INTRODUCTION. 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), Odontoglossum ringspot virus (ORSV), and Odontoglossum mosaic virus (CymMV, Potexvirus) are the most prevalent [3, 4], as they can be transmitted rapidly, contributing to a significant spread among susceptible plants [5]. 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. 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.
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.

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-ST1 (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.
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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Терміново серед навчальних колективів та науковців Київського національного університету імені Тараса Шевченка відбувається розповсюдження нових і вірусних інфекцій, які можуть призвести до зниження видового різноманіття, які навчальні колективи та науковці Київського національного університету імені Тараса Шевченка, а також до зниження видового різноманіття університету.

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ФІЛОГЕНЕТИЧЕСКИЙ 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 мозаики 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 (CP) вируса мозаики цимбидиума (CymMV), а также CP вируса кольцевой пятнистости одонтоглоссума (ORSV). Данные последовательности были получены в результате амплификации и секвенирования РНК CymMV и ORSV, выделенных из растений Phalaenopsis sp. коллекции Ботанического сада им. А. В. Фомина Киевского национального университета имени Тараса Шевченко. Полученные секвенсы сравнивали с последовательностями других известных изолятов CymMV и ORSV из ГенБанка. Было показано, что исследуемый изолят ORSV имеет подобие с другими изолятами 96–99 % для нуклеотидных и 93,4–98 % для гена CP 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, OT-ПЦР, филогенетический анализ.