



## Viruses in Flowering Ginger

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### Introduction

Growers of ornamental flowering ginger (*Alpinia purpurata* (Vieillard) K. Schumann) on O'ahu, Hawai'i, have recently reported outbreaks of disease causing severe dieback (Figure 1) that threaten their livelihood. Virus-like symptoms were observed on red ginger from all of the locations where dieback was reported. Recently, flowering ginger in Hawai'i has been found to host a number of plant-pathogenic viruses including badnaviruses and a potyvirus.

Virus-like symptoms can be found on flowering ginger plants infected with single or multiple combinations of viruses; however, only plants infected with the potyvirus *Banana bract mosaic virus* (BBrMV) show clear and easily identifiable symptoms. The symptoms of disease in red ginger infected with two badnaviruses, *Canna yellow mottle virus* (CaYMV) and *Banana streak virus* (BSV), are difficult to assess clearly, though virus-like symptoms in plants found to be infected with either badnavirus alone are well defined. However, there is not yet clear evidence for the causal agent of the recent dieback disease plaguing growers of red ginger. Careful examination of the disease etiology through the application of Koch's



Figure 1. "Dieback" field symptoms

postulates is necessary to link any of the previously mentioned viruses to the disease.

### Host

Flowering ginger, *Alpinia purpurata* (Vieillard) K. Schumann, is an herbaceous, perennial plant with bright red or pink floral bracts and inconspicuous white flowers. It is native to the Pacific Basin and was introduced to Hawai'i, where it became naturalized sometime prior to

1930.<sup>1</sup> Flowering ginger is a popular ornamental and cut flower for home use and commercial sale, and prized for its bright floral bracts and long shelf life.<sup>1</sup> Thick, cane-like stems emerge from rhizomes and are wrapped by leaf sheaths.<sup>1</sup> The leaves are alternate, lack petioles, and the leaf lamina are oblong with pointed apices.<sup>1</sup> Each shoot bears a compact, spike-like inflorescence with clusters of ovate or broadly obovate bracts that subtend small, tubular, white flowers.<sup>1</sup> Flowering ginger is a member of the *Alpinia* genus of the *Zingiberaceae* family of the *Zingiberales* order. The *Zingiberales* also include members of the *Canna* genus of the *Cannaceae* family as well as members of the *Musa* genus of the *Musaceae*

family. Plant species that are closely related to flowering ginger are important to growers because they may host plant pathogens and/or insect vectors of plant pathogens that may be transmitted to ginger.

## Viruses

### *BBrMV*

*Banana bract mosaic virus* (BBrMV), an RNA virus, is a member of the *Potyvirus* genus of the family *Potyviridae*. BBrMV was first reported in *Musa* spp. (banana) from the Philippines in 1979 and later reported from many other countries including India, Samoa, Sri Lanka, Thailand, Vietnam, Colombia, Ecuador and Hawai'i.<sup>2-7</sup> BBrMV can spread quickly and causes significant economic losses worldwide. BBrMV was first reported in Hawai'i in 2010 from a new host, flowering ginger.<sup>8</sup> Although BBrMV has not yet been reported to naturally infect banana in Hawai'i, the strain found in Hawai'i in flowering ginger has been shown to infect banana in greenhouse studies. BBrMV is transmitted by aphid vectors and in Hawai'i by the black banana aphid (*Pentalonia nigronervosa*).

### *CaYMV*

*Canna yellow mottle virus* (CaYMV), a DNA virus, is a member of the *Badnavirus* genus of the *Caulimoviridae* family. Some members of this genus have integrated their genomic sequences into their host's genome. These viruses are also termed endogenous pararetroviruses (EPRV). EPRV infection can produce synergistic effects in hosts if those hosts harbor dual or multiple infections with other plant pathogenic viruses. CaYMV was first reported infecting *Canna* spp. in Japan in 1979 and was later reported from the United States, Italy, the Netherlands, India, and Kenya.<sup>9-13</sup> CaYMV was reported infecting a new host, flowering ginger, in Hawai'i in 2017.<sup>14</sup> The episomal (non-integrated) form of CaYMV was found to be associated with severe symptoms on flowering ginger in Hawai'i.<sup>14</sup> It is unknown if CaYMV has integrated into the ginger genome. The vector of CaYMV has yet to be identified, though it may be a species of mealybug since many other badnaviruses are vectored by mealybugs.<sup>14</sup>

