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PHYLOGENETIC ANALYSIS OF CYMBIDIUM MOSAIC AND ODONTOGLOSSUM RINGSPOT VIRUSES ISOLATED FROM *PHALAENOPSIS* SP.

Cymbidium mosaic virus and Odontoglossum ringspot virus are the most common and widespread viruses in ornamental orchids. Infections caused by these viruses can lead to a decrease in the phenotypic diversity of orchid collections, and as a result – to their complete depletion. The aim of the study is to determine the origin of Ukrainian isolates of orchid viruses in the collection of protected soil from O.V. Fomin Botanical Garden of Taras Shevchenko National University of Kyiv. The properties of nucleotide and amino acid sequences of the coat proteins (CP) of Cymbidium mosaic virus (CymMV) and of Odontoglossum ringspot virus (ORSV) were investigated. RNAs of CymMV and ORSV were isolated from leaves of Phalaenopsis sp. collected from A.V. Fomin Botanical Garden of Taras Shevchenko National University of Kyiv, amplified through RT-PCR and sequenced. Obtained sequences were compared at nucleotide and amino acid levels with CymMV and ORSV isolates available in the GenBank. ORSV isolated in Ukraine shared 96-99 % and 93,4-98 % CP similarity to other known ORSV isolates at nucleotide and amino acid levels, respectively. CymMV isolated in Ukraine revealed approximately 77-97 % similarity for nucleotide sequences and 84-100 % for amino acid sequences to isolates from the GenBank. Phylogenetic analysis showed that studied ORSV and CymMV isolates may have common origin with some South Korean isolates.

Keywords: orchids, CymMV, ORSV, RT-PCR, phylogenetic analysis.

Introducton. Tropical and subtropical species of orchids are cultivated in many countries and remain one of the most important commercial crops in the world's floral industry. Viral diseases of orchids are known from the middle of XX century. These may significantly affect the quality of ornamental flowers, and, consequently, decrease their commercial and esthetical value. Currently, about 50 viruses infecting orchids has been described [1, 2]. Among them, Cymbidium mosaic virus (CymMV, Potexvirus) and Odontoglossum ringspot virus (ORSV, Tobamovirus) are the most prevalent [3, 4], as they can be transmitted rapidly, contributing to a significant spread among susceptible plants [5]. These viruses are transmitted exclusively in artificial ecosystems, mainly during vegetative propagation or mechanically. Viral infections affect physiological state of orchids grown in greenhouses [5]. Causing a loss of flowers' ornamentation, they can have negative impacts on developing of commercial floriculture [6, 7, 8].

In Ukraine, tropical orchids are mostly grown in greenhouses in the collections of botanical gardens. Cases of CymMV and ORSV infection among tropical orchids in the collections of botanical gardens of Ukraine have been known previously [9, 10], but phylogenetic analysis of detected CymMV and ORSV isolates have not been carried out.

The aim of the study is to determine the origin of Ukrainian isolates of orchid viruses in the collection of protected soil from O.V. Fomin Botanical Garden of Taras Shevchenko National University of Kyiv.

Materials and Methods. To investigate the molecular properties of CymMV and ORSV isolated in Ukraine, samples of *Phalaenopsis* sp. were collected from A.V. Fomin Botanical Garden of Taras Shevchenko National University of Kyiv. Virus identification was carried out using DAS-ELISA with polyclonal antisera specific to CymMV and ORSV (Prime Diagnostics, The Netherlands).

Total RNA was extracted using RNeasy Plant Mini Kit (Qiagen, Germany).

Reverse transcription polymerase chain reaction (RT-PCR) was accomplished using SuperScript II kit (Invitrogen, USA). To amplify coat protein (*CP*) gene of CymMV and ORSV, duplex RT-PCR was used in one reaction tube with the two pairs of primers: CymMV-F – 5'-ACAATAATTTGAAATAATCATGGGA-3', CymMV-R – 5'-AAAACCACACGCCTTATTAAGTTTG-3' (expected product size 716 bp); ORSV-F – 5'-ACGCACAATCTGATTCGTATTGAA-3', ORSV-R – 5'-TATCAACGTTATTTTCCTAAATAT-3' (expected product size 528 bp) [11].

