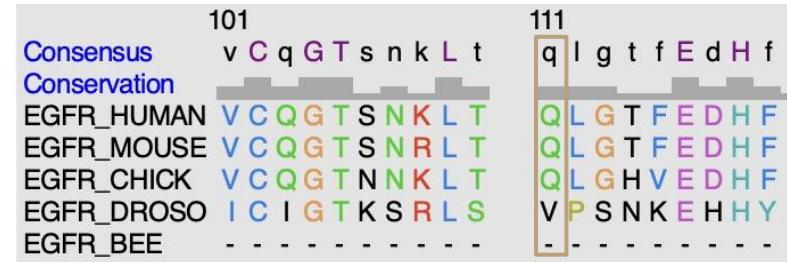
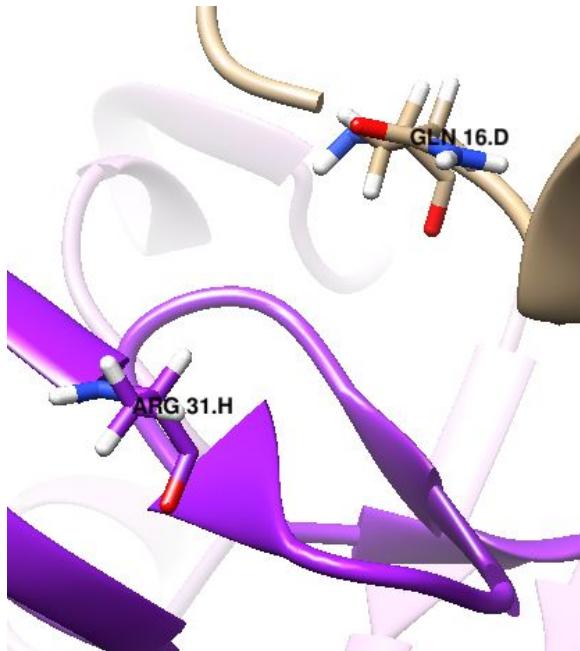


EGFR DI_L: OCHRE
EREG: PURPLE

3.2. ASYMMETRICAL DIMERIZATION



EREG Binding: Hydrogen Bonds – DOMAIN I (right)

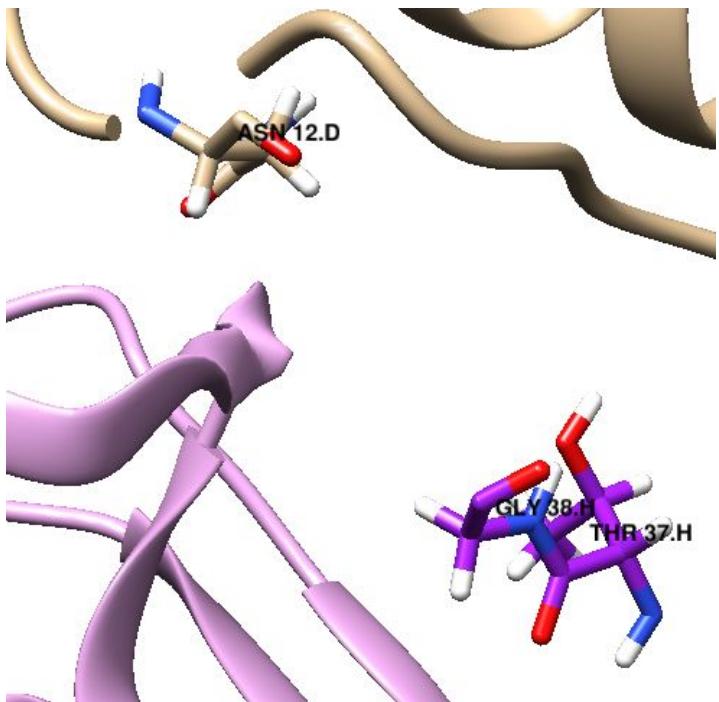


EGFR DI_R: OCHRE
EREG: PURPLE

3.2. ASYMMETRICAL DIMERIZATION



EREG Binding: Hydrogen Bonds – DOMAIN I (right)



	101	111
Consensus	v C q G T s n k L t	q l g t f E d H f l
Conservation		
EGFR_HUMAN	VCQGTSNKL	QLGTFEDHFL
EGFR_MOUSE	VCQGTSNRL	QLGTFEDHFL
EGFR_CHICK	VCQGTTNNKL	QLGHVEDHFT
EGFR_DROSO	ICIGTKSRL	VPSNKEHHYR
EGFR_BEE	- - - - -	- - - - -

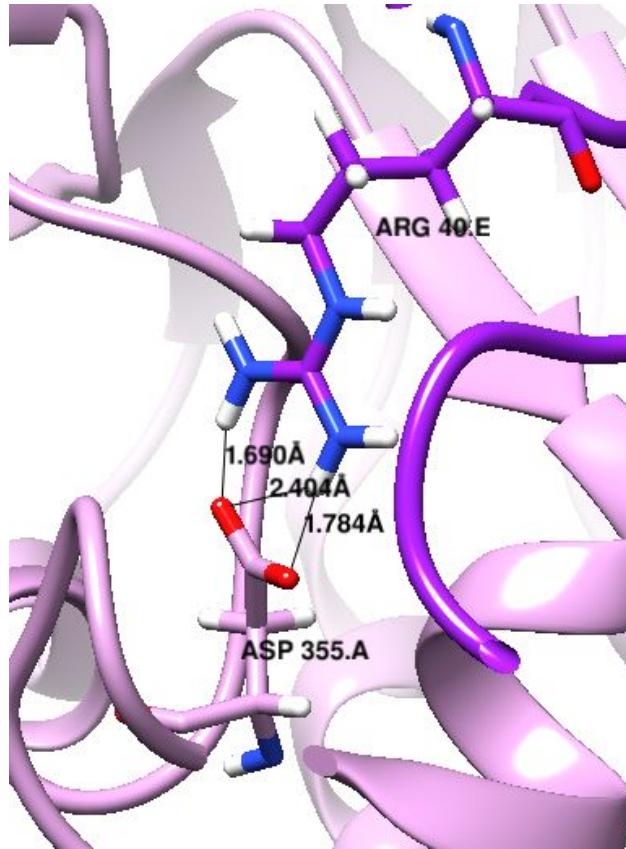
	91
Consensus	Y C R C E V G Y T G
Conservation	
EREG_MOUSE	FCRCEVGYTG
EREG_RAT	YCRCEVGYTG
EREG_HUMAN	YCRCEVGYTG
EREG_CHIMP	YCRCEVGYTG
EREG_CAT	YCRCEVGYTG
EREG_PIG	YCRCEVGYTG
EREG_CHICK	YCRCDVGFSG

EGFR DI_R: OCHRE
EREG: PURPLE

3.2. ASYMMETRICAL DIMERIZATION



EREG Binding: Salt bridge – DOMAIN III (left)



Consensus	F	s	G	d	q	f	t	y	t	-
Conservation	█	█	█	█	█	█	█	█	█	█
EGFR_HUMAN	F	R	G	D	S	F	T	H	T	-
EGFR_MOUSE	F	K	G	D	S	F	T	R	T	-
EGFR_CHICK	F	L	G	D	A	F	T	K	T	-
EGFR_DROSO	F	S	G	F	Q	D	V	Y	A	N
EGFR_BEE	F	Q	G	F	Q	H	V	Y	R	N

Consensus	v	R	C	E	H	F	F	L	T	V
Conservation	█	█	█	█	█	█	█	█	█	█
EREG_MOUSE	L	R	C	E	H	F	F	L	T	V
EREG_RAT	L	R	C	E	H	F	F	L	T	V
EREG_HUMAN	V	R	C	E	H	F	F	L	T	V
EREG_CHIMP	V	R	C	E	H	F	F	L	T	V
EREG_CAT	V	R	C	E	H	F	F	L	T	V
EREG_PIG	V	R	C	E	H	F	F	L	T	V
EREG_CHICK	V	R	C	V	H	S	E	L	-	V

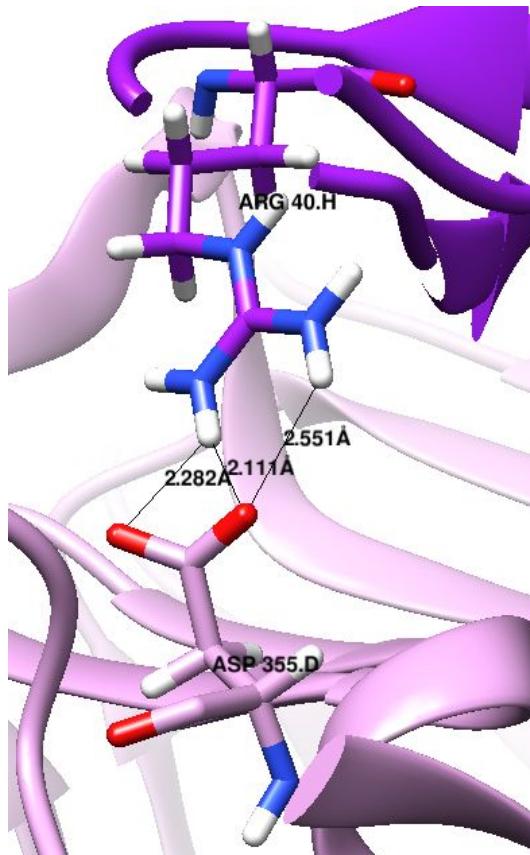
EGFR DIII: ORCHID
EREG: PURPLE

PDB: 5WB7

3.2. ASYMMETRICAL DIMERIZATION



EREG Binding: Salt bridge – DOMAIN III (right)



	451
Consensus	F s G d q f t y t -
Conservation	
EGFR_HUMAN	F R G D S F T H T -
EGFR_MOUSE	F K G D S F T R T -
EGFR_CHICK	F L G D A F T K T -
EGFR_DROSO	F S G F Q D V Y A N
EGFR_BEE	F Q G F Q H V Y R N

