primer pairs: ToCV-CP-F/ToCV-CP-R for amplifying the CP gene, and To-Hsp-F/To-Hsp-R (5'-GTCGACAATAATTAGCGGTCC-3'/5'-TTATTACT AATGGGCCGGACTC-3', unpublished) for amplifying the Hsp70 homologous gene. The amplicons with expected sizes were obtained from the inoculated plants but not the controls. The amplicon of the Hsp70 homologous gene was sequenced and submitted to GenBank (KX900413). The infection of some solanaceous (tomato, pepper, eggplant) and cucurbit (pumpkin) crops with ToCV has been previously reported in China (Sun et al. 2016; Zhao et al. 2013). Although ToCV can experimentally infect three leguminous plants: pea (Pisum sativum), dwarf French bean (Phaseolus vulgaris), and broad bean (Vicia faba) (Morris et al. 2006), no natural leguminous host of ToCV has been reported. To our knowledge, this is the first report of cowpea as a natural host of ToCV.

References:

Morris, J., et al. 2006. Eur. J. Plant Pathol. 114:265. Sun, X. H., et al. 2016. Plant Dis. 100:2543. Zhao, L. M., et al. 2015. J. Phytopathol. 162:627. Zhao, R. N., et al. 2013. Plant Dis. 97:1123. e-Xtra

First Report of physostegia chlorotic mottle virus on Tomato (Solanum lycopersicum) in Germany. Y. Z. A. Gaafar, Institute for Epidemiology and Pathogen Diagnostics, Julius Kühn-Institut, 38104 Braunschweig, Germany; M. A. M. Abdelgalil, Institute for Epidemiology and Pathogen Diagnostics, Julius Kühn-Institut, 38104 Braunschweig, Germany; and Faculty of Science, Fayoum University, Fayoum, Egypt; D. Knierim, Leibniz Institute DSMZ, German Collection of Microorganisms and Cell Cultures, Braunschweig, Lower Saxony 38124, Germany; K. R. Richert-Pöggeler, Institute for Epidemiology and Pathogen Diagnostics, Julius Kühn-Institut, 38104 Braunschweig, Germany; W. Menzel and S. Winter, Leibniz Institute DSMZ, German Collection of Microorganisms and Cell Cultures, Braunschweig, Lower Saxony 38124, Germany; and H. Ziebell, Institute for Epidemiology and Pathogen Diagnostics, Julius Kühn-Institut, Messeweg 11-12, 38104 Braunschweig, Germany. Y. Z. A. Gaafar and M. A. M. Abdelgalil contributed equally. This research was funded by a grant from the Federal Office of Food and Agriculture within the Euphresco network "2015-F-172: The application of Next-Generation Sequencing technology for the detection and diagnosis of non-culturable organisms: viruses and viroids". Y. Z. A. Gaafar is supported by the German Egyptian Research Long Term Scholarship (GERLS). Plant Dis. 102:255, 2018; published online as https://doi.org/10.1094/PDIS-05-17-0737-PDN. Accepted for publication 7 August 2017.

In September 2015, a tomato sample collected in the German state of Hesse was sent to the Julius Kühn-Institut for analysis. While the fruits showed marbling and discoloration, the leaf samples from this plant did not show any obvious symptoms. Transmission electron microscopy (TEM) revealed the presence of bullet-shaped virus particles indicating the presence of a rhabdovirus. However, immunosorbent electron microscopy using antiserum JKI-1073 for Eggplant mottled dwarf virus (EMDV) could not confirm EMDV infection. The virus was mechanically transmitted to Nicotiana benthamiana, N. clevelandii, and Chenopodium quinoa inducing yellowing and leaf deformation, while mechanical transmission to N. occidentalis (P1 and 37b) failed. Extraction of double stranded-RNA (dsRNA) followed by random-PCR (Froussard 1992), cloning of PCR products, and sequencing failed to reveal any virus sequences. Total RNA was extracted from infected N. benthamiana, followed by ribo-depletion, library preparation and submission for next-generation sequencing (NGS) using an Illumina MiSeq platform as described by Knierim et al. (2017). De novo assembly of the trimmed reads was done with Geneious v 10.1.3 (Biomatters LTD, NZ). Using MEGA BLAST, 13 contigs showed between 95.6 and 98.5% similarity with physostegia chlorotic mottle virus (PhCMoV) isolate PV-1182 (accession no. KX636164). The complete PhCMoV genome (13,321 nt length) was assembled by mapping reads to this reference genome and used to design PhCMoV-specific RT-PCR primers for detection (HZ-343 5'-CGGTGAGTGGGGCAACTAAT-3'/HZ-344 5'-AGCGATGGGGTCTAGTGTCT-3'). RT-PCR confirmed the presence of PhCMoV in the test plants resulting in amplicons of approximately 875 bp. In August 2016, similar symptoms on tomato fruits were observed by a different grower in Hesse. The presence of PhCMoV was confirmed by TEM and RT-PCR. Additionally, the PCR products were sequenced and showed 97% identity to KX636164. Surprisingly, reanalysis of a tomato sample from 2003 that was infected by a hitherto unknown rhabdovirus using NGS also confirmed infection with PhCMoV. This sample also originated from Hesse although the original grower is unknown. The complete genome of the 2003 PhCMoV sample was assembled following the same methods described above. Pairwise comparison between the genomes of 2015 and 2003 isolates resulted in 99.7% nucleotide identity and 96.9% when compared with KX636164. These findings indicate the presence of PhCMoV in tomato in

Germany for a long time albeit isolated occurrences in different production areas. PhCMoV was recently identified from Physostegia virginiana plants showing leaf deformation and severe chlorotic and mottle symptoms in Austria (Menzel et al. 2016). However, it is not known if there is a link between PhCMoV isolates infecting P. virginiana and tomato as the routes of transmission and dissemination are currently unknown. The sequences from this report were deposited in GenBank (accession nos. KY706238 and KY859866 [full-length sequences], KY882263 and KY882264 [partial sequences]). To our knowledge, this is the first host record of PhCMoV in tomato and a new country record for Germany.

