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Identification of Virus Infection on Native Orchids in Liwa Botanical Garden

Mahfut¹, H Shafira², S Wahyuningsih³, T T Handayani⁴, Sukimin⁵

^{1,3,4} Department of Biology, Faculty of Mathematics and Natural Sciences, University of Lampung, Jl. Sumantri Brojonegoro no 1, Bandar Lampung, Indonesia

² Graduate School of Biology, University of Lampung, Jl. Sumantri Brojonegoro no 1, Bandar Lampung, Indonesia

⁵ Liwa Botanical Garden. Way Mengaku, Balik Bukit, Lampung Barat, Lampung, Indonesia

email: mahfut.mipa@fmipa.unila.ac.id¹, haninshafira954@gmail.com²,
wahyu6125@yahoo.co.id³, tundjungtripenihandayani@fmipa.unila.ac.id⁴,
sukimin81@gmail.com⁵

Abstract. Liwa Botanical Garden is one of the regional botanical gardens in Indonesia with the theme "Indonesian Ornamental Plants". At the present, disease infections are still a major obstacle in efforts to preserve and develop the potential natural orchids. Based on previous research, it is known that some individual orchids exhibit symptoms of viral disease infections, namely mosaic, chlorotic, streak, and necrosis. This research was conducted to determine the identification of against virus infections in Liwa Botanical Garden through a collection of samples that showed symptoms of infection, analysis of disease symptoms, and analysis of the level of disease resistance. The results showed the response of native orchids in Liwa Botanical Garden to viral infections showed symptoms in the form of necrotic black and chlorotic patches, yellowing leaves on orchids *Coelogyne* sp., *Flickingeria* sp., *Calanthe* sp., *Trixspermum centipeda*, *Bulbophyllum* sp. The type of orchid that shows the most symptoms is *Flickingeria* sp. a total of 8 samples. The type of natural orchid in the Liwa Botanical Garden which is most vulnerable to being infected with a virus is *Flickingeria* sp. with a disease intensity of 42%, while the type of orchid most infected virus is *Coelogyne* sp. with an intensity of 15%. The results of this activity are expected to be basic information in efforts to protect plants against diseases to support the application of conservation of natural orchids in the Liwa Botanical Garden.

Keyword: identification, virus infection, orchid virus, natural orchid, liwa botanical garden.

1. Introduction

Liwa Botanical Garden is one of the regional botanical gardens in Indonesia with the theme "Indonesian Ornamental Plants". This botanical garden is located in Pekon Kubu Prabu, Liwa, West Lampung with an area of 86.68 ha. Liwa Botanical Garden was opened in 2017 under Forestry Service which is focused on the collection of Indonesian ornamental plants, one of which is natural orchids [1;14]. Orchids have a variety of flower colors such as red, white, yellow, orange, flame red, dark red,



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pink, and other color combinations. According to botanists from 30 thousand species of natural orchids in the world, 5,000 of them are found in Indonesia [2].

Until now, infectious diseases are still a major obstacle in efforts to preserve and develop the potential of natural orchids. Based on previous research [10] it is known that some individual orchids exhibit symptoms of viral disease infections, namely mosaic, chlorotic, streak, and necrosis. [3;4;5;6;7;8;9;11;12;13] reported that orchid plants were able to be infected with more than 25 types of viruses. *Odontoglossum ringspot virus* (ORSV) and *Cymbidium mosaic virus* (CymMV) are the types of viruses that are reported to infect the most and have the widest spread in the world. Viral infections cause a decrease in plant vigor and flower quality resulting in economic losses [6;11]. Based on this, it is necessary to conduct research related to "Identification of Disease and Efforts to Protect Natural Orchid Plants Against Virus Infection in the Liwa Botanical Garden" to determine the response of natural orchids to viral infections as well as the types of natural orchids that are most susceptible to virus infections in the Liwa Botanical Garden. The results of this activity are expected to be basic information in efforts to protect plants against diseases to support the application of conservation of natural orchids in the Liwa Botanical Garden.

2. Materials and Methods

2.1. Sample collection

Sample collection was carried out on orchid leaves in Liwa Botanical Garden which showed symptoms of being infected with a virus. All samples were tabulated and documented with photos.

2.2. Analysis of Disease Symptoms

This analysis is done by matching photos with the literature that has been previously reported. The literature used is [3;4;5;6;8;12].

2.3. Disease Intensity Analysis

This stage is done to determine the severity of the disease (disease severity). Disease intensity is the proportion of infected hosts to the total observed surface area of the host. Calculation of disease intensity analysis is done using the method and the scale of disease intensity refers to [10].