	101
Consensus	v R C E H F F L T V
Conservation	
EREG_MOUSE	L R C E H F F L T V
EREG_RAT	L R C E H F F L T V
EREG_HUMAN	V R C E H F F L T V
EREG_CHIMP	V R C E H F F L T V
EREG_CAT	V R C E H F F L T V
EREG_PIG	V R C E H F F L T V
EREG_CHICK	V R C V H S E L - V

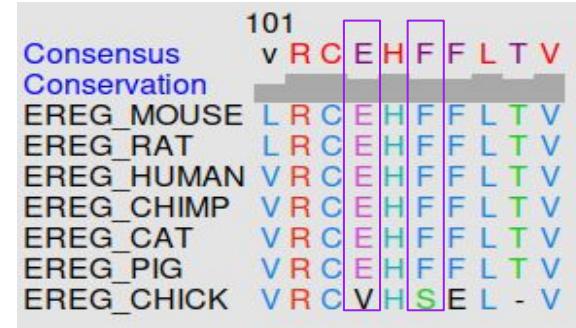
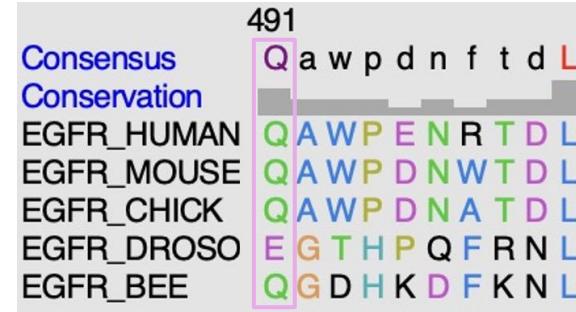
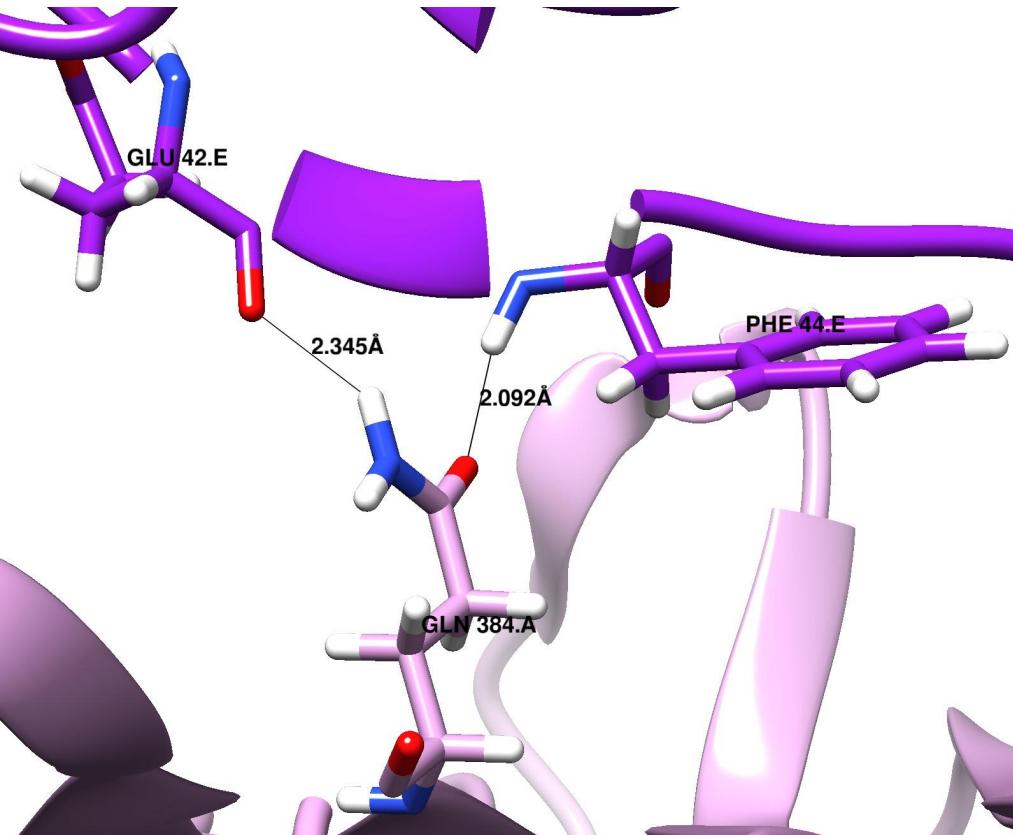
EGFR D_{III}_R: ORCHID
EREG: PURPLE

PDB: 5WB7

3.2. ASYMMETRICAL DIMERIZATION



EREG Binding: Hydrogen Bonds – DOMAIN III (left)



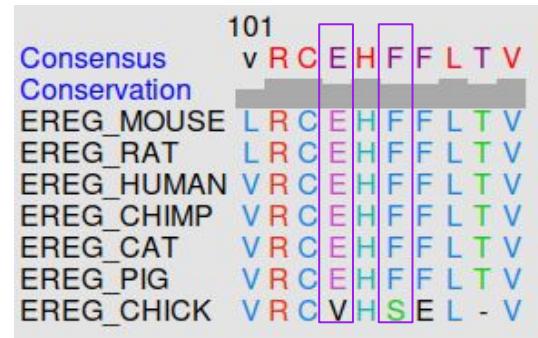
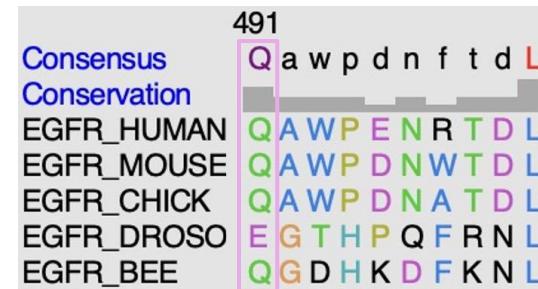
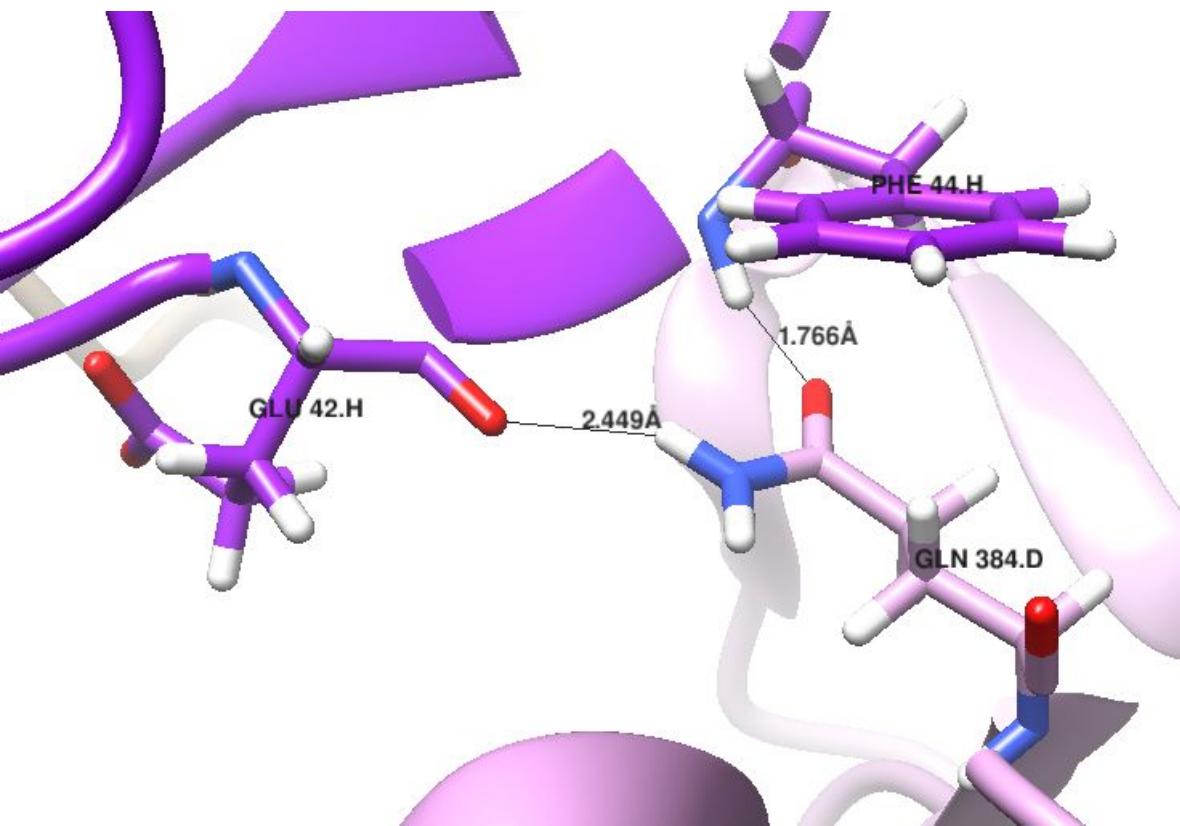
EGFR D3L: ORCHID
EREG: PURPLE

PDB: 5WB7

3.2. ASYMMETRICAL DIMERIZATION



EREG Binding: Hydrogen Bonds – DOMAIN III (right)



EGFR DIII_R: ORCHID
EREG: PURPLE

CONCLUSION



1

The functions of EGFR, such as intracellular signalling, represent a predominant role in many tissues.

2

Alterations in this protein have been reported to be responsible for several types of human cancer

3

EGFR is a relatively big protein, and it is very conserved in the animal kingdom.

4

Its ligands are also conserved and present high structural similarities. However, its binding can induce different receptor conformations, thus enabling variated cellular responses.