The amplified products from *Phalaenopsis* sp. samples were purified using MinElute Gel Extraction Kit (Qiagen, Great Britain). Resulting PCR products were sequenced using Applied Biosystems 3730x1 DNA Analyzer using Big Dye terminators, version 3.1 (Applied Biosystems, USA).

Phylogenetic analysis was carried out using MEGA 9 software.

Results and Discussion. Orchid plants from collection of A.V. Fomin Botanical Garden of Taras Shevchenko National University of Kyiv with symptoms of mosaic, spots, necrosis and chlorosis have been detected. Symptoms of mosaic on the leaves were an alternation of light and dark green areas, which formed a mosaic pattern. Symptoms of necrotic or annular spotting are the formation of small (0.5–2 mm) and medium-sized (2–4 mm) black necrosis of round and oval shape on the leaf blade, sometimes surrounded by a narrow border (Fig. 1). A fairly common symptom was yellowing of the leaf blade in combination with the formation of marginal leaf necrosis. Symptoms of mosaic, yellowing and ring necrosis can occur on plants under the influence of CymMV and ORSV [12, 13, 14].

CymMV and ORSV were detected by ELISA with appropriate antisera in all selected samples of orchid plants. It should be noted that mixed infection with CymMV + ORSV was detected in plants of *Phalaenopsis* sp.

Further leaves of *Phalaenopsis* sp. with the symptoms typical of CymMV and ORSV infection (Fig. 1) confirmed by DAS-ELISA were used for total RNA extraction.

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Fig. 1. Leaf of *Phalaenopsis sp.* with symptoms of chlorosis and necrosis

The total RNA was amplified in RT-PCR. Agarose gel electrophoresis revealed the presence of expected amplified products with corresponding molecular weight (Fig. 2).

Gene sequence of Ukrainian ORSV isolate (ORSV-PHA-Ukr) were compared with another known isolates of ORSV available from the GenBank. For this study, we used published 32 full-genomic sequences of ORSV.

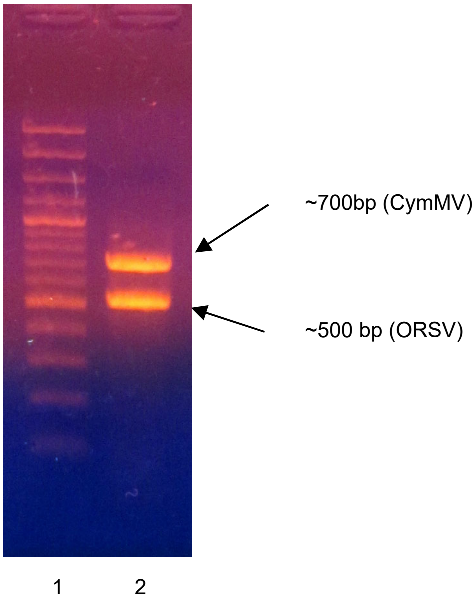

Fig. 2. Electrophoretic separation of RT-PCR products in agarose gel: 1 – DNA ladder (100 bp, Fermentas), 2 – products of amplification (arrows indicate cDNAs of CymMV and ORSV CPs)

The nucleotide and amino acid sequences of the CP gene/protein of ORSV-PHA-Ukr shared, respectively, 96-99 % and 93,4-98 % similarity with the other ORSV isolates. Our results are in line with literature data stating high homology of ORSV isolates [15, 16].

Since full genomic sequences of ORSV isolates were used for the analysis, we were able to compare CP gene sequence of ORSV-PHA-Ukr with that of other isolates (Table 1).

Table 1. Comparison of partial nucleotide and amino acid sequences of ORSV isolated in Ukraine with other virus isolates available from the GenBank

GenBank isolates	CP sequence	
	Nucleotide sequence, %	Amino acid, %
ORSV-SW8/Cymbidium (Australia)	98,7	97,4
ORSV-Taiwan-1 (Taiwan)	99,2	98,7
ORSV-Taiwan-2(Taiwan)	98,3	98,1
ORSV-Cy-1 (Japan)	98,7	98,1
ORSV-Cymbidium (USA)	99,2	98,7
ORSV-CR (USA)	99,2	98,7
ORSV-S1 (Singapore)	98,9	98,7
ORSV (Korea)	97,4	94,8

As shown in Table 1, the degree of CP gene similarity of ORSV-PHA-Ukr in comparison with other ORSV isolates from GenBank is quite high. This demonstrates high level of conservancy of ORSV genome (or at least its *CP* gene).

