



Collecting Plant Disease and Insect Pest Samples for Problem Diagnosis

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Most plants in Hawaii are prone to attack by various insect pest and plant disease organisms. Pest outbreaks and diseases must be identified accurately to enable their efficient management. In this article we describe the basic requirements for collecting, preserving and submitting plant insect and disease samples to the Agricultural Diagnostic Service Center (ADSC), a branch of the Cooperative Extension Service, College of Tropical Agriculture and Human Resources (CTAHR), University of Hawai'i at Mānoa. If you need to collect a soil sample to eliminate the possibility that the problem is due to physiological causes (nutrient deficiency or toxicity), see the CTAHR publication "Testing Your Soil: Why and How to Take a Soil-Test Sample," on the Web at <http://www.ctahr.hawaii.edu/oc/freepubs/pdf/SCM-9.pdf>.

Collecting plant samples

It is important to gather the best plant samples possible and to record all pertinent background information for the diagnostician. Following are general guidelines for collecting plant samples.

Examine the entire plant for symptoms

Diseased plants generally have been infected by one or more pathogenic microorganisms, although they may also have an abiotic disease that does not involve a plant pathogen. Plants affected often display a range of symptoms—visual signs of the infection. Often, not all symp-

toms of a particular disease will appear on any one plant within a diseased crop, and more than one plant organ may be affected by a given disease.

Examine all of the main plant organs for disease symptoms: roots, stems, leaves, and blossoms. Collect samples from various plant organs as needed. Plants may suffer from more than one disease simultaneously. Segregate different types of symptoms into different samples.

Collect several plant specimens

A single plant sample may not be enough to allow a correct diagnosis of the problem; several plant samples showing the range of symptoms may be needed. If possible, select samples with various stages of disease development (early and late stages). Samples should be as typical or representative of the overall problem as possible. The best plant tissues for diagnosis are the ones showing the symptoms in various stages of disease development, and adequate amounts of them are important, but submitting excessive amounts of leaves or soil should be avoided.

Do not collect dead plants or plant organs

Dead plants or plant organs may not be useful for diagnosis. Often their tissues have been invaded by decomposing fungi and bacteria and the original pathogens are no longer detectable. Always select plant samples from living tissues and focus your attention on plants or plant

organs that are in early stages of disease or are in the process of dying, and not already dead.

Collect the entire plant whenever possible

If the wrong part of a plant is submitted, a disease cannot be diagnosed with confidence. For example, some leaf symptoms of disease (wilting, for example) are the result of damaged or diseased roots that have rotted and are no longer functioning to support the plant; in such cases, a correct diagnosis often depends on having a sample of the roots. Plants should be carefully dug from the ground (not pulled out) so the root systems remain relatively intact. When the entire plant cannot be sampled, shipped, or submitted, collect the largest plant sample possible, or portions of each major plant organ (roots, stems, leaves, flowers). Branch specimens should include the diseased area and part of the healthy area and may be at least 8–12 inches long. Be sure that root samples are from the affected plants and not from adjacent weeds. If entire plants cannot be sampled, submit photographs of affected plants where possible.

Provide as much background and related information as possible

Good information contributes to a better understanding of the problem. A complete description of the problem and the crop's history should accompany the sample. Give the name of the plant submitted. Indicate when the problem appeared and when the sample was taken. Specify all fertilizers and pesticides used. Examine the growing site carefully and note the conditions. Make note of environmental conditions for the site such as elevation, flooding, previous crop history, etc. Indicate any observable pattern of disease occurrence (for example, in random patches, or uniformly throughout the crop). Review the worksheet on p. 7 (which is based on ADSC's Form 357) for information required. This information is helpful in making a distinction between damage caused by pests and damage caused by other factors.

Preserving plant samples

After collecting the samples, do not expose them to direct sunlight. Keep them cool and do not allow them to dry out or cook. Place samples in plastic bags in the shade or in a cooler until they are ready for delivery to the plant clinic. Leaves may be pressed between the pages of a book or magazine or wrapped in tissue.

Collecting insect samples

A good insect sample consists of a range of the organism's life stages, if possible, presented in well preserved, undamaged condition. Following are general guidelines for collecting insect samples.

Avoid touching insects with fingers

Some insects can injure humans. Handling insects can also cause damage to their bodies that may prevent their identification.

Collect different life stages of the insect

Sometimes insects cannot be properly identified unless a certain life stage is present. For example, adults may be needed for correct identification.

