ISSN: 1412-4718, eISSN: 2581-138x

Serdani AD · Sandy YA · Li'aini AS

Identification and characterization of pathogens causing diseases on *Begonia* at Eka Karya Bali Botanic Garden

Abstract. *Begonia* is one of the world's largest genera of flowering plants that spread in various tropical and subtropical regions worldwide. However, deforestation, overexploitation, climate change, and pathogen-causing diseases have threatened the diversity of begonia. Diseases on begonia need to be handled seriously because the level of spread and damage can result in the death of the plant. Thus, this study was conducted to identify the main pathogens causing diseases in the begonia which is expected to be basic information in determining the effective control treatment. As a result, begonia collections of Eka Karya Bali Botanic Garden were mainly infected by three pathogenic fungi (*Fusarium* spp., *Oidium begoniae*, and *Botrytis cinerea*) and one bacterium (*Xanthomonas begoniae*).

Keywords: Bacterial leaf spot · *Fusarium* wilt · Gray mold disease · Powdery mildew

Submitted: 8 November 2022, Accepted: 6 March 2023, Published: 17 April 2023
DOI: http://dx.doi.org/10.24198/kultivasi.v22i1.42764

Serdani AD[1] · Sandy YA[2] · Li'aini AS[3*]
[1] Off Main Campus Study Program of Agroecotechnology, Faculty of Agriculture, Blitar Islamic University, Jl. Majapahit No.2- 4, Blitar 66137, Indonesia
[2] Off Main Campus Study Program of Agroecotechnology, Faculty of Agriculture, Brawijaya University Jl. Pringgodani, Kec. Mojoroto, Kediri 64111, Indonesia
[3] Research Center for Plants Conservation, Botanical Garden, and Forestry, National Research and Innovation Agency, Jl. Ir. H. Juanda No.13 Kota Bogor 16122, Indonesia
*Correspondence: syafa.liaini@gmail.com

Jurnal Kultivasi Vol. 22 (1) April 2023
ISSN: 1412-4718, eISSN: 2581-138x

Introduction

Begonia (Begoniaceae) is one of the world's largest genera of flowering plants. Begoniaceae consists of 2000 species spread in various tropical and subtropical regions worldwide (Wang et al., 2020). Begonia is an erect or creeping herbaceous plant with male and female flowers in one or different individuals, watery stems, scattered leaves, clear petioles, toothed leaf edges, oval to elongated leaf shapes, protective leaves that fall off easily, asymmetrical leaf shape, and capsule-form fruit equipped with 3 seed wings measuring 0.03-0.04 mm (Siregar, 2005; Girmansyah, 2008; 2010).

Begonia is widely used as an ornamental plant (Siregar, 2017). This is because begonia has a variety of uniqueness, including beautiful flowers with attractive colors (white, yellow, orange, pink, and red); asymmetrical leaf shapes; various leaf sizes (±10 cm); and plant heights ranging from 15-30 cm. It's average size and high adaptability make begonia easy to maintain and grow as indoor ornamental plants (Wiriadinata and Girmansyah, 2001).

Today, the decline in begonia plant diversity continues to occur. This is due to deforestation, overexploitation, and global climate change (Widjaja et al., 2014). In addition, several diseases also threaten the diversity of begonia. The level of plant resistance to pathogens infection differs from one another. Some soil or airborne diseases are known to have a very detrimental attack rate on flowering plants like begonias; for example, *Sclerotiodes* disease, *Fusarium* spp., and so forth. Diseases that infect begonia need to be handled seriously because the level of spread and damage can result in the death of the plant (Wasito and Marwoto, 2003). Thus, the present study was conducted to identify the main pathogens causing diseases in the begonia collection at Eka Karya Bali Botanic Garden (Eka Karya BBG). The results of this study are expected to be basic information in determining disease control treatments for begonia.

Materials and Methods

Observations of disease symptoms in begonia were carried out at the greenhouse of Begonia Garden, Eka Karya BBG, from April to June 2019 at an average daily temperature of 21-22°C and 80-90% relative humidity (RH). Eka Karya BBG is located in Candikuning Village, Bedugul District, Tabanan Regency, Bali, at 1250-1450 m above sea level.