To determine possible origin of ORSV isolate sampled in Ukraine, phylogenetic analysis based on its amino acid sequence was conducted. Maximum Likelihood method was chosen as a discrete method, which was performed using Poisson model. As can be seen from the resulting phylogenetic tree (Fig. 3), ORSV-PHA-Ukr isolate and one of the isolates from the South Korea were clustered together suggesting their possible common origin.

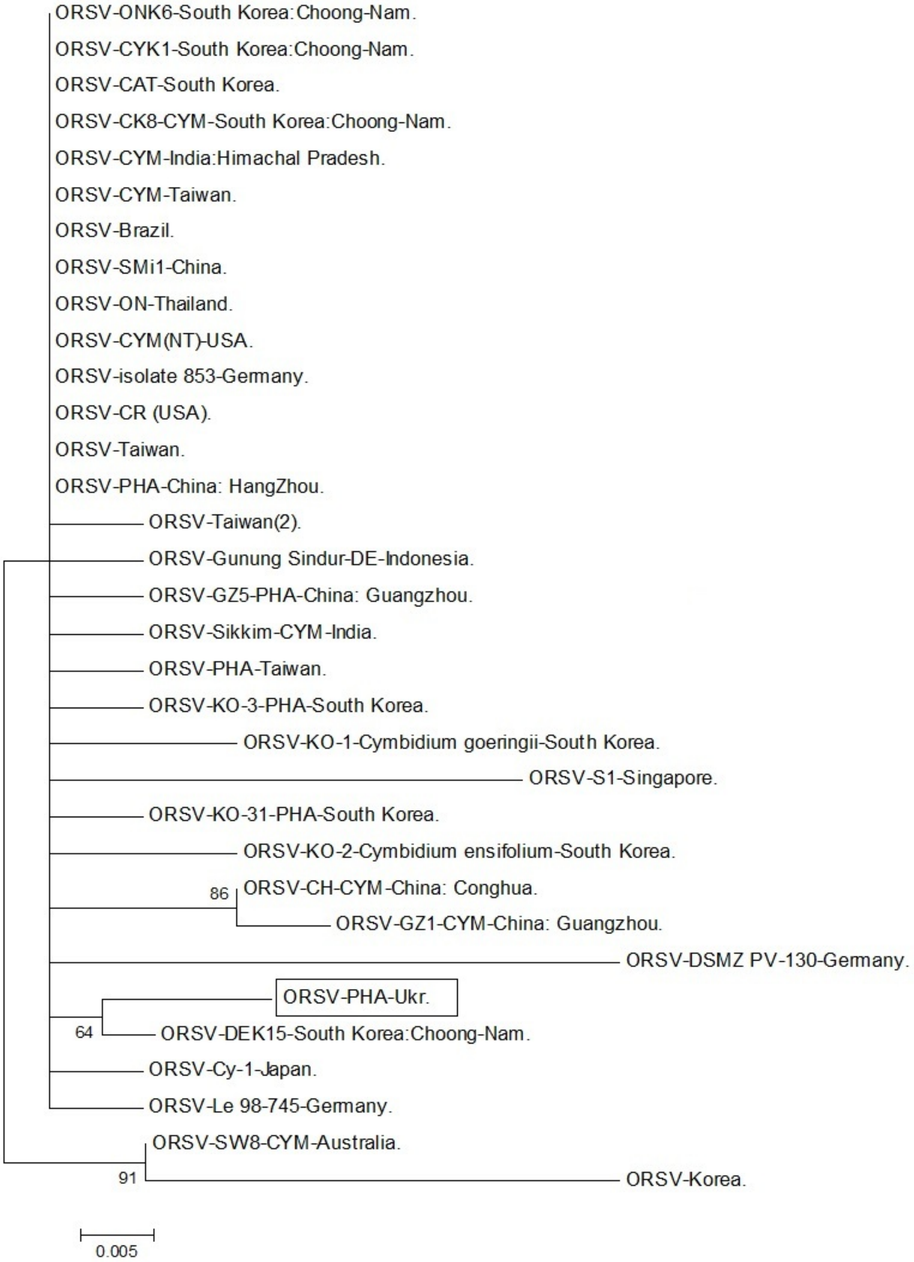

Fig. 3. Phylogenetic analysis of Ukrainian ORSV isolate based on amino acid sequence of coat protein. Phylogenetic tree (50 % majority rule consensus tree) is constructed using ML method (Poisson model, bootstrap 1000 replications)