BIBLIOGRAPHY

1. Ferguson KM, Berger MB, Mendrola JM, Cho HS, Leahy DJ, Lemmon MA. 2003. EGF Activates Its Receptor by Removing Interactions that Autoinhibit Ectodomain Dimerization. *Mol Cell*;11(2):507-17.
2. Freed DM, Bessman NJ, Kiyatkin A, Salazar-Cavazos E, Byrne PO, Moore JO, et al. 2017. EGFR Ligands Differentially Stabilize Receptor Dimers to Specify Signaling Kinetics. *Cell*;171(3):683-695.e18.
3. Garrett TPJ, McKern NM, Lou M, Elleman TC, Adams TE, Lovrecz GO, et al. 2002. Crystal Structure of a Truncated Epidermal Growth Factor Receptor Extracellular Domain Bound to Transforming Growth Factor α . *Cell*. 110(6):763-73.
4. He Y, He G, He T. 2021. Specifically Targeted Transport of Plasma Membrane Transporters: From Potential Mechanisms for Regulating Cell Health or Disease to Applications. *Membranes*. 27;11(10):736.
5. Hu C, Leche CA, Kiyatkin A, Yu Z, Stayrook SE, Ferguson KM, et al. 2022. Glioblastoma mutations alter EGFR dimer structure to prevent ligand bias. *Nature*. 602(7897):518-22.
6. Huang Y, Ognjenovic J, Karandur D, Miller K, Merk A, Subramaniam S, et al. 2021. A molecular mechanism for the generation of ligand-dependent differential outputs by the epidermal growth factor receptor. *eLife*;10:e73218.
7. Lemmon MA, Schlessinger J, Ferguson KM. 2014. The EGFR Family: Not So Prototypical Receptor Tyrosine Kinases. *Cold Spring Harbor Perspectives in Biology*. 1;6(4):a020768-a020768.
8. Ogiso H, Ishitani R, Nureki O, Fukai S, Yamanaka M, Kim JH, et al. 2002. Crystal Structure of the Complex of Human Epidermal Growth Factor and Receptor Extracellular Domains. *Cell*;110(6):775-87.
9. Wieduwilt MJ, Moasser MM. 2008. The epidermal growth factor receptor family: Biology driving targeted therapeutics. *Cell Mol Life Sci*. 65(10):1566-84.

PEM QUESTIONS

1. Where is the EGFR tyrosine containing tail located?

- a) In the transmembrane domain
- b) In the N-terminal tail
- c) In domain II of the extracellular portion
- d) In the intracellular C-terminal tail**
- e) EGFR does not have a Tyr-containing tail

2. Which EGFR family member does not have any known ligand but still dimerizes?

- a) Her3
- b) EGFR
- c) ErbB2**
- d) ErbB4
- e) All receptors have a known ligand that they need for dimerization

3. Which domain of EGFR contains the dimerization arm?

- a) Domain I
- b) Domain II**
- c) Domain III
- d) Domain IV
- e) EGFR does not dimerize

4. According to SCOP classification, which is the fold corresponding to domains I and III of the EGFR?

- a) Leucine-rich repeat**
- b) Knottin
- c) Greek key
- d) Beta barrel
- e) None of the above is true

5. How many disulfide bonds do EGFR ligands present intramolecularly?

- a) 2
- b) 8
- c) 0
- d) 5
- e) 3

6. EGF structure is divided into 3 loops. What does determine this division?

- a) They have different conformation
- b) They are determined by hydrogen bonds
- c) The division corresponds to their function
- d) They are determined by disulfide bonds
- e) The researchers that first crystallized the receptor decided so

7. EGF binds to EGFR by 3 sites. These correspond to:

- a) Site 1: the B-loop binds to the domain I of the receptor. Site 2: the A-loop binds to the domain III of the receptor. Site 3: the C-loop binds to the domain III of the receptor
- b) Site 1: the C-loop binds to the domain I of the receptor. Site 2: the A-loop binds to the domain II of the receptor. Site 3: the B-loop binds to the domain III of the receptor
- c) Site 1: the A-loop binds to the domain I of the receptor. Site 2: the A-loop binds to the domain III of the receptor. Site 3: the B-loop binds to the domain III of the receptor
- d) Site 1: the B-loop binds to the domain I of the receptor. Site 2: the A-loop binds to the domain II of the receptor. Site 3: the C-loop binds to the domain III of the receptor
- e) None of the above

8. Which are the domains involved in the tethered dimerization arm, which inhibits dimerization following EGF binding?

- a) Domain I and Domain II
- b) Domain III and Domain IV
- c) Domain II and Domain III
- d) Domain II and domain IV
- e) Domain III and domain V

9. EGF binding is found to induce an inhibited monomeric form of the receptor due to:

- a) Temperatures below 0° C
- b) Low pH conditions**
- c) High pH conditions
- d) Neutral pH conditions
- e) Temperatures above 100° C

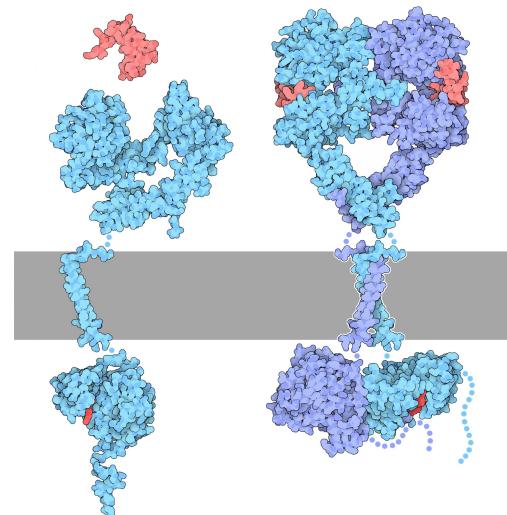
10. Which ligand of EGFR induces asymmetric dimerization?

- a) TGF-alpha
- b) EGF
- c) EREG**
- d) All of the above
- e) None of the above

Structural Biology

The Epidermal Growth Factor Receptor

Analysing the extracellular domain of EGFR and its ligands
EGF, TGF- α , and EREG binding



Source: PDB-101 Molecule of the month. June 2010, David Goodsell

Daniel Martín López, Guillem Miró Almuni, and Gisela Rusiñol Gálvez

EXTRA SLIDES

1. EGFR MSA

	1	11	21	31	41	
Consensus	-	-	-	-	-	
Conservation	-	-	-	-	-	
EGFR_HUMAN	-	-	-	-	-	
EGFR_MOUSE	-	-	-	-	-	
EGFR_CHICK	-	-	-	-	-	
EGFR_DROSO	M L L R R R N G P C	P F P L L L L L A	H C I C I W P A S A	A R D R Y A R Q N N	R Q R H Q D I D R D	
EGFR_BEE	-	-	-	-	-	
	51	61	71	81	91	
Consensus	-	-	-	-	-	
Conservation	-	-	-	-	-	
EGFR_HUMAN	-	-	M R P S G T A	G A A L L A L A A	L C P A S R A L E E	K - - - - - K
EGFR_MOUSE	-	-	M R P S G T A	R T T L V L L T A	L C A A G G A L E E	K - - - - - K
EGFR_CHICK	-	M G V	R S P L S A S G P R	G A A V L V L L L A	G V A L C S A V E E	K - - - - - K
EGFR_DROSO	R D R D R F L Y R S	S S A Q N R Q R G G	A N F A L G L G A N	G V T I P T S L E D	K N K N E F V K G K	
EGFR_BEE	-	-	-	-	-	
	101	111	121	131	141	
Consensus	v C q G T s n k L t	q l g t f E d H f l	s L q r m y n N C E	v V l g N L e i T y	v q r - N y D L s F	
Conservation	-	-	-	-	-	
EGFR_HUMAN	V C Q G T S N K L T	Q L G T F E D H F L	S L Q R M F N N C E	V V L G N L E I T Y	V Q R - N Y D L S F	
EGFR_MOUSE	V C Q G T S N R L T	Q L G T F E D H F L	S L Q R M Y N N C E	V V L G N L E I T Y	V Q R - N Y D L S F	
EGFR_CHICK	V C Q G T N N K L T	Q L G H V E D H F T	S L Q R M Y N N C E	V V L S N L E I T Y	V E H - N R D L T F	
EGFR_DROSO	I C I G T K S R L S	V P S N K E H H Y R	N L R D R Y T N C T	Y V D G N L K L T W	L P N E N L D L S F	
EGFR_BEE	-	-	-	-	-	
	151	161	171	181	191	
Consensus	L k t I q E V a G Y	v L I A l n t V e R	i p l e n L Q I I R	G n - l y e n s y a	l a v l s n y - t n	
Conservation	-	-	-	-	-	
EGFR_HUMAN	L K T I Q E V A G Y	V L I A L N T V E R	I P L E N L Q I I R	G N M Y Y E N S Y A	L A V L S N Y D A N	
EGFR_MOUSE	L K T I Q E V A G Y	V L I A L N T V E R	I P L E N L Q I I R	G N A L Y E N T Y A	L A I L S N Y G T N	
EGFR_CHICK	L K T I Q E V A G Y	V L I A L N M V D V	I P L E N L Q I I R	G N V L Y D N S F A	L A V L S N Y H M N	
EGFR_DROSO	L D N I R E V T G Y	I L I S H V D V K K	V V F P K L Q I I R	G R T L F S L S V E	E E K Y A L F V T -	
EGFR_BEE	-	-	-	-	-	
	201	211	221	231	241	
Consensus	- t g - I r e L p m	r n L q e i L n G a	V r f s N N p n L C	n m r T I q W r d I	v s n - s f m y n y	
Conservation	-	-	-	-	-	
EGFR_HUMAN	K T G - L K E L P M	R N L Q E I L H G A	V R F S N N P A L C	N V E S I Q W R D I	V S S - D F L S N M	
EGFR_MOUSE	R T G - L R E L P M	R N L Q E I L I G A	V R F S N N P I L C	N M D T I Q W R D I	V Q N - V F M S N M	
EGFR_CHICK	K T Q G L R E L P M	K R L S E I L N G G	V K I S N N P K L C	N M D T V L W N D I	I D T - S R K P L T	
EGFR_DROSO	- Y S K M Y T L E I	P D L R D V L N G Q	V G F H N N Y N L C	H M R T I Q W S E I	V S N G T D A Y - Y	
EGFR_BEE	-	-	- M Y N N Y N L C	H I R T I N W E E I	I T G P S A M Y S Y	