Isolation of pathogenic fungi was carried out at the Applied Botany Laboratory, Eka Karya BBG, while microscopic observations of pathogens were carried out at the Genetic Conservation Laboratory, Eka Karya BBG. The results of the macroscopic and microscopic disease observations were descriptively analyzed and compared with other previous studies.

The leaves of healthy and infected begonia were observed to determine the presence of disease symptoms. The macroscopic observations were made on leaf colors and disease symptoms.

Pathogen isolation was carried out in a sterile manner in a laminar airflow cabinet. The symptomatic leaves of begonia found in the Begonia Garden were cut into squares with a dimension of 1 cm × 1 cm, dipped in 5.25% NaOCl, then rinsed three times in sterile distilled water. After that, the leaves were placed in a petri dish containing potato dextrose agar and incubated at 26°C for three days. Every single colony of fungi was taken using a loop and transferred to a new media, then incubated at 26°C to obtain pure isolate. After three days, the single colony of fungi was taken using a loop and placed on an object glass, covered with a cover glass, then microscopically observed using an Olympus CX31 microscope.

Results and Discussion

Established in 1959, Eka Karya BBG has collected thousands of plants genera, including *Begonia*. This plant family (Begoniaceae) is grown in a greenhouse that stores about 300 species of *Begonia* (Fig. 1).

The observation showed that 4 types of diseases mainly infect the Begoniaceae collection of Eka Karya BBG as described below (Table 1).

***Fusarium* wilt.** *Fusarium* is a genus of fungi that causes disease in many plants. According to Trubus (2016), one of the species that infect begonia is *F. foetens*. This fungal infection causes dull green leaves with yellow spots, pale veins, and brown vascular tissue (Rosa & Moorman, 2018). Similarly, we found that begonia infected by *Fusarium* have

dull green to yellowing leaves, pale veins, and yellow to browning spots, as shown in Fig. 2a. In severe infections, the lower leaves of some begonia plants showed a wilting symptom (Fig. 2b). This is supported by Trubus (2016) and Rosa and Moorman (2018) which stated that severe infection of *Fusarium* turn leaves into yellow color and then wilt starting from the lower leaves then spread to the top of the plant, then the roots and stems rot, and the plant dies.

Fusarium spp. forms three types of asexual spores: microconidium, macroconidium, and chlamydospores. Chlamydospores are resistant spores. Most of the isolates of *Fusarium* spp. have white, purple, or pink colonies at the center of the colony. The colony will change from white to orange in isolates that form large amounts of sporodochium (Sutejo et al., 2008). Similarly, our isolate is spherical colonial with ivory white in color.

In *Fusarium* spp., conidium is formed on monophyly, long, and unbranched conidiophores as in *F. solani, F. sacchari,* and *F. verticillodes*, or formed on branched monophyalid conidiophores as in *F. heterosporous* (Fisher et al., 1983). In contrast, our isolate showed polyphyalid conidiophore as in *Fusarium* sp. found by Ngittu et al. (2014) (Fig. 3).

