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Bacterial Wilt of Edible Ginger in Hawai'i

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The rhizomes of edible ginger ('awapuhi Pākē, Zingiber officinale Roscoe) are a valued spice or fresh herb ingredient in international cuisine. They are used as edible flavorings in products such as candies, beverages and liqueurs, ice creams, baked goods, curry powder blends, sauces, pickles, and condiments. Practitioners of traditional medicine use ginger rhizomes to treat ailments such as nausea, motion sickness, migraine, and dyspepsia, and to reduce flatulence and colic.

Ginger root ranked 20th among Hawai'i's agricultural commodities in 2009, with a sales value of \$2.24 million from 50 harvested acres (Statistics of Hawai'i Agriculture 2009). This was a decline of about \$1.75 million from that reported for ginger in 2005–2006. It did not



A mature ginger plant with bacterial wilt.

or lands not previously cultivated with edible ginger. Pathogen-free ginger seed can be difficult to obtain, inhibiting new farmers from starting a farm. Furthermore, pathogen-contaminated shoes or tools can spread the disease to new areas, forcing ginger growers to quarantine fields and keep visitors out. Disease onset can be rapid and severe, causing great crop loss.

Bacterial wilt is a significant disease of many crops besides ginger root. These wilt diseases are caused by several subgroups of a bacterial pathogen, *Ralstonia solanacearum*. Historically, strains of *R. solanacearum* have been classified into 5 races based on their host ranges, and into 5 biovars based on their differential ability to produce acid from a set of specific carbohydrates. *R solanacearum* race 4

reach the top 20 in 2010 or 2011, the last year for which there are data available.

An important factor in the decline of ginger root acreage and sales since 2006 was bacterial wilt, a plant disease that devastates ginger. Farms with pathogen-infested soils cannot be replanted successfully. Epidemics of bacterial wilt cause major crop losses and discourage growers from farming edible ginger.

In Hawai'i, ginger growers farm this crop nomadically, moving from infested locations to wilt-free fields infects ginger in Hawai'i and much of Asia.

Here we discuss bacterial wilt of edible ginger in Hawai'i and suggest integrated practices for best management of the disease.

Pathogen

The pathogen is a soil- and water-borne bacterium, Ralstonia solanacearum race 4. Synonyms for Ralstonia solanacearum include Pseudomonas solanacearum (Smith 1896) Smith 1914; Burkholderia

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solanacearum (Smith 1896) Yabuuchi et al. 1992; and others. The pathogen is widely distributed in areas where edible ginger has been grown previously. The host range of R. solanacearum race 4 is restricted to edible ginger (Z. officinale).

Ralstonia solanacearum belongs to rRNA homology group II (non-fluorescent) within the pseudomonads. Ralstonia pickettii (a saprophyte or human facultative pathogen), Pseudomonas syzygii (causing Sumatra disease of cloves), and the Blood Disease Bacterium (causing blood disease of banana in Indonesia) are closely related. These bacteria may cross-react in detection procedures based on serology or DNA. Sub-classification of R. solanacearum is based on genetic fingerprinting methods into Division I (biovars 3, 4, and 5 from Asia) and II (biovars 1, 2A, and 2T from South America). Additional, proposed taxonomic divisions are based on nucleic acid sequence analysis into "phyllovars" and "sequevars." Clearly, this species is taxonomically intricate.

R. solanacearum spreads by infested soil adhering to hands, boots, tools, vehicle tires, and field equipment; in water from irrigation or rainfall; and within infected ginger rhizomes (Janse 1996). This bacterium infects

ginger roots and rhizomes through openings where lateral roots emerge or through wounds caused by handling, parasitic insects, or root-knot nematodes (Swanson et al. 2005). The pathogen survives in soils within infected plant debris and as free-living bacteria. Ginger crops can be completely lost to the disease in heavily infested soils.

