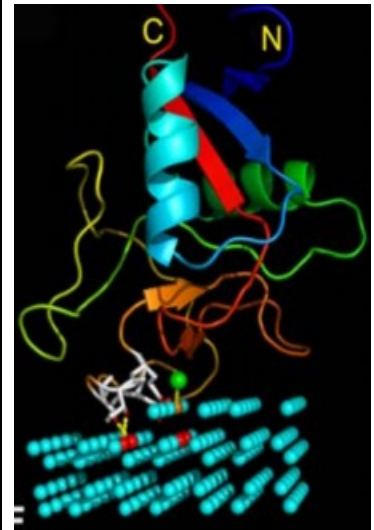
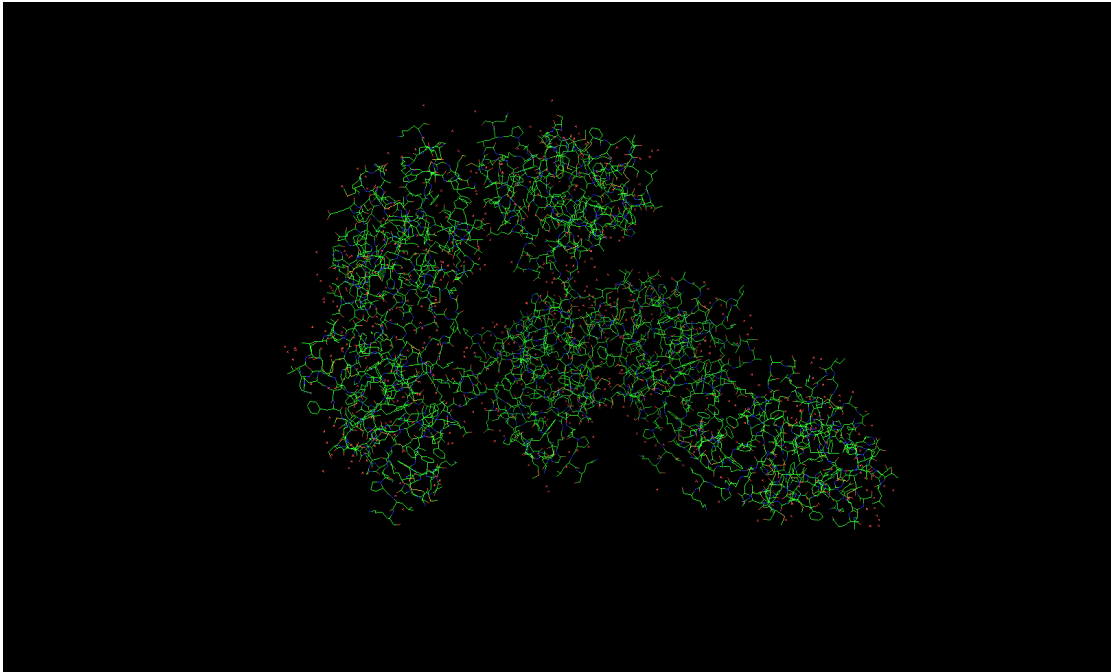


Weekly Report: June 24 -June 28

-Vatsa Shah

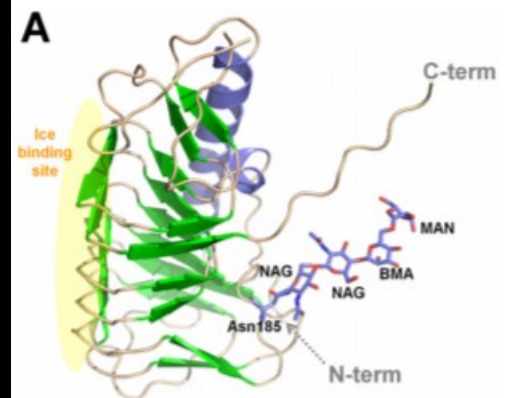
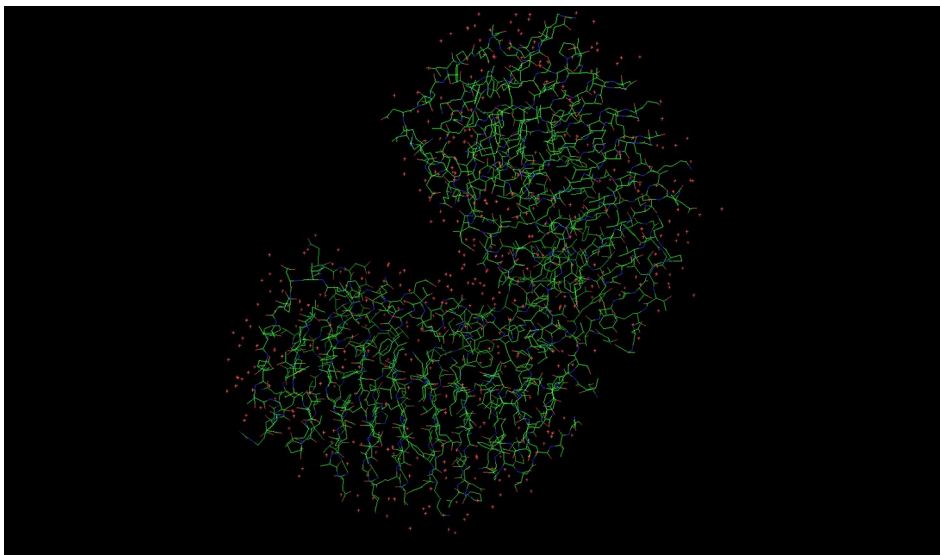
Learned some Basic properties of following AFP's and IBP's ->

1) 2PY2 / herringAFP / hAFP->



Above left image consist of 6 clusters of 2PY2 Protein all assymmetric in structure. Each cluster has one Ca^{+2} ligand which forms co-ordinate bond with water molecule of ice lattice. It Attaches to prism plane of lattice ice. Above right image shows one chain of hAFP binding to prism plane of ice. Ice binding site includes Asp94, Thr96, Thr98, and Glu99. When hAFP binds to ice, constrained water molecules are released (shown by “+” sign).

2) 3UYU / LeIBP ->



Above Left image consist of two chains of 3UYU protein.

Above right image shows IBS of LeIBP.

LeIBP is a member of a large class of ice-binding proteins.

Each chain has Glycerol (GOL) and Selenomethionine (MSE) as Ligands.

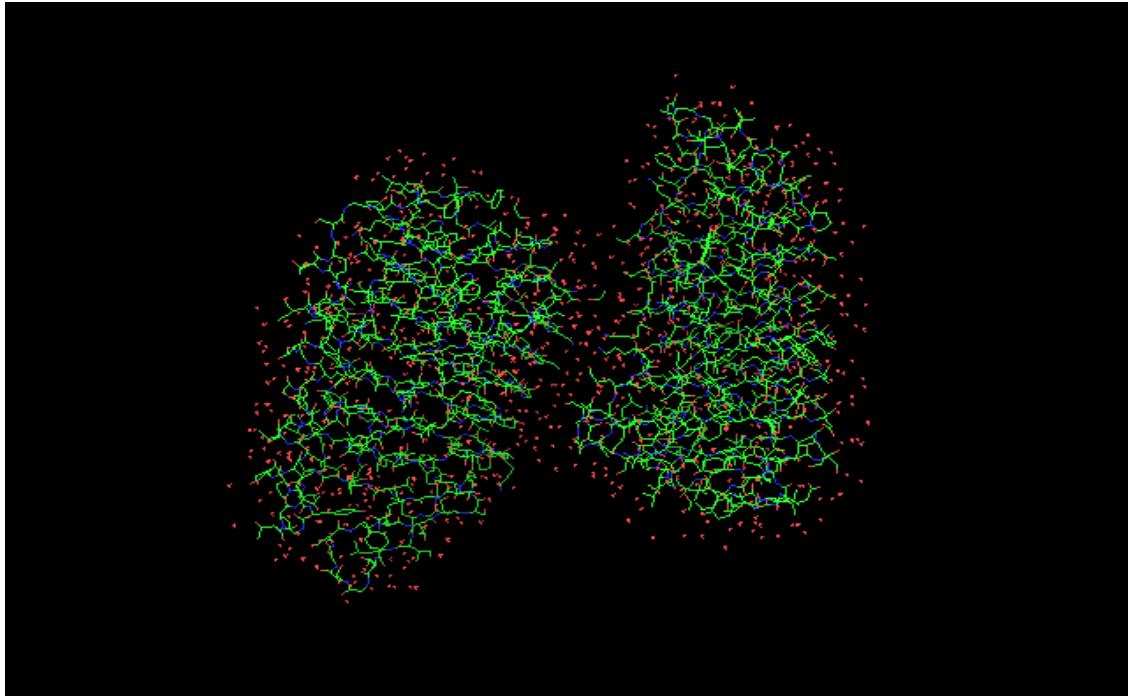
It has Cyclic structure.