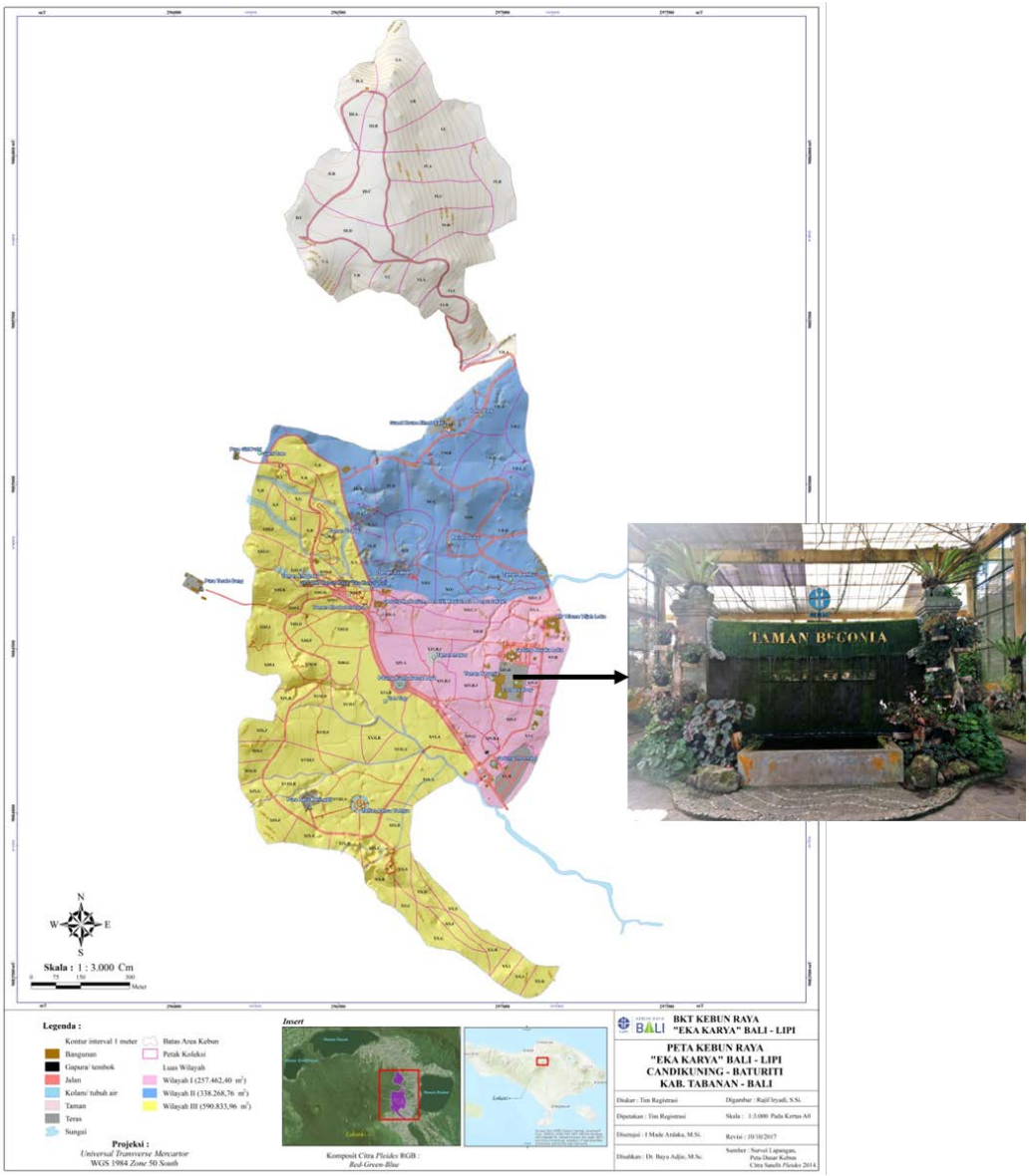


Figure 1. Location of Begoniaceae collection at Eka Karya Bali Botanic Garden (Map by the Registration Unit of Eka Karya BBG).

ISSN: 1412-4718, eISSN: 2581-138x

Table 1. Diseases found in the begonia collection of Eka Karya Bali Botanic Garden

Plant diseases	Causal agent	Symptom
Fusarium wilt	*Fusarium* sp.	• Dull green to yellowing leaves, • Pale veins, • Yellow to browning spots, • Wilting symptoms on the lower leaves.
Powdery mildew	*Oidium begoniae*	• White or gray spots spread all over the plant including leaves, stems, and flowers, • Young shoots and leaves to curl, • Pale green leaves, necrotic, and eventually fall when the infection is severe.
Gray mold	*Botrytis cinerea*	• Pale yellow spots on the leaves, • Brown spots and blight, • Silver-gray fungal mass is seen in dead plant tissue.
Bacterial leaf spot	*Xanthomonas begoniae*	• Circular or angular yellow spots, scattered, and stiff-like blisters on the leaves, • Brown or black irregular shape (V-shaped), surrounded by transparent yellow sores.

F. foetens spores can survive in the soil for up to 30 years, infecting plants through roots or lateral growth points. The spores spread through water flow, wind, cultivation activities, and equipment. The growth of fungal spores in the vascular tissue of plants inhibits the water supply for plants, so the stomata close, and the plants die. Infection is exacerbated when excessive watering without good drainage and air circulation (Trubus, 2016).