Disease symptoms and signs

R. solanacearum colonizes the xylem, the water-conducting elements of a ginger plant's vascular system, and causes wilt. The symptoms caused by this pathogen on ginger include the following:

- "Green wilt," the diagnostic symptom of the disease. This occurs early in the disease cycle and precedes leaf yellowing. Infected green ginger leaves roll and curl due to water stress caused by bacteria blocking the water-conducting vascular system of the ginger stems.
- Leaf yellowing and necrosis. Leaves of infected plants invariably turn yellow and then necrotic brown. The yellowing, however, should not be confused with another disease of ginger causing



An immature edible ginger plant showing the typical symptoms of bacterial wilt: "green wilt" (green ginger leaves roll and curl due to water stress caused by bacteria blocking the water-conducting vascular system of the ginger stems), leaf yellowing, and necrosis.



A field of young ginger plants affected by bacterial wilt disease. The wilting, yellowing plants are infected with *Ralstonia solanacearum* race 4.

similar symptoms known as Fusarium yellows. Plants infected by the fungus *Fusarium oxysporum* f. sp. *zingiberi* do not wilt rapidly, as they do when affected by bacterial wilt. Instead, *Fusarium*infected ginger plants are stunted and yellowed. The lower leaves dry out over an extended period of time. The differences between these two diseases can be clarified by comparing their descriptions in CTAHR publication "Diseases of Ginger (*Zingiber officinale*) in Hawaii" (http://www.ctahr.hawaii.edu/ oc/freepubs/pdf/C2-62.pdf).

- Plant stunting. Diseased plants grow poorly and may be stunted.
- Plant decline and death. Diseased plants can decline rapidly and die before harvest.
- Discolored rhizomes that are often rotted inside.
- Rhizomes and stem vasculature with a water-soaked appearance.
- Discoloration of vascular tissues.
- Soft rots (secondary diseases) caused by opportunistic bacterial plant pathogens in the genus *Erwinia* or *Pectobacterium*.

E.E. Trujillo (1964) accurately described the progress of disease symptoms for bacterial wilt of ginger:

The first symptoms of wilt are a slight yellowing and wilting of the lower leaves. The wilt progresses upward, affecting the younger leaves, followed by a complete yellowing and browning of the entire shoot. Under conditions favorable for disease development, the entire shoot becomes flaccid and wilts with little or no visible yellowing. However, the plant dries very rapidly and the foliage becomes yellow-brown in 3 to 4 days. Young succulent shoots frequently become soft and completely rotted and these diseased shoots break off easily from the underground rhizome at the soil line. The underground parts are also completely infected. Gravish-brown discoloration of the rhizomes may be localized if the disease is at an early stage of infection, or discoloration may be general if the disease is in an advanced stage. A water-soaked appearance of the central part of the rhizome is common. In advanced infections, the entire rhizome becomes soft and rots. Bacterial wilt of ginger can be distinguished from other rhizome rots of ginger by the condition of the rhizome and the



An edible ginger rhizome infected with *Ralstonia solanacearum* race 4, showing a discolored, necrotic center.



Discolored ginger rhizome and stems affected by bacterial wilt.



Growers should control other pests that damage ginger plants, such as the lesser corn stalk borer (*Elasmopalpus lignosellus*), which creates holes in the stems of ginger plants.



When an infected rhizome is suspended in water, the white, milky bacteria stream from the diseased tissue.

foliage. A better diagnostic feature is the extensive bacterial ooze that shows as slimy, creamy exudate on the surface of a cut made in the rhizome or on the above-ground stem of an infected plant.

Disease "signs" refer to the observable presence of a plant pathogen in or on infected host tissues. Signs can be useful in diagnosing the pathogen and the disease. These are signs of bacterial wilt of ginger:

- Bacterial streaming, i.e., large populations of bacteria that exude from the cut surface of infected plant tissue when observed with a microscope or observed macroscopically when a diseased ginger rhizome is suspended in a glass or beaker of water.
- Bacterial ooze from infected tissues, especially from infected rhizomes. Ooze is the emission of bacterial colonies from infected tissues, seen as moist, milky mounds collecting on the tissues' surfaces.

Pathogen detection and disease diagnosis

The bacterial wilt pathogen, the disease, or both can be detected using a number of different laboratory methods. These methods test several different sources for the pathogen, including soil, water, and infected plant



Bacterial ooze from an infected ginger rhizome.

tissue. The tests may be performed either before or after planting.