LeIBPs has dimeric right-handed β -helix fold, composed of three parts: a large coiled structural domain, a long helix region and a C-terminal hydrophobic loop region (243 PFVPAPEVV 251).

Aligned Thr/Ser/Ala residues are critical for ice binding within the Beta face of LeIBP.

Ice binding sites are Ser43, Thr65, Ser147, Ser171, Thr198, Thr216, and Ala234.

3) 3VN3 / TisAFP6->



Above image shows two chains of TisAFP6.

Each chain has Ethylene glycol (EDO) and 3-Iodo-Tyrosine (IYR) as Ligands.

It is an assymmetric Protein.

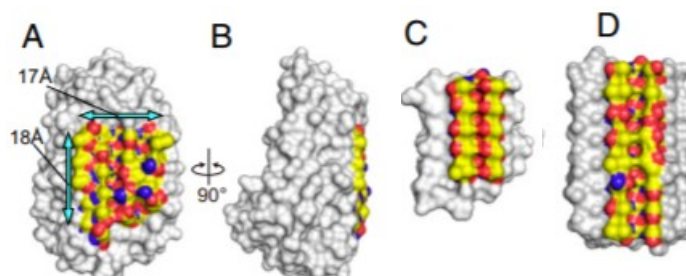
TisAFP6 forms a 52-Å-long semipear-shaped structure, whose principle constituent is a six-loop right-handed β -helix having a triangular cross-section.

TisAFP6 attaches to the basal plane of ice (β -sheet of the b-face).

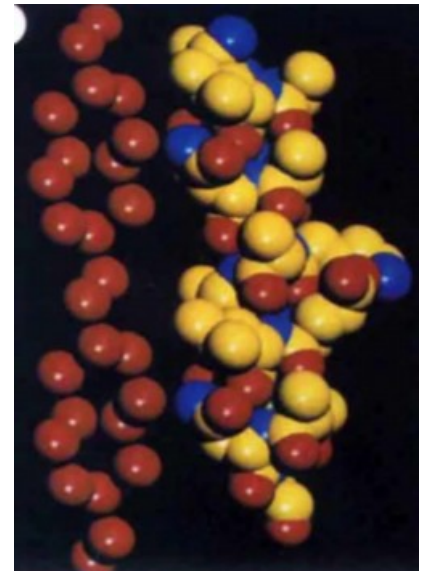
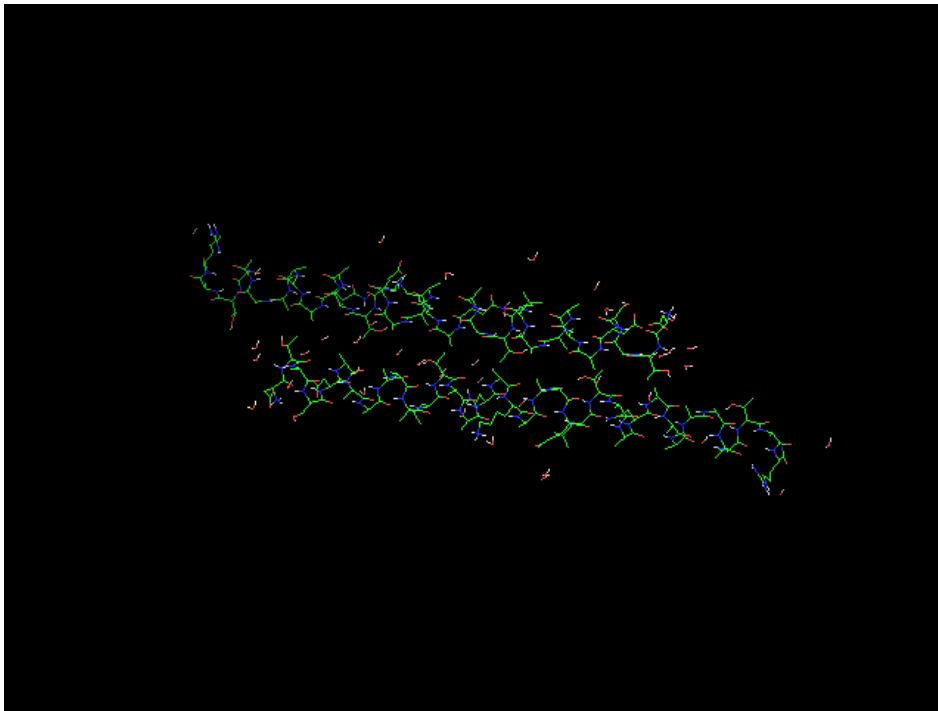
The ice-binding β -sheet of TisAFP6 is assembled from a set of 4- to 5-residue peptides located in the middle of five parallel β -strands; T20-G-V-S-T-V25 (β 1), T211-S-V-A-L-Q216 (β 6), T193-A-V-T-F-K198 (β 5), Q175-A-V-N-T-E180 (β 4), and G148-T-L-DV152 (β 3).

Below image highlights IBS of TisAFP6.

TisAFP6 shows TH activity of $\sim 0.3^\circ\text{C}$ at 0.11 mM.

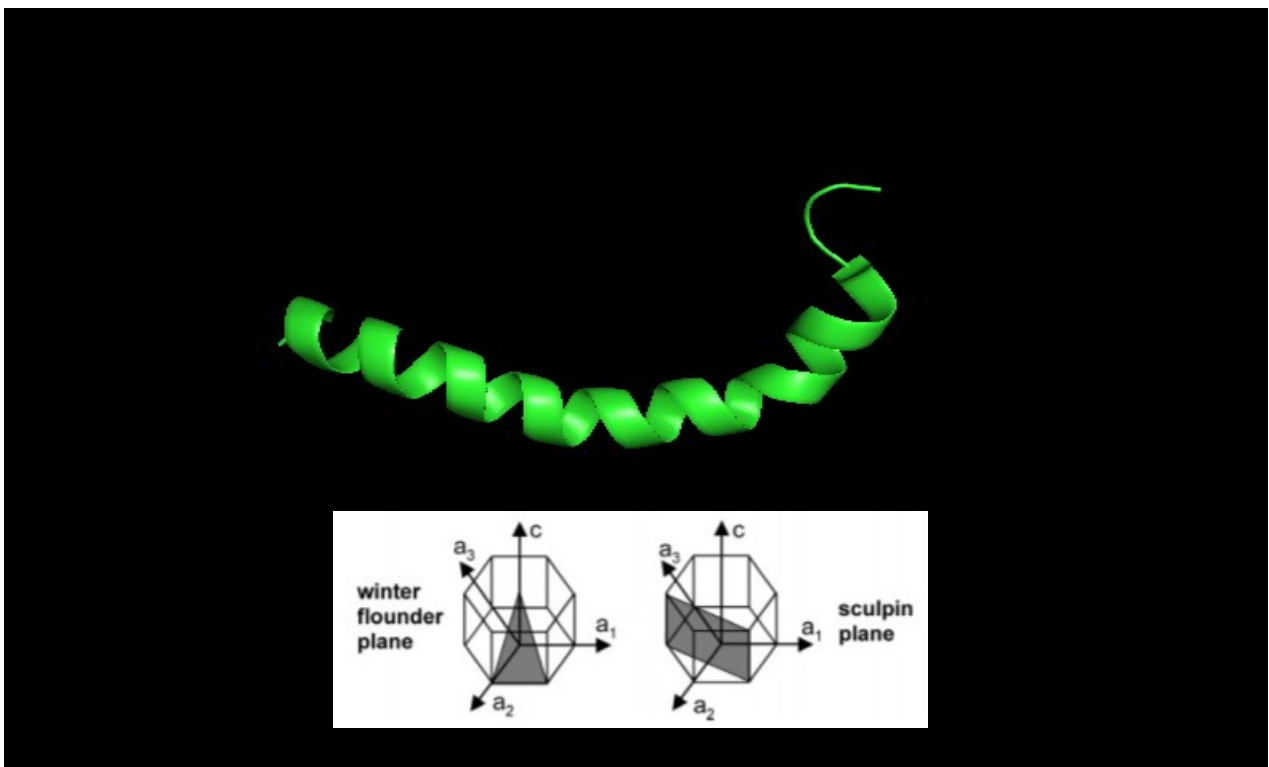


4) 1WFA / Winter Flounder AFP / Type I AFP->



Above Left image consist of two alpha-helical chain of 1wfa protein each having one NH₂ group residue. It is an assymetric Antifreeze protein. Residues 9 to 28 are major ice binding site.
Above Right image shows IBS of AFP (left surface of protein). Red dots represent water molecules. Binding specificity of the protein is along {0112} axes of the {2021} ice planes.