2.4. Disease Intensity Analysis

Determination of the level of resistance of orchids to disease follows the method of [10].

3. Results and Discussion

3.1. Sample collection

The sample collection was conducted from January 2nd week to February 2nd week 2020. Samples were randomly selected from orchid individuals that showed virus infection in the green house of the Liwa Botanical Garden. Based on the results of the collection obtained 16 samples of 5 types of orchids, namely *Coelogyne* sp., *Flickingeria* sp., *Calanthe* sp., *Trixspermum centipeda*, *Bulbophyllum* sp. The type of orchid that is most infected with the disease is *Flickingeria* sp. with a total of 8 samples. Overall data collection results are shown in **Table 1**.

Table 1. Collection of orchid leaf samples at Liwa Botanical Garden which shows a virus infection.

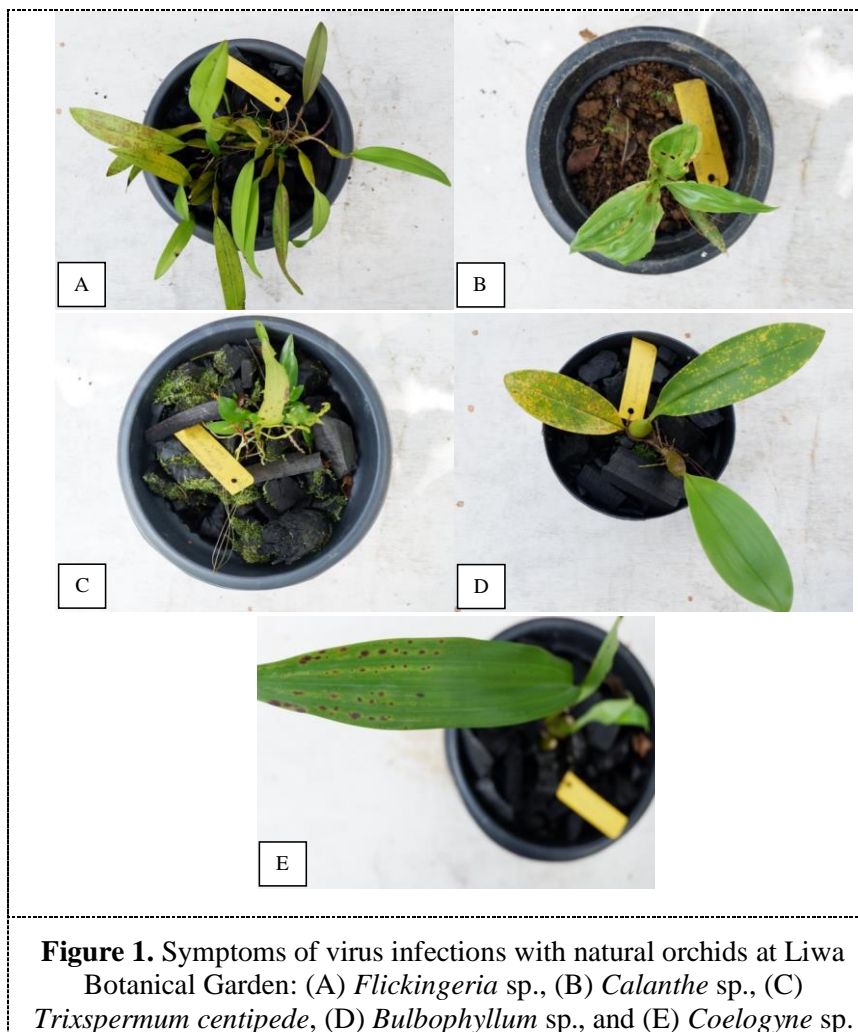
Species of Orchid	Number of Samples	Symptoms of Infection
<i>Coelogyne</i> sp.	2	Necrotic and chlorotic
<i>Flickingeria</i> sp.	8	Necrotic and streak
<i>Calanthe</i> sp.	2	Necrotic and chlorotic
<i>Trixspermum centipeda</i>	2	Mosaic
<i>Bulbophyllum</i> sp.	2	Necrotic and streak