### *BSV*

*Banana streak virus* (BSV), another DNA virus, is also a member of the *Badnavirus* genus of the *Caulimoviridae* family. Banana streak disease was first observed from Africa on the Ivory Coast in 1958, and the etiol-

ogy of the disease was confirmed, establishing BSV as the causal agent.<sup>15, 16</sup> Banana streak disease has been reported from more than 40 countries around the world including Africa, Asia, Australia, Europe, Oceania, and tropical America; essentially everywhere that bananas are grown.<sup>5</sup> BSV has integrated into the genome of some banana cultivars, although it is not yet known if the BSV isolate identified in flowering ginger in Hawai'i is integrated into the ginger genome. BSV isolates from around the world show a high degree of genetic heterogeneity as well as differing serological and biological properties.<sup>17</sup> We have identified the species of BSV in Hawai'i to be *Banana streak virus* 'Goldfinger' (BSGfV) through sequence analyses of PCR products generated from symptomatic ginger plants.

## Host Range and Disease Symptoms

### *BBrMV*

The known host range of BBrMV is limited to various *Musa* spp., small cardamom (*Elettaria cardamomum*) in India, and flowering ginger in Hawai'i.<sup>8, 18, 19</sup> Symptoms of BBrMV on flowering ginger include: mosaic, streaking, severe cupping of leaves, browning of flowers, and reduced bract size and shelf life<sup>20</sup> (Figure 2). Symptoms of BBrMV on banana include a distinctive mosaic pattern on the bracts and may also include spindle-like purple streaks on the bracts, midribs, peduncles, and fruits.<sup>3, 21, 22</sup>

### *CaYMV*

The known host range of CaYMV is limited to *Canna* spp. and flowering ginger in Hawai'i, though there is concern that this may expand to include other tropical plants.<sup>14</sup> Symptoms of flowering ginger infected with CaYMV include yellow mottling & necrosis of the leaves, vein streaking and stunting of leaves (Figure 3). Flowering ginger infected with both BBrMV and CaYMV exhibit very distinct and severe necrotic and stunting symptoms that are easily recognizable to the trained observer (Figure 4).

### *BSV*

The known host range of BSV is limited to *Musa* spp. in most parts of the world, and flowering ginger in Hawai'i.<sup>20</sup> Symptoms of flowering ginger infected with BSV have been observed as: streaking on the bracts, leaves, leaf sheaths, and stems, but further research is necessary to confirm these preliminary observations and any causative



Figure 2. BBrMV symptoms



Figure 3. CaYMV symptoms



Figure 4. BBrMV & CaYMV co-infection symptoms

link to infection with BSV (Figure 5). Symptoms of BSV on banana depend on the virus isolate and host cultivar, but they include inconspicuous chlorotic flecking, lethal necrosis, and, most commonly, discontinuous chlorotic dots or streaks that eventually become necrotic that run from the midribs to leaf margins.<sup>23</sup> BSV is transmitted by several species of mealybugs including mealybugs (*Planococcus citri* and *Pseudococcus* spp.). Other mealybug vectors include *Dysmicoccus* spp. in West Africa and South America, *Planococcus musae* in Nigeria, *Ferrisia virgata* in India, *D. brevipes*, *P. ficus* and *Paracoccus burnerae* in South Africa.

## Detection

### *BBrMV*

There are a number of molecular and serological diagnostic assays available for reliable detection of BBrMV. The unique symptoms produced by this virus in flowering ginger appears to be a reliable diagnostic for the trained observer. Molecular assays including reverse transcription polymerase chain reaction (RT-PCR) assays using BBrMV-specific primers or potyvirus-specific universal primers have been established.<sup>5,24</sup> A specific immunocapture reverse transcription loop-mediated isothermal amplification (IC-RT-LAMP) assay has also been developed to detect BBrMV.<sup>20</sup> Serological assays include commonly available commercial diagnostic kits from Agricultural Diagnostics (Agdia) that are based on an enzyme linked immunosorbent assay (ELISA). ELISA kits exist for both universal potyvirus detection and specific BBrMV detection. Other immunological technologies such as the ImmunoStrip<sup>®</sup> assay (Agdia) are also available for rapid potyvirus detection.