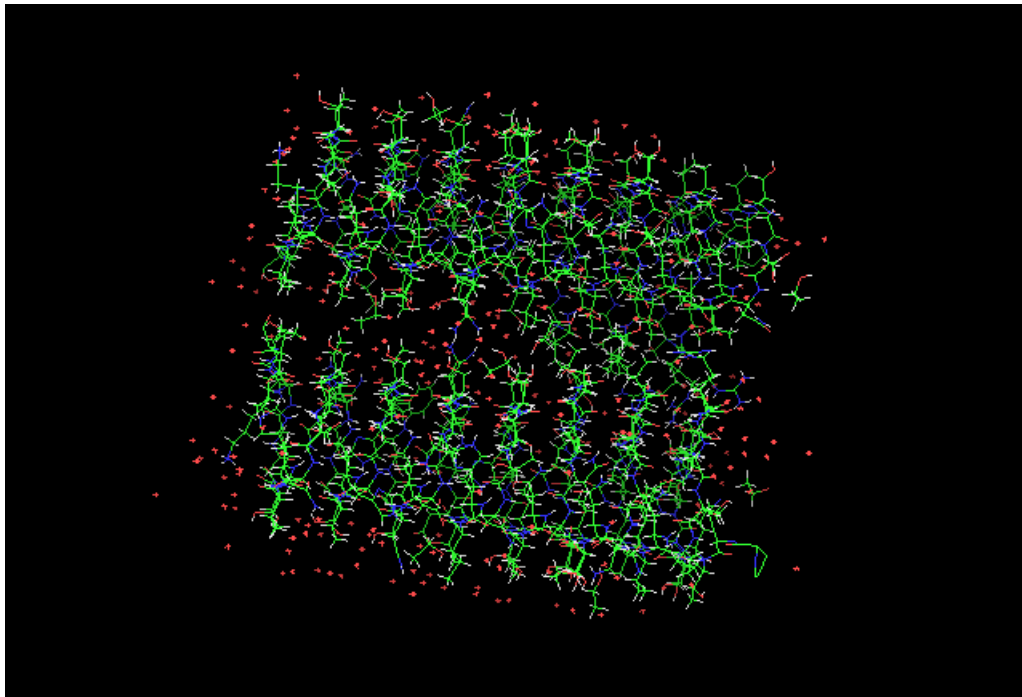
5) 1Y04 / Sculpin AFP->



This Protein accumulates at {2110} ice planes.

Contains an unusual sequence of N-terminal residues, together with two of the 11-residue repeats (residues 11-20 and 21-32) that are characteristic of the type I winter flounder AFP.

6) 3ULT / LpIBP ->

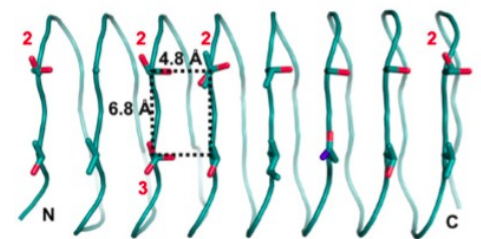


Above image consist of 2 Beta-helical chain of LpIBP.

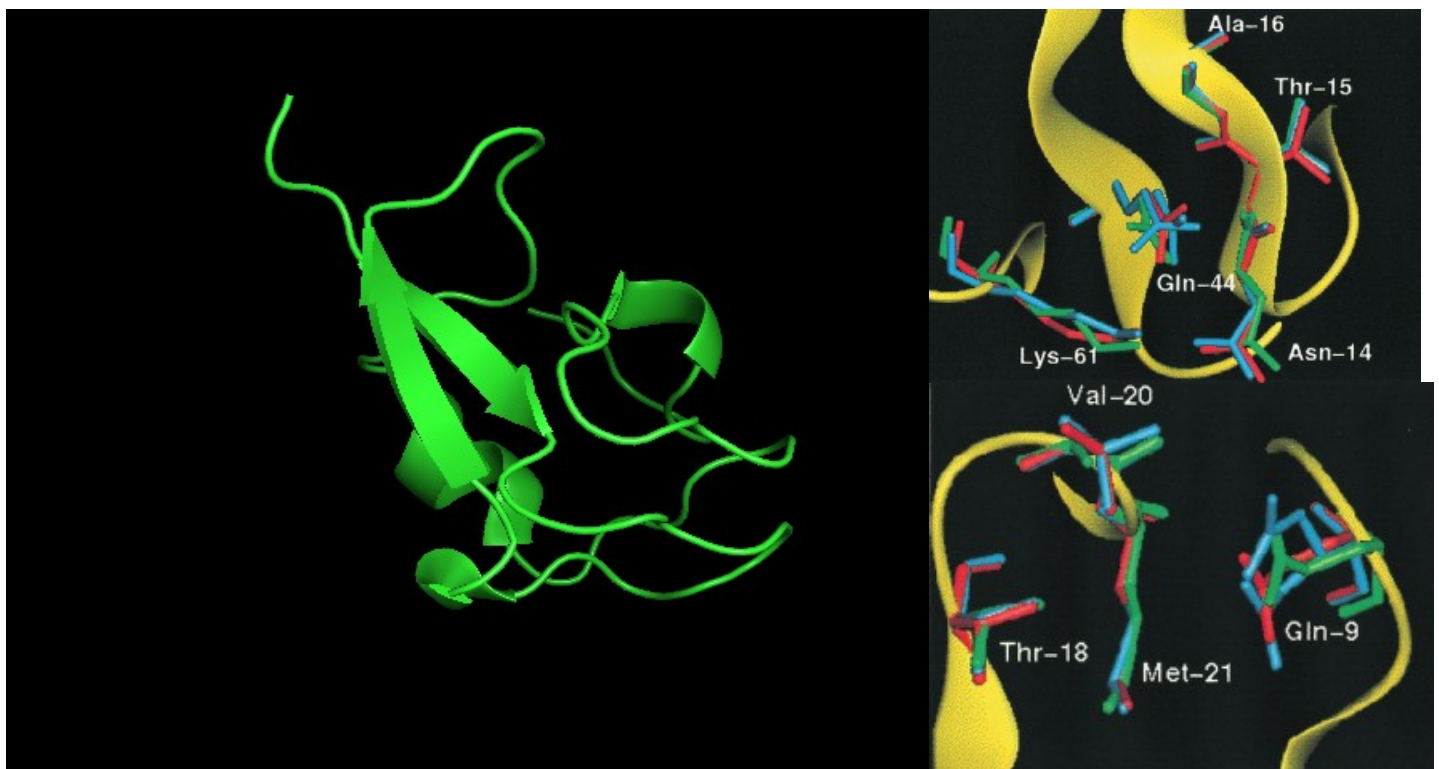
It has Ethylene glycol (EDO) and Ethanol (EOH) as Ligands.

LpIBP binds to the basal and primary-prism planes of ice.

Right side image show's the Ice binding site.(Red sticks are IBS).