- 1. Bioassay. A bioassay consists of exposing ginger plants growing in pots to field soils to see if bacterial wilt disease symptoms develop. Bioassays are useful because they are
 - reliable diagnostic tests,
 - very sensitive,
 - easy to set up,
 - fast, because symptoms develop rapidly, and
 - simple for farmers to interpret.

Bioassay procedure (outline, Anne Alvarez, personal communication):

- Collect samples of field soil from an area intended for ginger cultivation. Use appropriate soil-sampling methods, as described in the CTAHR publication "Testing Your Soil: Why and How to Take a Soil-Test Sample" at http:// www.ctahr.hawaii.edu/dnn/Portals/43/AS-4.pdf.
- ii. Obtain young, tissue-cultured ginger plants. The advantage of using tissue-cultured plants is that they are pathogen-free and symptoms



Severely rotten ginger rhizomes infected by bacterial wilt.

develop rapidly when they are exposed to *R*. *solanacearum*. Alternatively, one may use non-tissue-cultured ginger plants if they are also pathogen-free.

- iii. Transplant the tissue-cultured ginger plants into pots containing the field soil.
- iv. Raise the temperature to 80°F and keep the soil wet with frequent irrigation.
- v. Check the plants daily for the development of symptoms. They should occur 1–2 weeks after transplanting. If wilting develops, the test is positive for *R. solanacearum*. If desired, uproot the symptomatic plants and submit them to the UH-CTAHR ADSC to confirm the identity of the pathogen.
- 2. Symptoms and signs. Diagnosing the disease solely by its symptoms is not conclusive; other diseases can cause symptoms that closely resemble bacterial wilt. For a more dependable diagnosis, inspect the rhizomes for signs of the pathogen, bacterial ooze or bacterial streaming, together with the symptoms previously mentioned.



Secondary invaders including soft-rot bacteria can colonize affected tissues.

- 3. Immunological tests. These tests are based on serology, wherein antibodies designed to detect a specific antigen (i.e., the bacterial wilt pathogen) create a visible test reaction. The Immunostrip method (based on the Enzyme-Linked Immunosorbent Assay (ELISA) method) can detect *R. solanacearum* within 30 minutes. The Immunostrip user guide at https://orders.agdia.com/Documents/m182.pdf describes the testing procedure.
- 4. DNA tests. The Polymerase Chain Reaction (PCR) is a test for bacterial wilt based on the selective amplification of the 16S ribosomal rRNA gene from *R. solanacearum* (Seal and Jackson 1991). If a fragment of target DNA is amplified and appears as a band on an electrophoresis gel, *R. solanacearum* was present in the extracted sample. Growers can request a PCR analysis of soil or plant tissue samples submitted to the Komohana Cooperative Extension Service ADSC office (assay based on Shintaku et al. 1996). The analysis code for a PCR analysis is "D1" and the cost is currently \$12.00 per sample. The PCR test is not offered at other UH-CTAHR ADSC locations.

Other DNA-based tests such as 'LAMP' (loopmediated isothermal amplification) are possible (Kubota et al. 2008) but are not yet widely used by diagnostic laboratories.

Disease Management

The most efficient practices to manage bacterial wilt will integrate some or all of the following practices.

Site selection. The cultivation site should be well drained and without recent history of ginger cultivation (at least 10 years). The best sites have rainfall dispersed evenly throughout growing season, with a drier period near the end, before and during harvest. Excessive rainfall and waterlogging favor bacterial wilt and other soilborne diseases. Do not plant downslope from another ginger field if runoff water can carry the pathogen into your field. Before planting, test the soil in your field for the pathogen by either PCR or a plant bioassay.

Planting considerations. Avoid planting during very wet weather, as this promotes dispersal of the pathogen within fields on muddy boots, tools, and vehicles.