The selection of sampling locations is based on previous research by [10] reported viral infection in the collection of natural orchids at Liwa Botanical Garden in August and December 2019. The types of infected orchids were *Calanthe*, *Cymbidium*, and *Dendrobium* with symptoms of necrosis, chlorotic and streak. Other studies have also reported ORSV infections in *Phalaenopsis amboinensis* (KRB2) and *Phalaenopsis amabilis* (KRB12) from Bogor Botanical Gardens, *Phalaenopsis amabilis* (KRP18) and *Dendrobium salacence* (KRP20) from Purwodadi Botanical Gardens, and *Phalaenopsis modesta* J. Sm. (KRBp5) from the Balikpapan Botanic Garden [7]. ORSV can infect a variety of orchids including *Aranda* sp., *Dendrobium* sp., *Phalaenopsis* sp., and *Grammatophyllum* sp. Usually leaves arise in circles, lines, yellowish green or brown spots [3]. Furthermore [10] also reported the presence of CymMV virus infection in the orchid *Phalaenopsis* sp. and *Dendrobium* sp. in Java and Bali, there are symptoms of necrosis.

In this study, *Flickingeria* sp. is the type of orchid most infected with viral diseases. This shows that this type of orchid is the host most susceptible to infection with pathogenic viral diseases.

3.2. Analysis of Disease Symptoms

Based on the analysis of disease symptoms, viral infections in orchid samples of *Coelogyne* sp., *Flickingeria* sp., *Calanthe* sp., *Trixspermum centipeda*, *Bulbophyllum* sp. showing symptoms of viral infections that are necrosis, streak, and chlorotic. Overall samples showing symptoms of viral infection are presented on **Fig 1**.



On *Coelogyne* sp., showing symptoms of necrosis in the form of black patches on the leaves, which is different from the orchid *Flickingeria* sp., *Calanthe* sp., and *Trixspermum centipeda* which have symptoms of necrosis and chlorotic namely yellowing leaves. Whereas in *Bulbophyllum* sp. the symptoms shown are chlorotic. Overall symptoms of viral diseases that infect natural orchids in the Liwa Botanical Garden are presented on **Fig. 2**.