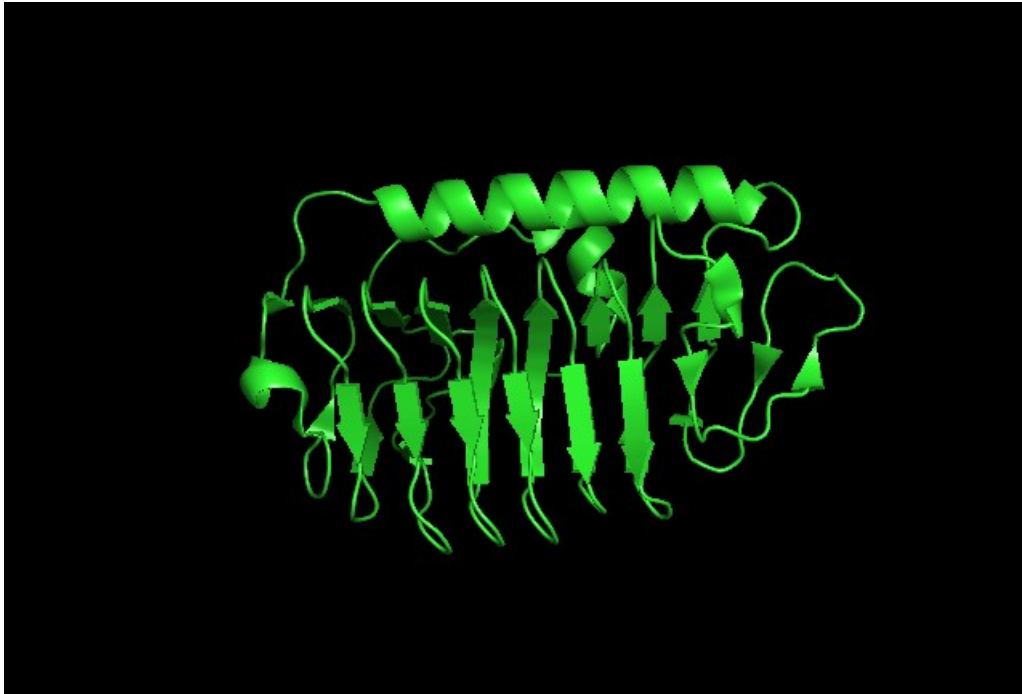


7) 1HG7 / Type III AFP->



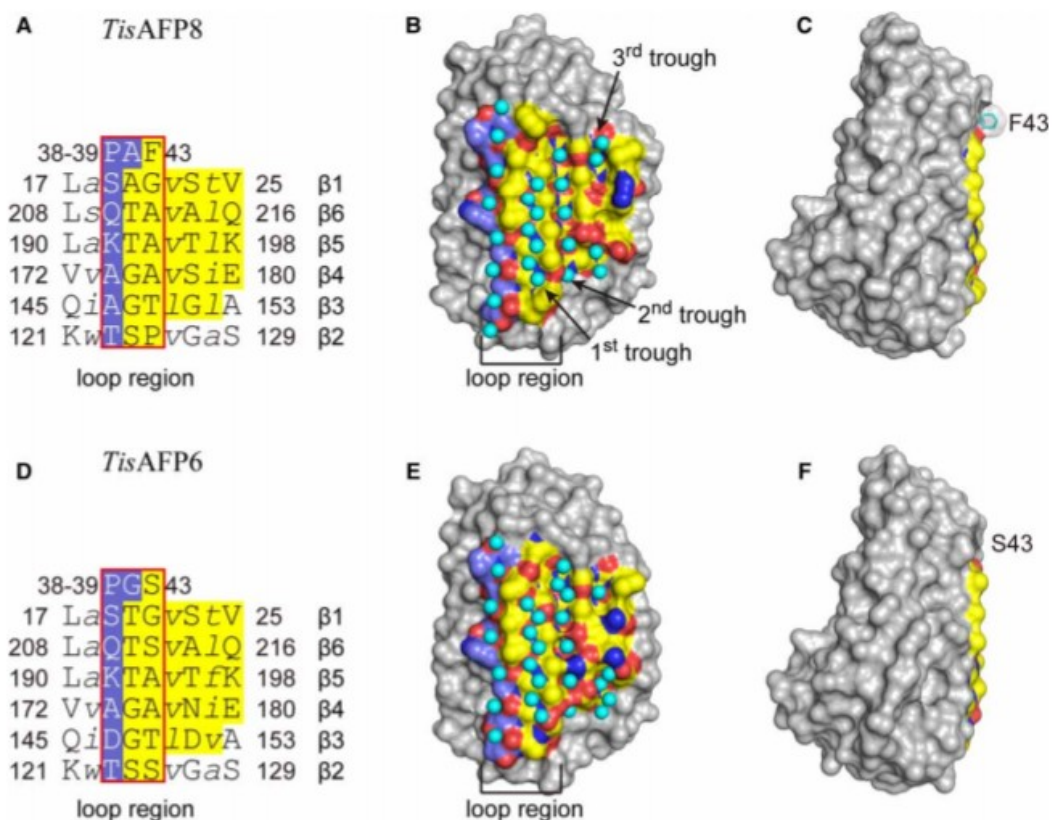
It has sulphate ion as Ligand. The residues Gln9, Thr15, Ala16, Thr18 and Gln44 of 1HG7 interact with water molecules from the (1 0 1 0) prism plane of the ice crystal and residue Asn14 interacts with an advanced water molecule in the conjunction region between the prism and basal (0 0 0 1) planes of the ice. Energetically favourable interactions of this protein with ice could be formed with several surface of ice crystal, with the lowest energy calculated for the (2 0 2 1) pyramidal ice plane. Above left image shows structure of protein. Above right image shows residues binding to ice.

8) 5B5H / TisAFP8->



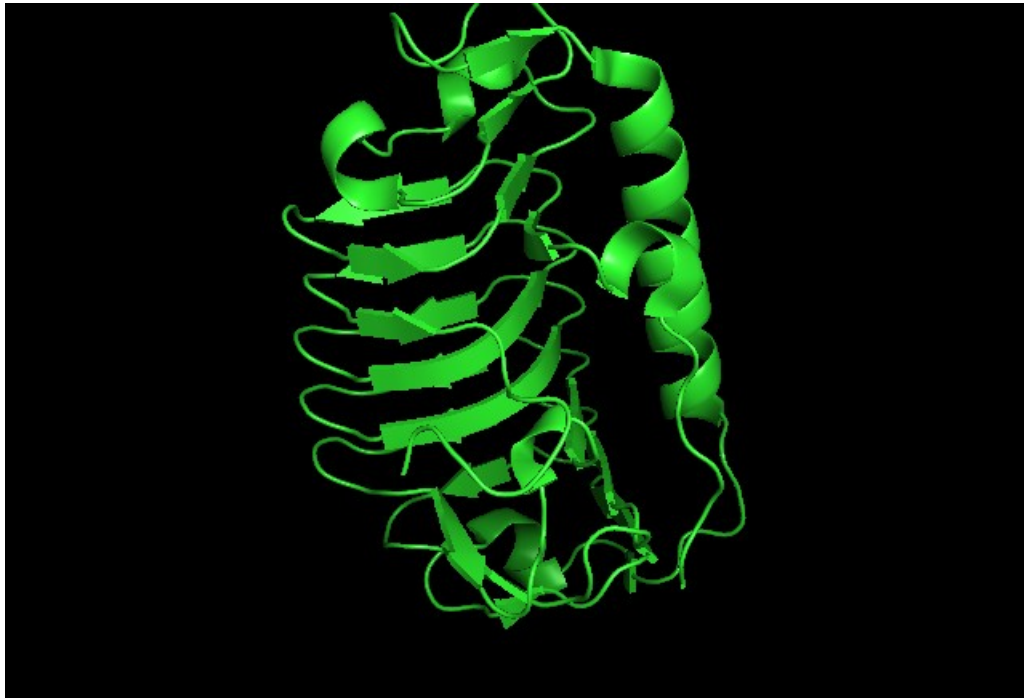
It has sodium ion (NA) and sulphate ion (SO₄) as ligands.

TisAFP8 exhibit's a TH activity of 2.0°C at 0.11 mM, which is almost 10-fold higher than that of TisAFP6 at the same concentration. The morphology and TH activity indicates that TisAFP8 is a member of hyperactive AFPs and possesses basal plane affinity, which is a hallmark of hyperactive AFPs.

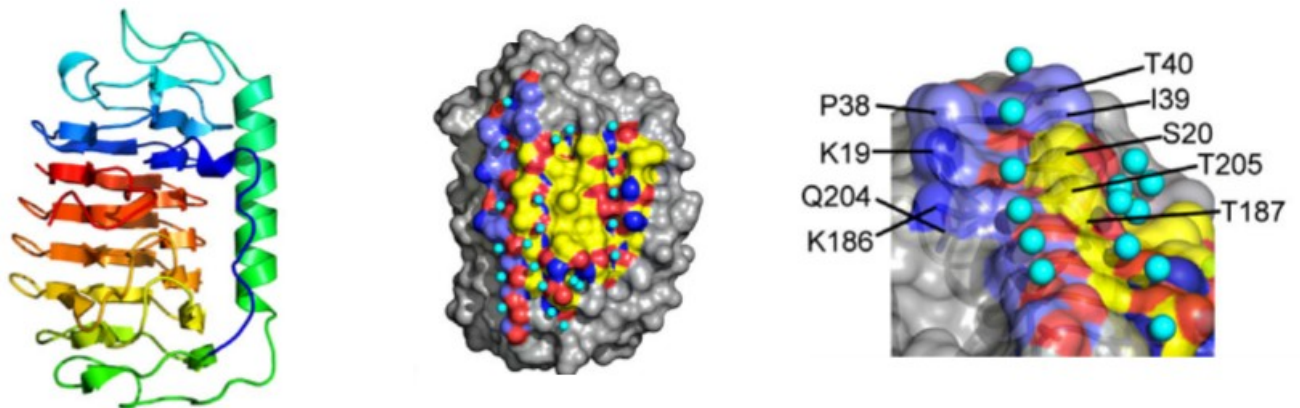


Above figure shows the difference between IBS of TisAFP6 and TisAFP8.

9) 3WP9 / ColAFP->



ColAFP binds to multiple planes of ice, including the basal plane. ColAFP binds to ice through a compound ice-binding site (IBS) located at a flat surface of the beta-helix and the adjoining loop region. ColAFP exhibited thermal hysteresis activity of approximately 4 °C at a concentration of 0.14 mM, and induced rapid growth of ice crystals in the hexagonal direction.