1. EGFR MSA

251	261	271	281	291
Consensus s y D F q s h e s s	C p K C d p S C p n	G - C W G a G e e N	C Q K l t K i i C a	q Q C s - G R C r G
Conservation				
EGFR_HUMAN S M D F Q N H L G S	C Q K C D P S C P N	G S C W G A G E E N	C Q K L T K I I C A	Q Q C S - G R C R G
EGFR_MOUSE S M D L Q S H P S S	C P K C D P S C P N	G S C W G G E E N	C Q K L T K I I C A	Q Q C S - H R C R G
EGFR_CHICK V L D F A S N L S S	C P K C H P N C T E	D H C W G A G E Q N	C Q T L T K V I C A	Q Q C S - G R C R G
EGFR_DROSO N Y D F T A P E R E	C P K C H E S C T H	G - C W G E G P K N	C Q K F S K L T C S	P Q C A G G R C Y G
EGFR_BEE V Y N F T S P E R A	C T P C D K S C E Q	G - C W G E G P E N	C Q K Y S K T N C S	P Q C W Q G R C F G
301	311	321	331	341
Consensus p s P s d C C H n q	C A a G C T G P r e	S D C l a C k k F r	D e A t C K d t C P	P l m l Y N P T T Y
Conservation				
EGFR_HUMAN K S P S D C C H N Q	C A A G C T G P R E	S D C L V C R K F R	D E A T C K D T C P	P L M L Y N P T T Y
EGFR_MOUSE R S P S D C C H N Q	C A A G C T G P R E	S D C L V C Q K F Q	D E A T C K D T C P	P L M L Y N P T T Y
EGFR_CHICK K V P S D C C H N Q	C A A G C T G P R E	S D C L A C R K F R	D D A T C K D T C P	P L V L Y N P T T Y
EGFR_DROSO P K P R E C C H L F	C A G G C T G P T Q	K D C I A C K N F F	D E A V S K E E C P	P M R K Y N P T T Y
EGFR_BEE P N P R E C C H L F	C A G G C T G P K Q	S D C I A C K N F F	D D G V C T Q E C P	P M Q K Y N P T T Y
351	361	371	381	391
Consensus q m d v N P E G K Y	s f G A T C V k k C	P - n y v v t D h G	s C V R s C p p D k	y e v e e e g - r k
Conservation				
EGFR_HUMAN Q M D V N P E G K Y	S F G A T C V K K C	P R N Y V V T D H G	S C V R A C G A D S	Y E M E E D G V R K
EGFR_MOUSE Q M D V N P E G K Y	S F G A T C V K K C	P R N Y V V T D H G	S C V R A C G P D Y	Y E V E E D G I R K
EGFR_CHICK Q M D V N P E G K Y	S F G A T C V R E C	P H N Y V V T D H G	S C V R S C N T D T	Y E V E E N G V R K
EGFR_DROSO V L E T N P E G K Y	A Y G A T C V K E C	P - G H L L R D N G	A C V R S C P Q D K	M D K G G E - - -
EGFR_BEE S W E P N P D G K Y	A Y G A T C V R R C	P - E H L L K D N G	A C V R S C P P K K	K A L N G E - - -
401	411	421	431	441
Consensus C k k C d G P C p K	v C n G i g i g e -	k - - l s i n A t N	I d s F K n C T a I	s G d i h I L p v a
Conservation				
EGFR_HUMAN C K C E G P C R K	V C N G I G I G E F	K D S L S I N A T N	I K H F K N C T S I	S G D L H I L P V A
EGFR_MOUSE C K C D G P C R K	V C N G I G I G E F	K D T L S I N A T N	I K H F K Y C T A I	S G D L H I L P V A
EGFR_CHICK C K C D G L C S K	V C N G I G I G E L	K G I L S I N A T N	I D S F K N C T K I	N G D V S I L P V A
EGFR_DROSO C V P C N G P C P K	T C P G V T V - -	— - - L H A G N	I D S F R N C T V I	D G N I R I L D Q T
EGFR_BEE C V P C D G P C P K	T C K G V E K - -	— - - V H S G N	I D S F K D C T I I	E G S I T I L D Q S
451	461	471	481	491
Consensus F s G d q f t y t -	- - - - - p P L	D P q k L e v f k T	V K E I T G F L I I	Q a w p d n f t d L
Conservation				
EGFR_HUMAN F R G D S F T H T	- - - - - P P L	D P Q E L D I L K T	V K E I T G F L L I	Q A W P E N R T D L
EGFR_MOUSE F K G D S F T R T	- - - - - P P L	D P R E L E I L K T	V K E I T G F L L I	Q A W P D N W T D L
EGFR_CHICK F L G D A F T K T	- - - - - L P L	D P K K L D V F R T	V K E I S G F L L I	Q A W P D N A T D L
EGFR_DROSO F S G F Q D V Y A N	Y T M G P R Y I P L	D P E R R E V F S T	V K E I T G Y L N I	E G T H P Q F R N L
EGFR_BEE F Q G F Q H V Y R N	F S F G K R Y E K M	H P D K L E V F S T	L K E I T G F L N I	Q G D H K D F K N L

1. EGFR MSA

501	511	521	531	541
Consensus h a F e N L E i I R	G R T k q h g q F s	- L A V V k l s i t	S L G L R S L K E I	S d G d v i l s g N
Conservation				
EGFR_HUMAN H A F E N L E I I R	G R T K Q H G Q F S	- L A V V S L N I T	S L G L R S L K E I	S D G D V I I S G N
EGFR_MOUSE H A F E N L E I I R	G R T K Q H G Q F S	- L A V V G L N I T	S L G L R S L K E I	S D G D V I I S G N
EGFR_CHICK Y A F E N L E I I R	G R T K Q H G Q Y S	- L A V V N L K I Q	S L G L R S L K E I	S D G D I A I M K N
EGFR_DROSO S Y F R N L E T I H	G R Q L M E S M F A	A L A I V K S S L Y	S L E M R N L K Q I	S S G S V V I Q H N
EGFR_BEE S Y F R N L E V I G	G R T L T E - Y F A	S L Y V V K T S L V	S F G L S S L K K I	Y S G S I A I L E N
551	561	571	581	591
Consensus k n L C Y A n t i N	W k k l f k t p e Q	K t k i i n N R n e	n d C k a d g h V C	d p l C S d e G C W
Conservation				
EGFR_HUMAN K N L C Y A N T I N	W K K L F G T S Q G	K T K I I S N R G E	N S C K A T G Q V C	H A L C S P E G C W
EGFR_MOUSE R N L C Y A N T I N	W K K L F G T P N Q	K T K I I M N N R A E	K D C K A V N H V C	N P L C S S E G C W
EGFR_CHICK K N L C Y A D T M N	W R S L F A T Q S Q	K T K I I Q N R N K	N D C T A D R H V C	D P L C S D V G C W
EGFR_DROSO R D L C Y V S N I R	W P A I Q K E P E Q	K V W V N E N L R A	D L C E K N G T I C	S D Q C N E D G C W
EGFR_BEE K N L C Y A Q S I N	W T R I K K S S E H	E S L L S N N R N E	S E C I K D G L V C	D E Q C S D E G C W
601	611	621	631	641
Consensus G P g P r q C I S C	k n f s r g r e C v	a k C n i l e g e p	r e f v e n s e C i	q C H p E C l p q -
Conservation				
EGFR_HUMAN G P E P R D C V S C	R N V S R G R E C V	D K C N L L E G E P	R E F V E N S E C I	Q C H P E C L P Q -
EGFR_MOUSE G P E P R D C V S C	Q N V S R G R E C V	E K C N I L E G E P	R E F V E N S E C I	Q C H P E C L P Q -
EGFR_CHICK G P G P F H C F S C	R F F S R Q K E C V	K Q C N I L Q G E P	R E F E R D S K C L	P C H S E C L V Q N
EGFR_DROSO G A G T D Q C L T C	K N F N F N G T C I	A D C G Y I S N - -	A Y K F D N R T C K	I C H P E C R - T -
EGFR_BEE G P G P A Q C L S C	K N F I L G N D C L	Q D C T A P G - - -	I Y Q A D E K T C K	V C H E E C D G S -
651	661	671	681	691
Consensus - - a - n - t C t G	p G p D h C i q C a	H y i D G P H C V k	e C P A g k y g e N	- - - - -
Conservation				
EGFR_HUMAN - - A M N I T C T G	R G P D N C I Q C A	H Y I D G P H C V K	T C P A G V M G E N	- - - - -
EGFR_MOUSE - - A M N I T C T G	R G P D N C I Q C A	H F I D G P H C V K	T C P A G I M G E N	- - - - -
EGFR_CHICK S T A Y N T T C S G	P G P D H C M K C A	H V R D G Q H C V S	A C P A G V L G E N	- - - - -
EGFR_DROSO - - - - - C N G	A G A D H C Q E C V	E C P K N K Y N D R	G V C R E C H A T C	- - - - -
EGFR_BEE - - - - - C I G	P N T D H C K K C K	E C P A S K Y N D N	G V C K S C H G N C	- - - - -
701	711	721	731	741
Consensus - - - - -	- - - - -	- - - - -	- - - - -	- d t l v W k Y a d
Conservation				
EGFR_HUMAN - - - - -	- - - - -	- - - - -	- - - - -	- N T L V W K Y A D
EGFR_MOUSE - - - - -	- - - - -	- - - - -	- - - - -	- N T L V W K Y A D
EGFR_CHICK - - - - -	- - - - -	- - - - -	- - - - -	- D T L V W K Y A D
EGFR_DROSO D G - C T G P K D T	I G I G A C T T C N	L A I I N N D A T V	K R C L L K D D K C	P D G Y F W E Y V H
EGFR_BEE V G G C E G P E N N	I G P N G C H S C D	K A I L N - D H V P	E G C L Q K K E S C	P D G Y Y Y E W V S