Collect multiple specimens

Collect several specimens of the insect.

Time of day matters

Many leaf-feeding insects (such as caterpillars) may hide from predators during daylight hours. It may be necessary to capture insects during twilight in the evening or early morning.

Preserving insect specimens

Most insects: Roaches, termites, bugs, beetles, flies, wasps, ants, maggots, spiders, etc. should be immersed in isopropyl ("rubbing") alcohol, which kills and preserves them.

Mites, scales, aphids, thrips: Send these in alive on some of the affected foliage or stems, collected as you would a plant specimen. Place in a plastic bag when collected. Refrigerate until sent.

Butterflies and moths: Kill the specimens by freezing, wrap lightly in tissue paper, and place in a crush-proof box. Careful handling is required because the pattern of scale coloration is often used in identification.

Caterpillars: Send in alive on some of the host plant tissues in a plastic bag. Refrigerate until sent.

Grubs: Send in alive in a pint or two of soil enclosed in a plastic bag. Refrigerate until sent.

Packaging plant and insect samples

It is important to package the samples properly to ensure they arrive in good condition at the plant clinic. Following are general guidelines for handling and packaging plant and insect samples.

Use plastic bags

For most samples including leaves, stems and roots, use plastic bags to prevent plant samples from drying out during transport. However, fleshy fruits, vegetables, or tubers in stages of decay should be wrapped individually in dry newspaper.

Do not add extra water

Do not add any water or moist paper towels. Moisture favors the growth of fungi and bacteria that decay plant tissues, which can confuse or obscure the diagnosis of the pathogen.

Label the samples

Write your name, telephone number, and name of the plant or sample number on the plastic bag, using an indelible marker. Or, write the information on a piece of paper and insert in the bag.

Segregate plant tissue and insects from soil

Keep soil off the foliage. Avoid contact of plant or insect samples and information labels with any soil that might be in the plastic bag (unless it is a soil-dwelling insect).

Tie off the root ball

Place the root system and accompanying soil in a plastic bag and tie it securely to the lower stem of the plant. This will prevent roots from drying out and will keep the soil secure. Place a second plastic bag over the aboveground part of the plant, if possible, to prevent it from drying out.

Submit samples as soon as possible

Decayed plant or insect samples are useless for an accurate disease diagnosis. Always plan to have samples arrive at the ADSC within one or two days of their collection, if possible, or take steps to inhibit the deterioration or decay of samples (i.e., by refrigeration).

Submitting samples

The UH-CTAHR Agricultural Diagnostic Service Center (ADSC) analyzes soils, plant tissues, and forages and identifies plant pests and pathogens, for a fee. The services are itemized on the Web at www.ctahr.hawaii.edu/adsc.

Samples of plants and insects may be hand-delivered or mailed to the nearest ADSC or Cooperative Extension Service (CES) office. Because samples submitted to CES offices have to be transported to ADSC for diagnosis, contact the CES office to determine which day of the week is the best to submit samples. Commercial operations with large numbers of samples for diagnosis (for example, for virus assay), are requested to contact ADSC in advance before submitting the samples so that ADSC staff will be ready to handle the request.

At the time of submission, clients are asked to fill out a form describing the sample. The form asks about plant parts affected and plant symptoms and is similar to the check-off lists on p. 6.

Samples are processed in the order they are received. Diagnosis is mailed to each client as soon as the work is completed. The CES offices can provide information on the ADSC fee schedule.

Special sampling considerations

Turf

Samples should be at least a 6" x 6" piece of the turf, including both root system and soil. If using a golf course cup cutter, please send at least two plugs. Collect samples from the border between healthy and diseased turf, so that about two thirds of the sample is diseased turf and one third of the sample is healthy-looking turf. Wrap the soil and roots in aluminum foil to keep the soil from shaking loose during transit, and place it in a plastic bag. Do not expose samples to direct sunlight or high temperatures. Fill out the sample submission form completely, making sure to list all fertilizer, herbicide, fungicide, and insecticide applications made in the last 30 days, as well as any major cultural practices (aerification, topdressing, etc.).

Woody plants

(This includes trees, landscape shrubs, vines, etc.)