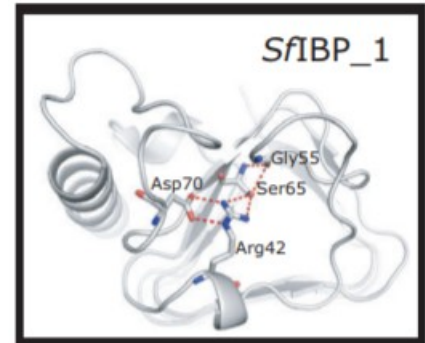
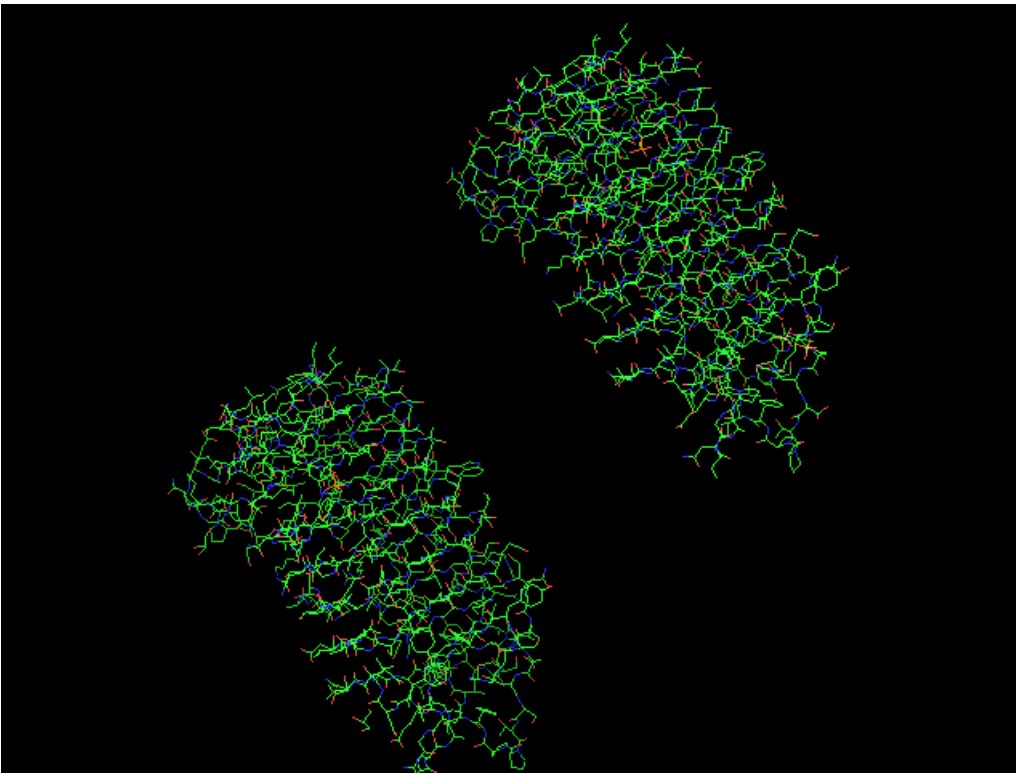


Above left image shows structure of ColAFP.

Above middle image shows IBS of ColAFP.

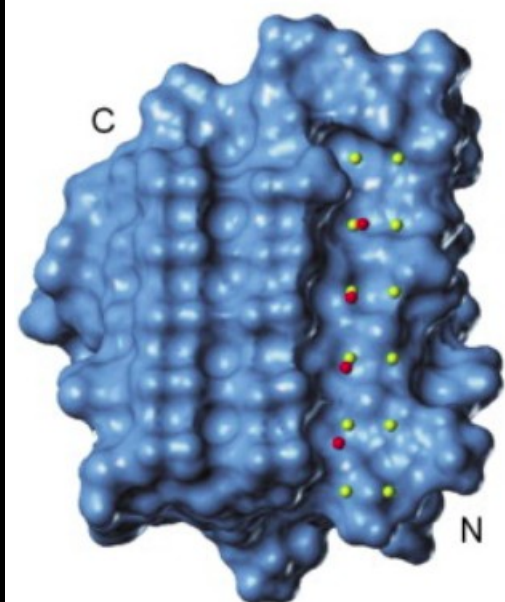
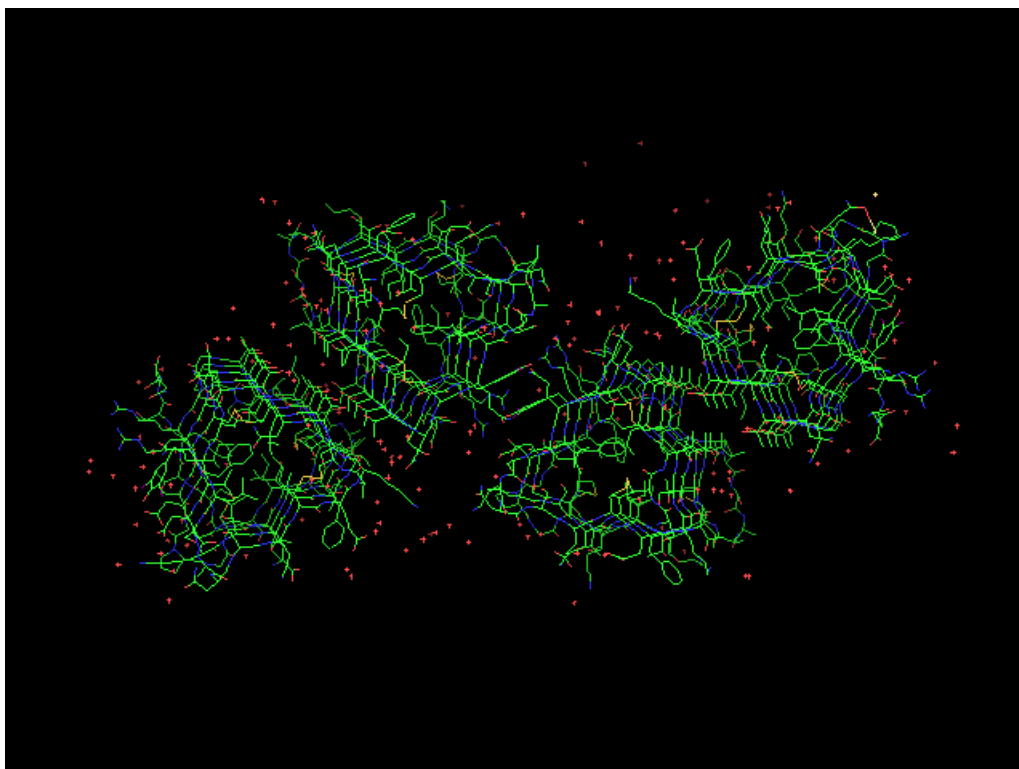
Above right image shows Ice-Binding residues of ColAFP.

10) 6BG8 / SfIBP_1->



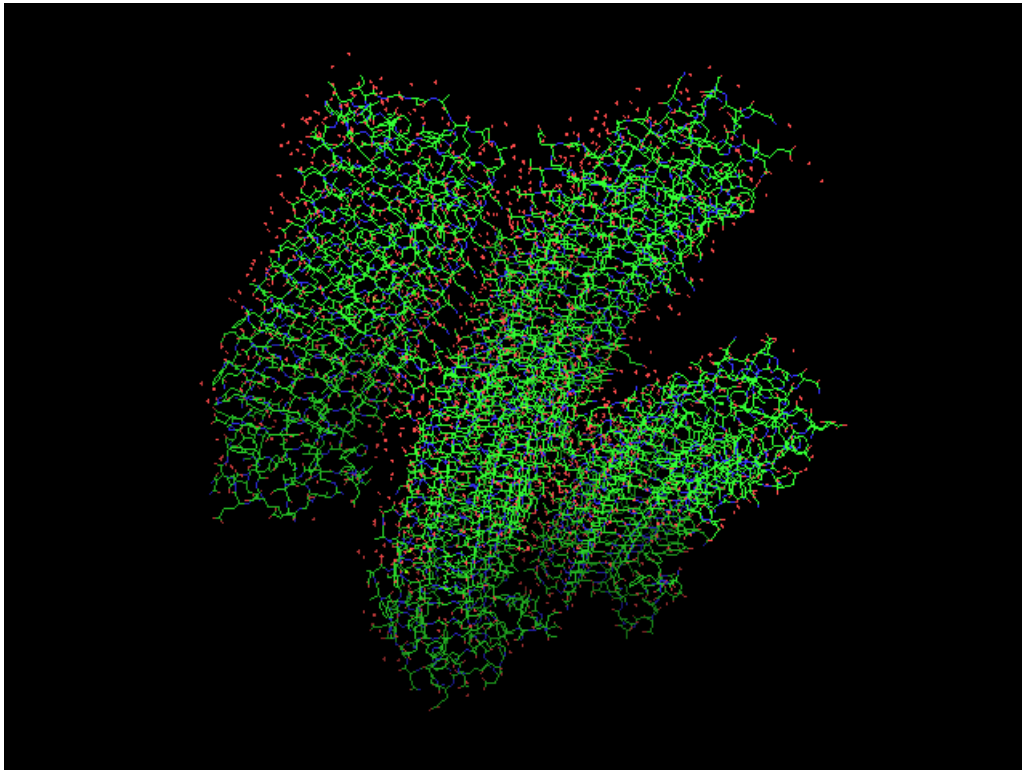
Above left image shows 2 chains of SfIBP and above right image shows Ice-binding residues of SfIBP. Each chain has 4 unique ligands namely phosphate ion (PO₄), chloride ion (CL), Glycerol (GOL) and Sulfate ion (SO₄). SfIBP_1 are able to bind to the basal plane of ice.

