Protein focus

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Killer toxin Protein (KP4): Harnessing the power of a killer toxin for good

Related links

InterPro website: Killer toxin, Kp4 family (IPR015131)

Various yeast strains produce cytotoxic proteins as a way of gaining a competitive advantage for limited nutritional resources over other strains. These proteins bind to the cell wall in a receptor-mediated way and are subsequently translocated to the cell membrane. Some killer toxin proteins are reported to cause death by blocking calcium channels affecting calcium transport thus leading to cell death[1]. These proteins are termed killer toxins and the organisms producing them are dubbed 'killer' strains. Scientists have taken advantage of the ingenious 'killer phenomenon' strategy from yeast to engineer transgenic plants expressing killer toxins to confer resistance to fungal infection in plants.

The protein KP4 has been successfully used in a variety of agricultural applications to confer disease resistance in plants. Current success stories involving the use of KP4's cytotoxic properties include

- genetically modified maize plants expressing KP4 which resistant to corn smut[2],
- disease resistant tobacco[3]
- disease resistant wheat[4]
- use of KP4 in controlling wine spoilage[5]

Other potentially exciting applications for KP4 include modelling the 'killer' behaviour of the protein in the development of antibiotics to control fungal infections[6].

So what do we know about this protein and what features make it attractive in genetic modification?

Killer toxin KP4

KP4 is produced by a fungus Ustilago maydis carrying a double stranded virus (UMV4) which encodes for this protein. U. maydis is a fungus responsible for the corn smut disease in maize[7] but not all strains of the fungus carry this virus hence not all U. maydis strains produce KP4. Fungal strains carrying the virus producing this toxin are able to kill other fungi of the same or different species while remaining unharmed themselves. KP4 acts by blocking fungal calcium channels[8]. Since calcium is an essential component in cell growth and meiosis, the net effect of KP4 is cell death caused by inhibited cell division. A similar effect has been observed in mammalian cells, where KP4 blocks voltage gated channels affecting calcium import in the cell. Studies have shown that this protein is very stable, showing resistance to degradation by solvents, high temperatures, and proteases. The crystal structure of KP4 has been solved[9].

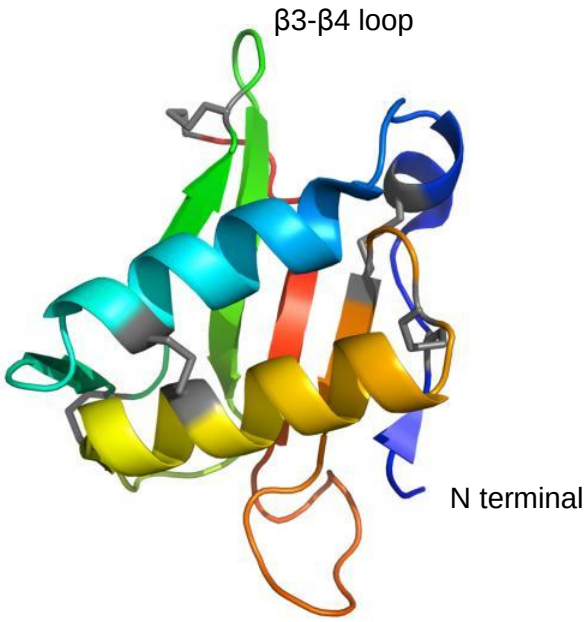
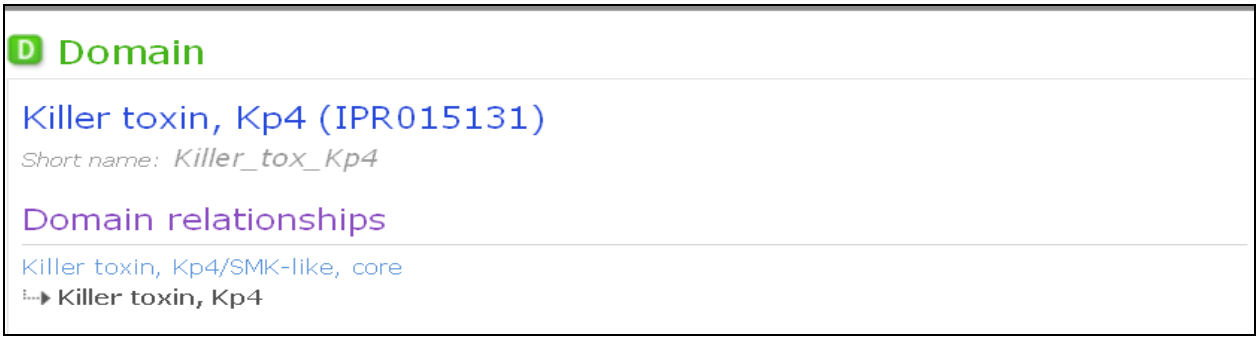