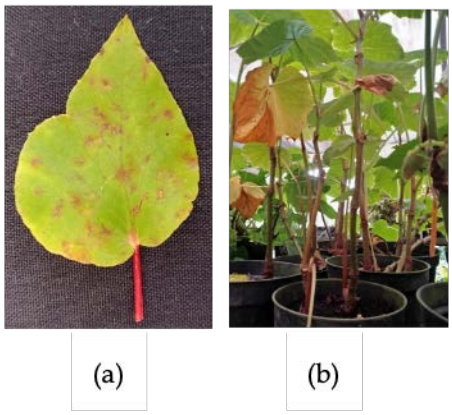


Figure 2. The infection symptoms of *Fusarium* sp. in a: *Begonia pisudoumuricata* leaf, b: *Begonia lempuyang ensis* plants in the nurseries (doc. ASL).

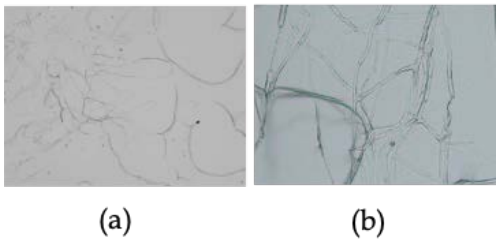


Figure 3. Microscopic cross-section of a: the chlamydospores of *Fusarium* sp. that infect *Begoniaceae* plant collections at 10x magnification (doc. ASL), b: the polyphyalid of *Fusarium foetens* that infect *Begoniaceae* plant collections at 40x magnification (doc. ASL).

Figure 4. The symptom of powdery mildew on *Begonia eka karya* plant collections (doc. ASL).

Figure 5. Microscopic cross-section of the conidiophores of *Oidium begoniae* that attack *Begoniaceae* plant collections at 40x magnification (doc. ASL).

Powdery mildew. Powdery mildew on begonia is caused by the fungus *Oidium begoniae* (Erysiphales: Erysiphaceae). *O. begoniae* is an obligate parasite (can only take nutrients from living hosts) that can cause defoliation, shoot death, and inhibit the growth of plant collections (Quinn, 1985). The infection starts from young leaves, then spreads to other parts of the plant (Kontaxis, 1985; Windham & Witte, 1998). Spores are produced in large numbers on the surface of the host so that white powder appears on the surface of the infected plant (Kontaxis, 1985; Windham & Witte, 1998). We found that powdery mildew on begonia collections of Eka Karya BBG showed a similar symptom as shown in Fig. 4. White or gray spots spread on the lower and upper surface of the leaves, stems, and flowers.

In addition, some previous studies showed that powdery mildew causes young shoots and leaves to curl, the leaves turn pale green, necrotic, and eventually fall when the infection is severe (Windham & Witte, 1998; Trubus, 2016). The infection in flowers results in deformed and low-quality flowers. Powdery mildew also prevents flowering in susceptible hosts (Windham & Witte, 1998). On older leaves with severe infection, powdery mildew symptoms are characterized by yellowish or brown spots and subsequently form necrotic spots that can reduce photosynthetic efficiency, resulting in leaf death and fall.

Similar to the previous study conducted by Putri et al. (2018) (Fig. 5a), from microscopic observations, it can be seen that the observed conidiophores of *Oidium* found from our begonia collections were elliptical and colorless (Fig. 5b). *Oidium* sp. is known as an obligate parasite that can only live on living tissue. Based on the observations, the white powdery layer is a collection of mycelium, conidium, and conidiophores of pathogenic fungi (Putri et al., 2018).

Oidium infection occurs through stomata (natural openings). Then, germinated conidia form haustoria which enter epidermal cells, and absorb nutrients contained in the epidermal cells (Boyce, 1961). Their conidia can be spread by wind, humans, cultivation equipment, or other infected plants (Kontaxis, 1985; Quinn, 1985). Furthermore, *O. begoniae* infects plants in the dry season, yet fog and high relative humidity with temperatures between 16-27°C play an important role in spore germination (Hansen, 2009).

