V. 바이러스 병학

E-01. The First Occurrence of Potato Virus T in Potato (*Solanum tuberosum*) in Korea. Young Il Hahm[1], Hyo Won Jung[1], Hong Soo Cheo[2] and Gwan Yong Shin[1]. [1]Department of Potato, National Alpine Agriculture Experiment Station, Pyoung-Chang 232-950, Korea. [2]Department of Plant Pathology, Agricultural Science and Technology Institute, Suwon, 441-707, Korea.

Potato virus T(PVT) was isolated from natually infected *Solanum tuberosum* cv. Superior on the field at Kangwon area in 1997 and also identified by indicator plant reactions, physical properties, electron microscope and serological test. The main symptoms caused by this virus in *S. tuberosum* cv. Superior are mild mosaic, stunting and necrotic patterns but, it causes very slight symptoms and sometimes latent. This virus induced systemic necrosis in *Chenopodium amaranticolor,* systemic mild mosaic in *C. quinoa*, systemic severe mosaic *in Datura stramonium*, systemic mottle *in Nicotiana debneyi* and local lesions in inoculated leaf of *Phaseolus vulgaris.* The thermal inactivation point of the virus was 60℃ in 10 min., the dilution end point was 10^{-3} and the longevity *in vitro* was 3 days at room temperature. The purified virus was the filamentous particles of 640㎚ in length with direct negative dipping method under electron microscope. The produced antiserum for diagnosis showed the specific titer with microprecipitin tests and agar gel diffusion tests. This is the first report of potato T trichovirus, from potato in Korea.

E-02. Isolation and Comparison of Two Strains of Soybean Mosaic Virus, SC and G5H. Yul-Ho Kim[1], Bong-Choon Lee[2], Ok-Sun Kim[1], Jae-Hwan Noh[1], Myoung-Ki Kim[1] and Dae-Joon Im[1]. [1]National Crop Experiment Station, RDA, Suwon, Korea 441-100. [2]National Horticultural Research Institute, RDA, Suwon, Korea 441-100.

Soybean mosaic virus (SMV)-G5H causes systemic necrosis on Hwanggeumkong. A newly-collected SMV-SC isolate induced severe mosaic symptom or systemic necrosis on Jangbaegkong. And SC isolate did not show the characteristic necrosis by G5H on Ogden. Among the 42 recommended soybean cultivars in Korea, 4 were resistant to G5H and 21 were resistant to SC. The nucleotide sequence similarity of the two isolates in the cylindrical inclusion protein (CIP) coding regions is below 70%. The results on virulence and nucleotide suquences of SMV-SC and G5H suggested that SMV-SC might be a different strain of SMV.

E-03. Effectiveness of Cross Protection by a Mild Strain of Zucchini Yellow Mosaic Virus in Cucumber and Squash. Gug-Seoun Choi[1], Jae-Hyun Kim[1], Yong-Moon Choi[1], Myong-Soon Lim[1] and Jang-Kyung Choi[2]. [1]National Horticultural Research Institute, R. D. A., Suwon, Korea 441-440. [2]Kangwon National University, Chunchon, Korea 200-701.

A mild strain of zucchini yellow mosaic virus, ZYMV-35, was derived from a virulent strain, ZYMV-C, in cucumber by high temperature treatment. ZYMV-35 and ZYMV-C distinctly differed in the degree of symptom severity on cucumber, squash, watermelon and gourd. The cucurbit plants infected with ZYMV-35

developed obvious leaf mosaic symptoms or symptomless. These symptoms were much milder than those induced by ZYMV-C. Cucumber and squash at the cotyledon stage were inoculated with ZYMV-35 and then challenged by sap-transmitted ZYMV-C after different periods. About 2 weeks of incubation were required following ZYMV-35 inoculation to provide cross protection against infection by ZYMV-C. No protection occurred if ZYMV-C was infected 3 days after ZYMV-35 inoculation and protection was intermediated if ZYMV-C infected cucumber and squash after 7-10 days. In vinylhouse experiments, ZYMV-35 provided protection in cucumber and squash against ZYMV-C. The fruits harvested from ZYMV-35 inoculated plants were distortion-free, marketable and indistinguishable from those harvested in the healthy plants. Cross protected plants produced 6-8% fewer marketable fruits than healthy plants but the plants inoculated with ZYMV-C did not produce marketable fruits.