1. EGFR MSA

Consensus	751	761	771	781	791
Conservation	a n - - -	- - a V C h l C H	p n C t - - -	- - - - -	- - - - -
EGFR_HUMAN	A G	H V C H L C H	P N C T	- - - - -	- - - - -
EGFR_MOUSE	A N	- - - N V C H L C H	A N C T	- - - - -	- - - - -
EGFR_CHICK	A N	- - - A V C Q L C H	P N C T	- - - - -	- - - - -
EGFR_DROSO	P Q E Q G S L K P L	A G R A V C R K C H	P L C E L C T N Y G	Y H E Q V C S K C T	H Y K R R E Q C E T
EGFR_BEE	P L E Q G P L K P L	A S K A V C R K C H	S R C K K C T G Y G	F H E H V C Q E C T	K Y K R G E Q C E D
Consensus	801	811	821	831	841
Conservation	- - - - -	- - - - -	- - y G C t G P G I	e g C p k c - - -	- - - - -
EGFR_HUMAN	- - - - -	- - - - -	- - Y G C T G P G L	E G C P T N - - -	- - - - -
EGFR_MOUSE	- - - - -	- - - - -	- - Y G C A G P G L	Q G C E V W P - - -	- - - - -
EGFR_CHICK	- - - - -	- - - - -	- - R G C K G P G L	E G C P N G - - -	- - - - -
EGFR_DROSO	E C P A D H Y T D E	E Q R E C F Q R H P	E C N G C T G P G A	D D C K S C R N F K	L F D A N E T G P Y
EGFR_BEE	E C P A D Y F A D A	N - K L C I P C F S	E C R G C F G P G P	N Q C Y K C R N Y K	I Y I D E D T D G -
Consensus	851	861	871	881	891
Conservation	- - - - -	- - - - -	- - - - -	- - - - -	- - g p k i P s I a
EGFR_HUMAN	- - - - -	- - - - -	- - - - -	- - - - -	- - G P K I P S I A
EGFR_MOUSE	- - - - -	- - - - -	- - - - -	- - - - -	- - S G P K I P S I A
EGFR_CHICK	- - - - -	- - - - -	- - - - -	- - - - -	- - - - - S K T P S I A
EGFR_DROSO	V N S T M F N C T S	K C P L E M R H V N	Y Q Y T A I G P Y C	A A S P P R S S K I	T A N L D V N M I F
EGFR_BEE	- N T T S F N C T E	T C T P E Y P H K I	F N P D S E - P Y C	S L E T A G - - - L	I E N E L Q P A I L
Consensus	901	911	921	931	941
Conservation	a G v v g g L L a l	I V V a l g I g l f	m R r R h i v r K r	T l r r l l q e r -	- E l v E P L t P s
EGFR_HUMAN	T G M V G A L L L	L V V A L G I G L F	M R R R H I V R K R	T L R R L L Q E R -	- E L V E P L T P S
EGFR_MOUSE	T G I V G G L L F I	V V V A L G I G L F	M R R R H I V R K R	T L R R L L Q E R -	- E L V E P L T P S
EGFR_CHICK	A G V V G G L L C L	V V V G L G I G L Y	L R R R H I V R K R	T L R R L L Q E R -	- E L V E P L T P -
EGFR_DROSO	I I T G A V L V P T	I C I L C V V T Y I	C R Q K Q K A K K E	T V K M T M A L S G	C E D S E P L R P S
EGFR_BEE	A G V A V F A L A F	L V V A A I I M Y F	W R V R A K A K E N	T V K M T M A L T G	L D D N E P L R P T
Consensus	951	961	971	981	991
Conservation	g e a p N q a k L R	I I K e t E f k K i	k v L G s G A F G t	V Y K G I W i P E G	E k V K I P V A I K
EGFR_HUMAN	G E A P N Q A L L R	I L K E T E F K K I	K V L G S G A F G T	V Y K G L W I P E G	E K V K I P V A I K
EGFR_MOUSE	G E A P N Q A H L R	I L K E T E F K K I	K V L G S G A F G T	V Y K G L W I P E G	E K V K I P V A I K
EGFR_CHICK	- - - - -	- - - - -	- - - - -	- - - - -	- - - - -
EGFR_DROSO	N I G A N L C K L R	I V K D A E L R K G	G V L G M G A F G R	V Y K G V W V P E G	E N V K I P V A I K
EGFR_BEE	G V K P N L A K L R	I I K E E E M R K G	G I L G Y G A F G N	V Y K G V W V P E G	E N V K I P V A I K

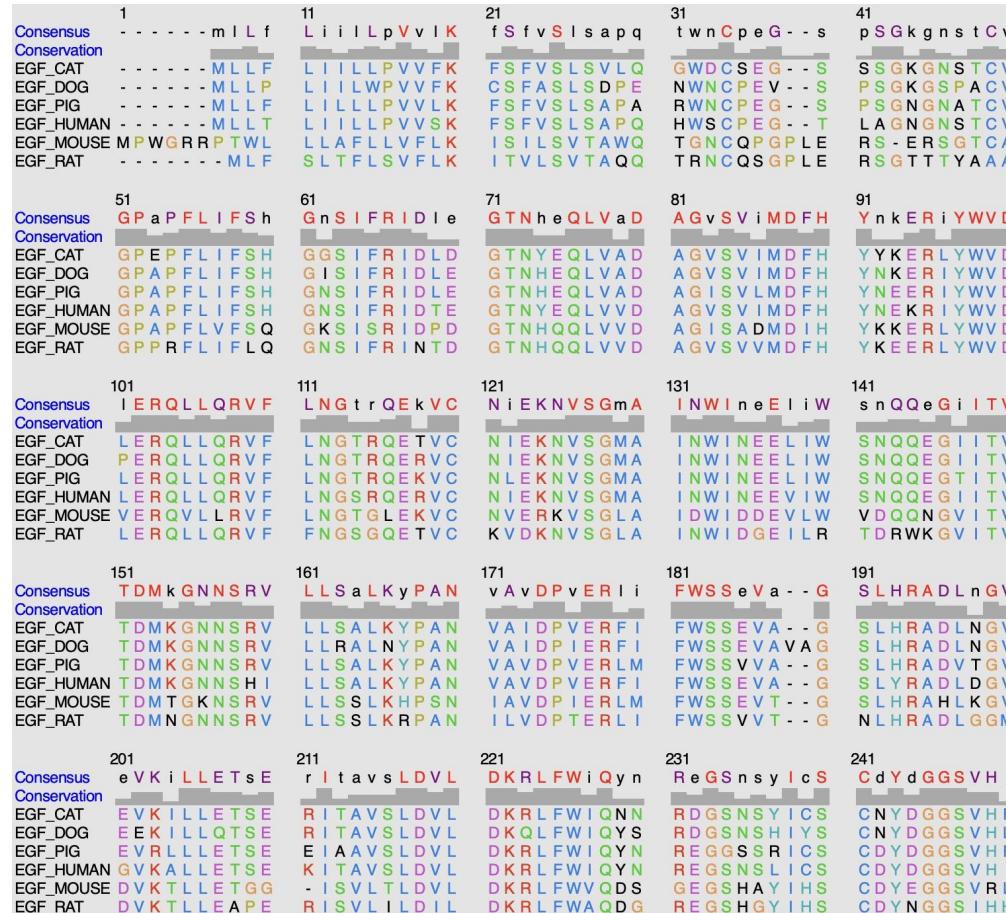
1. EGFR MSA

1101	1111	1121	1131	1141
Consensus q T P q h V K I T D	F G L A K L L g a e	e k e Y h A a G G K	m P I K W m A L E s	I l h R i y T h q S
Conservation				
EGFR_HUMAN K T P Q H V K I T D	F G L A K L L G A E	E K E Y H A E G G K	V P I K W M A L E S	I L H R I Y T H Q S
EGFR_MOUSE K T P Q H V K I T D	F G L A K L L G A E	E K E Y H A E G G K	V P I K W M A L E S	I L H R I Y T H Q S
EGFR_CHICK -	-	-	-	-
EGFR_DROSO Q T P S L V K I T D	F G L A K L L S S D	S N E Y K A A G G K	M P I K W L A L E C	I R N R V F T S K S
EGFR_BEE Q T P N C V K I T D	F G L A K L L D I N	E E Q Y K A A G G K	M P I K W L A L E C	I Q H R V F T H K S
1151	1161	1171	1181	1191
Consensus D V W a y G V T i W	E l m t f G s k P y	e g i P A s d i p s	i l E k G e r L p Q	P p I C T i D v Y m
Conservation				
EGFR_HUMAN D V W S Y G V T V W	E L M T F G S K P Y	D G I P A S E I S S	I L E K G E R L P Q	P P I C T I D V Y M
EGFR_MOUSE D V W S Y G V T V W	E L M T F G S K P Y	D G I P A S D I S S	I L E K G E R L P Q	P P I C T I D V Y M
EGFR_CHICK -	-	-	-	-
EGFR_DROSO D V W A F G V T I W	E L L T F G Q R P H	E N I P A K D I P D	L I E V G L K L E Q	P E I C S L D I Y C
EGFR_BEE D V W A F G V T I W	E V L T Y G G R P Y	E N V P A R N V P E	L L E K G E R L P Q	P A I C T I D V Y M
1201	1211	1221	1231	1241
Consensus i m v k C W m i D A	d s R P k F k e l i	- e F a k m a R D P	q R Y L a l i q G D k	r m h L P s y T d q
Conservation				
EGFR_HUMAN I M V K C W M I D A	D S R P K F R E L I	I E F S K M A R D P	Q R Y L V I Q G D E	R M H L P S P T D S
EGFR_MOUSE I M V K C W M I D A	D S R P K F R E L I	L E F S K M A R D P	Q R Y L V I Q G D E	R M H L P S P T D S
EGFR_CHICK -	-	-	-	-
EGFR_DROSO T L L S C W H L D A	A M R P T F K Q L T	T V F A E F A R D P	G R Y L A I P G D K	F T R L P A Y T S Q
EGFR_BEE I M I K C W M L D A	E S R P S F K E L A	E D F A K M S R D P	G R Y L A I K G D K	Y M R L P S Y T L Q
1251	1261	1271	1281	1291
Consensus d - - - i y r a a	m d e e d m e a v v	d a D E Y L i P q q	g f - - - p - - -	- - - - - - - - -
Conservation				
EGFR_HUMAN N - - - F Y R A L	M D E E D M D D V V	D A D E Y L I P Q Q	G F F S S P	- - - - - - - - -
EGFR_MOUSE N - - - F Y R A L	M D E E D M D E D V V	D A D E Y L I P Q Q	G F F N S P	- - - - - - - - -
EGFR_CHICK -	-	-	-	-
EGFR_DROSO D E K D L I R K L A	P T T D G S E A I A	K P D D Y L Q P K A	A P	-
EGFR_BEE D E K E M I R N L A	S A M D G P E A L V	D A D E Y L Q P K S	R A P I P P G L S A	S S T S G S P P N T
1301	1311	1321	1331	1341
Consensus - - - - -	- - - - -	- - - - -	- - - s t h R t p l	I s s l s a t s - -
Conservation				
EGFR_HUMAN - - - - -	- - - - -	- - - - -	- - - S T S R T P L	L S S L S A T S N N
EGFR_MOUSE - - - - -	- - - - -	- - - - -	- - - S T S R T P L	L S S L S A T S N N
EGFR_CHICK -	-	-	-	-
EGFR_DROSO - - - - -	- - - - -	- - - - -	- - - G P S H R T D C	T D E M P K L N - -
EGFR_BEE P V K P C W P N G K	P L A A D S P T P Q	N Q Q N W D R E L L	R Y G A N H R N G N	V S S H E P G N S A

