Chain forming the beta-helices are very different. Nevertheless, gpV, gp138 and PA0616 are water soluble and SDS-resistant proteins and show no obvious membrane affinity. They are unlikely to unfold upon interaction with the membrane during phage attachment.

GpV, gp138 and PA0616 contain a conserved cluster of histidines at the tip of the beta-helical domain. These histidines bind a Fe atom in the octahedral configuration. In addition to Fe, gpV also contains Ca and Cl near the spike’s apex.

The peculiar topology and thermodynamic stability of gpV, gp138 and PA0616 suggests that these proteins are used as rigid and sharp needles to breach the outer membrane of the host cell using the energy of the contractile sheath. These spikes appear to create an opening in the host cell membrane into which the tail tube in inserted for subsequent DNA release into the host cell.

MS29.P04

Structure of influenza B nucleoprotein and its functional characterization
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The Influenza viruses are classified into three types: A, B and C. While influenza A virus is widely recognized as the most devastating one, influenza B virus also causes severe damages, in particular substantial mortality among patients younger than 18 years old. Influenza B virus is prevalent in Hong Kong. It accounts for 32% of the 554 laboratory-confirmed cases from February 27, 2011 to April 2, 2011. Among the influenza viral proteins, nucleoprotein (NP) is the major component of the ribonucleoprotein complex, which is crucial for the transcription and replication of the viral genome. We have recently determined the crystal structure of influenza B NP to a resolution of 3.2 Å. Two NP molecules, namely chains A and B, are found in an asymmetric unit. Homologous to the structure of influenza A NP [1,2], influenza B NP is composed of the head and body domains and a tail loop. Influenza B NP forms a tetramer in the crystal structure with two A chains and two B chains, in contrast to the trimer observed in influenza A NP. The homo-tetramer formation is the result of tail loop insertion from one NP molecule to its neighboring NP. Another major role of NP is to bind the genomic RNA of the virus. The putative RNA-binding regions are exposed in the influenza B NP tetramer. Residues involved in oligomerization and in RNA binding have been studied biochemically by static light scattering and surface plasmon resonance. The functional significance of these residues towards the ribonucleoprotein activities of the virus has also been investigated. The structure-function relationship of influenza B NP has enriched the current knowledge on influenza NP and provides valuable information for the design of anti-viral agents.

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Keywords: influenza virus, nucleoprotein, structure

MS29.P05


Packing disorder: structure of soluble domains of Hepatitis A Virus 2B protein
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Hepatitis A Virus (HAV) is a common cause of acute hepatitis worldwide, transmitted by fecal-oral route. HAV is classified within Picornaviridae family but it has some distinct biological characteristics like its slow viral replication and not inducing cellular lysis. Picornavirus genome encodes for a large polyprotein, which is processed by viral proteinases into a variety of precursor and mature proteins. The polyprotein is organized in structural proteins, the P1 region, and non-structural proteins, P2 and P3 regions. The P2 region includes proteins 2A, 2B and 2C involved in the virus life cycle. The role of 2A in proper capsid assembly and the implication of 2B in viral RNA synthesis and in the release of the virus particles from the cell, make them an interesting focus of study. The C-terminal domain of 2B is predicted to be organized as a transmembrane helix which would allow its presence in cellular membranes.

We have obtained small tetragonal crystals from the P2 N-terminus (including 2A and the soluble part of 2B) and determined the X-ray structure up to 2.7Å resolution. Only the structure corresponding to the 2B region (145 amino acids) could be determined because the 2A polypeptide appeared disordered in the crystals. The asymmetric unit (a.u.) contains two 2B molecules, organized in two domains: the first one shows a pseudo β-barrel organization and the second is an α-helix bundle. The two molecules in the a.u. are connected through their respective β-hairpins, included in the β-domain. This connection is extended by the crystal contacts, forming a fiber-like crystal packing along C-axis. Thus, the crystal network consists of parallel fibers of 2B separated by big spaces, where the disordered 71 residues of 2A protein seem to be allocated.