The obtained nucleotide sequence of *CP* gene of CymMV isolated in Ukraine (CymMV-PHA-Ukr) was pairwise compared with the other 33 CymMV isolates from the GenBank. Basing on the mutations in monomorphic sites, CymMV isolates can be divided into two monophyletic clusters at the nucleotide level – subgroup A and subgroup B [17]. According to our data and basing on the nucleotide sequence, CymMV-PHA-Ukr isolate belongs to subgroup A and shares 88-97 % similarity with other CymMV isolates from subgroup A and 77-87 % with the CymMV isolates from subgroup B. The deduced amino acid sequence of CymMV-PHA-Ukr shared 84-100 % similarity with other CymMV isolates.

For the further research of CymMV-PHA-Ukr phylogenetic tree of nucleotide sequence was constructed using ML method. As shown on Fig. 4, CymMV isolate sampled in Ukraine may have common origin with some isolates from the South Korea.

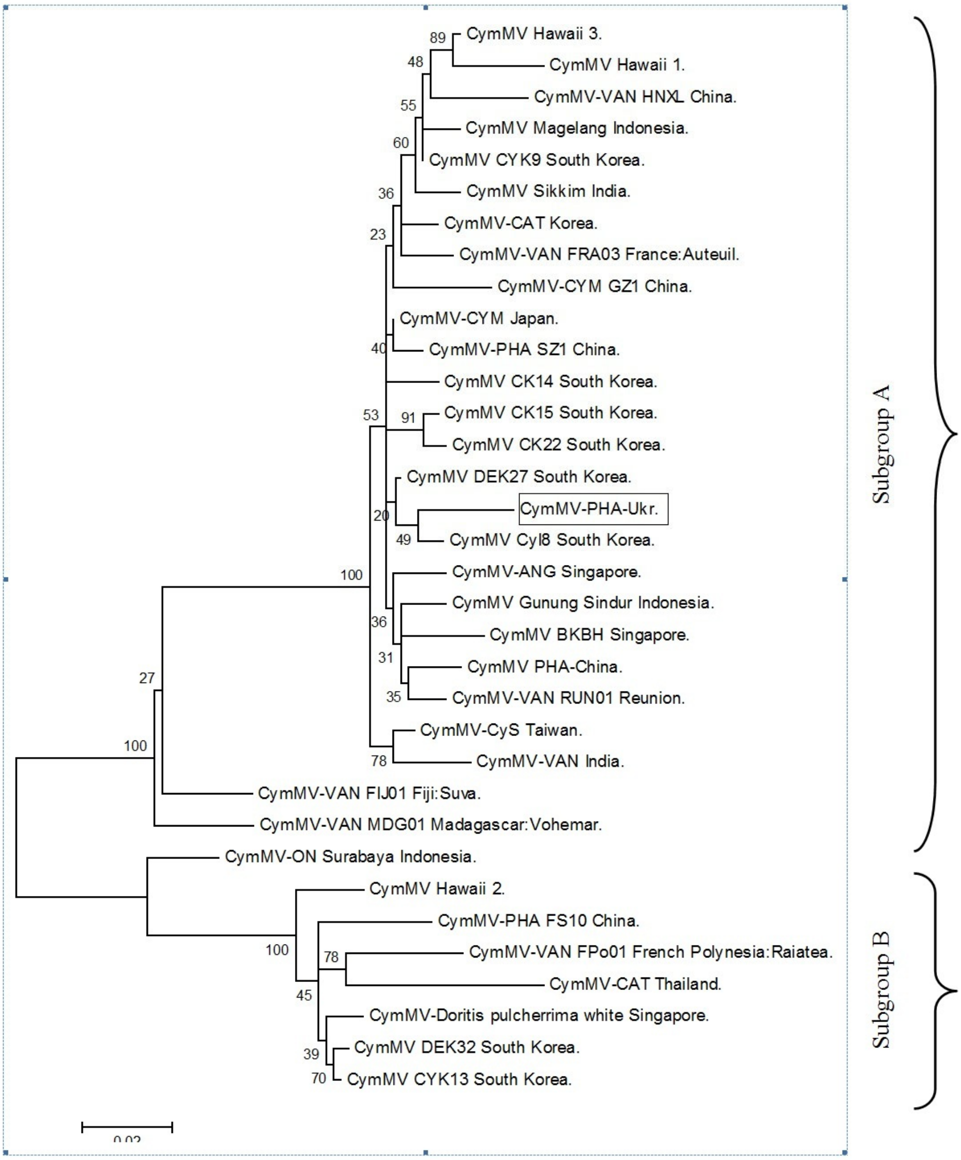

Fig. 4. Phylogenetic analysis of Ukrainian CymMV isolate based on nucleotide sequence of coat protein gene. The tree is constructed using ML method (Kimura 2-parameter model, bootstrap 1000 replications)

In conclusion, it was established that ORSV isolate collected from *Phalaenopsis* sp. in Ukraine shared high homology at the nucleotide and amino acid levels with other known ORSV isolates basing on CP gene/protein sequence. CymMV isolate sampled from orchids in Ukraine showed 77-97 % similarity to the virus isolates available from the GenBank. Isolates of both viruses (ORSV-PHA-Ukr and CymMV-PHA-Ukr) supposedly have common ancestors with the ORSV and CymMV isolates from the South Korea. Since the plants from tropical orchids' collections in botanical gardens in Ukraine were originally imported from eastern countries, we suggest that ORSV and CymMV may have invaded orchid collections with imported plant material, with subsequent spread in Ukraine.

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References