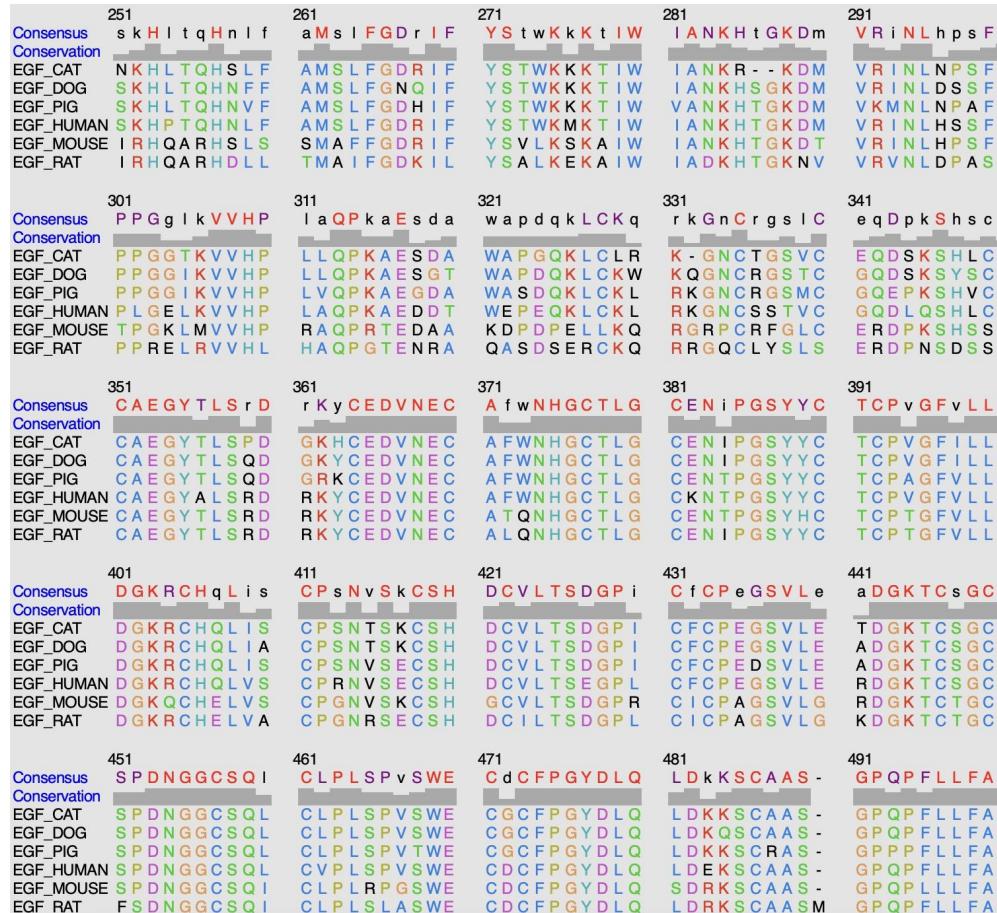
1. EGFR MSA

	1351	1361	1371	1381	1391
Consensus	- - - - n g	- - - - d	- - R Y s s D P t	- - - - d d	t f l p v p e y v n
Conservation					
EGFR_HUMAN	S T V A C I D R N G	L Q S C P I K E D S	F L Q R Y S S D P T	G A L T E D S I D D	T F L P V P E Y I N
EGFR_MOUSE	S T V A C I N R N G	- - S C R V K E D A	F L Q R Y S S D P T	G A V T E D N I D D	A F L P V P E Y V N
EGFR_CHICK	- - - - -	- - - - -	- - - - -	- - - - -	- - - - -
EGFR_DROSO	- - - - -	- - - R Y C K D P S	- - - - -	- - - - -	NS S T G D D E R D
EGFR_BEE	Q H G H Y T P P N G	H C G H V V G S D S	T A S R Y C S D P L	K M I D V R D C D V	T D D C F D G E V N
1401					
Consensus	q s v v p k r p a G s	I q l p l y h d q p	I y p a P s r q p h	y q n p h s m a - -	- p e y l n t m q p
Conservation					
EGFR_HUMAN	Q S V P K R P A G S	V Q N P V Y H N Q P	L N P A P S R D P H	Y Q D P H S T A V G	N P E Y L N T V Q P
EGFR_MOUSE	Q S V P K R P A G S	V Q N P V Y H N Q P	L H P A P G R D L H	Y Q N P H S N A V G	N P E Y L N T A Q P
EGFR_CHICK	- - - - -	- - - - -	- - - - -	- - - - -	- - - - -
EGFR_DROSO	S S A R E V G V G N	L R L D L P V D E D	D Y L M P T C Q P G	P N N N N N M N -	- N P N Q N N M A A
EGFR_BEE	S A H Q Q A Q V G N	L K L D L P L D E D	D Y L M P S P Q L P	T N T T Q Y M D L I	G D S K P T E M E P
1451					
Consensus	t c v n s g - - - y	a - w i q k g - - -	- S I D N P e Y q q	d f f p k e - k p n	g l f k g t t - - -
Conservation					
EGFR_HUMAN	T C V N S T F D S P	A H W A Q K G S H Q	I S L D N P D Y Q Q	D F F P K E A K P N	G I F K G S T - - -
EGFR_MOUSE	T C L S S G F N S P	A L W I Q K G S H Q	M S L D N P D Y Q Q	D F F P K E T K P N	G I F K G P T - - -
EGFR_CHICK	- - - - -	- - - - -	- - - - -	- - - - -	- - - - -
EGFR_DROSO	V G V A A G - - - Y	M D L I G V P - - -	V S V D N P E Y L L	N A Q T L G V G E S	P I P T Q T I G I P
EGFR_BEE	K R V N N G Y R K Y	P E F L T I Q G - K	T S L D N P E Y I M	S Q D E G P L T P Q	T I G I P T P D L E
1501					
Consensus	- - - a e g a e y l	r V a p p s S E - i	g a - - - - -	- - - - -	- - - - -
Conservation					
EGFR_HUMAN	- - - A E N A E Y L	R V A P Q S S E F I	G A - - - - -	- - - - -	- - - - -
EGFR_MOUSE	- - - A E N A E Y L	R V A P P S S E F I	G A - - - - -	- - - - -	- - - - -
EGFR_CHICK	- - - - -	- - - - -	- - - - -	- - - - -	- - - - -
EGFR_DROSO	V M G G P G T M E V	K V P M P G S E P T	S S D H E Y Y N D T	- - - Q R E L Q P L	H R N R N T E T R V
EGFR_BEE	K V L T N G T F G S	Q V R Q R S S E - E	E S D H E Y Y N D F	D R L E R E L Q P L	K P L R K N E T T V
1511					
1521					
1531					
1541					

