

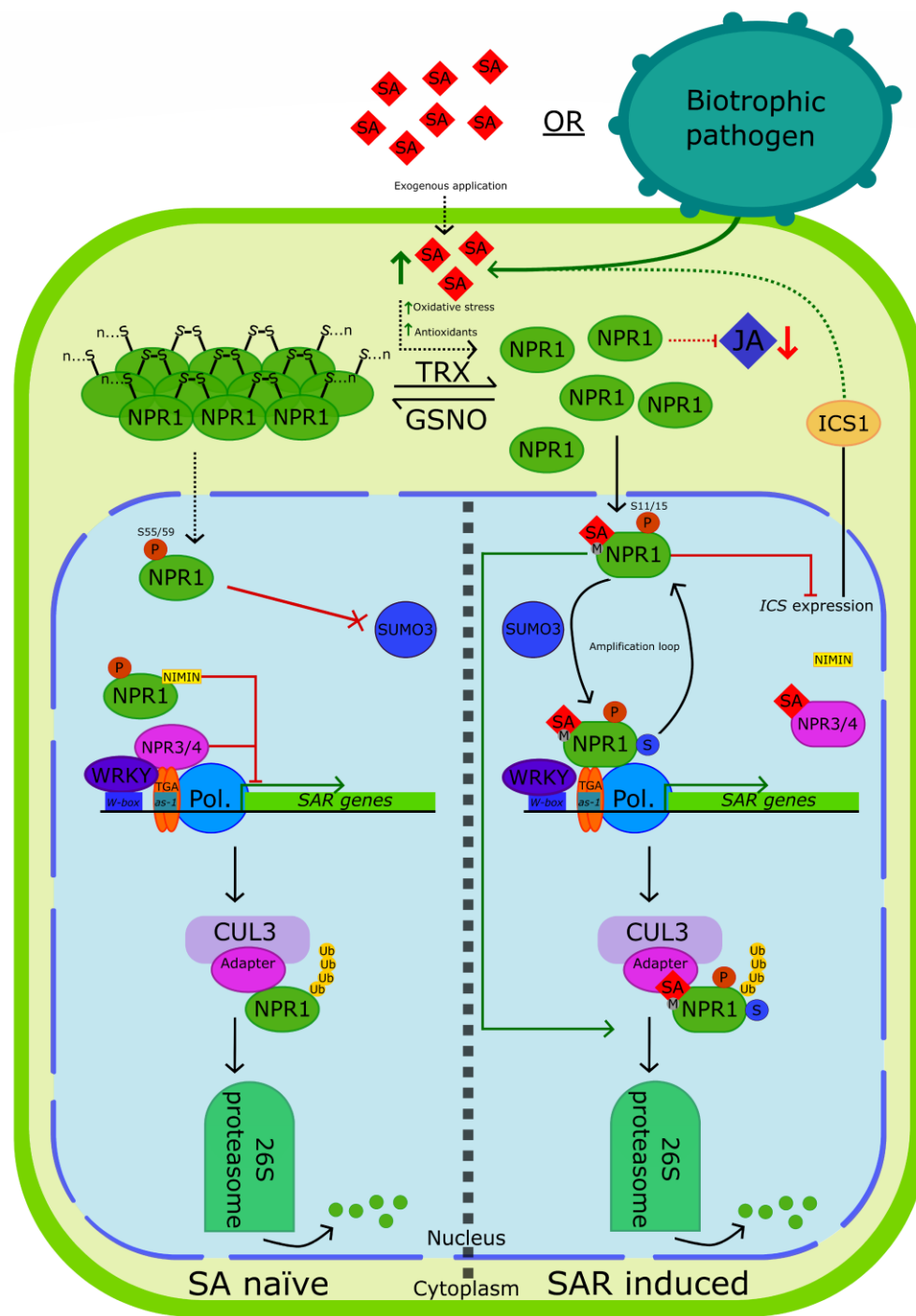
拟南芥水杨酸受体 NPR1的结构和功能研究

第14、15小组：林龙 陶奕含 李璐寒 刘静 左大庆 黄润洲

报告人：黄润洲

2020.01.05

Background



BLASTP

select all 17 sequences selected

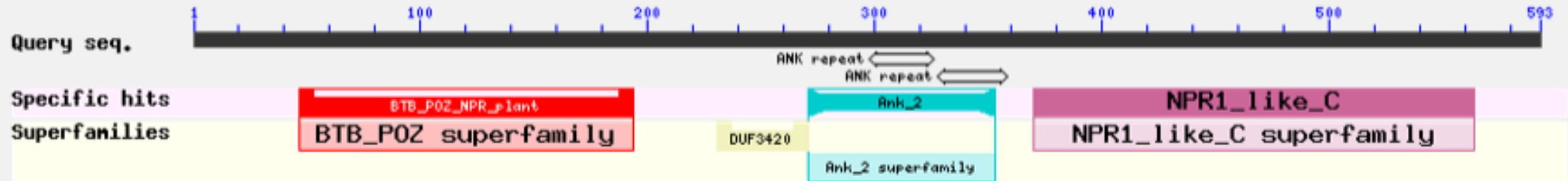
[GenPept](#) [Graphics](#) [Distance tree of results](#) [Multiple alignment](#)

	Description	Max Score	Total Score	Query Cover	E value	Per. Ident	Accession
<input checked="" type="checkbox"/>	RecName: Full=Regulatory protein NPR1; AltName: Full=BTB/POZ domain-containing protein NPR1; AltName: Full=Non-inducible immunity	1208	1208	100%	0.0	100.00%	P93002.1
<input checked="" type="checkbox"/>	RecName: Full=Regulatory protein NPR2; AltName: Full=BTB/POZ domain-containing protein NPR2 [Arabidopsis thaliana]	698	698	98%	0.0	62.29%	Q9SZI3.1
<input checked="" type="checkbox"/>	RecName: Full=BTB/POZ domain and ankyrin repeat-containing protein NPR1; Short=OsNPR1; AltName: Full=NPR1 homolog 1; Short=Os	457	457	92%	4e-154	46.37%	Q5D0W8.1
<input checked="" type="checkbox"/>	RecName: Full=BTB/POZ domain and ankyrin repeat-containing protein NPR1; Short=OsNPR1; AltName: Full=NPR1 homolog 1; Short=Os	457	457	92%	5e-154	45.84%	Q9FDY4.1
<input checked="" type="checkbox"/>	RecName: Full=Regulatory protein NPR3; AltName: Full=BTB/POZ domain-containing protein NPR3 [Arabidopsis thaliana]	360	360	87%	3e-116	39.07%	Q8L746.1
<input checked="" type="checkbox"/>	RecName: Full=Regulatory protein NPR4; AltName: Full=BTB/POZ domain-containing protein NPR4 [Arabidopsis thaliana]	358	358	88%	9e-116	38.99%	Q5ICL9.1
<input checked="" type="checkbox"/>	RecName: Full=BTB/POZ domain and ankyrin repeat-containing protein NPR1; Short=MhNPR1; AltName: Full=NPR1 homolog [Malus hup	358	358	84%	3e-115	40.98%	E7BQV0.2
<input checked="" type="checkbox"/>	RecName: Full=BTB/POZ domain and ankyrin repeat-containing protein NPR3; Short=OsNPR3 [Oryza sativa Japonica Group]	350	350	87%	2e-112	40.15%	Q75HA6.1
<input checked="" type="checkbox"/>	RecName: Full=BTB/POZ domain and ankyrin repeat-containing protein NPR2; Short=OsNPR2; AltName: Full=NPR1 homolog 2; Short=Os	347	347	86%	1e-110	39.37%	Q0JJ01.1
<input checked="" type="checkbox"/>	RecName: Full=Regulatory protein NPR6; AltName: Full=BTB/POZ domain-containing protein NPR6; AltName: Full=Protein BLADE-ON-PE	146	146	59%	8e-37	29.72%	Q9M1I7.1
<input checked="" type="checkbox"/>	RecName: Full=Regulatory protein NPR5; AltName: Full=BTB/POZ domain-containing protein NPR5; AltName: Full=Protein BLADE ON PE	131	131	56%	2e-31	28.65%	Q9ZVC2.1
<input checked="" type="checkbox"/>	RecName: Full=BTB/POZ domain and ankyrin repeat-containing protein NOOT2; AltName: Full=Protein NODULE ROOT 2; Short=MtNOOT	130	130	55%	3e-31	28.65%	A0A072VIM5.1
<input checked="" type="checkbox"/>	RecName: Full=BTB/POZ domain and ankyrin repeat-containing protein NOOT1; AltName: Full=Protein NODULE ROOT 1; Short=MtNOOT	130	130	55%	4e-31	29.09%	Q2HW56.1
<input checked="" type="checkbox"/>	RecName: Full=BTB/POZ domain and ankyrin repeat-containing protein COCH; AltName: Full=Protein COCHLEATA [Pisum sativum]	129	129	55%	5e-31	29.09%	G8GTN7.1
<input checked="" type="checkbox"/>	RecName: Full=BTB/POZ domain and ankyrin repeat-containing protein NPR5; Short=OsNPR5 [Oryza sativa Japonica Group]	125	125	55%	2e-29	28.87%	A2CIR7.1
<input checked="" type="checkbox"/>	RecName: Full=BTB/POZ domain and ankyrin repeat-containing protein NH5.2 [Oryza sativa Japonica Group]	122	122	55%	2e-28	27.60%	Q2QXZ2.1
<input checked="" type="checkbox"/>	RecName: Full=BTB/POZ domain and ankyrin repeat-containing protein NH5.1 [Oryza sativa Japonica Group]	121	121	55%	3e-28	28.42%	Q2RAQ5.1