Grey mold disease. This disease is caused by the fungus *Botrytis cinerea* (Helotiales: Sclerotiniaceae). *B. cinerea* infects plants in a cool environment with a temperature of 15°C, high relative humidity (93%), and low light, especially during the rainy season (Hausbeck & Moorman, 1996). The fungus can infect leaves, stems, crowns, buds, seeds, seedlings, tubers, and other parts of plants, except roots. Moreover, *B. cinerea* can cause the death of host cells, severe tissue damage, and decay and death of plant collections (van Kan, 2005). The conidia of *B. cinerea* can be transported by wind or water, and land on the surface of the host (Jarvis, 1977). Under optimal conditions, the disease cycle is capable of causing symptoms in only 3-4 days (van Kan, 2005).

Gray mold disease can be found in living plants with symptoms of brown spots and blight (Hausbeck & Moorman, 1996). The symptoms begin with pale yellow spots on the leaves. The spots then coalesce rapidly and widen, destroy cells and tissue, form blight symptoms (such as burning), and turn brown/black (Hausbeck & Moorman, 1996; Trubus, 2016). On the bloomed flowers, small rounded reddish-brown spots appear (Trubus, 2016). In advanced infections, plant tissue rots and dies (van Kan, 2005). These symptoms were also found in the begonia collection of Eka Karya BBG infected by *Botrytis*, as demonstrated in Fig. 6a-b. Meanwhile, in dead plant tissue, a silver-gray fungal mass is seen (Fig. 6c).

B. cinerea has hyphae shaped like bubbles bounded by white, gray, and brown partitions (Fig. 7). Then, it forms a branched and insulated mycelium. Furthermore, conidiophores appear perpendicular to the mycelium, insulated, and branched at the tips, and form a dichotomy or trichotomy. The older the conidiophores are, the

Jurnal Kultivasi Vol. 22 (1) April 2023
ISSN: 1412-4718, eISSN: 2581-138x

browner they are at the tips and lighter towards the branches. The tip of conidiophores swell to form an ampulla and there is a denticle as a place for the conidium to attach (Komalaningrat et al., 2018).

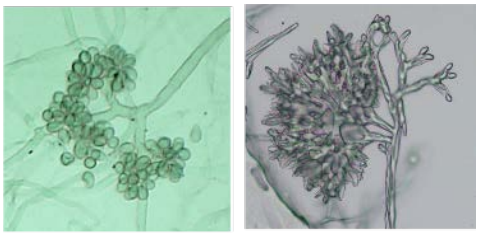
(a)

(b)

(c)

Figure 6. The symptoms of gray mold disease in *Begoniaceae* plant collections are a: brownish yellow spots on leaf surfaces of *Begonia albopicta*, b: silver fungal mycelium visible on dead leaf tissue, c: silver-grey fungal mycelium seen on a dead leaf (doc. ASL).

Figure 7. Conidia of *Botrytis cinerea* were assembled at the tip of the conidiophores at 40x magnification (doc. ASL).



(a) (b)

Figure 8. The infection symptoms of *Xanthomonas begoniae* causing bacterial leaf spot disease in *Begoniaceae* plant collections are a: the infection begins with brownish yellow spots starting from the leaf margins, b: the advanced infection with V-shaped "burns" symptom surrounded by yellow spots found on *Begonia argenteoguttata* (photo by ASL).

Bacterial leaf spot

Leaf spot disease is initiated by the bacterium *Xanthomonas begoniae* (Xanthomonadales: Xanthomonadaceae) which is carried by seeds. The further spread might be aided by splashing water, cultivation equipment, through insect's intermediary, or from the remaining infected plant collections. This disease can be very damaging to plant collections that flourish in nurseries and greenhouses with high humidity.

The bacterial infection of *X. begoniae* is indicated by circular or angular yellow spots, and scattered, and stiff-like blisters. The symptoms are first seen on the lower surface of the leaves, near the leaf margins or main vessels. The spots then coalesce, widen, and dry. The sores turn brown with an irregular shape (V-shaped in some hosts), surrounded by transparent yellow sores visible on both leaf surfaces. This pathogenic bacterium has typical symptoms: the leaves look water-soaked and withered, and there are spots of chlorosis and necrosis (Asrul et al., 2019). Similarly, the v-shaped spots found on the begonia collection were infected by *Xanthomonas* (Fig. 8). In *Xanthomonas* leaf blight, necrotic spots are brown (Schwartz & Gent, 2005). The advanced infection causes the leaves to wilt and fall.