Dieback: Collect a large handful of feeder roots and one quart of soil from the root zone of plants that have

representative, moderate symptoms. Do not submit dead plants for diagnosis. Place roots and soil together in a plastic bag and close it securely. Place several branches showing decline or dieback in a separate plastic bag. For smaller plants, submit an entire plant (confine the root ball in a plastic bag tied tightly to the stem). Place the entire plant in another plastic bag and close it securely. Be sure there is no water on the foliage surfaces (this causes deterioration during shipping).

Canker, swellings on trees or shrubs: Cut the branch several inches beyond each end of the affected area, including some live wood. Place the specimen in a plastic bag and secure the bag.

Greenhouse plants

Submit plants in their original pots whenever possible. Plant foliage should be dry, and the planting medium should be slightly moist but not soggy. Place the pot in a plastic bag and tie it securely to the stem above the collar to prevent spilling during transport. Wrap the foliage gently in newspaper, and enclose the entire plant and pot in a plastic bag.

Fleshy vegetables, fruits, mushrooms, harvested root crops

Do not place produce in plastic bags. Wrap each specimen in several layers of newspaper. Wrapped specimens

can be placed in paper bags. Collect several specimens. Package the specimens in a box to prevent them from being crushed. Avoid exposure to high temperatures or direct sunlight.

Small plants from fields, landscapes, or gardens

Dig up (do not pull) several complete plants, leaving soil and roots intact. Enclose the roots and soil (approximately one quart) in a plastic bag and secure the bag. Place the entire plant in another plastic bag and secure it. Keep soil off of foliage.

Nematodes

Nematodes are associated with plant roots and soil. Samples should consist of both soil and the roots of the affected plant (do not collect weed roots) collected at 6" depth. For large fields, collect multiple samples from a large area, bulk them together in a bucket, mix them together, and remove a representative subsample consisting of about of 2 pints of soil with roots. Place the sample in a plastic bag and keep it cool and away from direct sunlight.

Foliar nematodes

In wet environments, nematodes can invade and rot leaves, stems, buds, and flowers. These plant tissues should be collected, wrapped in tissue to prevent damage to the specimens, and placed in plastic bags.

Contact information

ADSC Web page

<http://www.ctahr.hawaii.edu/adsc>

ADSC at UH-Manoa

1910 East-West Rd.
Sherman Laboratory Rm 134
Honolulu, HI 96822
956-6706 (fax 956-2592)
adsc@ctahr.hawaii.edu

ADSC at CES-Hilo

Komohana Research and Extension Center
875 Komohana St.
Hilo, HI 96720-2757
981-5191 (fax 981-5211)
bushe@hawaii.edu

CES offices

Hawai'i	Hilo (Komohana)	875 Komohana St, Hilo 96720-2757	808-981-5199
	Kamuela	67-5189 Kamamalu Rd, Kamuela 96743-8439	887-6183
	Kona	79-7381 Mamalahoa Hwy, Kealahou 96750-7911	322-4892
Kaua'i	Lihue	State Office Bldg, 3060 Eiwa St, Ste 210, Lihue 96766-1881	274-3471
Maui	Kahului	310 W Kaahumanu Ave, Bldg 214, Kahului 96732-1617	244-3242
Moloka'i, Lāna'i	Hoolehua (Molokai)	P.O. Box 394, Hoolehua 96729-0394	567-6933
O'ahu	Honolulu	1955 East-West Rd, Rm 217, Honolulu 96822	956-7138
	Kaneohe	45-260 Waikalua Rd, Ste 101, Kaneohe 96744-3134	247-0421
	Urban Garden Center (UGC) ...	955 Kamehamea Hwy, Pearl City 96782-2501	453-6050
	Wahiawa	910 California Ave, Wahiawa 96786-2124	622-4185

To contact an office by e-mail, use the location name (or the one given above in parentheses); e.g., komohana@ctahr.hawaii.edu, lihue@ctahr.hawaii.edu, etc.