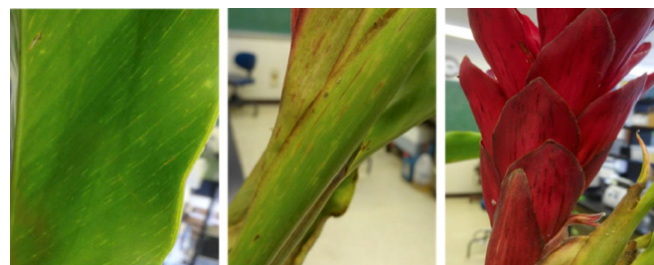


Figure 5. Possible BSV Symptoms



Figure 6a. Aphid and Mealybug Vectors



Figure 6b. Aphid and Mealybug Vectors (cont.)

### *CaYMV*

Currently, the only reliable diagnostic assays available for CaYMV are two molecular assays; a general badnavirus PCR assay using universal badnavirus primers, and a CaYMV-specific PCR assay that amplifies a fragment of open reading frame 3 (ORF3) of CaYMV.<sup>14</sup>

### *BSV*

There are molecular and serological assays available for detection of BSV. Molecular assays include a general badnavirus PCR assay using universal badnavirus primers and a BSGfV specific PCR assay for the species of BSV identified in Hawai'i.<sup>25,26</sup> Further, ELISA assays for BSV strains are available but of limited usefulness due to the high serological diversity of BSV species.<sup>17</sup> The most conclusive method for serological detection of BSV virions is immunosorbent electron microscopy (ISEM).<sup>27</sup>

### Transmission

#### *BBrMV*

BBrMV may be spread over long distances through the movement of virus-infected vegetative planting material. BBrMV is transmitted by several aphid species: *Pentalonia* spp. (Figure 6a), *Rhopalosiphum maidis*, *Aphis gossypii*, and *A. craccivora* in a nonpersistent manner.<sup>21</sup>

#### *CaYMV*

The vector of CaYMV is unknown, though it is likely to be a mealybug species, as most other badnaviruses are transmitted by mealybugs<sup>14</sup> (Figure 6b). CaYMV may

be spread over long distances through the movement of virus-infected vegetative planting material.

### *BSV*

Attempts to transmit BSV between banana plants by mechanical inoculation have been unsuccessful, which suggests that the spread of BSV on cutting tools or any other cultural operation is unlikely.<sup>16</sup> BSV is spread over long distances through vegetative propagation of infected planting materials such as rhizomes for flowering ginger or suckers and tissue cultured plantlets from infected banana plants.<sup>28</sup> In the field, mealybugs (*Pseudococcidae*) transmit BSV between bananas in a semipersistent manner, and observations suggest that virus spread between bananas is very slow.<sup>28</sup> There appears to be little difference in the transmission efficiency of BSV isolates observed between the two main mealybug vector species *Planococcus citri* and *Pseudococcus* spp.<sup>29-31</sup> Other mealybug species have been reported to transmit BSV including *Dysmicoccus* spp. in West Africa and South America, *Planococcus musae* in Nigeria, *Ferrisia virgata* in India, *D. brevipes*, *P. ficus*, and *Paracoccus burnerae* in South Africa.<sup>21,32,33</sup>