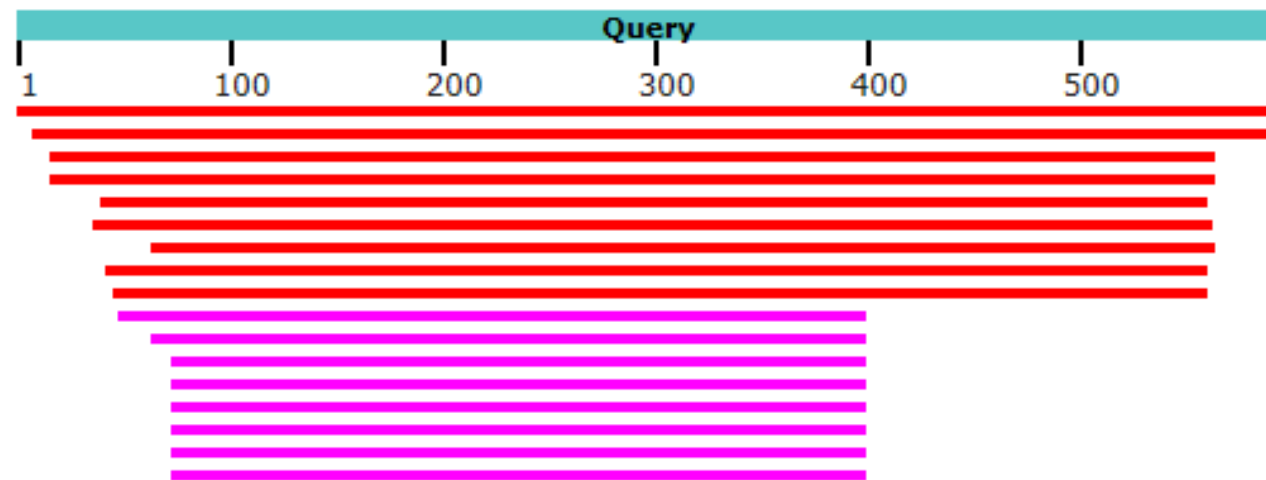
BLASTP

d ?

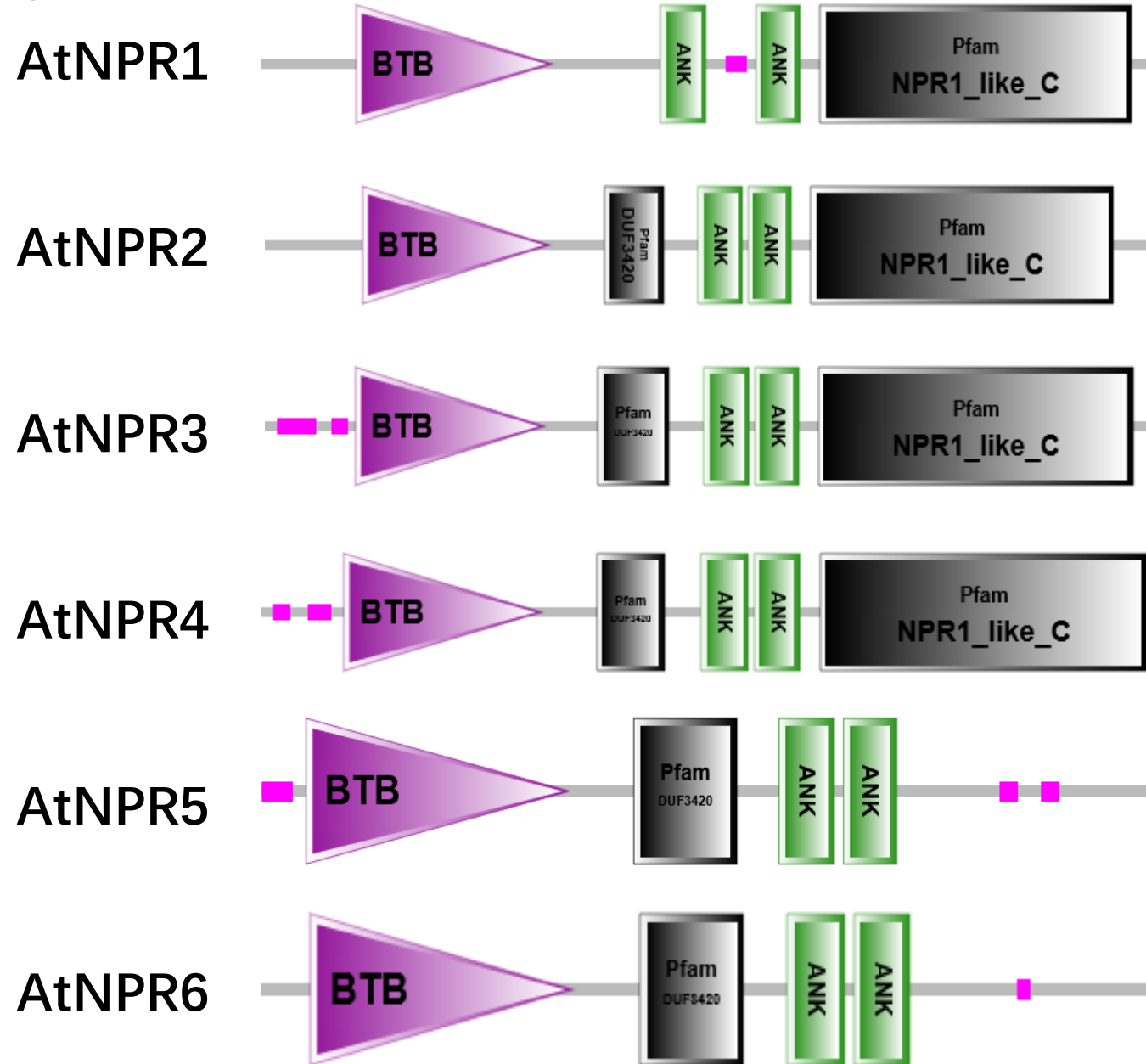
Putative conserved domains have been detected, click on the image below for detailed results.