References:

Froussard, P. 1992. Nucleic Acids Res. 20:2900. Knierim, D., et al. 2017. Arch. Virol. 162:291. Menzel, W., et al. 2016. Julius-Kühn-Archiv 454:283.

First Report of Grapevine geminivirus A in Diverse Vitis Species in Korea. Y. Jo and H. Choi, Department of Agricultural Biotechnology, College of Agriculture and Life Sciences, Seoul National University, Seoul, 08826, Republic of Korea; M. K. Song, J. S. Park, and J. W. Lee, Grape Research Institute, Chungbuk Agricultural Research and Extension Services, Okcheon, 29017, Republic of Korea; and W. K. Cho, Department of Agricultural Biotechnology, College of Agriculture and Life Sciences, Seoul National University, Seoul, 08826, Republic of Korea. Y. Jo, H. Choi, and M. K. Song contributed equally to this work. This work was partially supported by the National Research Foundation of Korea (NRF) grant funded by the Korea government Ministry of Education (No. NRF-2017R1C1B1007027) and the support of the "Cooperative Research Program for Agriculture Science & Technology Development" (Project No. PJ01186102) conducted by the Rural Development Administration, Republic of Korea. Plant Dis. 102:255, 2018; published online as https://doi.org/10.1094/PDIS-05-17-0771-PDN. Accepted for publication 25 August 2017.

Grape (Vitis sp.) is a popular fruit crop and host for a large number of viruses. So far, more than 70 different viruses have been identified from grapevines (Martelli 2014). Recently, grapevine geminivirus A (GGVA; family Geminiviridae) has been identified from two table grape (Vitis vinifera) cultivars, Black Beet and Nagano Purple (Al Rwahnih et al. 2017). Moreover, GGVA has been identified from grapevines in China (Fan et al. 2017). In 2016, we collected five grapevine leaf samples from 16 individual plants representing 12 different cultivars. All 15 different grapevine plants were grown at the Grape Research Institute, Okcheon, Korea, and Shine Muscat (V. labruscana Bailey  $\times$  V. vinifera) was obtained from a vineyard in Okcheon. Shine Muscat displayed leaf malformations, vein clearing, and yellowing, while the other cultivars did not display any observable disease symptoms. Sixteen different libraries for RNA sequencing without pooling were prepared and subjected to paired-end sequencing (2 × 100 bp) using Illumina's HiSeq 2000 followed by bioinformatics analyses, as described previously (Jo et al. 2016). Of 16 plants, 27 GGVA-associated contigs were identified from eight plants representing six cultivars: 188-08 (V. monticola × V. riparia) (two plants). Shine Muscat (two plants), Jaok (Vitis hybrid), Jungrang (Vitis hybrid), Jarang (V. vinifera), and Cabernet Sauvignon (V. vinifera). The numbers of reads associated with GGVA were 67 (188-08, #1 plant), 146 (188-08, #2 plant), 36 (Cabernet Sauvignon), 296 (Jaok), 118 (Jarang), 492 (Jungrang), 419 (Shine Muscat, #1 plant), and 171 (Shine Muscat, #2 plant). To confirm the results of RNA sequencing, we obtained five GGVA complete genome sequences (GenBank MF163261-65) from five cultivars by PCR using two abutting primer pairs (GGVAv950/GGVAc961 and GGVAv1402/GGVAc1438) and recovered the full GGVA genome, which was followed by Sanger sequencing (Al Rwahnih et al. 2017). The GGVA isolates 188-08 and Jaok shared 99% identity with isolate Tamar (KX618694.1), while isolates Cabernet Sauvignon, Jarang, and Shine Muscat shared 99% identity with known GGVA isolates: Koshu Sanjaku (KX570617.1), Scolokertek Kiralynoje (KX570618.1), and Black Beet (KX570609.1), respectively. The possible presence of GGVA in the other eight plants representing six cultivars—Okrang (Vitis hybrid), Campbell Early (V. labruscana), Chungporang (Vitis hybrid), Alicante Bouschet (V. vinifera), Cabernet franc (V. vinifera), and Chardonnay (V. vinifera)—was tested by PCR. PCR was conducted for 16 plants using GGVA-specific primers for amplifying the coat protein region of 414 bp in size, GGVA-1370F1 5'-TGTAAAGAGATCAGCCCAAATGTTTTC-3' (position 1,370 to 1,396), and GGVA-1783R1 5'-ATGCAATTTTCGTCTCCCTGCA-3' (position 1,783 to 1,762) based on the GGVA reference genome (NC\_031340.1). The PCR results demonstrated that all 16 plants were infected by GGVA, while RNA sequencing revealed the presence of GGVA in eight plants. Although a previous study reported a low rate of GGVA infection in the U.S.A. (Al Rwahnih et al. 2017), a previous study showed that GGVA was widely present in grapevines in China (Fan et al. 2017). Our results demonstrated that GGV can infect diverse Vitis