P18-10

GENOMIC CHARACTERIZATION OF A NOVEL MEMBER OF THE LUTEOVIRIDAE ASSOCIATED WITH ROSE SPRING DWARF DISEASE

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Virus and virus-like diseases occur wherever roses are grown. So far, viruses of the genera Ilarvirus and Nepovirus have been shown to be associated with specific rose diseases, but many diseases still have unknown etiologies. One of these is rose spring dwarf disease (RSD). A virus was transmitted by grafting from RSD-affected roses to Rosa multiflora plants. Double-stranded RNAs (dsRNAs) were recovered and used as templates for cDNA synthesis and generating a cDNA library. A nucleotide sequence of 5,738 nucleotides was identified. Computer-assisted analysis revealed five putative open reading frames (ORFs) and showed organization and sequence homology with viruses of the family Luteoviridae. ORF1 overlaps ORF 2 and are likely translated together via a -1 ribosomal frameshift event to give the virus-encoded RdRP. ORF 4 is contained in the same sequence as is ORF 3, but in a separate reading frame register. ORF 3 encodes for the coat protein (CP), and via a translational readthrough into ORF 5 the CP readthrough protein (RTP). Sequence analysis of RdRp, CP, and RTP indicated that the best hits for amino acid sequence similarities were Soybean dwarf virus (SbDV), Sugarcane yellow leaf virus (ScYLV), and Potato leafroll virus (PLRV), respectively. Based on our results we propose the name Rose spring dwarf associated virus (RSDaV).

P18-11

EXPRESSION OF THE MAIZE MOSAIC VIRUS (MMV) GLYCOPROTEIN IN INSECT CELLS

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Maize mosaic virus (genus *Nucleorhabdovirus*, family *Rhabdoviridae*) is transmitted in a persistent-propagative manner by *Peregrinus maidis* (Delphacidae, Hemiptera), the corn planthopper. Like other rhabdoviruses, the MMV genome encodes a surface glycoprotein that is likely involved in virus attachment and entry into host cells. To develop a better understanding of the role of the G protein in virus entry into arthropod vectors, we constructed plasmids to express different portions of the MMV glycoprotein gene from recombinant baculoviruses in SF21 cells. Two constructs were made: a soluble form of MMV G that contained the gene portions corresponding to the ectodomain (amino acids 21-530), and an insoluble form which included the ectodomain, transmembrane domain, and cytoplasmic tail (amino acids 21-592). For both protein constructs, the predicted MMV G protein N-terminal signal sequence was replaced with the baculovirus GP64 signal sequence. Interestingly, we found that both viruses produced a secreted form of the protein. This finding is consistent with expression of other rhabdovirus glycoprotein genes. Approximately 10% of the rabies and vesicular stomatitis Indiana virus G proteins are shed into the cell culture media as a truncated-soluble form. This is the first description of a plant-infecting rhabdovirus producing a truncated form of the G protein that is secreted. Both forms of MMV G will be useful tools for examining virus acquisition by planthoppers and identification of insect receptor molecules involved in uptake of rhabdoviruses.

P18-12

MOLECULAR ANALYSIS OF A CALIFORNIAN STRAIN OF *RUPESTRIS STEM PITTING ASSOCIATED VIRUS* FROM PINOT NOIR GRAPEVINE

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Rupestris stem pitting (RSP) is one of the diseases in the rugose wood complex (RW) and affects grapevines (*Vitis* spp.) worldwide. RSP is recognized as a wood marking pathogen disease and is characterized by production of pits below the point of inoculation on the woody cylinder of the indicator St. George (*V. rupestris* Steele). Infected plants become stunted and show slow decline. The etiology of RSP is not yet completely understood. *Rupestris stem pitting associated virus* (RSPaV), a single-stranded RNA positive sense virus in the genus Foveavirus has been reported to be associated with the RSP. A strain of RSPaV was obtained from V. vinifera, variety Pinot Noir (RSPaV-PN), its genome cloned, sequenced and characterized. Double-stranded RNA was purified from infected plants and used for constructing cDNA libraries. A total of 8,333nt have been sequenced excluding the Poly-A tail. The genome organization of RSPaV-PN is similar to other published isolates. The coat protein showed the highest nucleotide and amino acid identities of 85% and 93%, respectively compared to other RSPaV isolates. Phylogenetic analyses of fragments of the replicase and coat protein genes performed between RSPaV-PN and other 27 RSPaV isolates from different geographic regions showed that RSPaV-PN clustered with a few isolates. Specific primers were designed based on the RSPaV-PN open reading frame 1 sequence that encodes for the RdRp, and used in a field survey. An expected product of 503bp was identified in 51 samples from 286 tested. Sequences of the PCR products from positive samples showed that they had 96%-99% nucleotide and 95%-100% amino acid identities to RSPaV-PN sequences. When compared to the database these sequences showed nucleotide and amino acid identities of 73.6%-76.7% and 82.1%-86.2%, respectively.

P19-1

VIRAL AND CLINICAL CHARACTERIZATION OF A NOVEL VIRAL AGENT ISOLATED FROM PIGS WITH REPRODUCTIVE AND/OR NEUROLOGIC DISORDER

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Disease outbreaks characterized by reproductive failure and/or neurologic disorders, commonly referred to as "Porcine Reproductive and Neurologic Syndrome (PRNS)", were observed in many swine farms in Iowa and other states. Extensive laboratory investigation of clinical cases repeatedly resulted in the isolation of a cytopathic virus from sera, nervous and secondary lymphoid tissues, and fetal tissues which could not be recognized by antibodies specific for common swine viral pathogens in the U.S. A series of studies were conducted to further characterize this previously unrecognized virus tentatively designated as Virus X. Morphologically, the virion was enveloped and 50-60nm in size. The virus contained a single-stranded RNA genome. Antisera raised against Virus X in rabbits cross-reacted with BVDV and border disease virus (BDV) on Western immunoblot and immunofluorescence microscopy. The antisera; however, showed very minimal neutralizing activity against BDV and did not neutralize both types of BVDV. Potential NS3 region of the viral RNA was amplified by PCR from isolates of Virus X and its sequence showed a good homology (95%) with that of known pestiviruses at the deduced amino acid level, indicating that Virus X may be a novel pestivirus of swine. Clinically pregnant sows which were experimentally inoculated with an isolate of Virus X (ISUYP604671) at mid or late gestations developed clinical signs and pathological changes similar to field observations including the loss of pregnancy. In addition, caesarian-derived, colostrum-deprived young pigs at 4 weeks of age developed mild encepha-