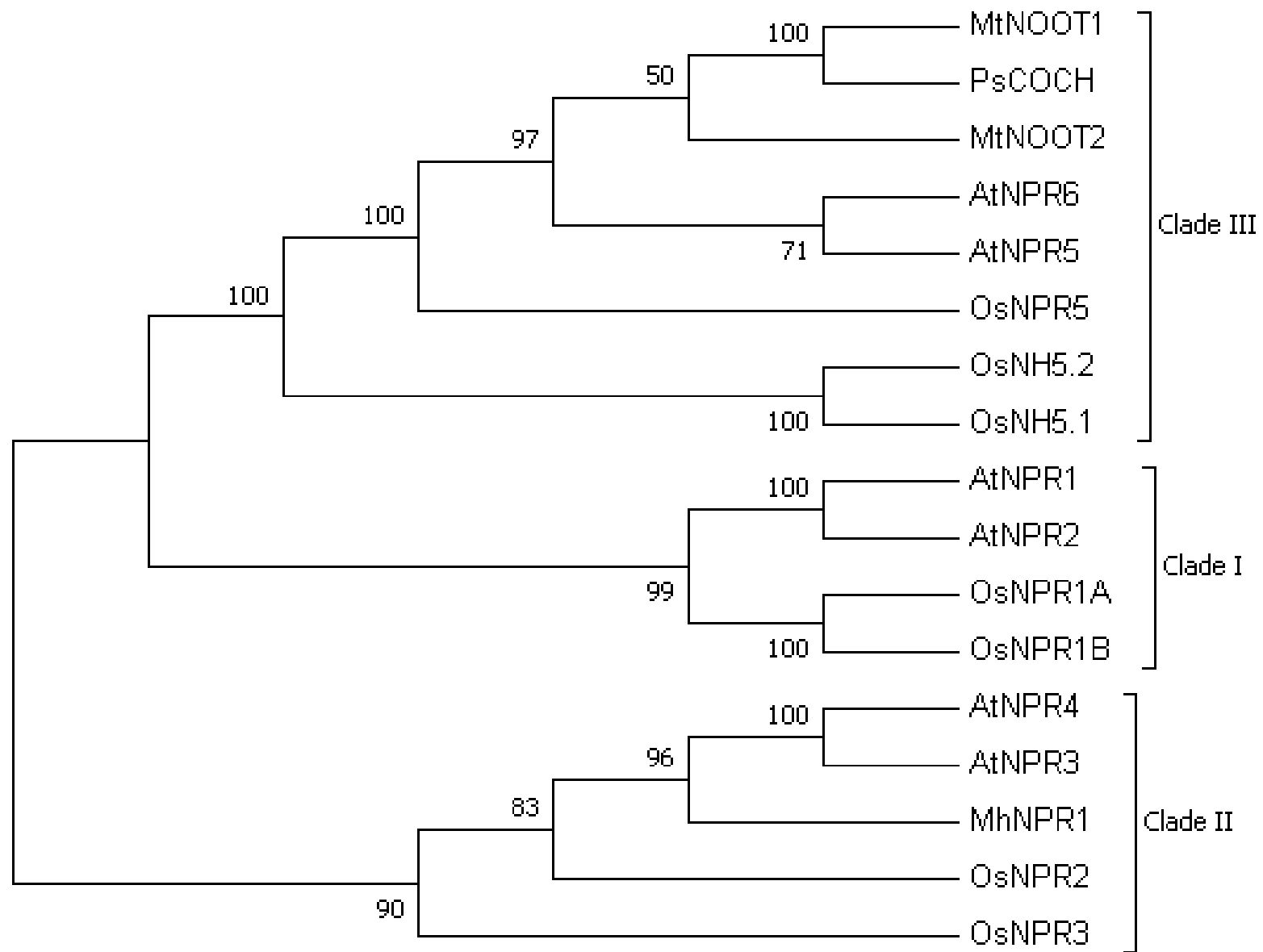
Distribution of the top 17 Blast Hits on 17 subject sequences