1. Viruses of orchids and their control / F. W. Zettler, G. C. Wistler, M. S. Elliot et al. // Plant. Disease. – 1990. – Vol. 74. – P. 621-626.

2. Lee Ch.-Hw. The Orchid-Infecting Viruses Found in the 21st Century [Electronic resource] / Ch.-Hw. Lee, Y.-X. Zheng, F.-J. Jan // Orchid Biotechnology III. – 2017. – P. 145-164. – Available from : https://doi.org/10.1142/9789813109223_0009

3. Bratsch S. A. Incidence of cymbidium mosaic, odontoglossum ringspot, and orchid fleck virus in orchids in Minnesota and production of antibodies for use in ELISA to detect orchid fleck virus / S. A. Bratsch, N. Olszewski, B. Lockhart // European Journal of Plant Pathology. – 2021. – Vol. 159, № 3. – P. 543-554.

4. First Report of Orchid Fleck Virus Associated with Citrus Leprosis Symptoms in Rough Lemon (Citrus jambhiri) and Mandarin (C. reticulata) the United States [Electronic resource] / A. Olmedo-Velarde, A. Roy, Ch. Padmanabhan et al. // Plant Disease, Sep. – 2021. – Available from : https://doi.org/10.1094/PDIS-12-20-2736-PDN

5. Cánovas S. E. First report of Cymbidium mosaic virus and Odontoglossum ring spot virus in Argentina [Electronic resource] / S. E. Cánovas, M. C. Ballari, C. F. Nome // Australasian Plant Dis. Notes. – 2016. – Vol. 11, № 2. – Available from : http://dx.doi.org/10.1007/s13314-015-0189-7.

6. Lopez-Hernandez M. S. Detection of Cymbidium mosaic virus (CymMV) and Odontoglossum ringspot virus (ORSV) from orchids in Mexico [Electronic resource] / M. S. Lopes-Hernandez, P. E. Palacios-Popo, R. De La Torre-Almaraz // Agrociencia [online]. – 2014. – Vol. 48, № 5. – P. 525-536. – Available from : https://www.redalyc.org/pdf/302/30232500006.pdf

7. Production of virus-free orchid Cymbidium aloifolium (L.) Sw. by various tissue culture techniques [Electronic resource] / Sh. Pradhan, T. Regmi, M. Ranjit, B. Pant // Heliyon. – 2016. – Vol. 2, № 10. – Article № e00176. – Available from : https://doi.org/10.1016/j.heliyon.2016.e00176.