### Management

#### *BBrMV*

Virus diseases are characteristically difficult to manage, often requiring eradication of the entire crop or relocation to a new geographical location to control losses. Prevention is paramount, and reduction of the

aphid vector populations may help to limit the spread of BBrMV. Insect predators of aphids in Hawai'i should be encouraged, and therefore it is not recommended to spray broad-spectrum insecticides for reduction of aphids as spraying of pesticides may cause a reduction in predators resulting in the aphids spreading the virus more rapidly through fields. Judicious use of contact insecticides is acceptable when aphids are observed. Exclusion of infected planting material and eradication of any possible reservoirs of virus inoculum or alternate hosts of BBrMV (potentially other members of Zingiberales) should be achieved through best management practices including exclusion and eradication. Exclusion and eradication can include (1) proper field sanitation (scouting to monitor insect populations and disease symptoms; removing, bagging, and destroying of infected plant materials; (2) avoiding movement of plants from infected areas; (3) verifying when buying or trading germplasm that plant propagation material is virus-free; (4) obtaining proper Hawai'i Department of Agriculture (HDOA) plant clearance when transporting plant materials between islands; and (5) rapid communication with local College of Tropical Agriculture and Human Resources (CTAHR) Extension agents and HDOA plant quarantine inspectors if virus-like symptoms are observed.

### ***CaYMV and BSV***

Management of CaYMV and BSV should be conducted as explained above for BBrMV, with some important exceptions including control of ants. It is imperative to control the presence and proliferation of ant species that “farm” mealybugs for their honeydew and effectively spread the slow-moving adult mealybugs and, therefore, any viruses they may harbor. Although adult mealybugs travel slowly, their juvenile instars are highly mobile. Previous field reports suggest that the natural spread of BSV by its mealybug vector (on banana) is limited and does not play a significant role in the disease epidemiology.<sup>34</sup> Therefore, it is crucial to scout for insect presence and control ant populations in the field.

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## **Further Resources**

### **Cooperative Extension Offices**

UH Mānoa - College of Tropical Agriculture and Human Resources: <https://cms.ctahr.hawaii.edu/ce/Find-Us>

### **O'AHU COUNTY**

#### **HONOLULU**

1955 East-West Rd, Honolulu, HI 96822

Phone: **(808) 956-7290**

#### **PEARL CITY**

955 Kamehameha Highway, Pearl City, HI 96782

Phone: **(808) 453-6050**

#### **WAHIAWĀ**

910 California Ave, Wahiawā, HI 96786

Phone: **(808) 622-4185**

### **HAWAI'I COUNTY**

#### **HILO**

875 Komohana Street, Hilo, HI 96720

Phone: **(808) 969-8201**

#### **KAMUELA**

67-5189 Kamamalu Road, Kamuela, HI 96743

Phone: **(808) 887-6183**

#### **KONA**

79-7381 Māmalahoa Highway, Kealahou, HI 96750

Phone: **(808) 322-4892**

### **MAUI COUNTY**

#### **MAUI**

310 Ka'ahumanu Ave., Bldg. 214, Kahului, HI 96732

Phone: **(808) 244-3242 x222**

#### **MOLOKA'I**

526 Hua'ai Rd, Ho'olehua, HI 96729

Phone: **(808) 567-6929**

### **KAUAI COUNTY**

#### **KAUAI**

State Office Building, 3060 Eiwa Street, Room 210, Līhu'e, HI 96766

Phone: **(808) 274-3471**

**Plant Quarantine Office**

Hawai'i Department of Agriculture: <http://hdoa.hawaii.gov/pi/pq/pqcontacts/>

**O'AHU COUNTY**

1849 Auiki Street, Honolulu, HI 96819

Phone: (808) 832-0566

Honolulu International Airport

Phone: (808) 837-8413

Toll-Free Pest Hotline: (808) 643-7378

Report A Pest After Hours: (808) 837-8092

**HAWAI'I COUNTY**Plant Inspection Office

95 Akahana St. Rm 9, Hilo, HI 96720

Phone: (808) 961-9393

Certified Nursery Program

16 East Lanikaula St, Hilo, HI 96720

Phone: (808) 974-4141

Keāhole Airport, Kailua-Kona

Phone: (808) 326-1077

**MAUI COUNTY**

635 Mua Street, Kahului, HI 96732

Phone: (808) 873-3962

**KAUA'I COUNTY**

4398A Pua Loke Street, Līhu'e, HI 96766

Phone: (808) 241-7135

**Plant Protection and Quarantine Offices**

United States Department of Agriculture  
Animal and Plant Health Inspection Office:  
[https://www.aphis.usda.gov/aphis/ourfocus/planthealth/sa\\_export/sa\\_ecs/hawaii](https://www.aphis.usda.gov/aphis/ourfocus/planthealth/sa_export/sa_ecs/hawaii)