11) 1L0S / CfAFP ->



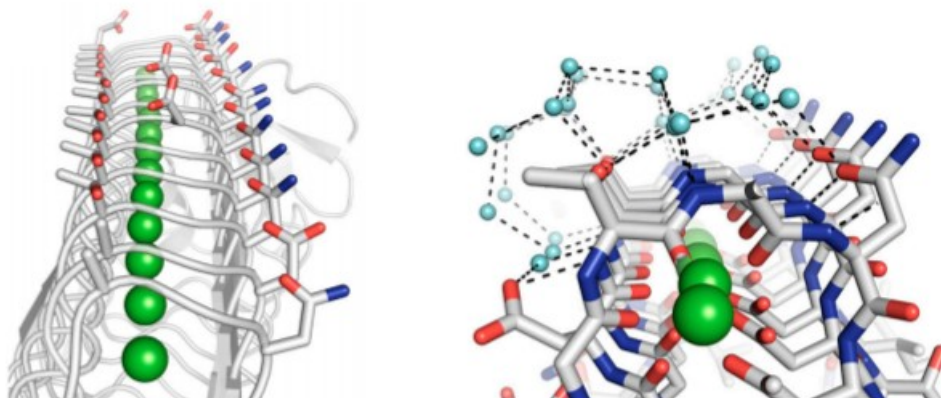
Above left image shows 4 chains of CfAFP and right image shows the Ice-binding surface of the CfAFP depicting bound water molecules (red) and ice lattice oxygen positions (yellow). Each chain has one cadmium ion (CD) as ligand and one modified residue 3,5-DIIODOTYROSINE (TYI). CfAFP has exceptionally regular β helix scaffold for the alignment of the Thr-X-Thr motifs in an orientation that appears ideal for binding to ice. CfAFP interacts with primary prism plane ice surface.

12) 3P4G / MpAFP ->

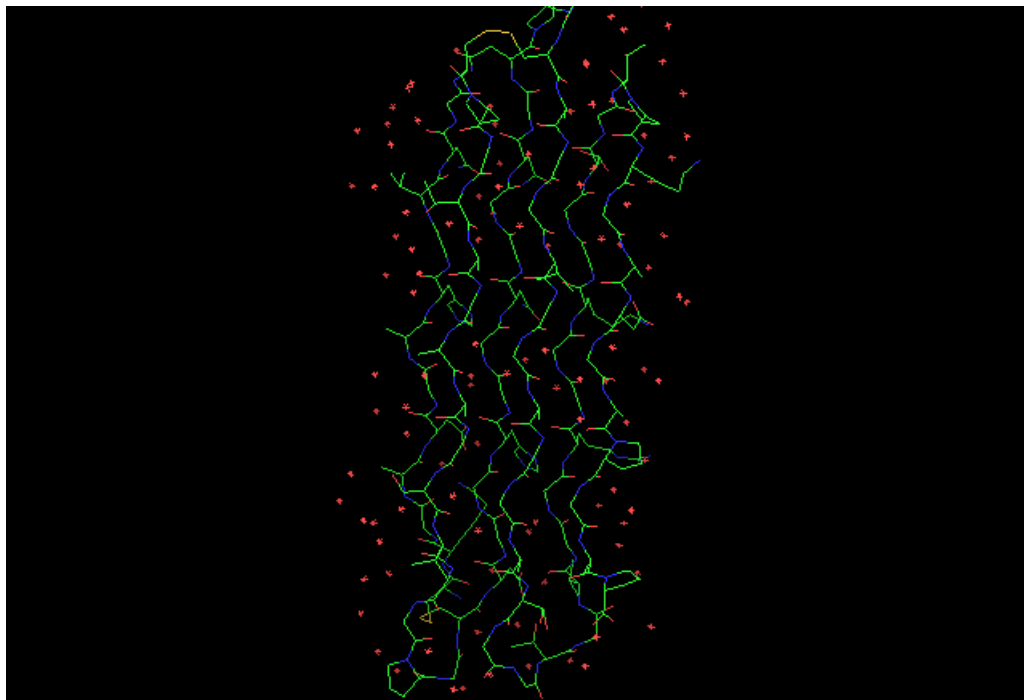


Above image shows four chains of MpAFP.

It has 3 types of ligands mainly acetate ion (ACT) (each on chain B and C), calcium ion (CA) (each on chain A,B,C and D), and Magnesium ion (MG) (each on chain A and B). MpAFP presents a long and flat IBS that runs the length of its Ca^{+2} -bound side (as shown in below left figure). It consists of the Thr and Asx (usually Asn) residues that project outward from the xGTGND Ca^{+2} -binding turns. These protein make an excellent three-dimensional match to both the primary prism and basal planes of ice. Bottom right figure shows binding of ice to IBS of MpAFP.



13) 2PNE / snow flea Antifreeze protein / sfAFP ->



Ice-binding surface encompasses residues 17-21, 47-51, 74-78.

The sfAFP molecule is made up of six antiparallel left-handed PPII helices, stacked in two sets of three, to form a compact brick-like structure with one hydrophilic face and one hydrophobic face.

1) image marks IBS of sfAFP.

2) image shows how this protein binds to the water molecules of ice.

3) image shows Cartoon of the backbone fold of sfAFP.

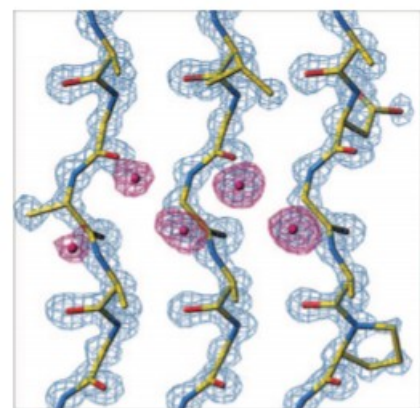
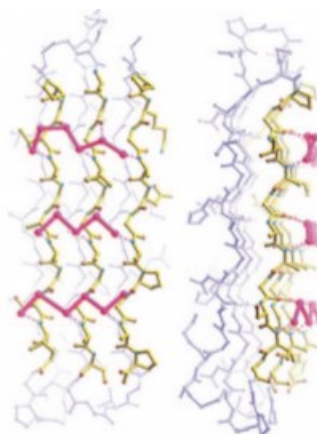
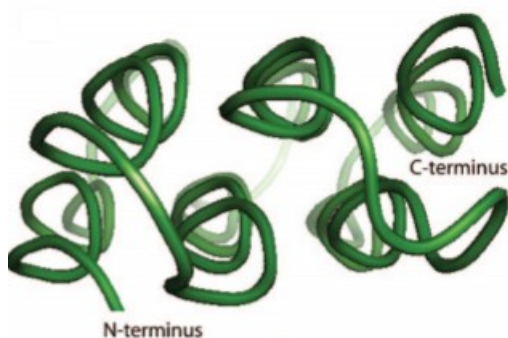
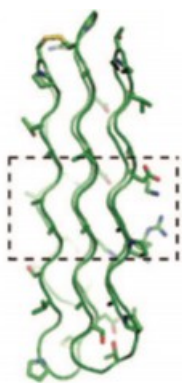
4) image shows Five ordered water molecules (magenta spheres) with electron density shown at 1σ .

