

Virology and Virus Diseases (J125-J167)

- J-125. Protective effect of two Japanese attenuated CMV strains, PV1-K and PV2-K against a Korean virulent CMV isolate, CMV-KP in Korean hot pepper cultivars.** J. H. Sung<sup>1</sup>, J. H. Park<sup>1</sup>, H. Y. Shin<sup>1</sup>, M. U. Chang<sup>1</sup>, H. Sayama<sup>2</sup>, and H. Atarashi<sup>2</sup>. <sup>1</sup>Dept. Biology, Yeungnam University, Gyongsan, 712-749, Korea, <sup>2</sup>Nippon Del Monte Corporation, R&D, Numata, 378-0016, Japan

Hot pepper seeds of 8 Korean cultivars; Buchon, Manita, Gunggangsán, Segae, chyungyang, Sintaeyang, Dabotap, Yomyong were sown in the greenhouse on 15th of July. Purified two Japanese attenuated CMV strains (CMV vaccines), PVI-K and PV2-K containing satellite RNAs were inoculated with cotton swabs 11 days after sowing to the pepper seedlings at the cotyledon stage. The Korean virulent CMV isolated from paprika (CMV-KP) was propagated on Xanthi-nc tobacco for a week and then the crude extract of the infected leaves was challenge-inoculated to the pepper seedlings 5 days after the vaccination. Virus symptoms appeared on leaves were evaluated 16 days after the challenge inoculation. The Korean CMV-KP induced severe mosaic symptoms in the hot pepper seedlings of 8 Korean cultivars, whereas the two CMV vaccines, PV1-K and PV2-K did either mild chlorosis or mild mosaic symptoms. The symptoms observed in both PV1-K+CMV-KP and PV2-K+CMV-KP treatments were either mild chlorosis or mild mosaic symptoms that were observed in the vaccine treatment (either PV1-K or PV2-K treatment). These results indicate that the pre-inoculation of both PV1-K and PV2-K strains to seedlings of Korean hot pepper cultivars protects the seedlings from the Korean virulent CMV-KP isolate.

- J-126. Adverse effect of two Japanese attenuated CMV strains, PV1-K and PV2-K on two Korean hot pepper cultivars.** H. Sayama<sup>1</sup>, H. Atarashi<sup>1</sup>, H. Y. Shin<sup>2</sup>, J. H. Sung<sup>2</sup>, J. H. Park<sup>2</sup>, and M. U. Chang<sup>2</sup>. <sup>1</sup>Nippon Del Monte Corporation, R&D, Numata, 378-0016, Japan, <sup>2</sup>Dept. Biology, Yeungnam University, Gyongsan, 712-749, Korea

Hot pepper seeds of two Korean cultivars, Manita and Buchon were sown in the greenhouse on 11th of April. Purified two Japanese attenuated CMV strains (CMV vaccines), PV1-K and PV2-K containing satellite RNAs were inoculated 21 days after sowing with cotton swabs to pepper seedlings of the cotyledon stage. The Korean virulent CMV isolate, CMV-KP was inoculated at the same time as inoculated with the vaccines. Virus symptoms appeared on leaves were scored 20 days after the inoculation. The plant height was measured 21 days after the inoculation (42 days after sowing) when the non-inoculated seedlings were ready to be transplanted to the field. Several plants from each of PV1-K, PV2-K and non-inoculated treatments (NT) were transplanted into the field to observe fruit shape and to measure the fruit weight. CMV-KP induced severe mosaic symptoms in both Manita and Buchon seedlings, whereas PV1-K and PV2-K did either mild chlorosis or mild mosaic symptoms in both cultivars. The plant height was reduced at the transplanting time 7% and 10% in PV1-K, and 11% and 14% in PV2-K, respectively, compared to that in NT. This result suggests that seedlings pre-inoculated with both PV1-K and PV2-K should be kept in

nurseries 5 to 10 days longer before transplanting in order to obtain vigorous seedlings which sustain in the field. The fruit shape seems to be not affected by the CMV vaccine treatments, however the fruit weight of Manita and Buchon was reduced 7% and 17% in PV1-K, and 13% and 17% in PV2-K, respectively, compared to that in NT. The reduction of the fruit weight and plant height may cause the reduction of yield. Cultural practices such as more fertilized application and higher plant density in the field may compensate for the yield reduction.

**J-127. Two attenuated potyviruses for cross protection against virulent isolates in cucumber.** Gug-Seoun Choi, Jae-Hyun Kim, Bong-Nam Chung, Hyun-Ran Kim, Jeong-Soo Kim, and Yong-Mun Choi. National Horticultural Research Institute, RDA, Suwon 441-440, Korea.