2. EGF MSA



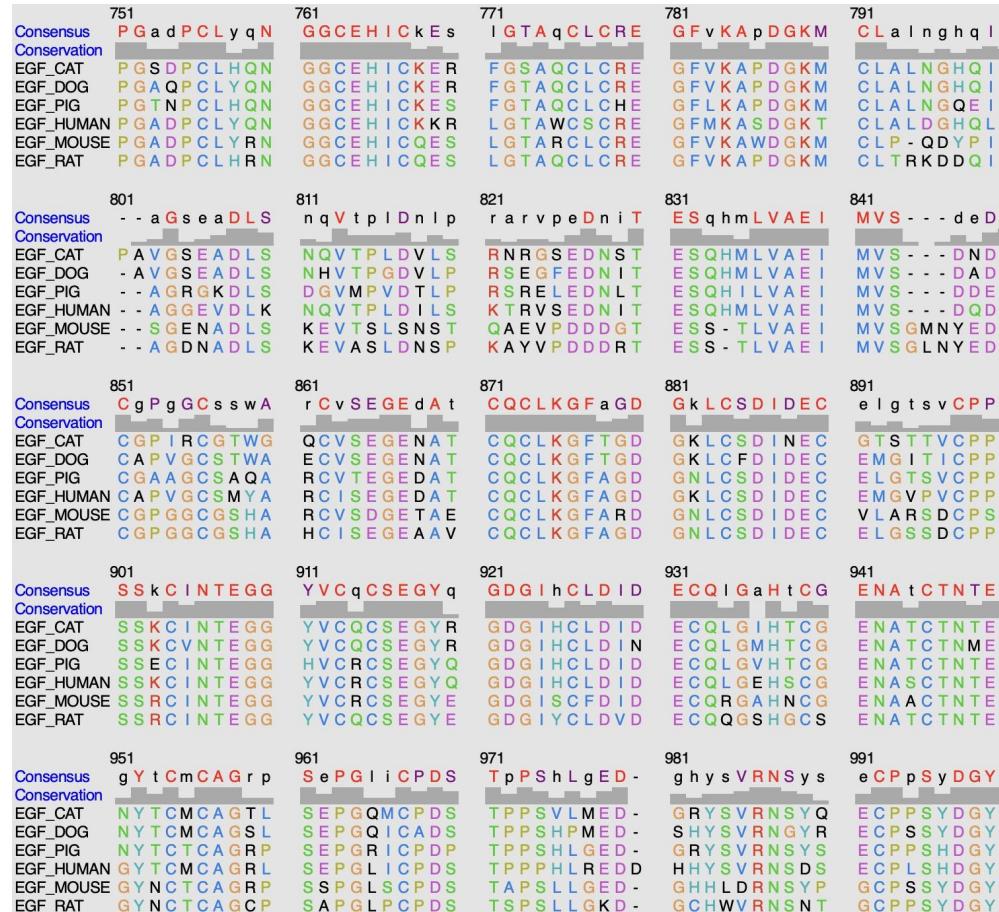
2. EGF MSA



2. EGF MSA

	501	511	521	531	541
Consensus	S Q D I R H M H F D	G T D Y g t L L S q	Q M G M V f A L D h	D P V E n K I Y F A	h T A L K W I E R A
Conservation					
EGF_CAT	S Q D I R H M H F D	G T D Y G S L L S Q	Q M G M V F A L D H	D P V E N K I Y F A	H T A L K W I E R A
EGF_DOG	S Q D I R H M H F D	G T D Y G T L L S Q	Q M G M V F A L D H	D P V E N K I Y F A	H T A L K W I E R A
EGF_PIG	S Q D I R H M H F D	G T D Y E T L L N Q	Q I G M V L A L D H	D P V E N K V Y F A	H T A L K W I E R A
EGF_HUMAN	S Q D I R H M H F D	G T D Y G T L L S Q	Q M G M V Y A L D H	D P V E N K I Y F A	H T A L K W I E R A
EGF_MOUSE	S Q D I R H M H F D	G T D Y K V L L S R	Q M G M V F A L D Y	D P V E S K I Y F A	Q T A L K W I E R A
EGF_RAT	S Q D I R H M H F D	G T D Y K T L L S R	Q M G M V F A L D Y	D P V E S K I Y F A	Q T A L K W I E R A
551					
Consensus	N M D G S Q R E R L	I e E g V D v P E G	L A i D W I g R k f	Y W T D s G K S I I	e G S D L n G K h r
Conservation					
EGF_CAT	N M D G S Q R E R L	I E E A V D V P E G	L A I D W I D R K F	Y W T D R G K A L I	E G S D L N G K H R
EGF_DOG	N M D G S Q R E R L	I E E G V D V P E G	L A I D W I D R K F	Y W T D S G K S L I	E G S D L N G K H R
EGF_PIG	N M D G S Q R E R L	F E E A V D V P E G	L A I D W I G R K F	Y W T D R G R S L I	E G S D L N G K Y R
EGF_HUMAN	N M D G S Q R E R L	I E E G V D V P E G	L A V D W I G R R F	Y W T D R G K S L I	G R S D L N G K R S
EGF_MOUSE	N M D G S Q R E R L	I T E G V D T L E G	L A L D W I G R R I	Y W T D S G K S V V	G G S D L S G K H H
EGF_RAT	N L D G S Q R E R R	I T E G V D T P E G	L A V D W I G R R I	Y W T D S G K S V I	E G S D L S G K H H
601					
Consensus	e I I I K E d I S q	P R G I A V H P m A	k R L F W T D m G i	n P R I E S S S L Q	G i g R l v I A S S
Conservation					
EGF_CAT	E I I I K E E V S Q	P R G I A V H P M A	K R L F W T D M G I	N P R I E S S S L Q	G i G R R V I A S S
EGF_DOG	E I I I K E D I S Q	P R G I A V H P M A	K R L F W T D M G I	N P R I E S S S L Q	G i G R L V I A S S
EGF_PIG	E I I I K E D I S Q	P R G I A V H P V A	K R L F W T D M G T	N P R I E S S S L Q	G i G R Q V I A S S
EGF_HUMAN	K I I T K E N I S Q	P R G I A V H P M A	K R L F W T D T G I	N P R I E S S S L Q	G L G R L V I A S S
EGF_MOUSE	R I I I Q E R I S R	P R G I A V H P R A	R R L F W T D V G M	S P R I E S A S L Q	G S D R V L I A S S
EGF_RAT	Q I I I K E S I S R	P R G I A V H P K A	R R L F W T D T G M	S P R I E S S S L Q	G S D R T L I A S S
651					
Consensus	d L v w P S G I T I	D Y L T D k L Y W C	D a K q S V I E M A	N L D G S K R q R L	a Q N D V G H P F a
Conservation					
EGF_CAT	D L V W P S G I T I	D Y L T D k L Y W C	D A K Q S V I E M A	N L D G S K R Q R L	A Q N D V G H P F A
EGF_DOG	D L V W P S G I T I	D Y V T D k L Y W C	D T K L S V I E M A	N L D G S K R Q R L	A Q N D V G H P F A
EGF_PIG	D L V W P S G I T I	D Y L T D k L Y W C	D A K Q S V I E M S	N L D G S R R Q R L	A Q N D V G H P F A
EGF_HUMAN	D L I W P S G I T I	D F L T D k L Y W C	D A K Q S V I E M A	N L D G S K R R R L	T Q N D V G H P F A
EGF_MOUSE	N L L E P S G I T I	D Y L T D T L Y W C	D T K R S V I E M A	N L D G S K R R R L	I Q N D V G H P F S
EGF_RAT	N L L E P S G I A I	D Y L T D T L Y W C	D T K L S V I E M A	D L D G S K R R R L	T Q N D V G H P F S
701					
Consensus	I A V F E D H V W F	S D W a m P S V I R	V N K R T G k N R V	R L r G S M L K P S	S L V V V H P L A K
Conservation					
EGF_CAT	I A V F E D H V W F	S D W T M P S V I R	V N K R T G K N R V	R L R G S M L K P S	S L V V V H P L A K
EGF_DOG	M A V F E D H V W F	S D W T M P S V I R	V D K R T G K N R V	R L R G S M L K P S	S L V V V H P L A K
EGF_PIG	V A V F E D H V W F	S D W T M P S V I R	V N K R T G Q N R V	R L R G S M L K P S	S L V V V H P L A K
EGF_HUMAN	V A V F E D Y V W F	S D W A I P S V I R	V N K R T G Q N R V	R L Q G S M L K P S	S L V V V H P L A K
EGF_MOUSE	L A V F E D H L W V	S D W A I P S V I R	V N K R T G Q N R V	R L Q G S M L K P S	S L V V V H P L A K
EGF_RAT	L A V F E D H V W F	S D W A I P S V I R	V N K R T G Q N R V	R L R G S M L K P S	S L V V V H P L A K