E-04. Cytoplasmic Inclusions Associated with Infection by Potato Viruses. Hyo Won Seo, Young Il Hahm and Hyo Won Chung. Potato Division, National Alpine Agricultural Experiment Station, RDA, Pyungchang 232-950, Korea.

For the identification of potato viruses we conformed the typical inclusions which can be characteristics of some viruses infection. Many proteinaceous structures double-stained by phosphotungstic acid (PTA) and osmium tetraoxide were observed in the symptomatic potato cell cytoplasm. Ultrathin section for electron microscopy revealed the presence of typical inclusions of unique morphology in the infected potato plants. Pinwheeled and laminated inclusions are commonly observed in cytoplasm of potato leave cells infected by potato virus Y (PVY) and potato virus X (PVX) respectively. Multi-layered cylindrical inclusions induced by potato virus Y are contained the virus particles longitudinally. Cell and organell disturbances in the parenchyma tissue were observed in the infected potato plants.

E-05. Cloning and Sequence Analysis of The Coat Protein of A New Carlavirus Isolated From Korean Native Lily. Ji Hwan Ryu[1], Hong Il Ahn[1], Ki Hyun Ryu[2], Seung Kook Choi[1], Won Mok Park[1] and Se Yong Lee[1]. [1]Graduate School of Biotechnology, Korea University, Seoul 136-701, Korea; [2]Department of Horticultural Science, Seoul Women's University, Seoul 139-774, Korea.

The 3,000 bp 3-terminal region of the genomic RNA of a new Korean isolate of carlavirus from a Korean native lily (*Lilium lancitoium*) was cloned and its nucleotide sequences were determined. The coat protein (CP) gene of the virus showed 72.0 % to 72.8 % nucleotide sequence identities and 86.9 % to 88.0 % amino acid sequence identities with those of the four strains (two Korean, one Netherlands, and one Japanese isolates) of lily symptomless virus (LSV). Interestingly, most of amino acid differences between the virus and LSV strains were located at the N-terminal region. Pairwise amino acid sequence comparison revealed that sequence identities of 22.0 % to 71.1 % between the virus and other 10 species of the known carlavirus. The 16 kDa protein gene of the virus shares 16.7 % to 72.9 % amino acid sequence identities with those of 9 other carlaviruses. The 25 kDa protein gene of the virus shares 30.7

% to 76.3 % amino acid sequence identities with those of 8 carlaviruses. The 7 kDa ORF was not found in the virus and this affected one nucleotide substitution in the start codon of the potential ORF. These data indicate that the virus is a quite distinct species of the *Carlavirus* genus and distinguished from the known LSV.

E-06. Identification of Potyviruses from Soybean Using The Polymerase Chain Reaction with Degenerate Primers. Jung Mo Koo[1], Chang Won Choi[1] and Ki Hyun Ryu[2]. [1]Department of Biology, Pai Chai University, Taejon, Korea 302-735, [2]Department of Horticultural Science, Seoul Women's University, Seoul, Korea 139-774.

Alignment of published potyvirus coat protein sequences revealed regions of extensive sequence conservation in the central core domain. Therefore, two different sets of primers were selected to amplify the DNA fragments on potyvirus-specific templates in combined assay of reverse transcription and the polymerase chain reaction (RT-PCR). A first set of degenerated primers, upstream primer (U341) and a downstream primer (D341), was used in the PCR to amplify cDNA representing a 341 nucleotide region of the coat protein gene in RNA purified from soybean mosaic virus and in total nucleic acid extracted from naturally infected soybean plants. Similarly, using a second set of primers (U335/D335), the expected amplification product of 335 bp was also detected from collected soybean plants.

E-07. Differentiation among Soybean Mosaic Virus by Biological, Serological Assays, and RT-PCR with Virus and Strain Specific Primers. Jung Mo Koo[1], Sun-Sub Hwang[1], Chang Won Choi[1], Ki Hyun Ryu[2], Won Mok Park[3]. [1]Department of Biology, Pai Chai University, Taejon, Korea 302-735, [2]Department of Horticultural Science, Seoul Women's University, Seoul, Korea 139-774, [3]Graduate School of Biotechnology, Korea University, Seoul 136-701, Korea.

