O-13. Identification and Differentiation of Cucumber Mosaic Virus Isolated from *Hydrangea macrophylla* for. *otaksa* Using PCR Techniques. Sun Jung Park[1]. Ju Hee Bang[1], Sang Yong Lee[1], Jang Kyung Choi[2] and Keum Hee Lee[3]. Department of Forest Resources Protection, Kangwon National University, Chunchon 200-701, Korea. [2]Department of Agricultural Biology, Kangwon National University, Chunchon 200-701, Korea. [3]Division of Overseas Pests, Department of Crop Protection, NIAST, RDA, Suwon 441-707, Korea.

An isolate of cucumber mosaic virus (CMV) was isolated from *Hydrangea macrophylla* for. *otaksa* (Sieb. et Zucc.) Wils. showing mosaic symptom, designated as CMV-Hm. Reverse transcription and polymerase chain reaction (RT-PCR) techiniques were used to identification and differentiation of CMV-Hm. RT-PCR used by two set of 20-mer primers, one was CMV-common primers and another was CMV subgroup I-specific primers designed in a conserved region of the 3' end of CMV RNA3, amplified about 490bp and 200bp DNA fragments from CMV-Hm, respectively. Restriction enzyme anaylsis of RT-PCR products using *Eco*RI and *Msp*I showed that CMV-Hm belonged to CMV subgroup I. But, CMV-Hm could be differentiated from other CMV subgroup I isolates using RNA fingerprinting by arbitrarily primed polymerase chain reaction (RAP-PCR) and single-stranded conformational polymorphism (SSCP).

O-14. Induction of Mild Strain CymMV and ORSV in Orchids. Dong Soo Jung, Bong Jin Koo and Moo Ung Chang. Dept. of Biology, Yeungnam University, Kyungsan 712-749, Korea.

Cymbidium Mosaic Virus (CymMV) and Odontoglossum Ringspot Virus (ORSV) variants which induced mild or symptomless pattern were selected to lessen virus diseases and to minimize damage in orchid cultivation. Plant sap was extracted from orchid infected with ORSV (Chang, et al., 1991) or CymMV (Chang, et al., 1991). After mutagen treatment like sodium nitrate, high temperature and ultraviolet, they were inoculated on *Chenopodium amaranticolor*. It was done repeatly 3-4 times with mild strain viruses to pick out genetically stable strains. Variants were chosen according to the procedure in virus-free *Phalenopsis* spp. and *Cymbidium* spp. prepared from tissue culture. Genome of the most mild strain ORSV and CymMV among variants was amplified by RT-PCR(Reverse Transcription-Polymerase Chain Reaction) with primers which were designed to refer to CymMV-K2(Koo, et al., 1998) and ORSV-Cy Korean isolate (Ryu, et al., 1995), respectively. They were cloned by pGEM-easy vetor and sequenced ABI prism 377 model (Perkin-Elmer) for comparision with CymMV-K2 and ORSV-Cy Korean isolate.

O-15. Amplification of Three Species of Cucumoviruses with A Genus-Specific Primers. Seung Kook Choi[1], Ki Hyun Ryu[2], Hae Ja Kim[3], Jang Kyung Choi[3] and Won Mok Park[1]. [1]Graduate School of Biotechnology, Korea University, Seoul 136-701, Korea. [2]Dept. Hort. Science, Seoul Women's University, Seoul 139-774, Korea. [3]Division of Biological Environment, Kangwon National University, Chunchon 200-701, Korea.

The pair of cucumovirus genus-specific primers that flank the coat protein gene were designed and used to amplify a DNA fragment of approximately 938 - 966 bp. The reverse transcription and polymerase chain reaction (RT-PCR) was used for detection and identification of three cucumoviruses (cucumber mosaic virus, CMV; peanut stunt virus, PSV; tomato aspermy virus, TAV) in various plants sources with a pair of primers, designed as CPTALL-3 and CPTALL-5. The primers specifically amplified the target size of DNA fragment in all the test cucumoviruses (CMV IA, IB and II, PSV and TAV). No DNA product of any length was produced when brome mosaic virus or tobacco mosaic virus RNA was used as templates. We could differentiate all the tested cucumoviruses by comparing PCR-restriction fragment length polymorphism with 6 different enzymes. This indicates that the designed primers are only specific for the cucumoviruses and useful for reliable information of identification of members of the *Cucumovirus* genus.