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Glossary of terms

- abiotic disease**—a disease that is not caused by a living organism (e.g., nutrient deficiency)
- bacterium (pl., bacteria)**—a prokaryotic, microscopic, single-celled organism
- blight**—sudden, severe, and extensive spotting, discoloration, wilting, or destruction of leaves, flowers, stems, or entire plants
- canker**—a plant disease characterized (in woody plants) by the death of cambium tissue and loss and/or malformation of bark, or (in non-woody plants) by the formation of sharply delineated, dry, necrotic, localized lesions on the stem; “canker” may also be used to refer to the lesion itself, particularly in woody plants
- chlorosis**—yellowing of normally green tissue due to chlorophyll destruction or failure of chlorophyll formation
- dieback**—progressive death of shoots, leaves, or roots, beginning at the tips
- disease**—abnormal functioning of an organism
- epidemic**—an increase of disease in a population; a general and serious outbreak of disease
- gall**—tumor-like, abnormal swelling or localized outgrowth, often roughly spherical, produced by a plant as a result of attack by a fungus, bacterium, nematode, insect, or other organism
- gumming, gummosis**—(pl. gummoses) pathologic condition characterized by excessive formation of gums; the products of cell degeneration
- host plant**—living plant attacked by or harboring a parasite or pathogen and from which the invader obtains part or all of its nourishment
- infection**—process in which an organism enters, invades, or penetrates and establishes a parasitic relationship with a host plant
- infest (n., infestation)**—to attack as a pest (used especially of insects and nematodes); to contaminate, as with microorganisms; to be present in large numbers
- leaf spot**—an area of dead tissue caused by the invasion of pathogens; leaf spots can increase in size either slowly and rapidly, depending on the pathogen
- mosaic**—disease symptom characterized by non-uniform coloration, with intermingled normal, light green and yellowish patches, usually caused by a virus; often used interchangeably with mottle
- nematode**—nonsegmented roundworm (animal), usually microscopic, parasitic on plants or animals, or free living in soil or water
- pathogen (adj., pathogenic)**—a disease-producing organism or agent
- parasite (adj., parasitic)**—organism that lives in intimate association with another organism on which it depends for its nutrition; not necessarily a pathogen
- petiole**—the stem of the leaf
- ring spot**—disease symptom characterized by yellowish or necrotic rings enclosing green tissue, as in some plant diseases caused by viruses; also formed by fungi that sporulate within a large spot, causing rings of spores alternating with areas of no spores
- root rot**—softening, discoloration, and often disintegration of plant tissue as a result of fungal or bacterial infection or nematode infestation
- rust**—a fungal disease of plants caused by a specialized group of Basidiomycetes that often produces spores of a rusty color
- scorch**—any symptom that suggests the action of flame or fire on the affected part, often seen at the margins of leaves
- shot-hole**—symptom in which dead tissue falls out of small lesions on leaves, giving the leaf the appearance of being hit by birdshot
- soft rot**—softening, discoloration, and often disintegration of plant tissue as a result of fungal or bacterial infection
- sooty mold**—black, nonparasitic, superficial fungal growth on sugars exuded from the plant or honeydew produced by aphids and other phloem-feeding insects
- stunt, stunting**—plants that grow very slowly or remain shorter than normal, often due to progressive reduction in the length of internodes or a decrease in the number of internodes
- yellow**—disease characterized by chlorosis and often stunting of the host plant
- virus**—a submicroscopic, intracellular, obligate parasite consisting of a core of infectious nucleic acid (either RNA or DNA) usually surrounded by a protein coat
- wilt**—drooping of leaves and stems from lack of water (inadequate water supply or excessive transpiration); vascular disease that interrupts normal water uptake
- Terminology sources:**
 Illustrated Glossary of Plant Pathology by C.J. D’Arcy, D.M. Eastburn, and G. L. Schumann, <http://www.apsnet.org/education/IllustratedGlossary>
 Plant Pathology by G. Agrios

Worksheet for gathering information necessary for diagnosis of plant and insect samples

Sample to be sent to:

- Insect clinic
- Plant disease clinic
- Soil analysis
- Don't know

Name and address

Date of sampling:

Area where sample was collected (e.g., Hilo)

Elevation

Collector of sample (if different from client)

Host common name

Host scientific name

Type of grower, growing situation (check all that apply)

- Commercial
- Greenhouse
- Potted
- House plant
- Home garden/landscape
- Agricultural field
- Grounds (commercial)
- Lawn/turf

Household pest, location found (check all that apply)

- At light bulbs
- Pantry
- In furniture
- Stored products
- Other

Pesticide history

List pesticides used

Rates of application (concentration)

Frequency or number of applications

Fertilizer history

List fertilizers used

Amount of fertilizers used

Frequency of fertilizer applications

Plants or area affected

No. of acres

No. plants

% of plants

Spatial pattern of damaged plants (check one)

- Uniform; widespread
- Random distribution
- Clustered

Rooting environment

Commercial potting medium (identify)