*Watermelon mosaic virus* (WMV) and *Zucchini yellow mosaic virus* (ZYMV) caused threatening the yield and quality of cucumber fruit, especially planted in late summer in Korea. Those viruses produced mosaic, yellowing and eventually distortion symptoms in the leaves of cucumber, and the fruits deformation by raised protuberances which made them unmarketable. To evaluate the potential of mild strains of WMV-M and ZYMV-M to protect of cucumber from severe isolates of these viruses, one was isolated from the leaf showing mild symptom in field-grown cucumber and the other was derived from a severe strain of ZYMV-C by heat treatment. In a preliminary attempt to evaluate the effectiveness of WMV-M and ZYMV-M for protecting cucumber against their respective severe strains in aphid-free greenhouse conditions, ratio of the marketable fruits was above 85% in the each trail treated with the attenuated viruses and healthy plants, and about 80% in the plants inoculated with mild strain and then challenge inoculation 2 weeks later with severe strain. However no fruit were marketable from the trails inoculated only with severe strain. In field experiments under severe epidemic conditions, increase in the marketable fruit numbers was up to 2.7 times in the protected plot by the attenuated viruses than those in the unprotected plot during about one month from the first harvest, however after the period the fruit marketability was significantly reduced.

**J-128. Multivirus protection by using recombinant antibody on tobacco.** Gun Soup Lee, Myung Hee Kwon, Eulyong Park, Hyung-Il Kim, Sukchan Lee, Ki Yoon Kim, Youn Lee, and Chanho Park. Department of Genetic Engineering, Sungkyunkwan University

Multivirus protection by using recombinant antibody on tobacco Virologists have been making numerous attempts to produce virus resistant plants by using the host plant proteins or virus proteins. Single-chain antibody fragments are recombinant proteins composed of variable domains of immunoglobulin fused by a flexible linker protein. The heavy chain variable region and light chain variable region of anti-ssDNA antibody were cloned by performing PCR in combination with designed primers. this recombinant scFv gene was cloned into plant expression vector to express in tobacco in order to study about geminivirus resistance with ssDNA genome. As the basic research for the study of bioassay about resistance in transgenic tobacco, we researched

biochemistry characterization in vitro. Theoretically, acquired protein by ssDNA specific monoclonal antibody composed of six types, which are the whole antibody, Fab, Fc and recombinant scFv, VH, VL proteins. To investigate ssDNA-catalytic affinity, ssDNA was treated with these purified proteins. Consequently, scFv protein had DNase activity. When scFv transgenic tobaccos were infected by ssDNA geminivirus that was protected to virus infection. Sufficient results were identified by southern hybridization and PCR. The investigation now being proceeded is to study virus resistance to the RNA virus. The results so far are virus protection

**J-129. Virulence differentiation of *Potato virus Y* groups occurring Solanaceae crops.** H. S. Choi<sup>1</sup>, J. W. Park<sup>1</sup>, S. H. Lee<sup>1</sup>, J. U. Cheon<sup>2</sup>, J. K. Choi<sup>3</sup> and Y. Takanami<sup>4</sup>. <sup>1</sup>National Institute of Agricultural Science and Technology(NIAST), Suwon, 441-707, Korea, <sup>2</sup>National Alpine Agricultural Experiment Station, Pyongchang, 232-950, Korea, <sup>3</sup>Department of Agricultural Biology, Kangwon National University, Chuncheon, 200-701, Korea, <sup>4</sup>Faculty of Agriculture, Graduate School of Kyushu University, Hakozaki, Fukuoka 812-8581, Japan

Two hundred eighty-one isolates of Potato virus Y (PVY) collected from red pepper, tobacco, potato, tomato, and paprika from 1999 to 2001 were classified into 4 groups (I, II, III and IV) based on biological, serological, cytopathological, and molecular properties. Group I induced vein-clearing symptoms in tobacco similar to those produced by the PVY<sub>o</sub> strain, and group II caused vein necrosis symptoms, which are typical symptoms for PVY<sub>n</sub> strain. Group III induced mosaic symptoms on tobacco, potato, tomato, and red pepper. Therefore, it did not belong to any of the previously described PVY strains. Group IV induced mosaic on tobacco, mild mosaic on tomato and red pepper, whereas no symptoms on potato. Peculiar inclusions of pinwheels, scrolls, laminated aggregates and helper components were observed in cells of *Nicotiana tabacum* cv.X-nc infected by the PVY groups III and IV. While, the inclusions of helper component were never observed in the group I and II. According to reactions to the different cultivars of *Lycopersicon esculentum*, Potato virus Y groups were divided into three groups (I, II, and III & IV). However, group I and II never infected *Capsicum annuum*, whereas group III and IV readily infected all varieties of the plant. In order to ascertain simple classification of the PVY groups, seven varieties of tomato, red pepper, and potato were selected as indicator plants based on virulence differentiation. Multiple alignment as well as cluster dendrograms of 3' noncoding region (3'-NCR) and a part of coat protein gene (CP) showed that group I belongs to the PVY<sub>o</sub> strain, group II and III to the PVY<sub>n</sub> strain, and group IV to the PepMoV. Based on analysis of the nucleotide of 3'-NCR, coat protein, and amino acid of coat protein, it seemed that Potato virus Y groups do not have any relationship in virulence differentiation and molecular phylogenesis.