APHIS, Plant Protection and Quarantine

300 Rodgers Blvd., #57, Honolulu, HI 96819

Phone: (808) 834-3240

**References**

1. Kobayashi, K.D., J. McEwen, and A.J. Kaufman. Ornamental Ginger, Red and Pink. College of Tropical Agriculture and Human Resources, Cooperative Extension Service, 8 (2007).
2. Magnaye, L.V., and R.V. Valmayor. BBTV, CMV and other viruses affecting banana in Asia and the Pacific. Food and Fertilizer Technology Center for the Asian and Pacific Region, Taipei City (1995).
3. Rodoni, B.C., Y.S. Ahlawat, A. Varma, J.L. Dale, and R.M. Harding. Identification and characterization of banana bract mosaic virus in India. *Plant Disease* **81**, 669–672 (1997).
4. Ferison, E., and C. Putter. FAO/IBPGR technical guidelines for the safe movement of *Musa* germplasm. *FAO/IBPGR Rome*, p 23 (1989).
5. Diekmann, M., and C. Putter. FAO/IPGRI technical guidelines for the safe movement of germplasm In: C.A.J. Putter (ed.) *Musa*, 2nd edn. Food and Agriculture Organization of the United Nations/International Plant Genetic Resources Institute, p 28 (1996).
6. Rodoni, B., J. Dale, and R. Harding. Characterization and expression of the coat protein-coding region of banana bract mosaic potyvirus, development of diagnostic assays and detection of the virus in banana plants from five countries in Southeast Asia. *Arch Virol J* **144**, 1725–1737 (1999).
7. Quito-Avila, D.F., et al. First report of *Banana bract mosaic virus* in 'Cavendish' banana in Ecuador. *Plant Disease* **97**, 1003–1003 (2013).
8. Wang, I.C., D.M. Sether, M.J. Melzer, W.B. Borth, and J.S. Hu. First report of *Banana bract mosaic virus* in flowering ginger in Hawaii. *Plant Disease* **94**, 921–921 (2010).
9. Yamashita, S., T. Natsuaki, Y. Doi, and K. Yora. 1985. *Canna yellow mottle virus*, a non-enveloped small-bacilliform virus in *Canna* sp. *Annals of the Phytopathological Society of Japan* **51**:642–646.
10. Lockhart, B., and L. Autrey. Occurrence in sugarcane of a bacilliform virus related serologically to banana streak virus. *Plant Disease* **72**, 230–233 (1988).
11. Marino, M.T., E. Ragozzino, B. Lockhart, R. Miglino, and D. Alioto. First report of *Canna yellow mottle virus* (CaYMV) in Italy and in the Netherlands. *Plant Pathology* **57**, 394 (2008).
12. Kumari, A., S. Kumar, and S.K. Raj. First report of *Canna yellow mottle virus* on *Canna* from India. *New Disease Reports* **29** (2014).
13. Agneroh, T.A., S.A. Bratsch, and B.E. Lockhart. First report of *Canna yellow mottle virus* in Kenya. *Plant Health Progress*, PHP-BR-14-0037 (2015).