Twenty one soybean cultivars collected from fields of Chungnam areas and several cultivars from different geographical locations, showing mosaic symptoms, were assayed for the presence of soybean mosaic virus (SMV). Crude sap of infected leaves was tested by the Ouchterlony gel diffusion using antiserum to SMV-G1. The precipitin bands were shown from 23 out of 33 cultivars. The crude sap was inoculated mechanically into several soybean cultivars to show the differential symptoms. A combined assay of reverse transcription and polymerase chain reaction (PCR) utilizing virus-specific primers, SMCP1 and SMCP2, was applied to 5 strains of soybean mosaic virus (SMV) from U.S.A. and numerous field isolates collected in Korea. The primers were designed to amplify the coat protein gene of SMV and identified Korean and U.S.A isolates. Another specifically designed assay using the strain-specific primers to amplify the unique regions in the cylindrical inclusion protein cistron, allowed discrimination of SMV-G2 and -G7 strains, respectively, among the isolates.

E-08. Cloning, Sequencing, and Expression of the Cymbidium Mosaic Virus-Kor. Coat Protein Gene.

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Cymbidium mosaic virus (CyMV) is one of the most prevalent and economically important orchid viruses. Like most of the polyadenylated monopartite positive-strand RNA virus, the open reading frame (ORF) coding for the coat protein (CP) of CyMV is located at the 3'end. The CP gene of CyMV-Kor were cloned, sequenced and translated in vitro. The sequence was determined from a cDNA clone that encompassing the 3'-region of the virus genome. It consisted of 748 nt and polypeptide chain of 220 amino acids. The CP of CyMV genome was expressed in *Escherichia coli* using an expression vector. The CyMV CP gene was encoding about 24 KDa protein. The translated product reacted with antibody raised against the CyMV particles.

E-09. Coat Protein Gene Analysis of Cymbidium Mosaic Virus (CymMV-K2) Mild strains.

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Four CymMV variants, CymMV-A, B, C, and D with mild or symptomless pattern in Chenopodium amaranticolor and Phalenopsis. spp. were selected by mutagen treatment for study of cross-protection. Coat protein gene of CymMV variants was cloned by RT-PCR (Reverse Transcription-Polymerase Chain Reaction) and sequenced by ABI prism 377 model (Perkin Elmer). As a result of sequence analysis, they were showed from 94% to 97% homology with CymMV-Korean type 2(Koo, et al., 1998), and from 91% to 96% homology with CymMV (Wong, et al., 1994). Variation of CP gene among variants brought about symptom change and important motif sequence directly concerned with symptom expression could be found out.

E-10. The Partial Sequencing of 3'-region of Garlic Mite-borne Mosaic Virus-Korean Strain (GMbMV-K).

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GMbMV-K (Koo, et al., 1998), novel classified rymovirus, was identified from the infected garlic plants in Uisong. GMbMV-K is known to be one of the major viruses in garlic. It showed that GMbMV-K infected garlic plants induced symptoms like streaks and malformations. Virions were purified from infected garlic leaves according to the sucrose-CsCl gradient method. After ultracentrifugation, GMbMV-K was collected from the bottom band and RNA purified with phenol:chloroform. Subsequently, RT-PCR have been done with oligo d(T)-15 and 5'-CCTGCTAAGCTATATGCTGA-3' primer. Next, amplified fragments were cloned and analysed with the automatic sequencer (Perkin-Elmer). In the data base search, cloned viral cDNA have 96 % homology with Japanese Garlic Virus and 94 % homology with Garlic Virus D, respectively.

E-11. Molecular Similarities of Tobacco Mosaic Virus-Induced Hypersensitive Cell Death and Copper-Induced Abiotic Cell Death in Tobacco.

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Hypersensitive cell death of plants during incompatible plant-pathogen interaction is one of the efficient defense mechanisms of plant against pathogen infection. To better understand the molecular mechanisms of plant hypersensitive response (HR), TMV-induced biotic plant cell death and $CuSO_4$-induced abiotic plant cell death were compare using several defense-related gene expressions as molecular markers. Five pathogenesis-related protein genes, two plant secondary metabolite-associated genes, two oxidative stress-related genes and one wound inducible genes isolated from tobacco were selected for monitoring gene expression during plant cell death. Northern blot analyses revealed that same set of defense-related genes were induced during both biotic and abiotic cell death of tobacco but with different time and magnitude. The expression of defense-related genes in tobacco plants were temporarily coincided with the time of cell death but with different patterns and magnitude. But, when cell suspension cultures were used, different patterns of defense-related genes are present in tobacco during pathogen and heavy metal-induced cell death. Expression of ten different defense-related genes of tobacco in relation to biotic HR and abiotic plant cell death were described.