**J-130. The effect of disease incidence to BaYMV by the physio-chemical property of BaYMV by the physio-chemical property of infected soil.** Jong Nae Hyun<sup>1</sup>, Yeon Kyu Hong<sup>1</sup>, Hyun Tae Kim<sup>1</sup>, Kee Do Park<sup>1</sup>, Soon Chul Kim<sup>1</sup>, and Key Woon Lee<sup>2</sup>. <sup>1</sup>National Yeongnam Agricultural Experiment Station, Milyang 627-803, Korea, <sup>2</sup>Department of Agricultural biology, Kyungpook National

University, Daegu 702-701, Korea

Barley Yellow Mosaic Virus (BaYMV) is responsible for one of the most important disease in malting Barley in Korea. It is transmitted by soil borne fungus, *Polymyza graminis*. To estimate the occurrence pattern of BaYMV by the physio-chemical property of soil collected from farmer's field, this study was conducted. The rate of disease occurrence to BaYMV investigated at 19 spots of 10th regions. The sample soil was collected by 20 pots among an infected field and analyzed the soil characters in 2002. The rate of disease occurrence to BaYMV was about 79%, 60%, 65% in Sacheon, Kosung and Hadong areas, respectively, which is malting Barley growing regions but Pohang, Yeongduk, Euesung, Gunwee, which are growing Covered Barley doesn't infected BaYMV. The tested of RT-PCR was showed that the BaYMV, BMMV were detected in Kosung and BaYMV, BMMV, SBWMV, were identified in Milyang but the others regions was infected only to BaYMV. The physio-chemical property of soil collected from infected soil show various range such as pH(4.98~8.05), EC(0.44~2.7%), OM(1.61~6.85), P2O5(58~519), K(0.22~1.82), Ca(2.3~9.7), Mg(0.5~2.6), Na(0.14~0.43). The correlation coefficient between physio-chemical property of the infected soil and infection rate shows significant to pH at 5% level. Although OM and Ca were high scores, it doesn't significant at 5% but it need to be more study in future

**J-131. Characteristics of *Bean common mosaic potyvirus* infecting mungbean in Korea.** H. S. Choi<sup>1</sup>, J. W. Park<sup>1</sup>, S. H. Lee<sup>1</sup>, J. U. Cheon<sup>2</sup>, and J. S. Kim<sup>3</sup>.  
<sup>1</sup>National Institute of Agricultural Science and Technology(NIAST), Suwon, 441-707, Korea, <sup>2</sup>National Alpine Agricultural Experiment Station, Pyongchang, 232-950, Korea, <sup>3</sup>National Horticultural Research Institute(NHRI), Suwon, 441-707, Korea

A virus causing yellow ringspot, yellow mosaic, and malformation symptoms was prevalent on mungbean plants in Korea. The causal virus was identified as bean common mosaic virus-peanut stripe strain(BCMV-PStV) and characterized based on biological, cytopathological, and molecular properties. In host range studies, the isolate caused mosaic on *Chenopodium quinoa*, *Nicotiana benthamiana*, *Phaseolus vulgaris*, and *Vicia faba*, whereas no symptoms on peanut. Virus inclusion patterns were similar to those produced by members of *Potyvirus* subdivision I with the scroll, pinwheel inclusions. In order to ascertain their taxonomic identity, The 709 nucleotide region consisting of a part of the coat protein gene and 3'-untranslated region (UTR) of the isolate was cloned and sequenced. Multiple alignment as well as cluster dendrograms showed that the isolate belongs to the BCMV-PStV subgroup.

**J-132. A novel strain of *Cucumber mosaic virus* isolated from water chickweed (*Stellaria aquatica*).** Jae-Hyun Kim<sup>1</sup>, Gug-Seoun Choi<sup>1</sup>, Jang -Kyung Choi<sup>2</sup>, Su-Young Chae<sup>1</sup>, Bong-Nam Chung<sup>1</sup>, Hyun-Ran Kim<sup>1</sup>, Jeong-Soo Kim<sup>1</sup>, and Yong-Mun Choi<sup>1</sup>. <sup>1</sup>National Horticultural Research Institute, RDA, Suwon 441-440, Korea, <sup>2</sup>Division of Biological Environment, Kangwon National University, Chunchon 200-701, Korea.

A strain of *Cucumber mosaic virus* (CMV) was isolated from a weed, water chickweed (*Stellaria aquatica*), growing in cultivated pepper field Chunchon of Korea. This isolate, Sa-CMV, was differentiated from other CMVs based on biological properties and nucleotide sequence analysis of the coat protein (CP) gene. Sa-CMV showed different reactions to all the tested plants except *Capsicum annuum* and *Cucumis sativus*, when compared with those of Mf-(subgroup I) and PaFm-CMV (subgroup II). Remarkably in *Nicotiana tabacum* cvs. Samsun, Xanthi-nc and Ky-57, Sa-CMV induced local necrotic ring spot on the inoculated leaves and wave pattern and mosaic on the upper leaves. RNA analysis, serology and RT-PCR of CP gene showed that Sa-CMV belonged to subgroup I of CMV. However, restriction enzyme analysis of the cDNA using Msp I showed that Sa-CMV was distinct from that of Mf-CMV. Based on comparison of the CP gene nucleotide and deduced amino acid sequences between other CMV strains, Sa-CMV was closely related to Mf-CMV with 93.7% and 97.2% identity, respectively.