1

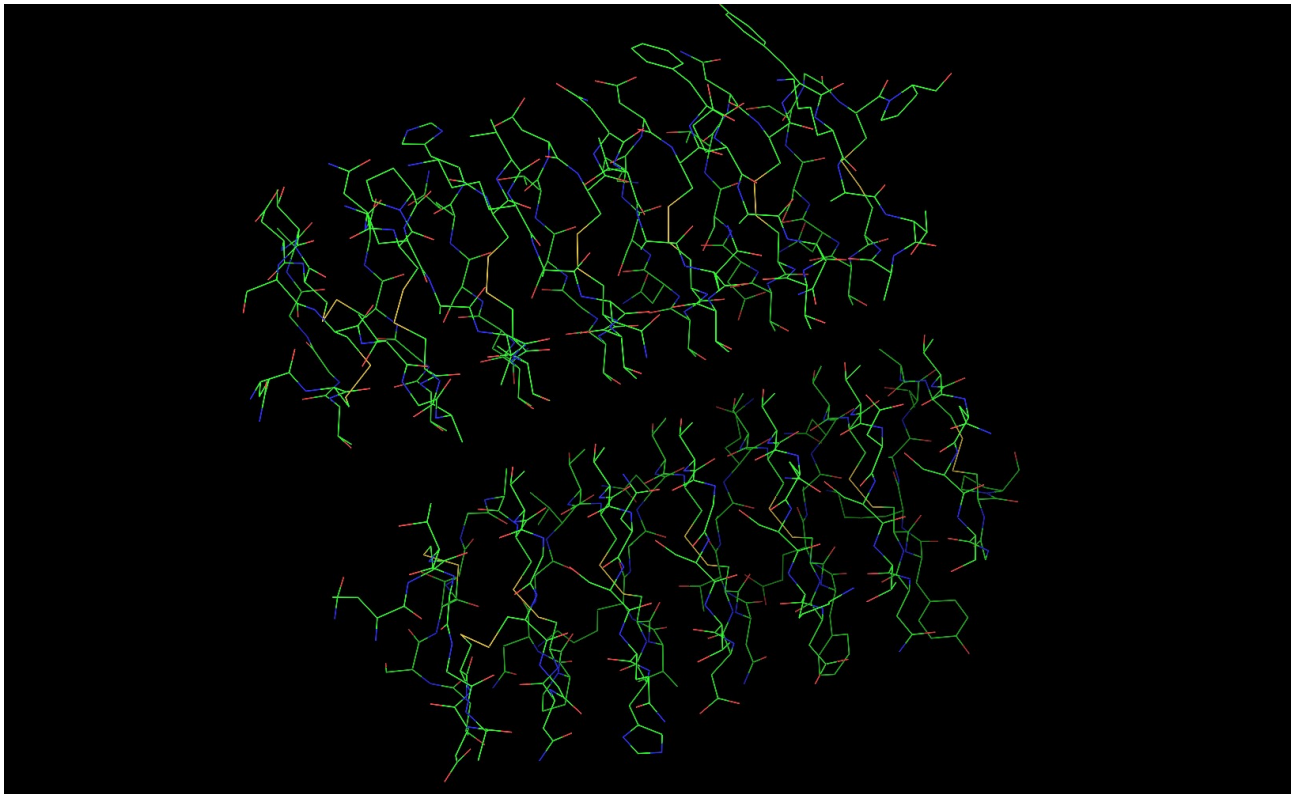
2

3

4



14) 1EZG / TmAFP ->

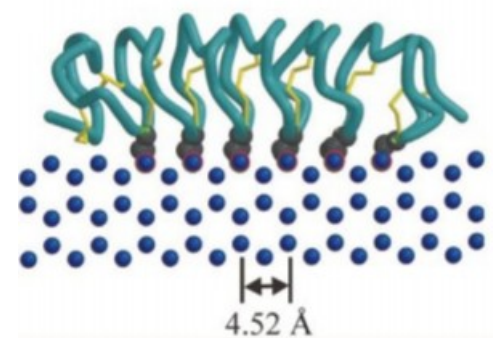
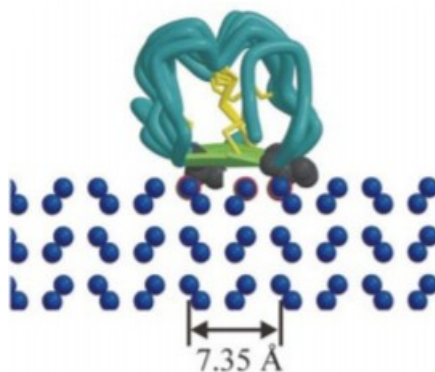
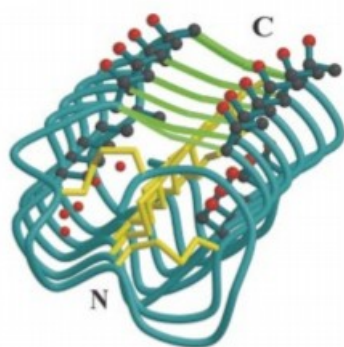


Above image shows two chains of TmAFP.

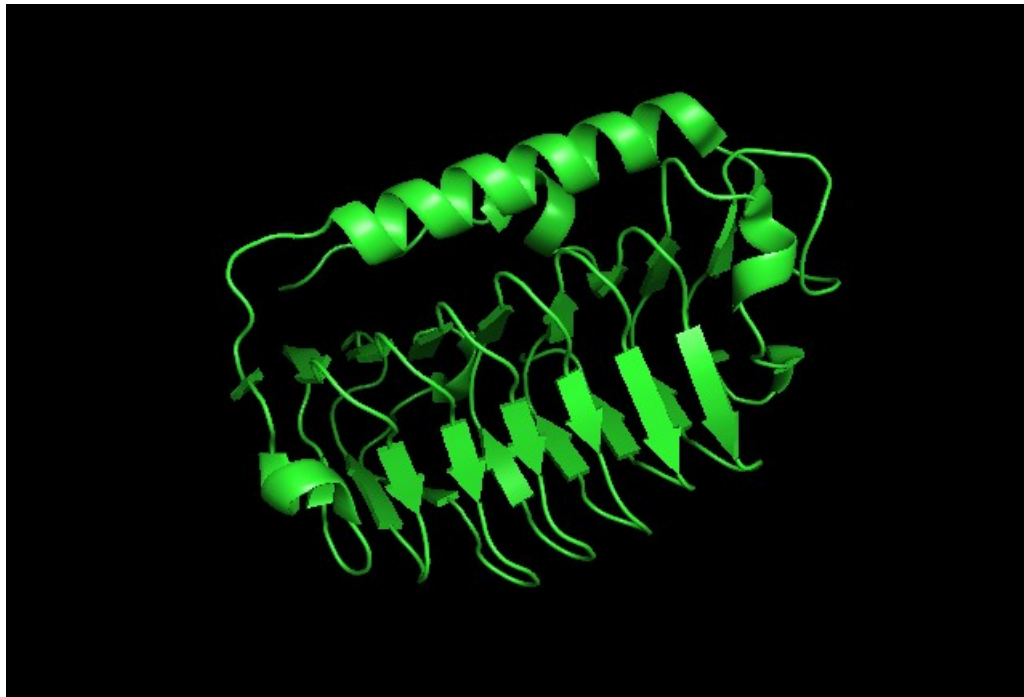
The two-dimensional array of threonine side chains makes a remarkably good match to the repeated spacing between oxygen atoms in the ice lattice on the primary prism plane, and a reasonable match to the basal plane.

Bottom left image shows one chain of TmAFP. Middle image shows end-on view of the beta-helix, showing the occupation of ice oxygen atoms (blue) on the prism plane of ice by three ranks of oxygen atoms from threonine and bound water (red perimeter). Right image shows side view of the b-helix, showing the occupation of ice oxygen atoms on the prism plane of ice by threonine from six loops.

The ice binding residues are Thr(27,39,51,63)-Cys(28,40,52,64)-Thr(29,41,53,65).



15) 4NU2 / FfIBP->

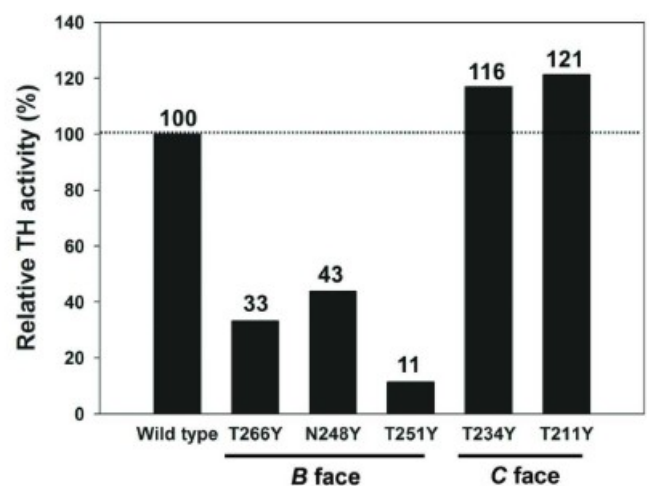
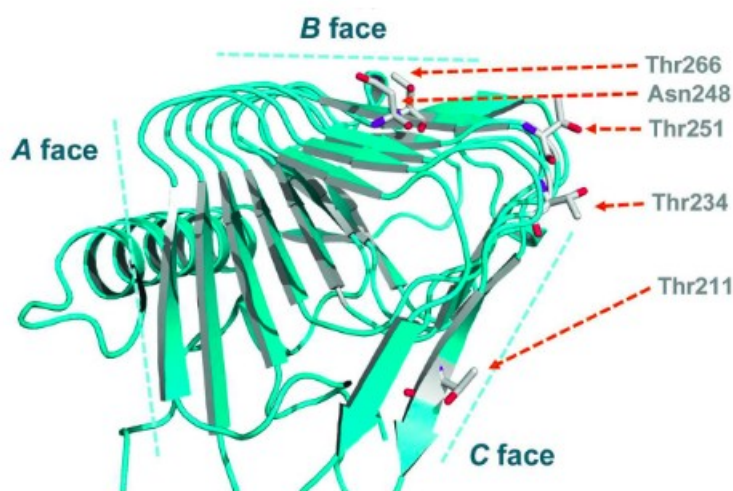


The ice-binding site of FfIBP is composed of a T-A/G-X-T/N motif, which is similar to the ice-binding residues of hyperactive antifreeze proteins.