Site, soil, and bed preparation. Use clean, pathogen-free tractors and equipment. Hose tractor down with clean water (not obtained from streams located below existing ginger fields) before entering field to be planted in ginger. Preferably spray with 10% bleach solution, particularly tractor wheels and blades. Prepare soil by plowing and harrowing so that the site and soils drain well after rainfall. Proper soil and bed preparation are essential for ginger production. Prior to planting, soils are typically plowed to a depth of 45–60 cm. Lime is incorporated to adjust the pH, and furrows are cut with a hand tiller to a depth of 30–45 cm, spaced about 150 cm apart. Fertilizer or organic amendments are placed at the bottom of the furrows and incorporated prior to seeding.

Plant pathogen-free seed. Growing disease-free ginger starts with planting pathogen-free seed. Consult these CTAHR publications for more information: 1) "Producing Bacterial Wilt-free Ginger in Greenhouse Culture" (2004) at www.ctahr.hawaii.edu/dnn/Portals/43/SCM-8.pdf or 2) "A Simplified Method of Multiplying Bacterial Wilt-Free Edible Ginger (*Zingiber officinale*) in Pots" (2013) at http://www.ctahr.hawaii.edu/oc/freepubs/pdf/PD-93. pdf; or watch the video at http://www.ctahr.hawaii.edu/



Mature field of ginger suffering from bacterial wilt.

ginger wilt/. Bleach can be used to sterilize the surface of ginger seeds: dip them in a 10% bleach solution (1 part commercial bleach to 9 parts water) for 10 minutes. However, this may not eliminate bacterial infections within the rhizomes. A hot water treatment, consisting of exposing seeds to a constant 50°C temperature for 10 minutes, is effective in controlling nematodes but also may not be effective for disease organisms such as *R. solanacearum* that are already present inside the rhizome.

Hilling, cultivation, and drainage. Hill the planting rows to promote aeration of the roots and allow adequate soil drainage. Hilling is often done by hand, even on commercial farms. The ginger crop, as it grows from the bottom of the furrows, is hilled 3–5 times during the growing season. This results in raised hilled beds that allow proper development of the rhizomes. Hilling of the ginger row, which is done at 6-week intervals, allows the rhizome to grow vertically. Ensure adequate soil drainage and diversion ditches to prevent the runoff of infected water sources into fields that are downslope from an infected field.

Limit site traffic and pathogen dispersal. Strictly limit non-farm vehicles and visitors into ginger fields, as the pathogen can be in soils on truck tires or boots. Do not bring dirty tools to the farm from other locations. Disinfest boots by steeping into a 10% bleach solution before entering a ginger field. Erect fences to deter pigs and other animals from entering a ginger field.

Product name	Active ingredient (%)	Formulation	Application
Admire® Pro Systemic Protectant	Imidacloprid (42.8%)	Flowable concentrate	Soil
Amtide [®] Imidacloprid 2F Insecticide	Imidacloprid (22.6%)	Emulsifiable concentrate	Soil
Couraze [®] 4F Insecticide	Imidacloprid (40.4 %)	Emulsifiable concentrate	Soil

Table 1. Insecticides registered in Hawai'i for use on ginger.

Note: Be aware that some pesticides affect non-target organisms and take this into consideration when using them. All pesticides should be used in accordance with label instructions.

Composting. Organic soil amendments (mulch, compost) promote microbial activity that may suppress *R. solanacearum* by competition, antibiosis, or both.

Crop rotation and intercropping. Rotate ginger with crops that are not hosts of the bacterial wilt pathogen, such as sweetpotato and taro. Do not intercrop or rotate ginger with solanaceous crops, including tomatoes, peppers, and eggplant. The pathogen can reproduce in these crops and build up levels of bacterial wilt in the field. Ginger may also be rotated with grain crops such as corn or upland rice. Other possible crops for use in rotation or intercropping include green onion, soybean, sweet corn, cabbage, and Sunn hemp.

Other pests. Control other pests that damage ginger plants, such as the lesser corn stalk borer (*Elasmopalpus lignosellus*) and the banana moth (*Opogona sacchari*), which may be controlled with applications of *Bacillus thurigiensis* (Bt). Injuries are infection courts for *R. solanacearum*, and some insects create wounds that favor the bacterial wilt pathogen. Refer to Table 1 for insecticides registered for use on ginger.