**J-133. Identification of viruses infecting *Vicia faba* L. in Korea.** Seon Gi Jeong<sup>1</sup> and Myong Gi Chung<sup>2</sup>. <sup>1</sup>Plant Environment Division, GyeongNam Agriculture Research and Extension Services, Jinju 660-360, Korea, <sup>2</sup>Department of Biology, Gyeongsang National University, Jinju 660-701, Korea

A preliminary survey of viruses was conducted from April to May in 2001 and March to May in 2002, and 95 samples, showing virus-like Symptoms, were collected in the broad bean (*Vicia faba* L.) production areas. The 95 isolated ; 66 of Bean common mosaic virus(BCMV), 13 of Broad bean wilt virus(BBWV), 9 of coinfectd BCMV+BBWV and 7 of unkown viruses were collected from at Namhae, Gyeongnam province. To confirm seed-born transmission of those viruses, ELISA analysis was done with young broad bean leaves collected in Namhae. And then the isolates were further by electron microscope. Symptoms of the viruses on indicator plants were various. BCMV and BBWV were showing symptoms ; vein banding, vein clearing and mosaic, malformation, stunt, necrotic ring symptoms, respectively.

**J-134. Identification of *Potato virus Y* shallot isolate from shallot (*Allium ascalonicum*) in Korea.** Young-Gyu Lee<sup>1</sup>, Young-Nam Yoon<sup>1</sup>, Young-Il Hahm<sup>1</sup>, Jeong-Uk Cheon<sup>1</sup>, Su-Heon Lee<sup>2</sup>, and Key-Woon Lee<sup>3</sup>. <sup>1</sup>National Alpine Agricultural Experiment Station, RDA, Pyeongchang, 232-955, Korea, <sup>2</sup>Plant Pathology Division, NIAST, RDA, Suwon 441-707, Korea, <sup>3</sup>Department of Agricultural Biology, Kyungpook National University, Taegu 702-701, Korea

*Potato virus Y* shallot isolate (PVY-shallot) was identified from *Allium ascalonicum* (shallot) showing yellow stripe on shallot collected from Gangneung area, eastern part Korea. Electron microscopic examination of negatively stained preparation revealed that PVY-shallot were filamentous particles of 780 §— in length. Inclusion bodies including pinwheels, scrolls and laminated aggregates were observed in the cytoplasm of infected *A. ascalonicum* and *N. tabacum* cv. Samsun leaf tissues under the electron microscope. DAS-ELISA determined that the virus was serologically related to *Potato virus Y*. PVY-shallot produced vein clearing in leaves of *Nicotiana tabacum*

and systemic mosaic in *N. glutinosa*, *N. occidentales* and *N. clevelandii*. Symptom reaction on indicator plants of PVY-shallot was identical with PVY. The PVY-shallot could be detected with RT-PCR using PVY detectable primer set designed to amplify about 759 bp of the partial CP gene of PVY.

**J-135. Molecular identification of a *Carlavirus* and a *Nepovirus* from daphne plant.** K. H. Ryu and B. Y. Lee. Plant Virus GenBank, Seoul Women's University, Seoul 139-774, Korea

Natural virus infection of *Daphne odora* plants induced chlorosis and warped systemic symptoms on their leaves. There is no sequence information of *Daphne*-infecting virus so far. To detect and identify causal virus(es) in the plants, cDNA library was constructed by using in vitro polyadenylated total nucleic acids extracted from partially purified virus preparations. Totally twenty three independent cDNA clones out of 105 clones were sequenced partially or entirely. BLAST search analysis revealed that they were originated from virus origin, and they could be divided into two subgroups ; carlaviral subgroup and nepoviral subgroup. This indicates the plants were infected at least two viruses of carla- and nepovirus and the symptoms were complex. Independent cDNAs contained different regions of virus coding regions such as coat protein (CP), 11kDa, 12kDa, 25kDa and replicase. Molecular analysis of *Daphne*-infecting viruses showed that the plants contained unknown carlaviruses and nepoviruses. In the case of carlaviral cDNAs, selected 20 cDNA clones encoding CP gene was 56 to 69 % identical at the amino acid level with *Helenium virus S*, *Garlic latent virus* and *Chrysanthemum virus B*. The cDNAs of 25 kDa protein had 57 to 62 % amino acid identical with Hop latent virus. For the nepovirus, 3 cDNA clones were derived from the viral RNA2, and they had 50 to 84 % amino acid identities to *Cycas necrotic stunt virus*. Therefore, the results suggest that *Daphne* plants were infected with several complex mixture of both carlaviruses and nepoviruses.