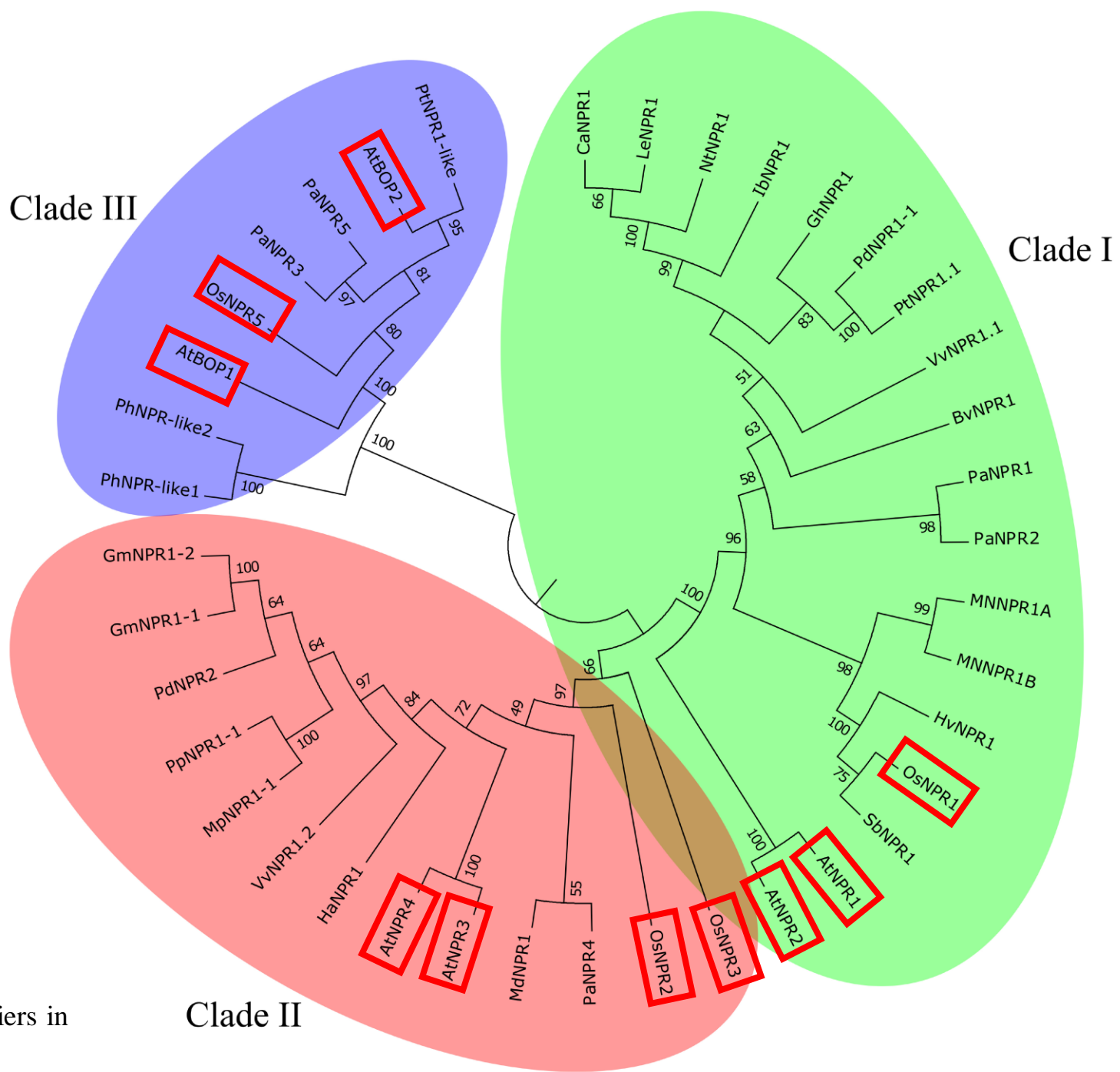


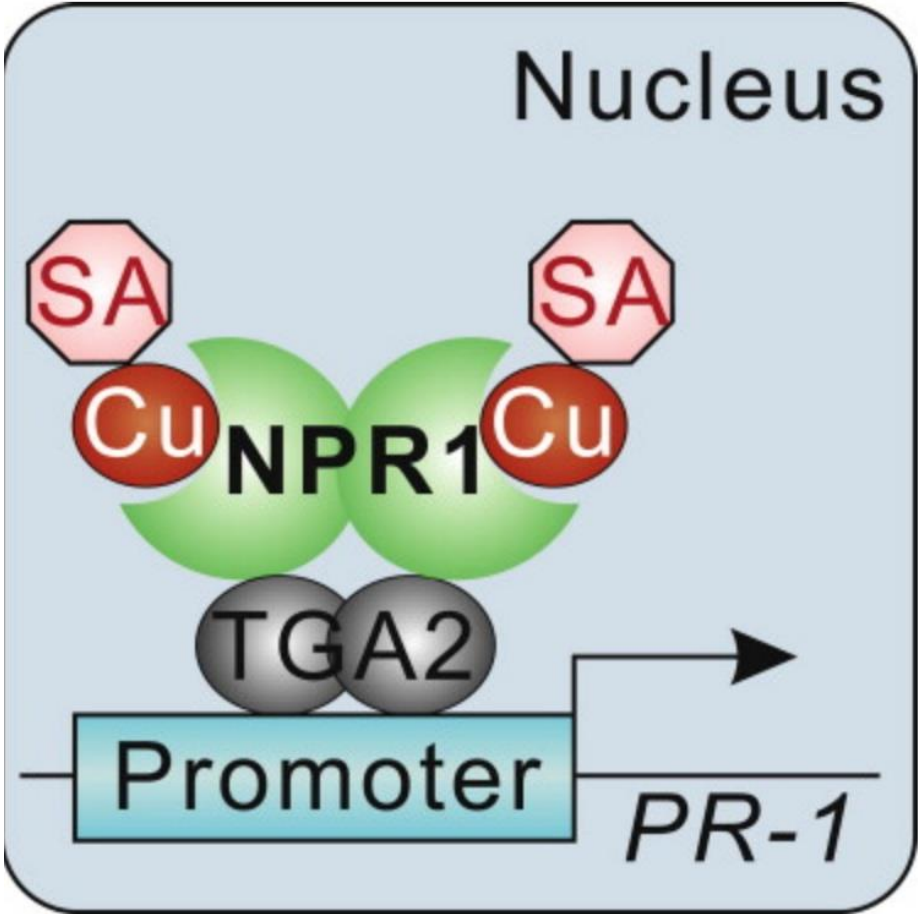
Domain & Motif



MP Tree








C82 & C216 are crucial for oligomerization

Amino acid modifications

Feature key	Position(s)	Description	Actions	Graphical view	Length
Modified residue ⁱ	11	Phosphoserine 1 Publication			1
Modified residue ⁱ	15	Phosphoserine 1 Publication			1
Disulfide bond ⁱ	82	Interchain (with C-216); in linked form			
Modified residue ⁱ	156	S-nitrosocysteine 1 Publication			1
Disulfide bond ⁱ	216	Interchain (with C-82); in linked form			

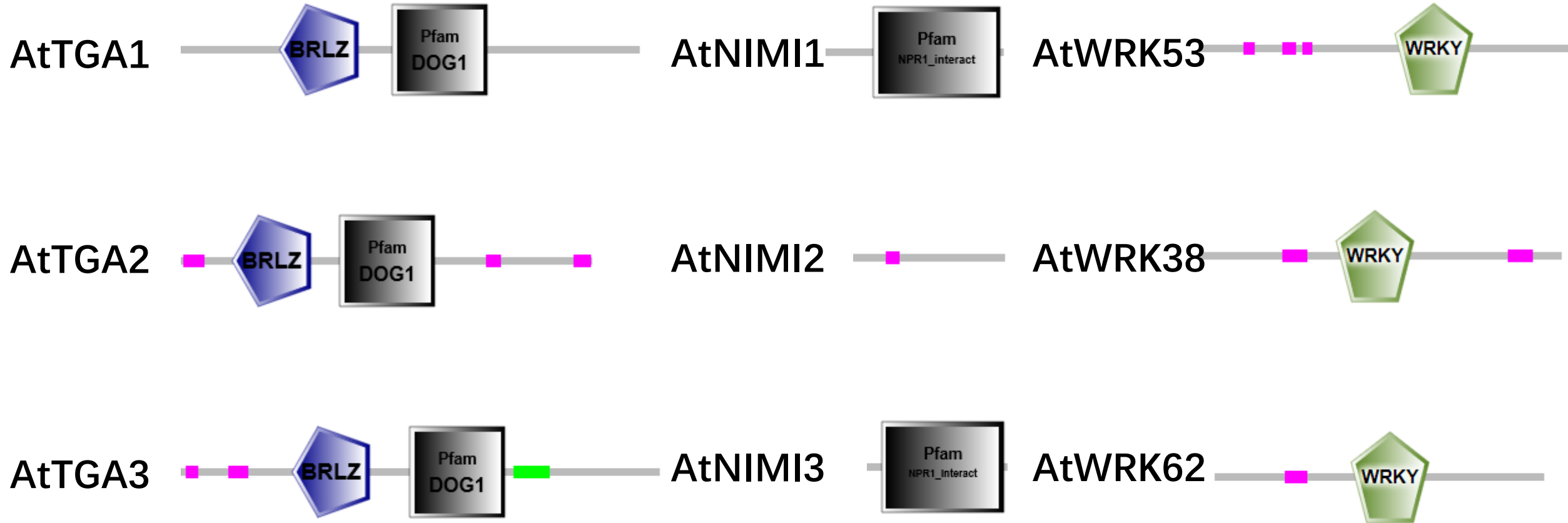
A bipartite NLS in C-terminus

Motif

Feature key	Position(s)	Description	Actions	Graphical view	Length
Motif ¹	537 – 554	Nuclear localization signal Sequence analysis	Add BLAST		18

KRLQKKQRYMEIQETLKK

TGA family & NIMIN family & WRKY family



```

#
# Aligned_sequences: 2
# 1: TGA1_ARATH
# 2: NIMI1_ARATH
# Matrix: EBLOSUM62
# Gap_penalty: 10.0
# Extend_penalty: 0.5
#
# Length: 89
# Identity:      16/89 (18.0%)
# Similarity:   35/89 (39.3%)
# Gaps:         10/89 (11.2%)
# Score: 43.5
#
#=====
TGA1_ARATH      71 MFDQEASTSRHPDKIQRRLAQNREAARKSRLRKKAYVQQLETSRLKLIQL      120
      |...|...|:.....:|:....|.....:      .:.|||:.
NIMI1_ARATH    16 MSKDENVESKETIRVDKRVREDEEEEEKKI-----DTFFKLIKH      55

TGA1_ARATH     121 EQELDRARQQGFYVGNIDTNSLGFSETMNPGLIAAFEME      159
      .||...:|:.....:|:.....|.....:|:..|
NIMI1_ARATH    56 YQEARKRRREELAENSGVRRKSNNGGERSGIVVPAFQPE      94

#-----
#-----

```

```

#=====
#
# Aligned_sequences: 2
# 1: TGA1
# 2: WRKY53
# Matrix: EBLOSUM62
# Gap_penalty: 10.0
# Extend_penalty: 0.5
#
# Length: 18
# Identity:      7/18 (38.9%)
# Similarity:   11/18 (61.1%)
# Gaps:         0/18 ( 0.0%)
# Score: 34.0
#
#
#=====

```

```

TGA1          236 RPSDLLKVLPHFDVLT 253
              :|.|:|:.|...|.||
WRKY53       245 KPKDILESLSNLTVRT 262

```

Structure Prediction by Phyre2

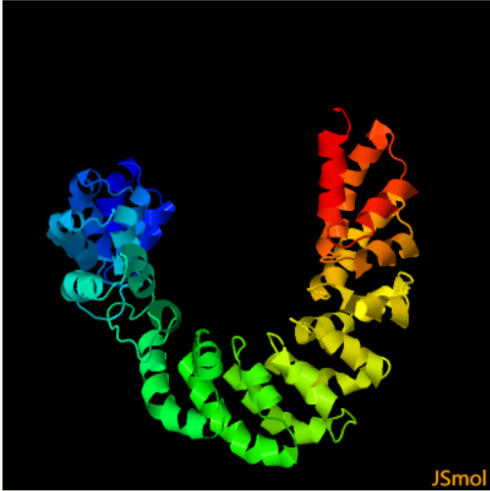
Phyre2

Date	Wed Jan 1 15:04:06 GMT 2020
Unique Job ID	26547dde30b4d1fe
Sequence	MDTTIDGFAD ... Download FASTA
Job Type	normal
Job Expiry	

[Download zip of all results](#)

Summary

Top model



Model (left) based on template [c4cj9A](#)


Top template information

PDB header:transcription
Chain: A: **PDB Molecule:**burrh;
PDBTitle: burrh dna-binding protein from burkholderia rhizoxinica in2 its apo form

Confidence and coverage

Confidence:	100.0%	Coverage:	66%
-------------	---------------	-----------	------------

390 residues (66% of your sequence) have been modelled with 100.0% confidence by the single highest scoring template.