Hydroponics

If rooted in soil, specify type

- Lava
- Red or brown, deep soil
- Rocky
- Other

Plant parts affected (check all that apply)

- | | |
|-----------------------------------|--|
| <input type="checkbox"/> Buds | <input type="checkbox"/> Rootlets |
| <input type="checkbox"/> Seeds | <input type="checkbox"/> Large rots |
| <input type="checkbox"/> Stem | <input type="checkbox"/> Growing tips |
| <input type="checkbox"/> Trunk | <input type="checkbox"/> Fruits or nuts |
| <input type="checkbox"/> Bark | <input type="checkbox"/> Bulbs or corms |
| <input type="checkbox"/> Tubers | <input type="checkbox"/> Branches, large |
| <input type="checkbox"/> Blossoms | <input type="checkbox"/> Branches, terminal |
| <input type="checkbox"/> Petioles | <input type="checkbox"/> Leaves, upper surface |
| | <input type="checkbox"/> Leaves, lower surface |

Plant symptoms (check all that apply)

- | | |
|--|--|
| <input type="checkbox"/> Galls | <input type="checkbox"/> Malformation |
| <input type="checkbox"/> Wilting | <input type="checkbox"/> Tip burn |
| <input type="checkbox"/> Stunting | <input type="checkbox"/> Discoloration |
| <input type="checkbox"/> Gumming | <input type="checkbox"/> Sudden collapse |
| <input type="checkbox"/> Yellowing | <input type="checkbox"/> Slow decline |
| <input type="checkbox"/> Root rot | <input type="checkbox"/> Leaf scorch |
| <input type="checkbox"/> Dieback | <input type="checkbox"/> Blight |
| <input type="checkbox"/> Shot hole | <input type="checkbox"/> Canker |
| <input type="checkbox"/> Leaf spot or blossom blight | <input type="checkbox"/> Mosaic |
| <input type="checkbox"/> Leaf fall or blossom drop | <input type="checkbox"/> Ringspot |
| <input type="checkbox"/> Fruit spot | <input type="checkbox"/> Rust |
| <input type="checkbox"/> Fruit rot | <input type="checkbox"/> Soft rot |
| <input type="checkbox"/> Mildew | <input type="checkbox"/> Sooty mold |

History of problem (check one):

- First time
- Recurrent

Symptom appearance (check one)

- Rapid
- Gradual

Summary description

Further comments

Examples of symptoms of disease infection and pest infestation (see Worksheet, p. 7)

algal leaf spot
avocado



fruit rot
anthracnose fungus, cucurbit



fruit rot
anthracnose fungus, papaya



aphids
tended by ants, oleander



canker
fungus, shower tree



chlorosis (yellow leaves)
root-knot nematode, noni



chlorosis (interveinal)
citrus



dieback
koa



erinoise mite
lychee



fruit rot
Phytophthora, papaya



fruit spot
Asperisporium fungus, papaya



galls
erineum mites, hibiscus



gummosis
shower tree



leaf blight (black leaf streak)
Sigatoka fungus, banana



leaf spot
Cercospora fungus, coffee



leaf spot
Cercospora fungus, lettuce



leaf spot
Asperisporium fungus, papaya



mosaic
cucumber mosaic virus, 'awa



necrosis (internal stem)
Verticillium fungus, naupaka



postharvest rot
Rhizopus fungus, noni



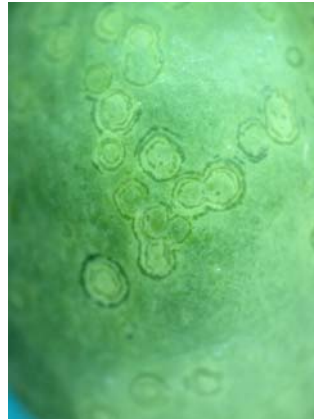
powdery mildew
fungus, papaya



powdery mildew
fungus, poinsettia



ringspot
papaya ringspot virus



root-knot
galls from nematodes, beans



root-knot
galls from nematodes, noni



root rot
Pythium, 'awa (kava)



rust
fungus, 'ōhi'a



rust
fungus, heliotrope



rust
fungus, plumeria



scab (fungal scab)
Elsinoe fawcettii, citrus



scorch
abiotic symptom, avocado



soft rot
bacteria, orchid



sooty mold
fungus, noni



sooty mold
fungus, palm



whitefly
tomato



wilt and dieback
Verticillium fungus, naupaka