**J-136. Genome stability and quasispecies of *Kyuri green mottle mosaic virus* populations during serial passaging in systemic hosts.** M. Y. Youn<sup>1</sup>, B. E. Min<sup>1</sup>, S. U. Kim<sup>2</sup>, W. M. Park<sup>3</sup>, and K. H. Ryu<sup>1</sup>. <sup>1</sup>Plant Virus GenBank, Division of Environmental and Life Sciences, Seoul Women's University, Seoul, 139-774, Korea, <sup>2</sup>Korea Research Institute of Bioscience and Biotechnology, Taejeon, 305-600, Korea, <sup>3</sup>Graduate School of Biotech., Korea University, Seoul 136-701, Korea.

The effects on pathogenicity and population diversity of genetically identical plant RNA virus, *Kyuri green mottle mosaic virus* (KGMMV), during infections and serial passage of systemic hosts, zucchini squash and cucumber plants, were examined. An initial target plants were inoculated with in vitro transcripts from a cloned full-length cDNA of KGMMV. This initial viral populations on cucumber and zucchini plants were transferred 5 times in parallel populations in the same host species over a period of 90 - 100 days. Viral genomic RNAs were then isolated from systemically infected leaf tissues of serial passages in the two host plants. RT-PCR products sized 386 bp long in replicase and 570 bp of coat protein genes of the virus were cloned for

sequence analysis. One hundred twenty five unique mutations of cucumber passages were detected from a total 120 clones (56,843 bases) sequenced, indicating a mutation frequency at  $2.20 \times 10^{-3}$  per nucleotide. One hundred and three unique mutations of zucchini passages were detected from a total 117 clones (55,797 bases) sequenced, demonstrating a mutation frequency at  $1.85 \times 10^{-3}$  per nucleotide. Comparison of the overall mutation frequencies in the two systemic host passages showed similar levels of population variation exist in the two hosts. Serial passages resulted in no significant change in diversity levels through 5 consecutive passages, consistent with the replicating population reaching equilibrium. Individuals in the population had a similar average genetic distance from the consensus sequences. This is the first report of quasispecies and population dynamics for cucurbit-infecting *Tobamovirus*.

**J-137. Complete nucleotide sequence and genome analysis of *Lily symptomless virus*.** S. A. Choi, B. Y. Lee, and K. H. Ryu. Plant Virus GenBank, Division of Environmental and Life Sciences, Seoul Womens University, Seoul, 139-774, Korea

*Lily symptomless virus* (LSV), a species of the genus *Carlavirus*, is the most prevalent virus infecting lily plants in the world. Viral RNA was isolated from *Lilium longiflorum* 'Georgia. Diagnostic RT-PCR using coat protein (CP) detecting primers and protein analysis by western blot revealed 32 kDa viral CP for LSV. Genomic RNA of the virus sized about 8.4kb long was used for template for construction of cDNA library in order to determine entire nucleotide sequence for LSV. For coverage of 5-end region, rapid amplification of cDNA end (RACE) technique was applied and obtained respective 500 bp cDNA clones containing the 5- terminal sequences. LSV genomic RNA consists of 8,408 nucleotide long comprising six open reading frames coding for proteins of 223 kDa (viral replicase), 25 kDa (triple gene block (TGB)-1), 12 kDa (TGB-2), 7 kDa (TGB-3), 32 kDa (CP) and 16 kDa from the 5 to 3 ends, respectively. Non coding regions of LSV were 67 bp and 49 bp nucleotides at the 5 and 3, respectively. The nucleotide sequence of the virus in the CP region was 87.3% identical with Japanese isolate and 99.3% with other isolates. Complete amino acid sequence of replicase was compared to that of other Carlaviruses (*Blueberry scorch virus*, *Hop latent virus* and *Aconitum latent virus*) and showed 45% to 68% similarities.

**J-138. Complete sequence analysis and a full-length cDNA clone construction of *Tobacco rattle virus* RNA2.** S. H. Choi<sup>1</sup>, J. K. Choi<sup>2</sup>, M. U. Chang<sup>3</sup>, and K. H. Ryu<sup>1</sup>. <sup>1</sup>Plant Virus GenBank, Department of Horticultural Science, Seoul Women's University, Seoul 139-774, Korea, <sup>2</sup>Division of Biological Environment, Kangwon National University, Chunchon 200-701, Korea, <sup>3</sup>Department of Biology, Yeungnam University, Gyongsan, 712-749, Korea

