the central RNAbinding groove and the key residues for this interaction are mainly located in this groove. RNA is strongly bent at each NP–NP interface and is largely solventinaccessible in the tetramer structure. The dimensions of the groove allow accommodation of ssRNA and further analysis showed that the majority of residuenucleotide interactions occur with the ribose and the phosphate moiety, suggesting a non sequence-specific ssRNA interaction. During the simulation time, the globular core domain did not reveal any loss of secondary structure, increase of radius of gyration or persistent increments on RMSD values, which supports the model quality. The RMSF calculations indicate the Nterminal arm as a very flexible region. Most of the key residues are conserved among all tospoviruses. Copies of the NP form oligomers that interact with the viral RNAs to build ribonucleoprotein complexes (RNPs) that are proposed to be transported via plasmodesmata and are templates for RNA replication and transcription. The proposed model may shed light on the mechanisms of RNP shaping and allow the identification of essential amino acid residues as potential targets for tospovirus control strategies.

PIV145 - NGS STRATEGY REVEALED THREE PUTATIVE MEMBERS OF A NEW GENUS IN THE POTYVIRIDAE FAMILY NATURALLY INFECTING STYLOSANTHES

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Nextgeneration sequencing (NGS) is quickly emerging as the goto tool for plant virologists when sequencing whole virus genomes and undertaking plant metagenomic studies for new virus discoveries. Two Stylosanthes sp. samples were shipped to sequencing through the use of NGS and three novel potyviruses, preliminrly named as Poty 1, Poty 2 and Poty 3, were discovered naturally infecting Stylosanthes plants. These samples were collected in experimental fields of Embrapa Beef Cattle in Mato Grosso do Sul, showing typical leaf mosaic symptons. To obtain a viral enriched fraction, the leaves were ground in phosphate buffer, filtred and centrifuged through a sucrose cushion. Viral RNA was extracted using RNeasy Mini Kit following the manufacturer's instructions. The RNA samples were pooled and sequenced at Macrogen INc. (Korea) using Illumina HiSeq 2000 technology. Based on the results from the consensus NGS, primers were designed for whole genome and these viruses were confirmed in the infected samples by RTPCR. The 3' ends were confirmed by using oligodT primers with specific forward to each virus. The 5' ends were confirmed with the techniques of the SMART PCR and RACE. The complete genomes were determined to comprise of 9213 nucleotides for Poty 1, 9197 nucleotides for Poty 2 and 9425 nucleotides for Poty 3 (excluding the polyA tails). The complete virus genomes and CP sequences were compared with sequences available in GenBank. The highest nucleotide identities of 43%, 39% and 56% were determined compared to other potyviruses, respectively. The genomes were deduced to encode a single open reading frame (polyprotein) on the plus strand. Phylogenetic analysis based on the whole genome sequences and coat protein amino acid sequences showed that the new viruses found are most closely related to the Blackberry virus Y (Poty 1 and Poty 2) and the Rose yellow mosaic virus (Poty 3). The biological features of these new potyviruses are currently being investigated.

PIV151 - HIGH INCIDENCE OF MIXED DNA AND RNA VIRUS INFECTIONS IN COMMON BEAN IN CENTRAL BRAZIL

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Common bean is one of the most important protein food source consumed worldwide, mainly in Africa, South and Central Americas. In Brazil, there are several diseases affecting bean fields, including viral diseases, such as the Bean golden mosaic virus (BGMV), responsible for losses that can reach 100%. During the winter crop season in 2016 a very high incidence of viruslike symptoms of mosaic, leaf curling and deformation, and plant dwarfing was reported by farmers in central areas of Brazil. Bean plants were collected in commercial farms in Luziânia, Cristalina and experimental plots in Goiânia and Brasília. Total DNA was extracted using

CTAB method. Total RNA was obtained using Trizol® reagent. Samples were tested for the presence of DNA and RNA viruses that are commonly found infecting beans in Brazil. Begomoviruses BGMV, Macroptilium yellow spot virus (MaYSV), and Macroptilium yellow net virus (MaYNV) were detected by PCR using specific primers. Detection of the RNA viruses Cowpea mild mottle virus (CPMMV Carlavirus), Bean rugose mosaic virus (BRMV Comovirus) and a new, yet not fully characterized rhabdovirus (Bean associated rhabdovirus BAR), recently found by our group was performed by RTPCR with specific primers. For the samples tested thus far, a high incidence of mixed infection, usually with three viruses was detected in all sampled areas. BGMV and CPMMV were present in 100% of the plants while BAR had an incidence of 60100%, varying according to the region where the samples were collect. BRMV was detected in a few samples while MaYSV and MaYNV were not identified in any of the samples. This widespread of mixed infection is causing extensive yield losses in the bean crop and will likely impact on availability and influence market prices.

PIV155 - A PLANT VIRUS COAT PROTEIN AS A CARRIER PROTEIN FOR MEDICAL INTEREST EPITOPES IN BACULOVIRUS/INSECT CELL SYSTEM

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The baculovirus expression vector system (BEVS) has been widely used to produce a large number of recombinant proteins and is becoming one of the most powerful, robust, and costeffective system for the production of proteins. The success of the system is duet to the intrinsic security and the high yields of protein expression. BEVS can be used for the production of virus-like particles (VLPs). The VLPs can be obtained basically the by expression of recombinant capsid proteins in a variety of heterologous systems, that promote the selfassembly of proteins into structures similar virus particles. VLPs have antigenicity similar to that of the native virus, but they lack genetic material, thus are not infectious. Tomato blistering mosaic virus (ToBMV) is a plant virus infecting plants from the genus Solanum and, in this work, candidate as a carrier protein for medical interest epitopes. Therefore, we are assessing the assembly of tymoviruslike particles (tVLPs) using BEVS. In this work, the potential use of tVLPs for displaying biopharmacological epitopes with medical interest and one epitope of Chikungunya virus (CHIKV) envelope protein 2 (E2) was selected. CHIKV is a mosquitoborne viral disease that causes headache, fever and severe joint pain. Since 2004, this virus is affecting thousands of people around the world. Importantly, there is no available serological kit for CHIKV detection produced in Brazil. Therefore, we will use the tVLPs displaying the CHIKV E2 epitope to generate diagnostic kits for human serum. For this purpose, we are analyzing the deletion construct of the first 23 a.a. of ToBMV CP whether this deletion will interfere with the correct assembly of the tVLPs. If not, we will replace these 23 a.a. by CHIKV epitope. At first, the deletion mutant gene of ToBMV CP was cloned to pFastBac1 vector. The constructs were sequenced and the DH10Bac strain of Escherichia coli that contains Bacmed was transformed with selected clones to generate recombinant baculoviruses by prokaryotic transposition and viral DNA transfection into insect cell. The protein expression of this construct is now on evaluation by immunoblotting using specific antiCP antibody.

PIV164 - A NEW PUTATIVE GEMYCIRCULARVIRUS DETECTED IN COMMON BEAN IN BRAZIL

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Genomoviridae is a recently created family of circular, singlestranded DNA viruses. The family is composed of one genus, Gemycircularvirus with a sole recognized species, Sclerotinia gemycircularvirus 1. The representative isolate, Sclerotinia sclerotiorum hypovirulence associated DNA virus 1, (SsADV1) was discovered in 2010, infecting the fungus S. sclerotiorum. There are more than one hundred SsADV1like putative viruses described in different hosts and environmental samples, including water from rivers, treated and