You may wish to submit your sequence to [Phyrealarm](#). This will automatically scan your sequence every week for new potential templates as they appear in the Phyre2 library.

[3D viewing](#)
[Interactive 3D view in JSmol](#)

For other options to view your downloaded structure offline see the [FAQ](#)

Colour by Secondary Structure Colour Rainbow N->C
[Close JSmol](#)













Sequence analysis

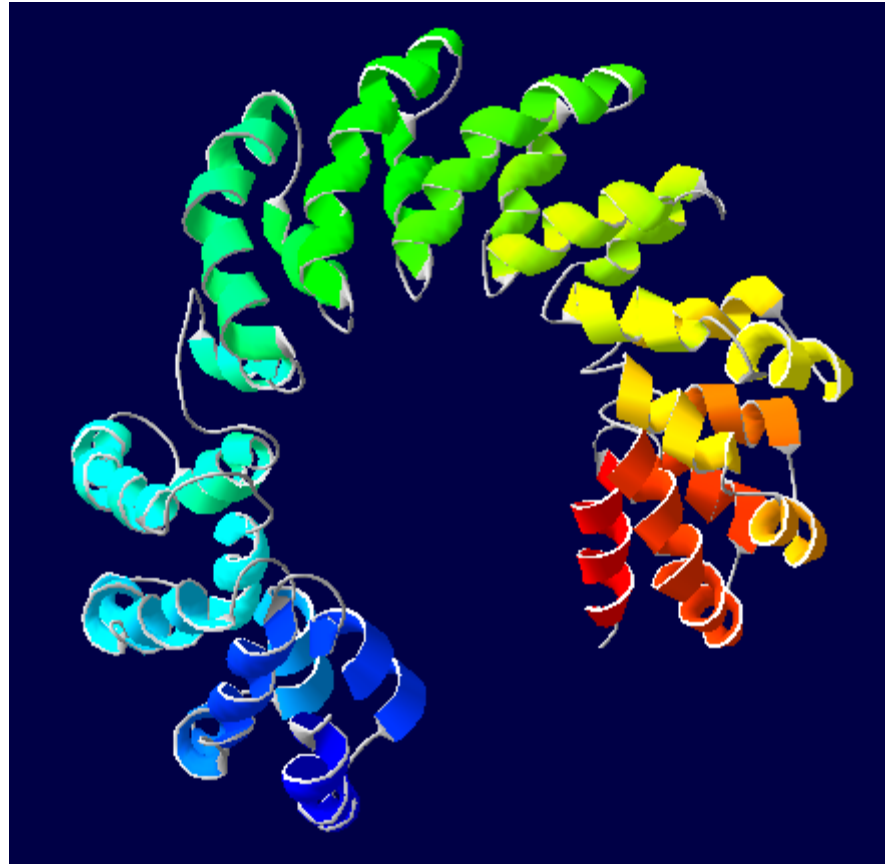
[View PSI-Blast Pseudo-Multiple Sequence Alignment](#) [Download FASTA version](#)

Secondary structure and disorder prediction [Show]

Residue 11-420

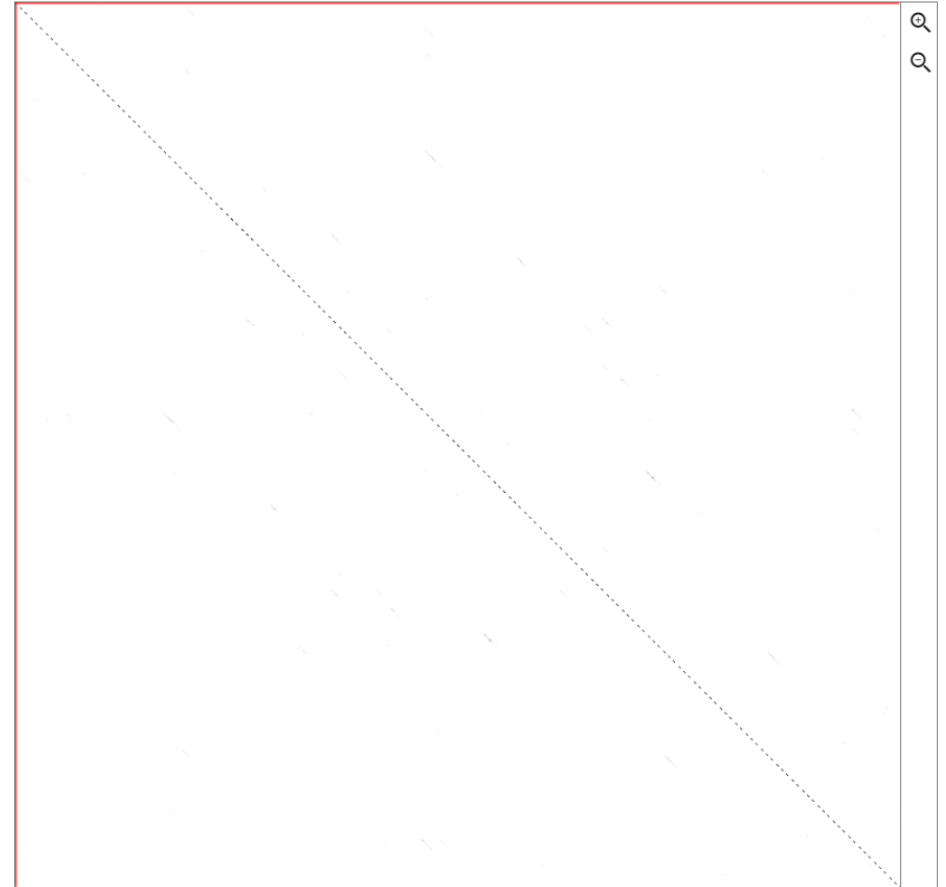
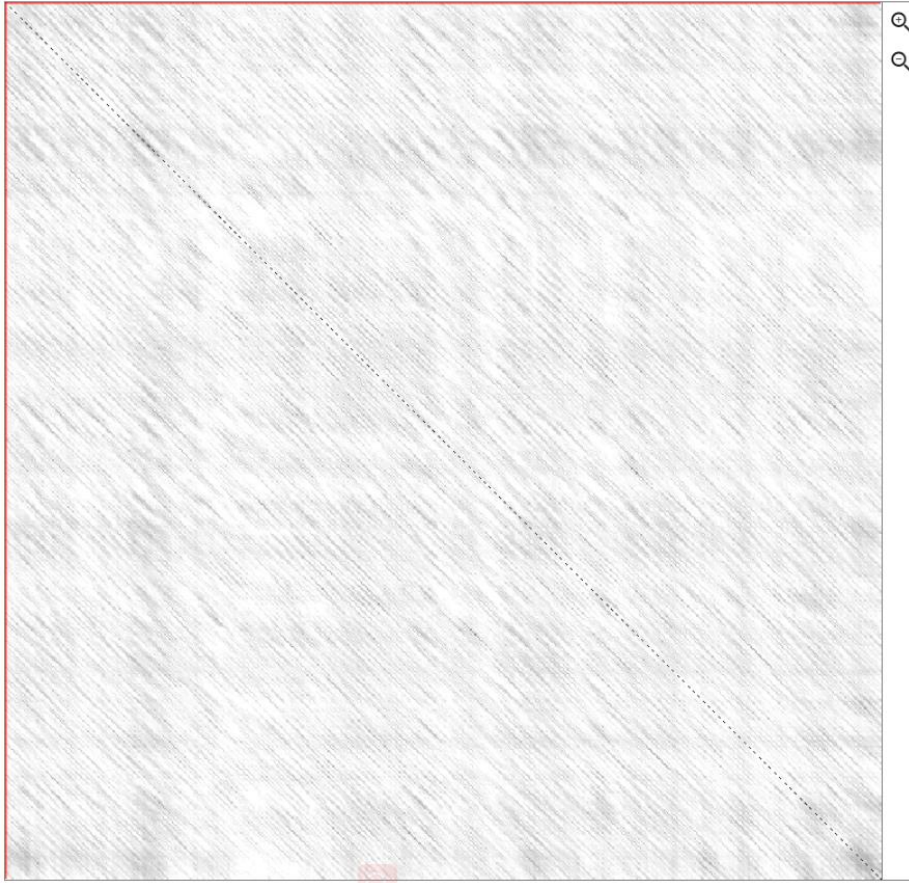
Domains and Repeats