The complete nucleotide sequence of RNA2 for a gladiolus isolate of *Tobacco rattle virus* (TRV-G), a type species of the Tobravirus genus, has been determined. TRV is one of major pathogens worldwide with broad host ranges. Several strains of TRV genome information indicates RNA2 molecule is quite variable; TRV RNA2 is 2-4kb long and contains three or four open reading frame (ORF)s coding for proteins of

viral coat protein (CP) and two or three ORFs related to nematode vector transmission. TRV-G RNA2 encoded proteins for 22kDa viral CP and other 3 ORFs(23kDa, 8kDa and 16kDa proteins). TRV-G CP consisted of 603bp(201 a.a), whereas those of other known TRV strains consisted of from 591 to 633 bp. Sequences of the TRV-G RNA2 CP exhibit low similarity to those of the known TRV 8 strains; 39.3 to 99.5 % at the amino acid level. Nucleotide sequence of 3NTR was more conserved than that of 5NTR. Strains of TRV could be divided into 3 subgroups(subgroup I, II and III) based on the phylogenetical analysis. Phylogenetic tree analyses of the coding regions and NTRs demonstrated that TRV-G was clustered together with Oregon strain (TRV-ORY) into the subgroup III, and may be originated from TCM strain. Full-length cDNA of TRV-G RNA2 was directly amplified by RT-PCR using the 5'-end primer containing a SP6 RNA promoter and virus-specific 3'-end primer and cloned into the pUC19. Capped in vitro transcripts from the RT-PCR amplicons as well as from the full-length clone are under investigation for their infectivity. Infectious transcript system TRV RNA2 can be useful for plant-TRV interaction study as well as foreign material transport study.

**J-139. Completed sequence of a Korean isolate of *Barley yellow mosaic virus*.**

Min Kyung Choi<sup>1</sup>, Kui Jae Lee<sup>1</sup>, Young-Hoon Lee<sup>2</sup>, Hyung-Moo Kim<sup>1</sup>, and Wang Hyu Lee<sup>1</sup>. <sup>1</sup>Faculty of Biology Resource Science Chonbuk National University, Jeonju, 561-756, Korea, <sup>2</sup>Rural Development Research Management, Bureau Research Planning Division, Suwon 441-707, Korea

The complete sequence of RNA of an isolate of barley yellow mosaic virus(BaYMV-HN) from Haenam Province Korea were determined. The sequences resembled those of an isolate from Japan(99.6% identical nucleotides for RNA1; 99.4% for RNA2) more closely than one from Germany (93.6 and 90.4% respectively). BaYMV-HN had two RNAs, RNA1(7.6Kb) and RNA2(3.5Kb). This virus contains one large open reading frame(ORF) which is open at the 5' end and terminates with a UAA stop codon at position 3538. Two RNAs, RNA1(7.6Kb) and RNA2(3.5Kb) of BaYMV-HN isolate was compared with four BaYMV strains, BaYMV-J from Japan, BaYMV-C from China, BaYMV-G from Germany and BaYMV-UK from United Kingdom. Molecular differences among strains were compared with sequence RNA regions coding for the CP(RNA1) and the N-terminus of the P2 Protein(RNA2). The 5' end of the coat protein (CP) (RNA1) among strains show nucleotid sequence homology in the range of 95% to 100% and the N-terminus of the P2 protein (RNA2) among strains show nucleotid sequence homology in the range of 88% to 100%. The P2 fragment was more variable than the CP and phylogenetic analysis of both regions showed the Asian and European isolates formed distinct clusters.

**J-140. Completed nucleotide sequence and genomic organization of *Barley mild mosaic virus* Korean strain (BaMMV-Kor).**

Min Kyung Choi<sup>1</sup>, Kui Jae Lee<sup>1</sup>, Jeong Chul Park<sup>2</sup>, Jae Hwan Seo<sup>2</sup>, and Wang Hyu Lee<sup>1</sup>. <sup>1</sup>Faculty of Bioresources Science, Chonbuk National University, Jeonju 561-756, Korea, <sup>2</sup>National Honam Agricultural Experiment Station, Iksan 570-080, Korea

The 5' -terminal half of RNA 1 and whole of RNA 2 of barley mild mosaic



bymovirus Kor strain(BaMMV-Kor)were sequenced to give together with the published data its complete genomic sequence. BaMMV-Kor RNA1 and RNA2 consist of 7263 and 3516 nucleotides, excluding the 3' poly A tails, respectively. RNA 1 encodes a single large polyprotein of 2258 amino acids(Mr 256K), containing eight putative functional proteins, and RNA 2 also encodes one polyprotein of 891 amino acids(Mr 98K), containing two functional proteins. These functional proteins are arranged in the same manner as in RNA amino acid sequence homology(25~28%)exists between the proteins of the two viruses. The BaMMV-Kor proteins showed less amino acid sequence homology(18~32%)with the corresponding proteins of potyviruses or rymoviruses than with those of BaYMV. Comparisons of the BaMMV-Kor proteins with the corresponding proteins of other partially sequenced BaMMV isolates show identity between the 3' non-coding regions(NCRs) in RNA 1 or RNA 2 of BaMMV-Kor and other BaMMV isolates, but only 68~72% identity between the 5' NCRs in RNA 2 of BaMMV-Kor and other BaMMV isolates.