8. Chung B.-N. Viral infection of tissue cultured orchids and evaluation of damages / B.-N. Chung, J.-Y. Yoon, M. S. Kim // Plant. Pathol. J. – 2010. – Vol. 26, № 2. – P. 194-197.

9. Korotieieva H. V. [Viral diseases in the orchid collection of Academician O.V. Fomin Botanical Gardens of Taras Shevchenko Kyiv National University] / H. V. Korotieieva, V. P. Polischuk // Scientific Notes of NaUKMA. – 2001. – Vol. 19, Part 2. – P. 399-400.

10. Spreading of virus infection in the orchid collection in Ukraine / V. Polischuk, G. Korotyeyeva, A. Bysov, A. Lavrentieva // Plant science. – Sofia, 2007. – Vol. XLIV, № 3. – P. 213-216.

11. Rapid detection of cymbidium mosaic virus by the polymerase chain reaction (PCR) / S. T. Lim, S. M. Wong, C. Y. Yeong et al. // J. Virol Methods. – 1993. – Vol. 41, № 1. – P. 37-46.

12. Pearson M. N. Further Observations on the Effects of Cymbidium Mosaic Virus and Odontoglossum Ringspot Virus on the Growth of Cymbidium Orchids [Electronic resource] / M. N. Pearson, J. S. Cole // Journal of Phytopathology. – Vol. 131, № 3. – P. 193-198. – Available at : https://doi.org/10.1111/j.1439-0434.1991.tb01187.x

13. RT-PCR detection of Odontoglossum ringspot virus, Cymbidium mosaic virus and Tospoviruses and association of infections with leaf-yellowing symptoms in Phalaenopsis [Electronic resource] / K. Yamane, K. Oyama, E. Iuchi et al. // Journal of Phytopathology. – 2008. – Vol. 156. – P. 268–273. – Available from : http://dx.doi.org/10.1111/j.1439-0434.2007.01352.x

14. Hu J. S. Detection of Cymbidium Mosaic Virus, Odontoglossum Ringspot Virus, Tomato Spotted Wilt Virus, and Potyviruses Infecting Orchids in Hawaii [Electronic resource] / J. S. Hu, S. Ferreira, M. Q. Xu // Department of Plant Pathology. – University of Hawaii, 1993. – P. 464-468. – Available from : https://www.apsnet.org/publications/PlantDisease/BackIssues/Documents/1993Articles/PlantDisease77n05_464.PDF

15. Yoon J.-Y. High sequence conservation among Odotoglossum ringspot virus isolates from orchids / J.-Y. Yoon, B.-N. Chung, S.-K. Choi // Virus Genes. – 2011. – Vol. 42, № 2. – P. 261-267.

16. The complete sequence of a Singapore isolate of odontoglossum ringspot virus and comparison with other tobamoviruses / C. G. Chng, S. M. Wong, P. H. Mahtani et al. // Gene. –1996. – Vol. 171, № 2 . – P. 155-161.

17. Evidence that Cymbidium mosaic virus (CymMV) isolates divide into two subgroups based on nucleotide diversity of coat protein and replicas genes / M. Moles, H. Delatte, K. Farreyrol et al. // Arch. Virol. – 2007. – Vol. 152. – P. 705-715.

References (Scopus)

1. Zettler FW, Wistler GC, Elliot MS, Ko N-J. Viruses of orchids and their control, Plant. Disease; 1990; 74: 621-626.

2. Chia-Hwa Lee, You-Xiu Zheng, and Fuh-Jyh Jan The Orchid-Infecting Viruses Found in the 21st Century Orchid Biotechnology III. March 2017; 145-164 :https://doi.org/10.1142/9789813109223_0009

3. Bratsch SA, Olszewski N, Lockhart B. Incidence of cymbidium mosaic, odontoglossum ringspot, and orchid fleck virus in orchids in Minnesota and production of antibodies for use in ELISA to detect orchid fleck virus. European Journal of Plant Pathology. 2021; 159 (3): 543-554.