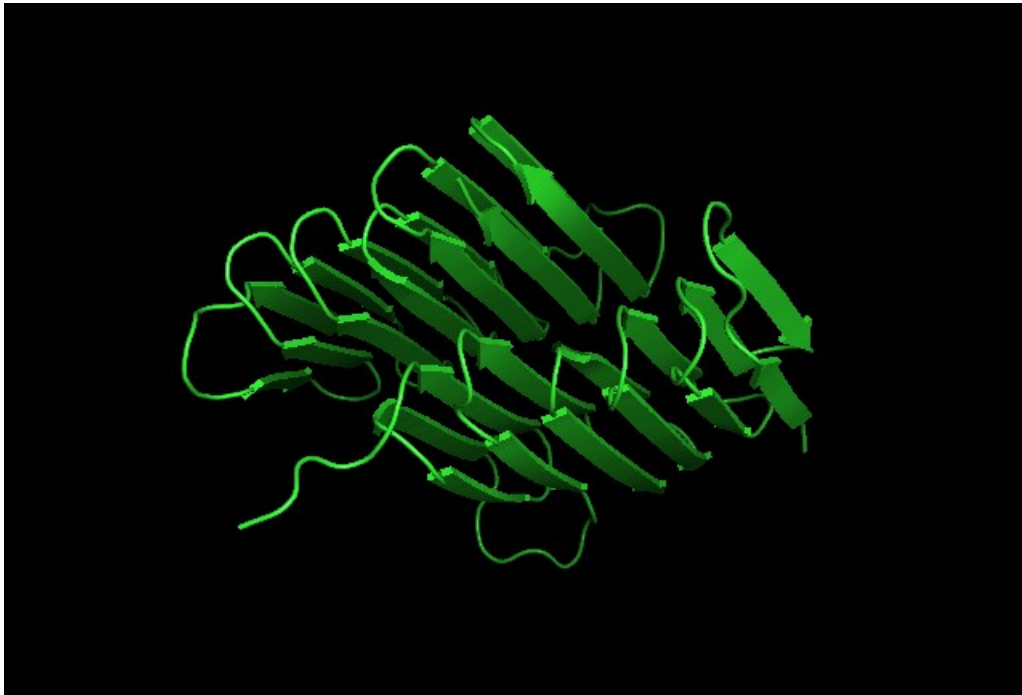
Bottom left image shows the three possible IBS planes of the FfAFP.

It also shows the Ice-Binding residues at B and C face of the protein.

Bottom right image shows the graph between percentage relative Thermal hysteresis vs the ice-binding residue at different faces of the protein.



16) 4DT5 / RiAFP->

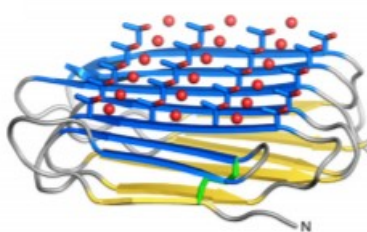


Above image shows two chains of RiAFP.

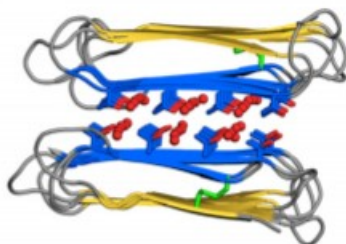
The protein has sulphate ion (SO₄) on chain A and B, and Glycerol (GOL) ligand on chain A. Ordered crystallographic waters on the RiAFP surface match several planes of hexagonal ice.

- 1) Image shows overall fold of RiAFP . Beta-Strands are shown in blue and yellow. The threonine side chains and coordinated water molecules (red) on the IBS are shown in ball-and-stick representation. The disulfide bond is indicated in green.
- 2) Image shows RiAFP dimers in the crystallographic asymmetric unit, with the ice-binding surfaces packed face-to-face.
- 3) Image shows Ice-Binding surface of RiAFP.
- 4) The thin core of RiAFP spans less than 6 Å and is tightly packed with interdigitating Ala, Ser, and Thr residues. Image shows end-on and 180° view of the core residues in RiAFP contributing to the capping motifs.

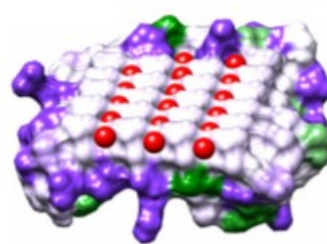
1



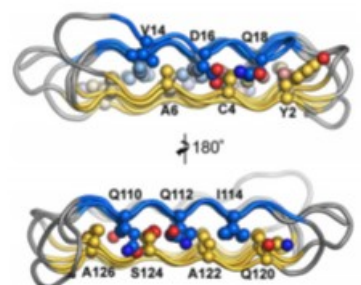
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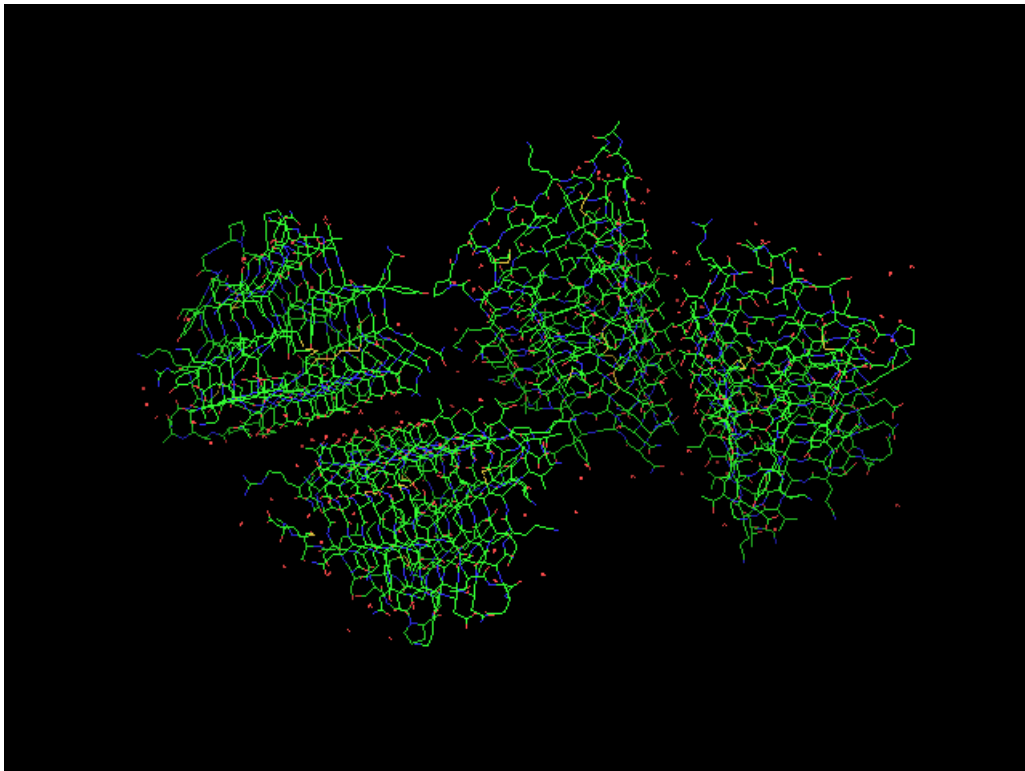
3



4



17) 1M8N / CfAFP->



Above Image consists of 4 chains of CfAFP.

The repetitive Thr-Xaa-Thr array forms the ice-binding face of the protein.

Below left image shows Ribbon representation of CfAFP-501 -helical crystal structure

Below right image shows Putative Thr array ice-binding platform of CfAFP. Two Val and one Ile occupy positions within the left of Thr residues. All residues are in the same rotameric configuration to form a binding site that enables interaction with ice through surface complementarity. Yellow color represents carbon and red color represents oxygen.