14. Zhang, J. et al. Characterization of *Canna yellow mottle virus* in a new host, *Alpinia purpurata*, in Hawaii. *Phytopathology* **107**, 791–799 (2017).
15. Lockhart, B. and D. Jones. Banana mosaic. In: D.R. Jones (ed) Diseases of Banana, Abaca and Enset. *CAB International Wallingford* pp 256–274 (2000).
16. Lockhart, B. Purification and serology of a bacilliform virus associated with banana streak disease. *Phytopathology* **76**, 995–999 (1986).
17. Lockhart, B., and N. Olszewski. Serological and genomic heterogeneity of banana streak badnavirus: Implications for virus detection in *Musa* germplasm. In: J. Ganry (ed) Breeding Banana and Plantain for Resistance to Diseases and Pests. *CIRAD/INIBAP Montpellier*, 105–113 (1993).
18. Sharman, M., J.E. Thomas, and R.G. Dietzgen. Development of a multiplex immunocapture PCR with colourimetric detection for viruses of banana. *J Virol Methods* **89**, 75–88 (2000).
19. Siljo, A., A.I. Bhat, C.N. Biju, and M.N. Venugopal. Occurrence of *Banana bract mosaic virus* on cardamom. *Phytoparasitica* **40**, 77–85 (2011).
20. Zhang, J., et al. Deep sequencing of banana bract mosaic virus from flowering ginger (*Alpinia purpurata*) and development of an immunocapture RT-LAMP detection assay. *Arch Virol* **161**, 1783–1795 (2016).
21. Selvarajan, R., and K. Jeyabaskaran. Effect of *Banana bract mosaic virus* (BBrMV) on growth and yield of cultivar Nendran (plantain, AAB). *Indian Phytopathol* **59**, 496–500 (2006).
22. Thomas, J.E., A.D. Geering, C.F. Gambley, A.F. Kessling, and M.White. Purification, properties, and diagnosis of banana bract mosaic potyvirus and its distinction from abaca mosaic potyvirus. *Phytopathology* **87**, 698–705 (1997).
23. Tripathi, S., B.L. Patil, and R. Verma. Viral diseases of banana and their management. In: Plant Viruses: Evolution and Management. (ed. R.K.G.e.a.) (Springer Science+Business Media, Singapore; 2016).
24. Zheng, L., B.C. Rodoni, M.J. Gibbs, and A.J. Gibbs. A novel pair of universal primers for the detection of potyviruses. *Plant Pathology* **59**, 211–220 (2010).
25. Harper, G., G. Dahal, G. Thottappilly, and R. Hull. Detection of episomal banana streak badnavirus by IC-PCR. *J Virol Methods* **79**, 1–8 (1999).
26. Geering, A.D., L.A. McMichael, R.G. Dietzgen, and J.E. Thomas. Genetic diversity among *Banana streak virus* isolates from Australia. *Phytopathology* **90**, 921–927 (2000).
27. Ahlawat, Y.S., R.P. Pant, B.E.L. Lockhart, and M. Srivastava. Association of a badnavirus with citrus mosaic disease in India. *Plant Disease* **86** (1996).
28. Daniells, J., J. Thomas, and M. Smith. Seed transmission of banana streak virus confirmed. *Infomusa* **4** (1995).
29. Dahal, G., et al. Relationship between natural occurrence of banana streak badnavirus and symptom expression, relative concentration of viral antigen, and yield characteristics of some micropropagated *Musa* spp. *Plant Pathol.* **49**, 69–79 (2000).
30. Kubiriba, J., J. Legg, W. Tushemereirwe, and E. Adipala. Vector transmission of *Banana streak virus* in the greenhouse in Uganda. *Annals of Applied Biology* **139**, 37–43 (2001).
31. Matile-Ferrero, D., and D. Williams. Recent outbreaks of mealybugs on plantain (*Musa* spp.) in Nigeria including a new record for Africa and a description of a new species of *Planococcus* (Ferris) (Homoptera: Pseudococcidae). *Bulletin de la Societe Entomologique de France* **100**, 445–449 (1995).
32. Meyer, J.B., G.G.F. Kasdorf, L.H. Nel, and G. Pietersen. Transmission of activated-episomal *Banana streak OL (badna)virus* (BSOLV) to cv. Williams banana (*Musa* sp.) by three mealybug species. *Plant Disease* **92**, 1158–1163 (2008).
33. Wambulwa, M.C., F.N. Wachira, L.S. Karanja, S.M. Kiarie, and S.M. Muturi. The influence of host and pathogen genotypes on symptom severity in banana streak disease. *African Journal of Biotechnology* **12**, 27–31 (2013).
34. Lockhart, B. Banana streak badnavirus infection in *Musa*: Epidemiology, diagnosis and control. *Food and Fertilizer Technology Center Technical Bulletin*. Food and Fertilizer Center Taipei **143**, 11 (1995).