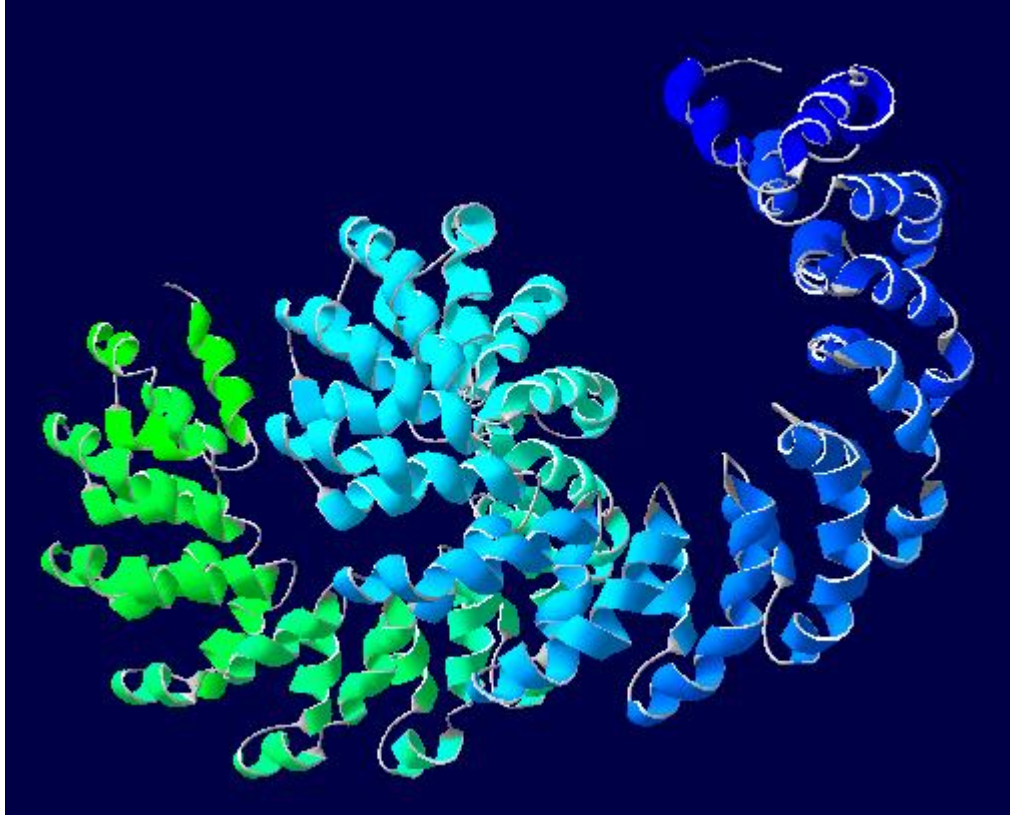
Feature key	Position(s)	Description	Actions	Graphical view	Length
Domain ⁱ	65 – 144	BTB 📌 PROSITE-ProRule annotation ▼	 Add  BLAST		80
Repeat ⁱ	265 – 295	ANK 1	 Add  BLAST		31
Repeat ⁱ	297 – 324	ANK 2	 Add  BLAST		28
Repeat ⁱ	328 – 357	ANK 3	 Add  BLAST		30