Fig 1- Structure of virally encoded KP4 toxin from U. maydis [10] *(PDB entry 1kpt)*

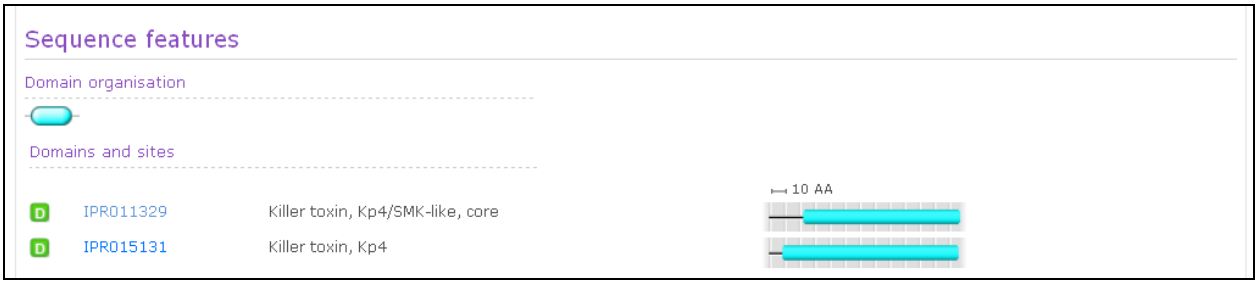
The protein has 5 disulphide bonds and is characterised by having a double split beta-alpha-beta motif with two beta-alpha-beta cross-overs. The protein exists as a monomer in solution. The solved structure depicts a dimer in the crystal unit cell belonging to the alpha-beta sandwich family of proteins.

What InterPro tells us

Investigating KP4 in InterPro, we see the database contains an entry for a KP4 domain, which is a subclass of the broader domain entry Killer toxin KP4/MSK-like core.



An example protein matching this entry is killer toxin KP4 from Ustilago maydis (UniProtKB accession Q90121). Information about this protein in InterPro includes a sequence features section where we can find information on the domains that the protein is predicted to possess.



The KP4/SMK-like core domain is found in salt mediated killer toxins of yeast, as well as KP4. The major distinction of this group of toxins is that firstly they exist as a single polypeptide and secondly that they possess two left-handed split beta/alpha/beta motifs. More information on other toxins containing this core domain can be found in InterPro entry IPR011329.

The killer toxin KP4 domain is found specifically in KP4 proteins. Mousing over this domain allows us to establish its location along the protein sequence.

Structural information about the region from 9-126 amino acids on the protein sequence is described by CATH and SCOP entries, corresponding to the crystal structure described in the structure section above.

The Gene Ontology terms provided by InterPro for this protein suggest that the protein is located in the extracellular region and is involved in pathogenesis.

InterPro: using protein signatures to predict protein function

The KP4 protein from U. maydis matches InterPro entry IPR015131 based on the protein signatures contained within that entry. Other proteins found in the same entry and therefore predicted to have a similar function, include a killer toxin from the blue fungus, a predicted calcium inhibitor from Metarhizium acridum, and several uncharacterised proteins. This information opens up the intriguing possibility that these proteins may be suitable candidates for transgenic exploitation in a similar manner to KP4[11].

Summary

The successful application of KP4 in molecular pharming has begun to open up possibilities of this toxin's use in other industries. Like with all new innovations, careful monitoring of the effects of transgenic crops is always paramount both in assessing their potential risks to human, as well as evaluating their propensity to upset the delicate balance that is our ecosystem.

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