Biofumigation. The use of volatile oils to kill or suppress a pathogen is known as biofumigation (Paret 2010). These oils are a natural component of certain green-manure crops such as mint, palmarosa, and lemongrass. When these plants are turned or plowed into the soil several months before planting, they decompose and release the essential oils. These oils are toxic to the bacterial wilt pathogen and can severely reduce its population in the field soil. However, the oils are very expensive if bought pre-processed, not naturally occurring as a component of green-manure crops, and in this case their use may not be economically feasible.

On-time harvest. Harvesting on time minimizes crop exposure to the pathogen.

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References

- Alvarez, AM. 2005. Diversity and diagnosis of *Ralstonia* solanacearum. In Bacterial Wilt Disease and the *Ralstonia solanacearum* Species Complex. C. Allen, P. Prior and AC Hayward, eds. St. Paul, MN: American Phytopathological Society (APS Press), 437–447.
- Alvarez, AM; Trotter, KJ; Swafford, MB; Berestecky, JM; Yu, Q; Ming, R; Hepperly, PR; and Zee, F. 2005. Characterization and detection of *Ralstonia* solanacearum strains causing bacterial wilt of ginger in Hawaii. Bacterial Wilt Disease and the *Ralstonia* solanacearum complex. C. Allen, P. Prior and AC Hayward, eds. St. Paul, MN:.American Phytopathological Society (APS Press), 471–477.
- Janse, J. 1996. Potato brown rot in Western Europe history, present occurrence and some remarks on possible origin, epidemiology and control strategies. *Bulletin OEPP/EPPO* 26: 679–695.

- Kubota, R.; Vine, BG; Alvarez, AM; and Jenkins, DM. 2008. Detection of *Ralstonia solanacearum* by loop-mediated isothermal amplification. *Phytopathology* 98:1045–1051.
- Kutin, RK; Alvarez, A; and Jenkins, DM. 2009. Detection of *Ralstonia solanacearum* in natural substrates using phage amplification integrated with real-time PCR assay. *Journal of Microbiological Methods* 76:241–246.
- Nelson, S. 2013. Bacterial wilt of edible ginger website. http://www.ctahr.hawaii.edu/gingerwilt/ (accessed 23 September 2013).
- Paret, ML; Cabos, R; Kratky, BA; and Alvarez, AM. 2010. Effect of plant essential oils on *Ralstonia solanacearum* race 4 and bacterial wilt of edible ginger. *Plant Disease* 94:521–527.
- Paret, ML; Kubota, R; Jenkins, DM; and Alavrez, AM. 2010. Survival of *Ralstonia solanacearum* race 4 in drainage water and soil and detection with immunodiagnostic and DNA-based assays. *Hort-Technology* 20:539–548.
- Paret, ML, deSilva, AS; and Alvarez, AM. 2011. Detection of *Ralstonia solanacearum* with an immunostrip assay; its specificity and sensitivity. *Indian Phytopathology* 61:518–522.

- Paret, ML; deSilva, AS; and Alvarez, AM. 2009. Bioindicators for *Ralstonia solanacearum* race 4: plants in the Zingiberaceae and Costaceae families. *Australasian Plant Pathology* 38:6–12.
- Statistics of Hawaii Agriculture 2009. 2011. Hawaii United States Department of Agriculture Department of Agriculture, Agricultural Development Division National Agricultural Statistics Service. http:// www.nass.usda.gov/Statistics_by_State/Hawaii/ Publications/Annual_Statistical_Bulletin/2009.pdf (accessed 7 August 2013).
- Shintaku, M; Kaneshiro, T; and Enriques, C. 1996. Detecting *Pseudomonas solanacearum* in edible ginger using the polymerase chain reaction. *J. Haw. Pac. Agric.* 7:11–19.
- Swanson, JK; Yao, J; Tans-Kersten, JK; and Allen, C. 2005. Behavior of *Ralstonia* solanacearum race 3 biovar 2 during latent and active infection of geranium. *Phytopathology* 95:136–14.
- Trujillo, EE. 1964. Diseases of Ginger (*Zingiber officinale*) in Hawaii. http://www.ctahr.hawaii.edu/oc/freepubs/pdf/C2-62.pdf (accessed 8 August 2013).