The arrangement of plant collection at the Indonesia Botanical Garden is based on taxonomy, use, and origin (Li'aini & Kuswantoro, 2023). In this case, at Eka Karya BBG, the begonia plant collection is planted in a greenhouse. This condition is profitable for phytopathogens. Moreover, the environmental conditions of Eka Karya BBG, which have low

temperatures and high humidity, are suitable for the development of phytopathogen, especially pathogenic fungi that are mainly found to infect the begonia plant collection. The interaction between susceptible plants and virulent pathogens in a suitable environment for the growth of pathogens will cause plant diseases. Therefore, safe control techniques that can suppress the growth of pathogens are needed.

Several control techniques that can be used include sterilizing seeds (before planting) and planting equipment (Wati et al., 2021). Moreover, the planting equipment for healthy and infected plants should be separated. Another way is by eradicating or destroying diseased plants. Botanical pesticides and biocontrol agents can also be used to control plant diseases. A previous study showed that *C. aeruginosa* extract contains curcuminoids that play the role of an effective antifungi (Sari & Li'aini, 2020). Furthermore, *Bacillus amyloliquefaciens* was found to be a potential agent to control *Xanthomonas* (Li'aini et al., 2017), while Trichoderma asperellum showed an effective control against pathogenic fungi.

Conclusions

We found four microorganisms causing diseases on the begonia collection of Eka Karya BBG. There was *Fusarium* wilt, powdery mildew caused by *Oidium begoniae*, grey mold caused by *Botrytis cinerea*, and bacterial leaf spot caused by *Xanthomonas begoniae*. This information can be used to determine the effective disease control treatment in begonia collections, especially in the Indonesian Botanical Gardens.

Acknowledgment

Our gratitude is delivered to Ayyu Rahayu and Cokorda Istri Meyga Semarayani from Research Center for Plants Conservation, Botanical Gardens, and Forestry, National Research and Innovation Agency (BRIN), Indonesia, for the help in documenting the phytopathogens, and Ketut Sumertana from Directorate of Scientific Collection Management, BRIN, for identifying the begonia species and the insight on diseases control methods on begonia

References

Asrul A, Arwiyanto T, Hadisutrisno B, Widada J. 2019. Karakterisasi patogen hawar daun bakteri secara fenotipik pada bawang merah (*Allium cepa* L. kelompok Aggregatum). Agroland:Jurnal Ilmu-ilmu Pertanian, 26(1): 58-68.

Boyce JS. 1961. Forest Pathology Third Edition. McGraw Hill Book Company, Inc. New York.

Fisher NL, Marasas WFO, Toussoun TA. 1983. Taxonomic importance of microconidia chains in *Fusarium* section *Liseola* and effects of water potential on their formation. Mycologia, 75(4): 693-698.

Girmansyah D. 2008. Keanekaragaman jenis *Begonia* (Begoniaceae) liar di Jawa Barat. Berita Biologi, 9(2): 195-203.

Girmansyah D. (2010). Keanekaragaman *Begonia* (Begoniaceae) dari kawasan Gunung Watuwila dan Gunung Mekongga, Sulawesi Tenggara. Berita Biologi, 10(1): 33-41.

Hansen MA. 2009. Powdery Mildew of Ornamental Plants. Virginia Polytechnic Institute and State University. Virginia, USA.

Hausbeck MK, Moorman GW. 1996. Managing *Botrytis* in greenhouse-grown flower crops. Plant Disease, 80: 1212-1219.

Jarvis WR. 1977. *Botryotinia* and *Botrytis* species: taxonomy, physiology and pathogenicity-a guide to the literature. Canada Department of Agriculture.

Komalaningrat DA, Tondok ET, Widodo. 2018. Identitas spesies *Botrytis* pada tanaman hortikultura di Jawa Barat, Indonesia. Jurnal Fitopatologi Indonesia, 14(6): 205–214.