Structure predicted by Phyre2

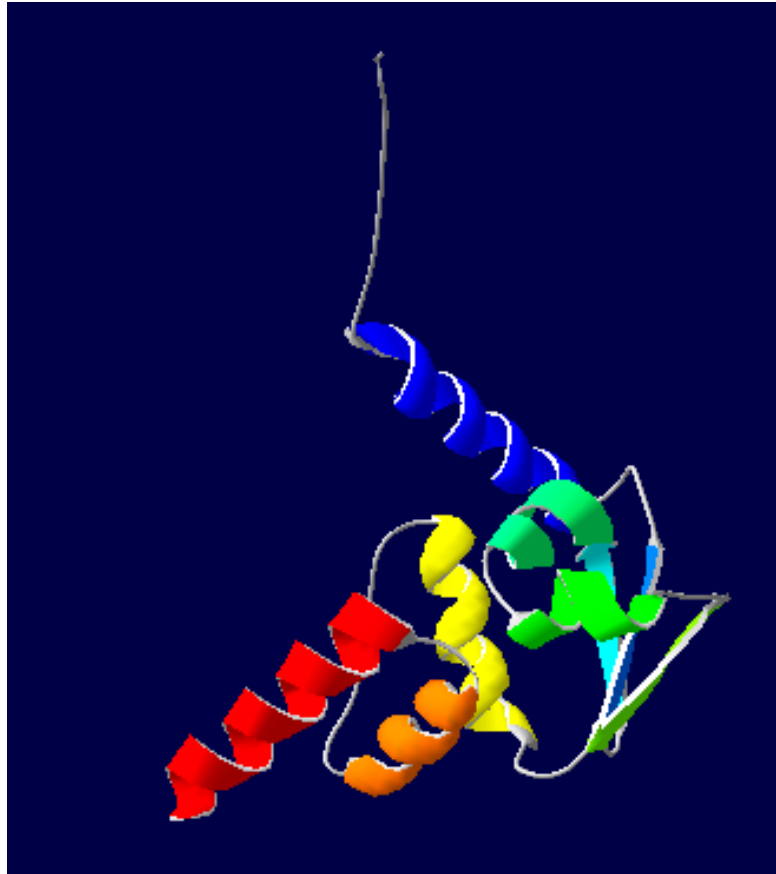
No repeat sequence



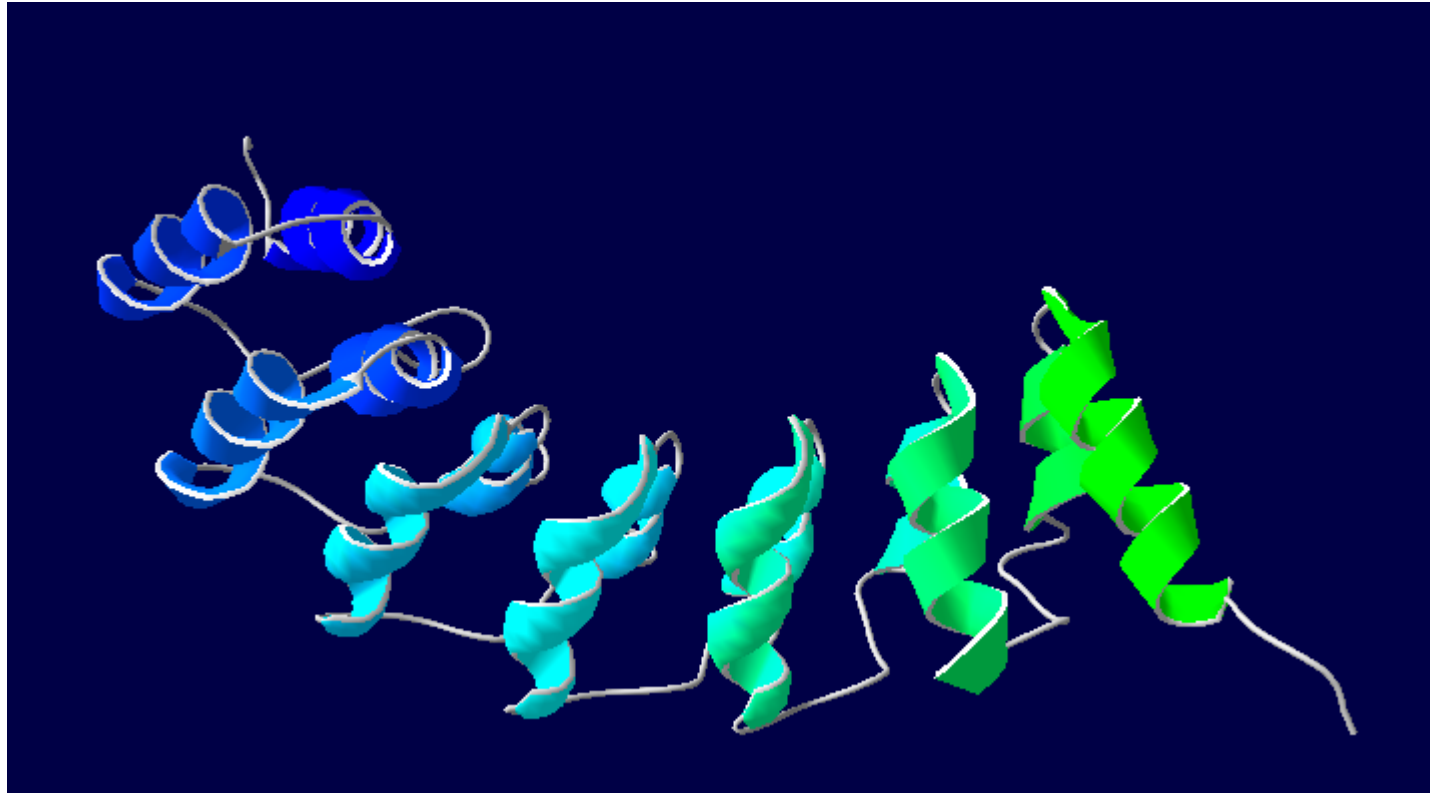


Template: A TAL Effector

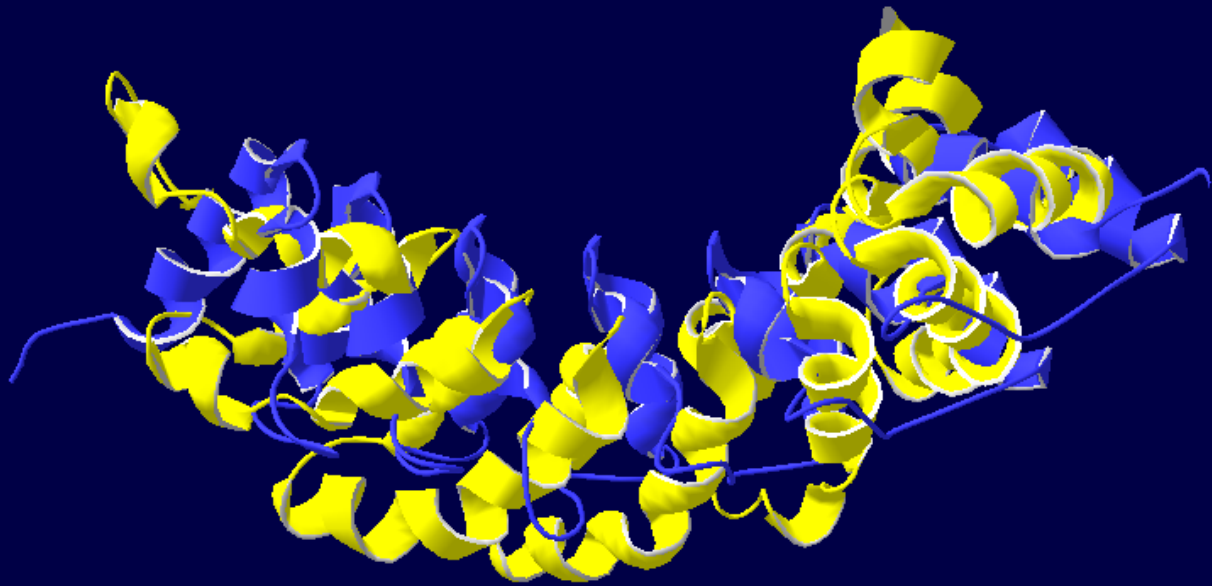




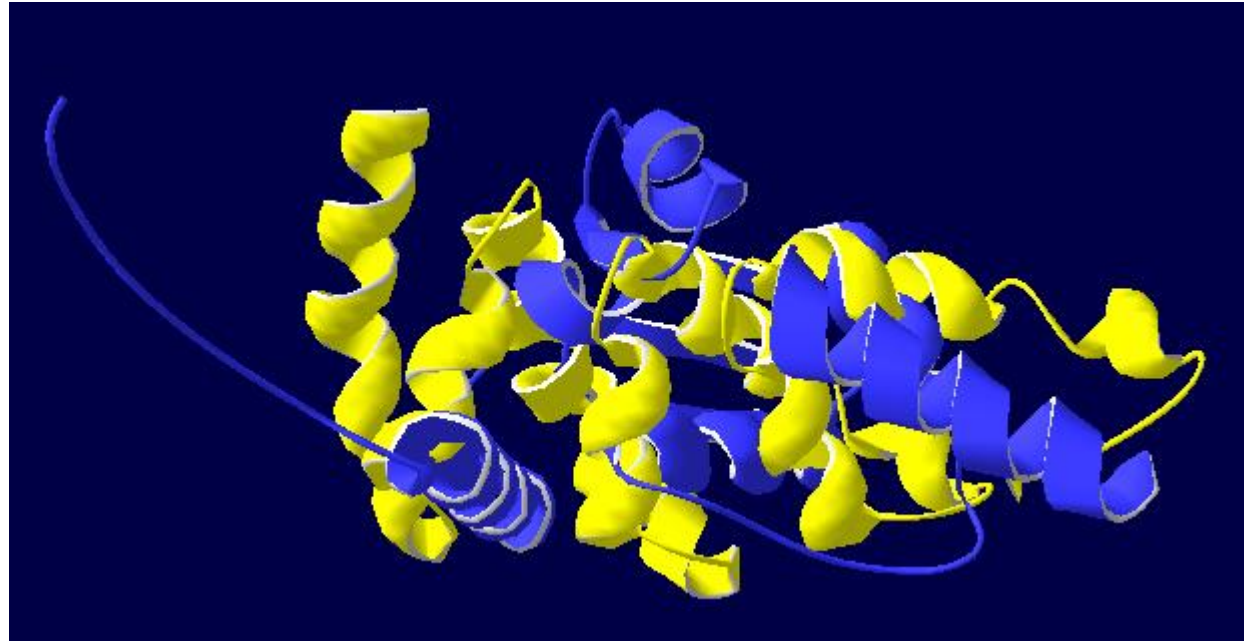
BTB domain from human promyelocytic leukemia zinc finger protein



Ankyrin repeat from 4O60, an artificial ankyrin repeat protein



Fitting TALE repeat & ankyrin repeat



Fitting TALE repeat & BTB domain

Structure Prediction by Swissprot

NPR1 Created: today at 16:27

Summary Templates 50 Models 1

Model Results

Oligo-State: Monomer
Ligands: None
GMQE: 0.31
QMEAN: -6.68

Global Quality Estimate
QMEAN: -6.68
C β : -3.47
All Atom: -3.20
solvation: -1.98
torsion: -5.46

Local Quality Estimate
Local Quality Estimate graph showing Predicted Local Similarity to Target vs Residue Number (180-480).

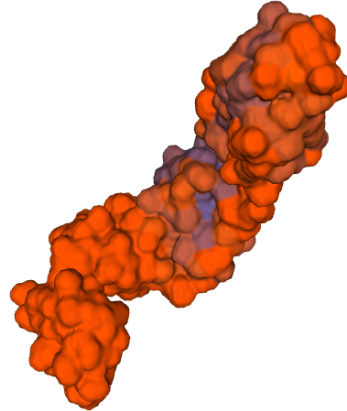
Comparison
Comparison graph showing Predicted Local Similarity to Target vs Protein Size (Residues).