4. Olmedo-Velarde A, Roy A, Padmanabhan Ch, Nunziata S., Nakhla MK and Melzer MJ First Report of Orchid Fleck Virus Associated with Citrus Leprosis Symptoms in Rough Lemon (Citrus jambhiri) and Mandarin (C. reticulata) the United States.Plant Disease . Sep. 2021. https://doi.org/10.1094/PDIS-12-20-2736-PDN

5. Cánovas SE, Ballari MC and Nome CF. First report of Cymbidium mosaic virus and Odontoglossum ring spot virus in Argentina. Australasian Plant Dis. Notes. 2016. 11: 2. http://dx.doi.org/10.1007/s13314-015-0189-7.

6. Lopez-Hernandez MS, PALACIOS-POPO P.E., DE LA TORRE-ALMARAZ R [Detection of Cymbidium mosaic virus (CymMV) and Odontoglossum ringspot virus (ORSV) from orchids in Mexico]. Agrociencia [online]. 2014, vol.48, n.5, pp.525-536. ISSN 2521-9766.

7. Pradhan Sh, Regmi T, Ranjit M, Pant B, Production of virus-free orchid Cymbidium aloifolium (L.) Sw. by various tissue culture techniques, Heliyon, Volume 2, Issue 10, 2016, e00176, ISSN 2405-8440, https://doi.org/10.1016/j.heliyon.2016.e00176.

8. Chung B-N, Yoon J-Y., Kim MS. Viral infection of tissue cultured orchids and evaluation of damages, Plant. Pathol. J., 2010; 26(2); 194-197.

9. Korotieieva HV, Polischuk VP. [Viral diseases in the orchid collection of Academician O.V. Fomin Botanical Gardens of Taras Shevchenko Kyiv National University] [Scientific Notes of NaUKMA]. 2001;19,(2): 399-400. Ukrainian

10. Polischuk V, Korotyeyeva G, Bysov A, Lavrentieva A Spreading of virus infection in the orchid collection in Ukraine. Plant science. Sofia. 2007; XLIV (3). : 213-216.

11. Lim S.T., Wong S.M., Yeong C.Y., Lee S.C., Goh CJ. Rapid detection of cymbidium mosaic virus by the polymerase chain reaction (PCR). J Virol Methods. 1993 Jan; 41(1):37-46. doi: 10.1016/0166-0934(93)90161-j. PMID: 8432762.

12. Pearson MN., Cole JS Further Observations on the Effects of Cymbidium Mosaic Virus and Odontoglossum Ringspot Virus on the Growth of Cymbidium Orchids, Journal of Phytopathology, 2008; 31 (3):193-198. Available at: https://doi.org/10.1111/j.1439-0434.1991.tb01187.x

13. Yamane K, Oyama K, Iuchi E, Ogawa H, Suzuki T and Natsuaki T. RT-PCR Detection of Odontoglossum ringspot virus, Cymbidium mosaic virus and Tospoviruses and Association of Infections with Leaf-Yellowing symptoms in Phalaenopsis. Journal of Phytopathology 2008; 156: 268–273. http://dx.doi.org/10.1111/j.1439-0434.2007.01352.x

14. Hu JS, Ferreira S, Xu MQ, Detection of Cymbidium Mosaic Virus, Odontoglossum Ringspot Virus, Tomato Spotted Wilt Virus, and Potyviruses Infecting Orchids in Hawaii, Department of Plant Pathology. 1993:464-468. https://www.apsnet.org/publications/PlantDisease/BackIssues/Documents/1993Articles/PlantDisease77n05_464.PDF

15. Yoon JY, Chung BN, Choi SK. High sequence conservation among Odotoglossum ringspot virus isolates from orchids, Virus Genes, 2011; 42(2): 261-267.

16. Chng CG, Wong S M, Mahtani P H, Loh C S, Goh C J, Kao M C, Chung M C, Watanabe Y. The complete sequence of a Singapore isolate of odontoglossum ringspot virus and comparison with other tobamoviruses. Gene, 1996; 171: 155–161

17. Moles M, Delatte H, Farreyrol K, Grisoni M. Evidence that Cymbidium mosaic virus (CymMV) isolates divide into two subgroups based on nucleotide diversity of coat protein and replicas genes, Arch. Virol, 2007; 152: 705-715.