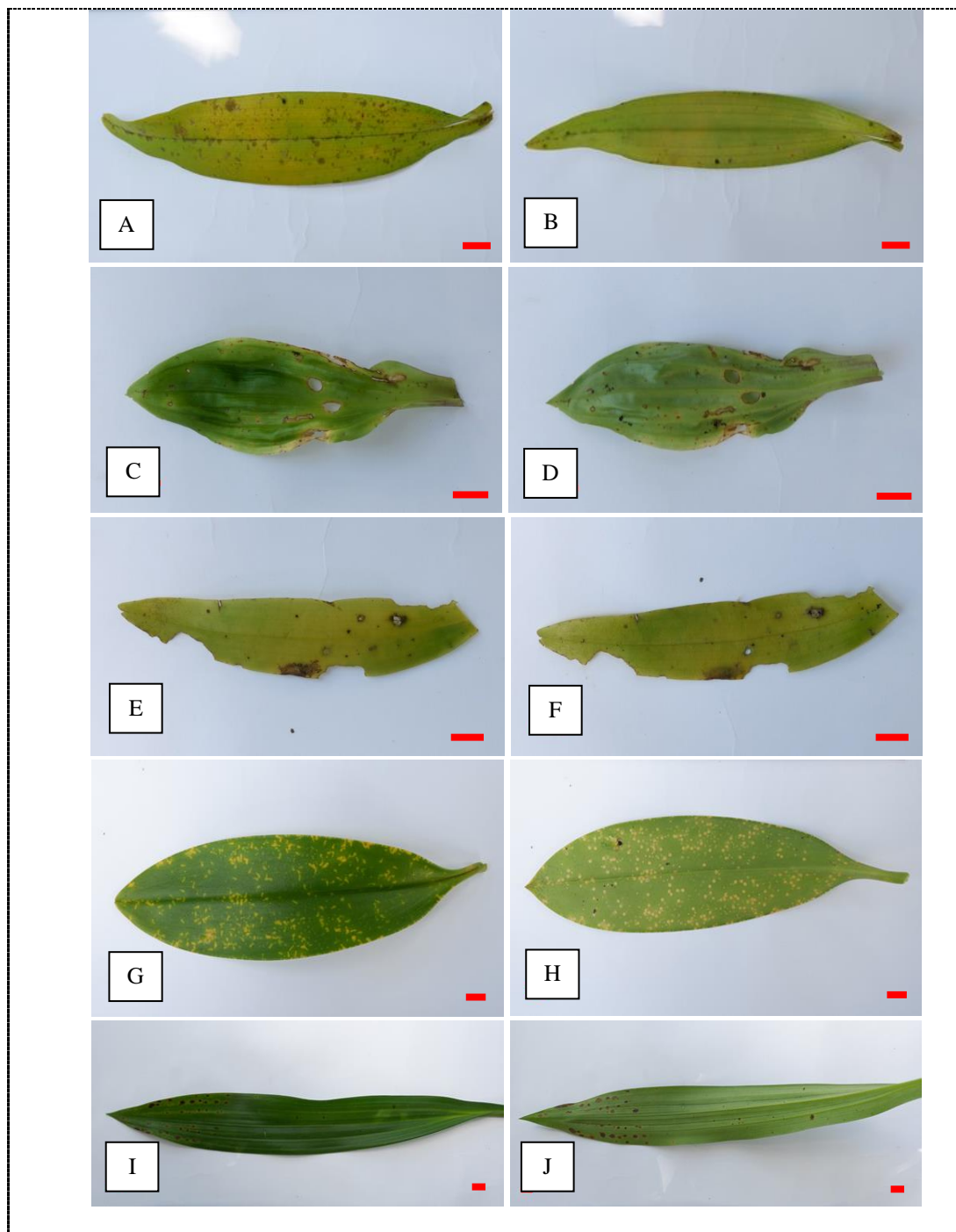


Figure 2. Symptoms of virus infection at Liwa Botanical Garden:
 (A, B) : Necrotic and chlorotic on *Flickingeria* sp.
 (C, D) : Necrotic and streak on *Calanthe* sp.
 (E, F) : Necrotic and chlorotic on *Trixspermum centipeda*
 (G, H) : Mosaic on *Bulbophyllum* sp.
 (I, J) : Necrotic and streak on *Coelogyne* sp.
 (A, C, E, G, I): Top surface of the leaf, (B, D, F, H, J): Surface under the leaf. Bar: 1 cm

Necrotic is characterized by physical damage or death to cells or tissues. Some of the symptoms that include necrotic types are necrose, rot, die back, and cancer (dead bark dries with a clear border) [3;4;8;12]. Chlorotic symptoms are symptoms caused by damage to chloroplasts which results in parts

of the plant which are normally green to yellow. Chlorotic symptoms are often associated with necrotic where chlorotic surrounds necrotic called "hello". Whereas curling leaf is a leaf symptom that shows changes in the shape of the edges in the form of curling [3].

3.3. Disease Intensity

Disease intensity analysis results obtained as a percentage as showed on **Table 2**.

Table 2. Analysis of mixed diseases in natural orchids on Liwa Botanical Garden.

No.	Species of Orchid	Disease Intensity
1	<i>Coelogyne</i> sp.	15%
2	<i>Flickingeria</i> sp.	42%
3	<i>Calanthe</i> sp.	40%
4	<i>Trixspermum centipeda</i>	20%
5	<i>Bulbophyllum</i> sp.	26%

Based on the analysis of the intensity of viral diseases in natural orchids in Liwa Botanical Garden the highest percentage was obtained in the orchid *Flickingeria* sp. with a percentage of 42%, this is because as many as 8 samples of a total of 15 leaves showed symptoms of the virus. Next is the orchid *Calanthe* sp. with a percentage of 40%, in the orchid *Bulbophyllum* sp. 26%, *Trixspermum centipeda* 20%, and *Coelogyne* sp. 15%.

3.4. Plant Resistance

The results of the analysis of the level of resistance to virus infection on native orchids in Liwa Botanical Garden are showed on **Table 3**.

Table 3. Analysis of the level of plant resistance to virus infections on Liwa Botanical Garden.

No.	Species of Orcid	Plant Resistance Level
1	<i>Coelogyne</i> sp.	Resistance
2	<i>Flickingeria</i> sp.	Susceptible
3	<i>Calanthe</i> sp.	Resistance
4	<i>Trixspermum centipeda</i>	Resistance
5	<i>Bulbophyllum</i> sp.	Resistance

Based on the results of the analysis it can be seen that the orchid *Flickingeria* sp. has a vulnerable level of resilience because this type of orchid is the host most susceptible to infection with pathogenic viral diseases. Percentage obtained on the type of orchid *Flickingeria* sp. as much as 42%. Mahfut et al. (2019) explained that the susceptibility level is resistance level in orchids infected with the disease more than 40%. This shows that the overall collection of natural orchid samples at Liwa Botanical Garden is capable of being infected by pathogens but a small portion of the cells supports the growth and development of pathogens so that they cause less disease.

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