Kontaxis D. 1985. Managing powdery mildew on begonia. California Agriculture, 39(3): 16-16.

Li'aini AS, Kuswantoro F. 2023. Notes on edible Poaceae collected at Eka Karya Bali Botanic Garden. AIP Conference Proceedings 2583, 090006.

Li'aini AS, Lin YH, Huang TC, Sulistyowati L. 2017. Application of *Bacillus amyloliquefaciens* to control black rot disease on cabbage caused by *Xanthomonas campestris* pv. *campestris*. Journal of Plant Medicine, 59(3):39-44.

Ngittu YS. 2014. Identifikasi genus jamur *Fusarium* yang menginfeksi eceng gondok (*Eichhornia crassipes*) di Danau Tondano. Pharmacon, 3(3): 156-161.

Putri MM, Nurahmah Y, Anggraeni I. 2018. Identifikasi penyakit yang menyerang bibit

sengon (*Paraserianthes moluccana* (Miq.), Barneby & JW Grimes) di persemaian dan pengendaliannya. Jurnal Sains Natural, 7(1): 31-38.

Quinn JA. 1985. Growth and sporulation of *Oidium begoniae*: development of a deterministic model. Annals of Applied Biology, 107(2): 163-178.

Rosa C, Moorman WG. 2018. Diseases of begonia. Handbook of florists' crops diseases. Springer. Cham, Switzerland.

Sari, ARK, Li'aini AS. 2020. Efektivitas antifungi ekstrak *Curcuma aeruginosa* terhadap patogenisitas *Colletotrichum capsici* pada tanaman cabai merah. Jurnal Hortikultura, 30(2): 1-12.

Schwartz HF, Gent DH. 2005. Diseases: *Xanthomonas* leaf blight of onion. Gardening Series No. 2.951. Colorado State University, USA.

Siregar HM. 2005. Begonia: Kebun Raya Bali. LIPI Press. Indonesia.

Siregar HM. 2017. The conservation of native, lowland Indonesian *Begonia* species (Begoniaceae) in Bogor Botanic Gardens. Biodiversitas, 18(1): 326-33.

Sutejo AM, Priyatmojo A, Wibowo A. 2008. Identifikasi morfologi beberapa spesies jamur *Fusarium*. Jurnal Perlindungan Tanaman Indonesia, 14(1): 7-13.

Trubus. 2016. Hama dan penyakit tanaman: deteksi dini dan penanggulangan. Trubus info kit Vol. 9. Redaksi Trubus. Depok.

van Kan JAL. 2005. Infection strategies of *Botrytis cinerea*. In VIII International Symposium on Postharvest Physiology of Ornamental Plants 669. pp. 77-90.

Wang WG, Randi A, Wang CXL, Shen JY, Ma XD, Shi JP, Xu T, Zhang SZ. 2020. *Begonia daunhitam*, a new Begonia (Begoniaceae) species from West Kalimantan, Indonesia. Taiwania, 65(1): 27-32.

Wasito A, Marwoto B. 2003. Pengujian keefektifan gliokompos terhadap pertumbuhan dan perkembangang tanaman krisan. Jurnal Hortikultura, 13(4): 229- 235.

Wati C, Arsi, Karenina T, Riyanto, Nirwanto Y, Nurcahya I, Melani D, Astuti D, Septriarini D, Purba SRF, Ramdan EP, Nurul D. 2021. Hama dan Penyakit Tanaman. Yayasan Kita Menulis. Medan.

Widjaja EA, Rahayuningsih Y, Rahayoe JS, Ubaidah R, Maryanto I, Walujo EB, Semiadi G. 2014. Kekinian Keanekaragaman Hayati Indonesia 2014. LIPI Press. Jakarta.

Windham MT, Witte WT. 1998. Naturally occurring resistance to powdery mildew in seedlings of *Cornus florida*. Journal of Environmental Horticulture, 16(3): 173-177.

Wiriadinata H, Girmansyah D. 2001. Potensi Begonia liar sebagai tanaman hias. Prosiding Seminar Sehari: Menggali Potensi dan Meningkatkan Prospek Tanaman Hortikultura Menuju Ketahanan Pangan. LIPI-Kebun Raya Bogor. Bogor, November 5, 2000.