**J-141. Biological and molecular characterization of *Pepper mottle virus* isolated from paprika.** G. S. Choi<sup>1</sup>, J. Y. Yoon<sup>2</sup>, J. H. Kim<sup>3</sup>, J. K. Choi<sup>3</sup>, and K. H. Ryu<sup>2</sup>. <sup>1</sup>National Horticultural Research Institute, Suwon 441-440, Korea, <sup>2</sup>Division of Environmental and Life Sciences, Seoul Women's University, Seoul 139-774, Korea, <sup>3</sup>Division of Biological Environment, Kangwon National University, Chunchon 200-701, Korea.

*Pepper mottle virus* (PepMoV) was isolated from paprika (*Capsicum annum* var. *grossum*) plants in Hwasung and Kyungki province in Korea. Symptoms of PepMoV was divided largely in two groups which are vein banding (Vb) and vein clearing (Vc) patterns, respectively, on the diseased paprika leaves. In host range tests, PepMoV-Vb1 was showed a typical mosaic symptom to the upper leaves of *Nicotiana tabacum* cv. White Burley and Ky57 plants, but PepMoV-Vc1 was revealed necrotic spots on the inoculated leaves of the plants. A set of primers was designed to detect PepMoV and RT-PCR products for the two isolates were cloned and their sequences were determined. Nucleotide sequences of two PepMoV isolates showed over 96% identities with the known PepMoV California strain (PepMoV-C). Full-length cDNAs of PepMoV-Vb and -Vc were synthesized by RT-PCR and their genome structure was analysed by RFLP analysis. Here we discussed the biological properties and molecular characteristics of PepMoV-Vb and -Vc from paprika and genome structures of full-length cDNAs of the two PepMoV isolates were compared to that of PepMoV-C.

**J-142. Molecular characterization of pear necrotic leaf spot virus and its fungus vector in Korea.** Hyekyung Shim<sup>1</sup>, Hyeongjin Jee<sup>2</sup>, Jiyeon Kim<sup>1</sup>, Honggeun Lee<sup>1</sup>, Soyeon Kim<sup>1</sup>, and Sukchan Lee<sup>1</sup>. <sup>1</sup>Department of Genetic engineering, Sungkyunkwan University, Suwon, 440-746, Korea, <sup>2</sup>Plant Pathology Division, National Institute of Agricultural Society and Technology, Rural Development Administration, Suwon, 441-707, Korea

Pear necrotic leaf spot virus (PNLSV) belongs to *Capillovirus* genus and infects pome fruits. Symptoms occur on PNLSV-infected pear leaves as reddish brown and

irregularly necrotic spots. PNLSV is transmitted by grafting and mechanical inoculation into susceptible hosts but natural vector of PNLSV has not been reported. But we have made observations on the spread of SDV in the field. Transmission occurs from tree-to-tree and tree-to susceptible herbaceous plants along but not across ditches in the field. Based on these observations, we consider it likely that PNLSV is soil-borne. In order to ascertain this possibility, various fungi were isolated and cultured from PNLSV-infected plants and nearby there and obtained 79 isolates. By means of RNA dot-blot hybridization, 5 isolates were sorted out and one of them was used for bioassay. Fungi infected kidney bean also showed typical symptoms and viral coat protein gene was detected. It was identified that this fungal vector belongs to *Phenicillium* by morphological and molecular markers. For the purpose of certification of transmission mode, fungal and host plant genomic DNA was amplified using specific primer sets. The N-terminal part, partial replicase region and coat protein gene of virus genome was amplified in both genomic DNA. Regardless of virus infection, interestingly, viral genome parts were detected in several host plants and fungi.

**J-143. Characterization of *Melon necrotic spot virus* isolated from muskmelon.**  
Gug-Seoun Choi, Jae-Hyun Kim, Su-Young Chae, Hyun-Ran Kim, Bong-Nam Chung, Jung-Su Kim, and Yong-Mun Choi. National Horticultural Research Institute, RDA, Suwon 441-440, Korea.

A severe disease of muskmelon (*Cucumis melo* cv. Alsnight), grown on rockwool in plastic house, was characterized by leaf and stem necrosis followed by death of the plants. In 2001, an isolate of *Melon necrotic spot virus*-MM (MNSV-MM) of the genus Carmovirus was identified as the casual agent of the disease on the basis of biological reactions and partial nucleotide sequence analyses of coat protein (CP) gene. MNSV-MM induced necrotic local lesions on the mechanically inoculated leaves and systemic necrotic spots on the upper leaves of melon cvs. Alsnight, Rui<sup>2</sup>, Party, Imperial, and Seolhang. However, watermelon and cucumber showed only necrotic lesions on the inoculated leaves. DsRNAs extracted from the melon infected with MNSV-MM were separated into three components. Molecular sizes of the dsRNAs were estimated at approximately 4.5, 1.8, and 1.6kbp. The amplified DNA products of MNSV-MM by RT-PCR showed approximately 1.2kbp. The DNA was digested to three fragments by Msp<sup>1</sup> treatment. Complementary DNA copies of the genomic RNA of MNSV-MM were cloned and the region deduced to encode the CP was sequenced. The putative CP coding region, located near 3' end of the genome, consisted of 1,170 nucleotides and had the potential to encode a 390 amino acid protein. The nucleotide and amino acid sequence of MNSV-MM CP gene were 84.0 to 94.6% and 90.8 to 94.9% identical to other MNSV isolates of GeneBank Database, respectively. The occurrence of MNSV is reported for the first time in Korea.