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ФІЛОГЕНЕТИЧНИЙ АНАЛІЗ ІЗОЛЯТІВ ВІРУСІВ МОЗАЇКИ ЦИМБІДІУМУ ТА КІЛЬЦЕВОЇ ПЛЯМИСТОСТІ ОДОНТОГЛОСУМУ, ВИДІЛЕНИХ ІЗ ОРХІДНИХ *PHALAENOPSIS* SP.

Віруси мозаїки цимбідіуму та кільцевої плямистості одонтоглосуму є найбільш патогенними та широко поширеними вірусами в колекціях декоративних орхідних. Інфекції, спричинені цими вірусами, можуть призвести до зниження видового різноманіття колекцій і, як наслідок, їх повного виснаження. Метою дослідження було визначення походження українських ізолятів вірусів орхідних у колекції захищеного ґрунту Ботанічного саду імені О. В. Фоміна Київського національного університету імені Тараса Шевченка. Вивчено нуклеотидні та амінокислотні послідовності гена капсидного білка (CP) вірусу мозаїки цимбідіуму (CymMV) та CP вірусу кільцевої плямистості одонтоглосуму (ORSV). Ці послідовності були отримані в результаті ампліфікації і сиквенування РНК CymMV та ORSV, виділених із інфікованих рослин Phalaenopsis sp. колекції захищеного ґрунту Ботанічного саду ім. О. В. Фоміна Київського національного університету імені Тараса Шевченка. Отримані сиквенси порівнювали з послідовностями відомих ізолятів CymMV та ORSV із ГенБанку. Було встановлено, що досліджуваний ізолят ORSV має подібність з іншими ізолятами 96–99 % та 93,4–98 % для CP на нуклеотидному й амінокислотному рівнях, відповідно. Виділений ізолят CymMV мав ступінь подібності з іншими ізолятами CymMV 77–97 % для нуклеотидних та 84–100 % – для амінокислотних послідовностей. Побудоване філогенетичне дерево показало, що обидва досліджувані віруси, CymMV та ORSV, мають спільне походження з деякими корейськими ізолятами.

Ключові слова: орхідні, CymMV, ORSV, ЗТ-ПЛР, філогенетичний аналіз.

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ФИЛОГЕНЕТИЧЕСКИЙ АНАЛИЗ ВИРУСОВ МОЗАИКИ ЦИМБИДИУМА И КОЛЬЦЕВОЙ ПЯТНИСТОСТИ ОДОНТОГЛОССУМА, ВЫДЕЛЕННЫХ ИЗ ОРХИДНЫХ *PHALAENOPSIS* SP.

Вирусы мозаики цимбидиума и кольцевой пятнистости одонтоглоссума являются наиболее патогенными и широко распространенными вирусами в коллекциях декоративных орхидных. Инфекции, вызванные этими вирусами, могут привести к снижению видового разнообразия коллекций и, как следствие, их полному истощению. Целью исследования было определение происхождения украинских изолятов вирусов орхидных в коллекции защищенного грунта Ботанического сада имени А. В. Фомина Киевского национального университета имени Тараса Шевченко. Было проведено исследование нуклеотидных и аминокислотных последовательностей гена капсидного белка (СР) вируса мозаики цимбидиума (CymMV), а также СР вируса кольцевой пятнистости одонтоглоссума (ORSV). Данные последовательности были получены в результате амплификации и сиквенирования РНК CymMV и ORSV, выделенных из растений Phalaenopsis sp. коллекции Ботанического сада им. А. В. Фомина Киевского национального университета имени Тараса Шевченко. Полученные сиквенсы сравнивали с последовательностями других известных изолятов CymMV и ORSV из ГенБанка. Было показано, что исследуемый изолят ORSV имеет подобие с другими изолятами 96–99 % и 93,4–98 % для гена СР на нуклеотидном и аминокислотном уровнях, соответственно. Выделенный изолят CymMV был подобен другим изолятам CymMV на 77–97 % для нуклеотидных и на 84–100 % – для аминокислотных последовательностей. Построенное филогенетическое дерево показало, что исследуемые вирусы, CymMV и ORSV, имеют общее происхождение с некоторыми корейскими изолятами.

Ключевые слова: орхидные, CymMV, ORSV, ОТ-ПЦР, филогенетический анализ.