Keywords: hepatitis A, 2B protein, packing

MS29.P06


Fab’-induced folding of intrinsically disordered HIV-1 Tat
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The human immunodeficiency virus type 1 (HIV-1) is the agent responsible for acquired immunodeficiency syndrome (AIDS). Besides the canonical gag/pol/env retroviral genes, HIV-1 codes for additional accessory and regulatory proteins that act at different stages of the viral replication cycle. Among regulatory proteins, the transcriptional activator protein Tat contributes to the transactivation of viral genes.
We have expressed a recombinant form of the coat protein of satellite tobacco necrosis virus (STNV) in *E. Coli* using a codon-optimized gene, and shown it assembles spontaneously into capsids closely resembling the wild-type virus. The T=1 virus-like particles (VLPs) package the recombinant RNA transcript, and conditions have been established for disassembly and reassembly in *vitro*. *In vivo* in *E.coli* VLP assembly is dependent on the presence of the coat protein N-terminal helix, and *in vitro* it requires RNA. We have solved the X-ray crystal structure of the VLP refined it to R_{wp} 17.4/20.7% at 1.4Å resolution. It is very similar to wild-type STNV structure with no evidence of ordered RNA. Icosahedral symmetry constraints were removed to reveal small differences between subunits, presumably owing to crystal packing.

We collected low resolution X-ray data in the range 140-6Å, and the 60-fold averaged electron density map clearly shows well ordered RNA fragments lodged near the inside surface of the capsid, close to basic clusters of N-terminal triple helices that extend into the interior of the particle. The RNA consists of a 3 bp helical stem, with a single unpaired base at the 3' end and probably consists of a number of short stem-loops, where the loop region is disordered. The arrangement of the RNA is different from that observed in other satellite viruses.

Using immobilised coat protein monomers placed under reassembly conditions with 'free' coat protein subunits, we have prepared a range of partially assembled coat protein species for RNA aptamer selection. SELEX directed against the RNA-binding faces of the STNV coat proteins resulted in the isolation of a clone, B3, that matches the STNV1 genome in sixteen out of twenty-five (16/25) nucleotide positions, including across a statistically significant 10/10 stretch. This latter region folds into a stem loop displaying the loop motif ACAa, and for B3 this region binds STNV coat protein. Analysis of the other aptamer sequences reveals that the majority can be folded to stem loops displaying versions of this motif. Using a sequence/secondary structure search motif we then analysed the genomic sequence of STNV1 identifying 30 stem loops displaying the sequence motif. The implication is that there are many stem loops in the genome carrying essential recognition features for binding the STNV coat protein. The predicted genomic RNA fold is only 8/30 such stem loops are thermodynamically favoured, implying that assembly may occur on nascent RNA transcripts where folding can be controlled kinetically.

**Keywords:** virus, RNA, recognition

**MS29.P08**

Structure of PBl-PB2 subunit interface of influenza A virus RNA polymerase

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Influenza is an infectious disease caused by the influenza virus. The symptoms such as a high temperature, chills, headache and so on are evident throughout the incubation period of about three days. Usually the symptoms disappear in a few weeks as the virus is cleared by the immune system, and there are no long-term consequences to health. However, sometimes highly pathogenic influenza viruses arise and cause pandemics, which may result in a large loss of human life. Recently a highly infectious H1N1 type virus appeared and caused a world-wide pandemic. It was also reported that humans had been

**Keywords:** virus, RNA, recognition

**MS29.P07**

Structure and RNA recognition in recombinant STNV capsids

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Influenza is an infectious disease caused by the influenza virus. The symptoms such as a high temperature, chills, headache and so on are evident throughout the incubation period of about three days. Usually the symptoms disappear in a few weeks as the virus is cleared by the immune system, and there are no long-term consequences to health. However, sometimes highly pathogenic influenza viruses arise and cause pandemics, which may result in a large loss of human life. Recently a highly infectious H1N1 type virus appeared and caused a world-wide pandemic. It was also reported that humans had been