**J-144. Several properties of an isolate of *Dasheen mosaic virus* isolated from *Colocasia esculenta* Schott in Korea.** Soon Bae Kwon<sup>1</sup>, He Jin Jeong<sup>2</sup>, Ju Yeon Yoon<sup>3</sup>, Su Jeong Heo<sup>1</sup>, Ki Hyun Ryu<sup>3</sup>, and Jang Kyung Choi<sup>2</sup>. <sup>1</sup>Reginal Crop Experiment Station, Gangwon Province Agricultural Research and Extension Service, Chuncheon, <sup>2</sup>Division of Biological Environment, Kangwon

National University, Chunchon 200-701, Korea, <sup>3</sup>Plant Virus GenBank, Division of Environmental and Life Sciences, Seoul Women's Univ. Seoul.

Taro (*Colocasia esculenta* Schott) originated in India and adjacent areas of southeast Asia and is now widely cultivated in Asia and Oceania. In some Pacific Coast countries, this crop is one of the main sources of starchy food. In Korea, the crop has been cultivated constantly over 4,000 tons/year. In this study, *Dasheen mosaic virus* (DsMV-Kr), causing mosaic symptoms was isolated from taro, and its properties were characterized. The local lesion symptom was found in inoculated leaves of *Tetragonia expansa* and systemic mosaic symptoms on *Zantedeschia aethiopica*, while 11 species of indicator plants did not infect. In an electron microscope, DsMV particles were long filamentous type, with about 850 nm in length and 12 nm in width. Molecular weight of the viral CP was about 34 kDa in SDS-PAGE. Gel double diffusion test revealed that DsMV was not serologically related to *Zantedeschia mosaic virus* (ZaMV) which has been reported an Araceae-infecting another potyvirus. The coat protein (CP) gene of DsMV(-Kr) was cloned and their nucleotide sequences were determined. The sequence of the 3'-terminal 1,199 nucleotides, which consisted of the 3'-terminus of the partial nuclear inclusion body (Nib), coat protein (CP) genes and 3'-non coding region. Similarity at the nucleotide and amino acid levels of CP and 3'-NCR were from 51.0 to 91.5% and from 49.1 to 93.9%, respectively, when compared with those of other potyviruses. Phylogenetic analysis of selected potyviral CP sequences indicates that the virus is an isolate of DsMV and distinguishable from other potyviruses. These results indicate a new strain of DsMV, and the name Kr strain of DsMV is hereby proposed.

**J-145. A novel strain of *Cucumber mosaic virus* isolated from water chickweed (*Stellaria aquatica*).** Jae-Hyun Kim<sup>1</sup>, Gug-Seoun Choi<sup>1</sup>, Jang Kyung Choi<sup>2</sup>, Soo-Young Chae<sup>1</sup>, Bong-Nam Chung<sup>1</sup>, Hyun-Ran Kim<sup>1</sup>, Jeong-Soo Kim<sup>1</sup>, and Yong-Mun Choi<sup>1</sup>. <sup>1</sup>National Horticultural Research Institute, RDA, Suwon 441-440, Korea, <sup>2</sup>Division of Biological Environment, Gangwon National University, Chunchon 200-701, Korea.

A strain of *Cucumber mosaic virus* (CMV) was isolated from a weed, water chickweed (*Stellaria aquatica*), growing in cultivated pepper field Chunchon of Korea. This isolate, Sa-CMV, was differentiated from other CMVs based on biological properties and nucleotide sequence analysis of the coat protein (CP) gene. Sa-CMV showed different reactions to all the tested plants except *Capsicum annuum* and *Cucumis sativus*, when compared with those of Mf-(subgroup I) and PaFm-CMV (subgroup II). Remarkably in *Nicotiana tabacum* cvs. Samsun, Xanthi-nc and Ky-57, Sa-CMV induced local necrotic ring spot on the inoculated leaves and wave pattern and mosaic on the upper leaves. RNA analysis, serology and RT-PCR of CP gene showed that Sa-CMV belonged to subgroup I of CMV. However, restriction enzyme analysis of the cDNA using Msp I showed that Sa-CMV was distinct from that of Mf-CMV. Based on comparison of the CP gene nucleotide and deduced amino acid sequences between other CMV strains, Sa-CMV was closely related to Mf-CMV with 93.7% and 97.2% identity, respectively.