Template: 5y4d.1.A
Seq Identity: 19.23%
Coverage: [Progress bar]
Description: Ankyrin-1, Ankyrin-2, Ankyrin-2

Model-Template Alignment

```
Model_01 MDTTIDGFADSYEISSTSFVATDNTDSSIVYLAAEQVLTGPDVSAQLQLLSNFSFVDFSDPDDFYSDAKLVLSGDRVSVFHRVCLVSARSSFFKSAALAAKKEKDSNNTAAVKLEKEIAKDYEVGFDSDVVT 130
5y4d.1.A
Model_01 VLAVVYSSRRVPPPKGVSECADENCCHVACRPVDFMLEVLYLAFIFKIPELITLYQRHLLDQVVDKVVIEDLVLILKLANICGKACMKLLDRCKEIVKSNVDMVSLKSLPEELVKEIIDRRKELGLEV 260
5y4d.1.A LHLAARESHVGLVQELLSGSSVDSATKRGNTLHIASLDCAEVVKVLRVCGANINACSQNGFTLYMRAQENHEDVVEYLENGANQ
Model_01 PKVKKHYSNVHKALDSDDIELVKLLKEDH-----TNLDDACALHFAYVGNVKTATDLLKLDLADVNRNPRGYTVLHVAAAMRKEFPQLILSL 348
5y4d.1.A STATEDGFTLAVALCOHQAVAILLNDTKGKVLFPALHIAARRDCKSAALLLQNDHNADVQSKMIVNVTESGFTLHIAAREYNNVATILLRGAAVDFTRNGFTLHVASERENNMVKLL 323
Model_01 LEKGASASEATLEGRALMIKQA-----TMAVECNNIPEQCKHSLK--GRLCVEILEQEDKRE-----QIPRDVPPSPFAVAADELKMTLI-----DLENRVLAQ 437
5y4d.1.A LIRGGQIDAKTRDGLPLHCAARSGHQVVELLRGAPLLAATKNGLSLHMAASDHVECVKHELLHRAFPVDDVTLDYLLALHVAAACGHQVTELELLKRRANPNARALNGFTLHIAACNRRQVME 453
Model_01 RLFPTPE-----AQAAMEIAEMKGTCEFIVTSLPDRDLTGTKRTSPGVKIAAPPRILEEHQSRRLRALSKTVELGKRRFFRCSAVLDQIMNCEDLTQLACGEDDTEAKRLQKQRYMEIQETLKKAFS 557
5y4d.1.A LRVYGASIQAITESGLTTHVAAAGHNVIVLLLC--N--GASFPDVTNIRGETLHMA----- 509
Model_01 EDNLELGNSSLLTDSSTSKSTGKRSNRKLSHRRR 593
5y4d.1.A
```

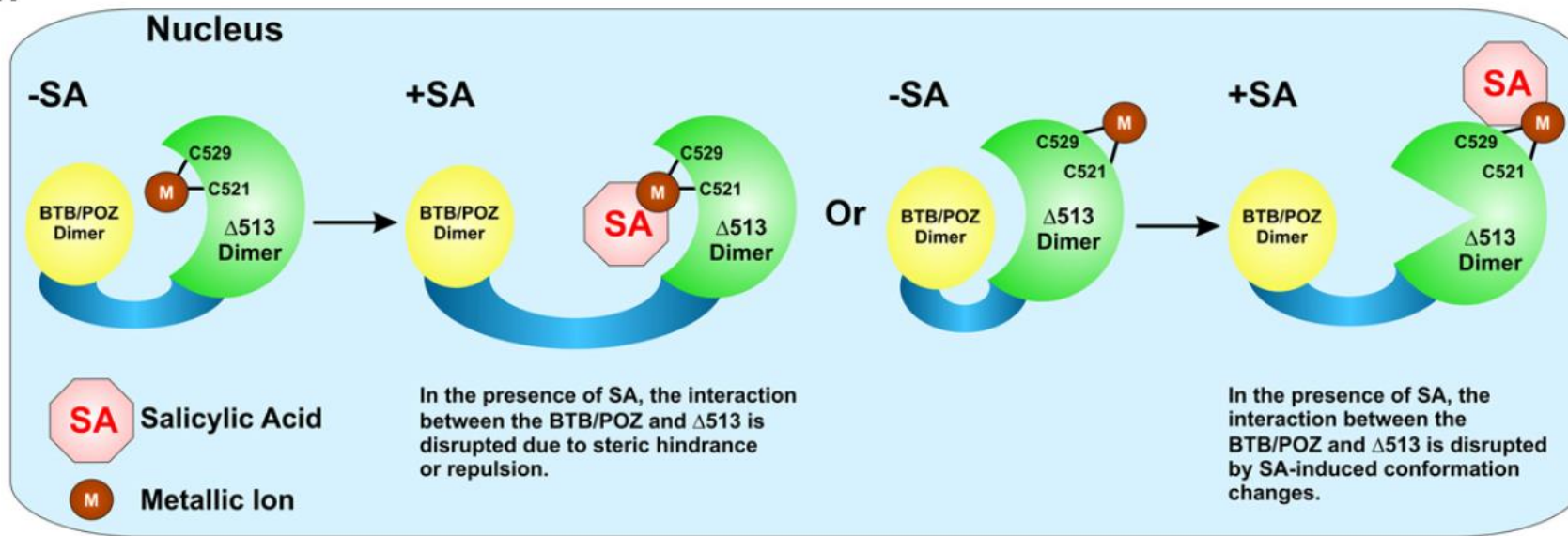
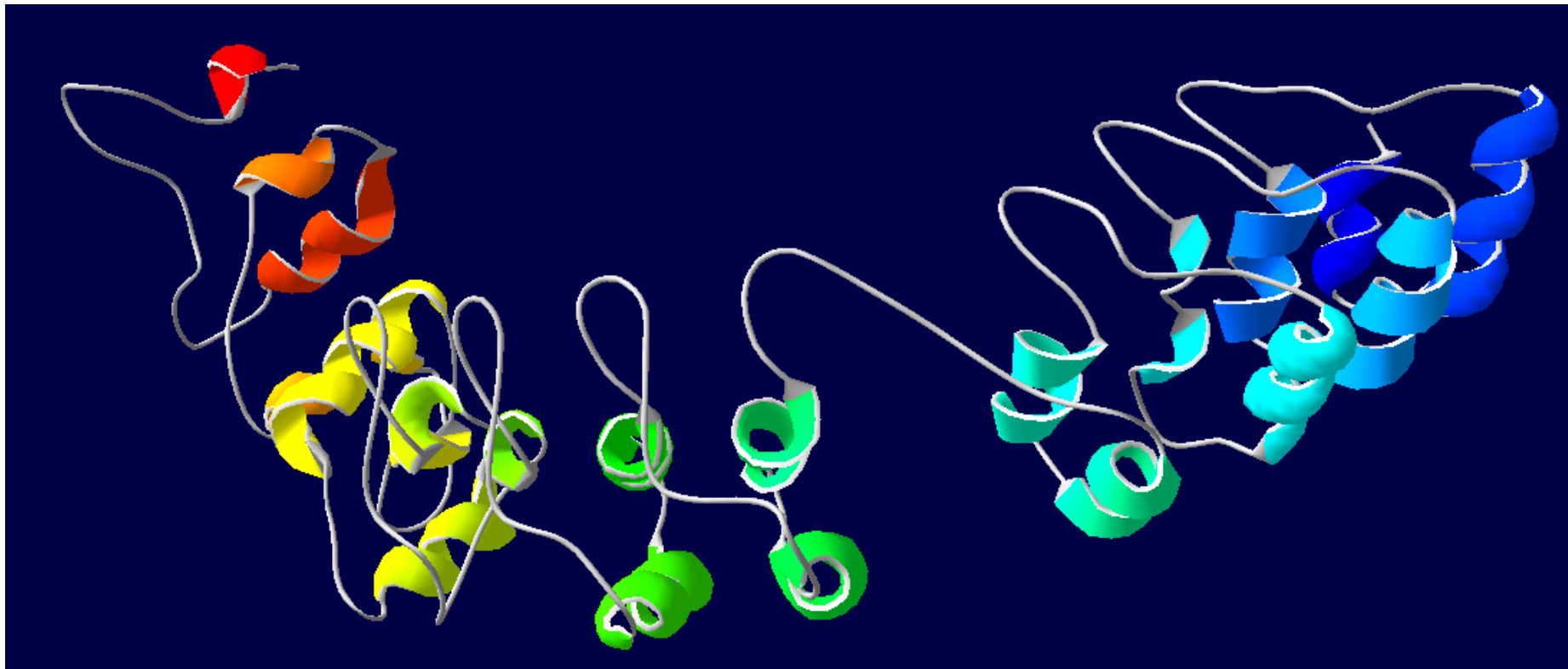
FastStone Capture



NGL Surface [Icons]

1 593

Residue 170-487



Conclusion

- 拟南芥中的NPR家族，根据序列和功能，可分为3个亚家族
- NPR蛋白C端的极性或负电残基参与SA的结合
- NPR1的S82和S216间的二硫键导致寡聚化
- NPR1与TGA蛋白、WRKY蛋白的结合，可能存在共同的机制
- NPR1目前能可靠预测的区域，只有中间的ankyrin repeat

谢谢大家