2. EGF MSA



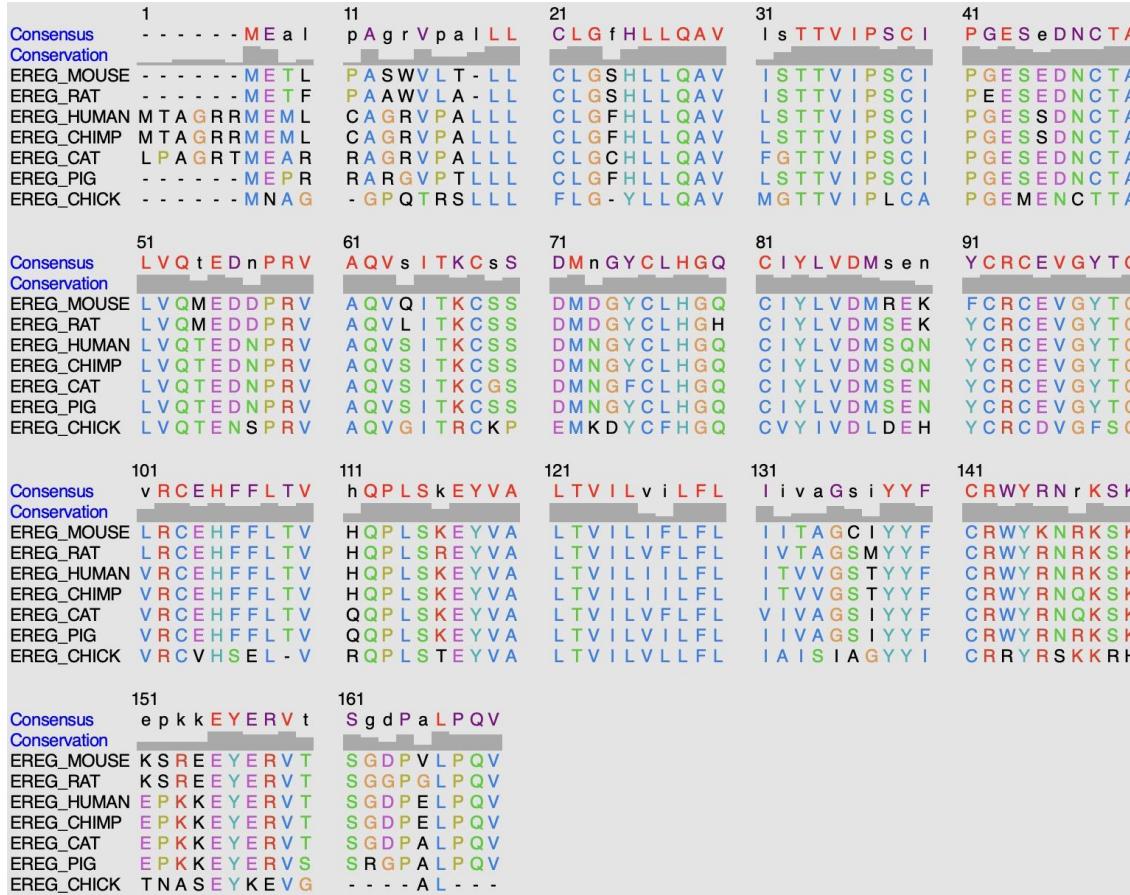
2. EGF MSA

1001	1011	1021	1031	1041
Consensus L y g G VC M Y I E	a v d r Y a C N C V	f G Y v G E R C Q h	R D L k w W E L R H	A G q G q q h q v t
Conservation				
EGF_CAT L Y N G V C M Y I E	A V D R Y A C N C V	F G Y V G E R C Q H	R D L K - W E L R H	A G Q G R Q R Q V T
EGF_DOG L Y N G V C M Y I E	A V D R Y A C N C V	F G Y V G E R C Q H	R D L K - W E L R H	A G Q G R Q R Q V A
EGF_PIG L H G G V C M Y I E	A V D S Y A C N C V	F G Y V G E R C Q H	R D L K W W E L R H	A G L G R Q W N V T
EGF_HUMAN L H D G V C M Y I E	A L D K Y A C N C V	V G Y I G E R C Q Y	R D L K W W E L R H	A G H G Q Q Q K V I
EGF_MOUSE L N G G V C M H I E	S L D S Y T C N C V	I G Y S G D R C Q T	R D L R W W E L R H	A G Y G Q K H D I M
EGF_RAT L N G G V C M Y V E	S V D R Y V C N C V	I G Y I G E R C Q H	R D L R W W K L R H	A D Y G Q R H D I T
1051	1061	1071	1081	1091
Consensus V V A V C V V a L V	L L L L L G I W G A	h y Y R T q K I I S	k n p K N P y E E s	S r e v s s s r P a
Conservation				
EGF_CAT V V A V C V V A L V	L L L L L G L W G A	H C Y R T K K L P S	K N L K N P Y E E S	S R E - - V S R P T
EGF_DOG A V A V G V V V L V	L L L L L G L G G A	H C Y R T K K L S S	K N L K N P Y E E P	S R E G S S S R P S
EGF_PIG V V A V C V V V L V	L L L L L G L W G A	H Y Y R T Q K L L S	K N P K N P Y E E S	G R D V S G I R P A
EGF_HUMAN V V A V C V V V L V	M L L L L S L W G A	H Y Y R T Q K L L S	K N P K N P C D E P	S R D V R S R R P A
EGF_MOUSE V V A V C V M A L V	L L L L L G M W G T	Y Y Y R T R K Q L S	E S S K K P S E E S	S G S V S S S G P N
EGF_RAT V V S V C V V A L A	L L L L L G M W G T	Y Y Y R T R K Q L S		S S N V S S S N G P D
1101	1111	1121	1131	1141
Consensus d s - e A g m a S c	P Q P W F V V i k E	H Q n l k N G s q P	a A g k d G a a a D	v g q f s S l e - G
Conservation				
EGF_CAT D S - E A G M A S C	P Q P W F V V I K E	H Q N L R N G S Q S	M A L K D S E A A D	V S Q F S S R E T G
EGF_DOG D S - E A R M A S F	P Q P W F V V I K E	H Q N L R N G S Q P	M A L K D G E S A D	V S Q F S S P E P G
EGF_PIG D G - E A G M S S C	P Q P W F V V I K E	H Q N L R N G S Q P	G A P K D G L G A D	V G Q F S S L E P G
EGF_HUMAN D T - E D G M S S C	P Q P W F V V I K E	H Q D L K N G G Q P	V A G E D G Q A A D	- - - - - G
EGF_MOUSE S S S G A A V A S C	P Q P W F V V L E K	H Q D P K N G S L P	A D G T N G A V V D	A G L S P S L Q L G
EGF_RAT S S - G A G V S S G	P Q P W F V V L E E	H Q Q P K N G R L P	A A G T N G A V V E	A G L S S S L - - -
1151	1161	1171	1181	1191
Consensus S v q p t S w R q e	p q p l y - m g t e Q	g C c i P p S S d k	G - g p q g m e - s	f h I P S y e a q -
Conservation				
EGF_CAT S V Q L P S R R N E	P Q V Y - M G A E Q	G C C I P P S S D K	G P G P H G M Q W G	F H L P S C E A Q P
EGF_DOG S V K R T S W R N E	H Q L Y - K D T E Q	G C C T P P S S N R	G T G S Q S M E Q S	F S V P S Y E A Q P
EGF_PIG S L Q P T S W R K E	P Q M Y - M D T E Q	G C C I P S S S D K	G S G P Q G I G Y S	F H L P S Y G A R S
EGF_HUMAN S M Q P T S W R Q E	P Q L C G M G T E Q	G C W I P V S S D K	G S C P Q V M E R S	F H M P S Y G T Q T
EGF_MOUSE S V H L T S W R Q K	P H I D G M G T Q Q	S C W I P P S S D R	G - - P Q E I E G N	S H L P S Y R P - -
EGF_RAT				
1201	1211	1221	1231	
Consensus i a l G v E K p h S	L I S A n p l l q q	R A p D p P h Q m e	I t Q	
Conservation				
EGF_CAT I A L G V E K P Q S	L L S A N P I L Q Q	R A S D L P H Q K K	L T Q	
EGF_DOG I A L G V E K P L Q S	L L S A K P L L Q Q	R A P D P P H Q M K	L I Q	
EGF_PIG I A V G V E K S H S	L L S A N P L R Q Q	R A P D P P H Q M E	L T Q	
EGF_HUMAN L E G G V E K P H S	L L S A N P L W Q Q	R A L D P P H Q M E	L T Q	
EGF_MOUSE - V G P E K L H S	L Q S A N G S C H E	R A P D L P R Q T E	P V Q	
EGF_RAT				

3. TGF- α MSA

Consensus	M V P s a G Q I A L	11 f A L G I I I A v C	21 Q A L E N S T S p L	31 S - D p P v A A A V	41 V S H F N d C P D S
Conservation					
TGFA_RAT	M V P A G Q Q L A L	L A L G I L V A V C	Q A L E N S T S P L	S - D S P V A A A V	V S H F N K C P D S
TGFA_MOUSE	M V P A T G Q Q L A L	L A L G I L L A V C	Q A L E N S T S P L	S - D S P V A A A V	V S H F N K C P D S
TGFA_PIG	M V P S A G Q F A L	F A L G I L L A V C	Q A L E N S T S P L	S A D P P I A A A V	V S H F N D C P D S
TGFA_HUMAN	M V P S A G Q Q L A L	F A L G I V L A A C	Q A L E N S T S P L	S A D P P V A A A V	V S H F N D C P D S
TGFA_SHEEP	M V P S A G Q Q L A L	F A L G I F L A V C	Q A L E N S T S P L	S - D P P V A A A V	V S H F N D C P D S
TGFA_RABBIT	- - - - -	- - - - -	- - - - -	- V	V S H F N Q C P D S
Consensus	H T Q f C F H G T C	61 R F L V Q K P A	71 C V C H S G Y V G a	81 R C E H A D L L A V	91 V A A S Q K K Q A I
Conservation					
TGFA_RAT	H T Q Y C F H G T C	R F L V Q E E K P A	C V C H S G Y V G V	R C E H A D L L A V	V A A S Q K K Q A I
TGFA_MOUSE	H T Q Y C F H G T C	R F L V Q E E K P A	C V C H S G Y V G V	R C E H A D L L A V	V A A S Q K K Q A I
TGFA_PIG	H S Q F C F H G T C	R F L V Q E D K P A	C V C H S G Y V G A	R C E H A D L L A V	V A A S Q K K Q A I
TGFA_HUMAN	H T Q F C F H G T C	R F L V Q E D K P A	C V C H S G Y V G A	R C E H A D L L A V	V A A S Q K K Q A I
TGFA_SHEEP	H T Q F C F H G T C	R F L L Q E E K P A	C V C H S G Y V G A	R C E H A D L L A V	V A A S Q K K Q A I
TGFA_RABBIT	H T Q F C F H G T C	R F L V Q E D K P A	C V C H S G Y V G A	R C E H A D L L A -	- - - - -
Consensus	T A L V V V s I V A	101 L A V L I I T C V L	111 I H C C q v R K H c	121 e w c r a l - c r h	131 e k p s a l l k g r
Conservation					
TGFA_RAT	T A L V V V S I V A	L A V L I I T C V L	I H C C Q V R K H C	E W C R A L V C R H	E K P S A L L K G R
TGFA_MOUSE	T A L V V V S I V A	L A V L I I T C V L	I H C C Q L R K H C	E W C R A L V C R H	E K P S A L L K G R
TGFA_PIG	T A L V V V S I V A	L A V L I I T C V L	I H C C Q V R K H C	E W C R A L I C R H	E K P S A L L K G R
TGFA_HUMAN	T A L V V V S I V A	L A V L I I T C V L	I H C C Q V R K H C	E W C R A L I C R H	E K P S A L L K G R
TGFA_SHEEP	T A L V V V T I V A	L A V L I I T C V L	I H C C E V R K H -	- - - - -	- S V V V P - -
TGFA_RABBIT	- - - - -	- - - - -	- - - - -	- - - - -	- - - - -
Consensus	t a c c h s e t v v	151 t A C C H S E T V V			
Conservation					
TGFA_RAT	T A C C H S E T V V				
TGFA_MOUSE	T A C C H S E T V V				
TGFA_PIG	T A C C H S E T V V				
TGFA_HUMAN	T A C C H S E T V V				
TGFA_SHEEP	- - - - -				
TGFA_RABBIT	- - - - -				

4. EREG MSA



5. EGF UNIPROT vs. PDB sequence

	951	961	971	981	991
Consensus	-	-	-	-	-
Conservation	-	-	-	-	-
EGF_HUMAN_PDB	-	-	-	-	-
EGF_HUMAN_UNIPROT	C P D S T P P P H L	R E D D H H Y S V R	E C P L S H	D G Y C L H D G V C	M Y I E A L D K Y A
	1001	1011	1021	1031	1041
Consensus	C N C V V G Y I G E	R C Q Y R D L K W W	E	-	-
Conservation	-	-	-	-	-
EGF_HUMAN_PDB	C N C V V G Y I G E	R C Q Y R D L K W W	E	-	-
EGF_HUMAN_UNIPROT	C N C V V G Y I G E	R C Q Y R D L K W W	E L R H A G H G Q Q	Q K V I V V A V C V	V V L V M L L L S