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Prevention and Control of Pests and Diseases

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Summary

A well-controlled growth environment with plants that are not unduly stressed is essential for *Arabidopsis* molecular biology research. Even if they do not kill the plants outright, insect pests and microbial pathogens can cause subtle changes in gene expression or plant metabolism that affect experimental results. Therefore, regular scouting for infestations, frequent cleaning of plant growth areas, proper disposal of dead or diseased plant material, and controlled access to the greenhouses or growth chambers will help to make experiments more reproducible. Powdery mildew, a fungal pathogen, and arthropod pests, including aphids, thrips, fungus gnats, and spider mites, are the most common greenhouse problems. Biological control methods such as parasitoid wasps and *Bacillus thuringiensis* crystal toxin can be used to contain some insect infestations. However, if an infestation gets out of hand despite reasonable precautions, insecticide or fungicide spraying by a licensed applicator may be necessary. Bacterial and viral infections of *Arabidopsis*, though they do occur, tend to be less common and can usually be controlled by maintaining optimal growth conditions and promptly disposing of dead or diseased plant material.

Key Words: Fungus gnats; thrips; aphids; spider mites; powdery mildew; *Botrytis*.

1. Introduction

Most *Arabidopsis* experiments require healthy plants that are free from pests and diseases. As *Arabidopsis* plants are grown from seed, have a short life cycle, and are often raised in growth chambers rather than greenhouses, it is possible to maintain an almost entirely pest-free growth environment. However, pests including insects, mites, and pathogens do sometimes find their way into *Arabidopsis* growth facilities. Early recognition, treatment, and continued monitoring are key to preventing a pest outbreak from getting out of control.

Many greenhouse pests, including thrips, aphids, spider mites, and powdery mildew, can infest *Arabidopsis* ***(1)***. Therefore, common greenhouse practices used in the care of other plant species can be applied equally well to preventing pest and pathogen infestations of *Arabidopsis*. This chapter focuses on the monitoring and management of pests and pathogens that are frequently found on Arabidopsis. It also includes a general description of plant symptoms, life cycles of the most common pests, and some of the available treatment options. Some specific chemical treatments and biological control organisms are discussed, but this does not represent an exhaustive list, nor is this chapter an endorsement for any particular chemical manufacturer. The specific treatment strategy adopted to control a particular pest depends on many factors and it is not possible to give a general approach that will apply to all *Arabidopsis* growth situations.

2. Pest Prevention Using Good Horticultural Practices

Healthy, vigorously growing plants will generally be less prone to pests and diseases than weak or stressed plants. Attention to the plants and their growth environment minimizes the

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risk of a pest outbreak *(2)*. It is much easier to prevent a pest problem than it is to contain one after it gets started, and early detection will make any outbreak that does happen much easier to manage. Here, we outline good cultural practices that will help to prevent pest outbreaks.

2.1. Growth Area

1. Keep the growth area clean. Start with a clean shelf or greenhouse bench. Used soil and old plant debris can harbor insects or fungal spores from previous plantings. Therefore, before planting, clean the bench area thoroughly.
2. Periodically schedule a time when the growth area can be completely emptied out. Wash all surfaces with a greenhouse disinfectant such as Zerotol™ (Biosafe Systems), Physan 20™ (Maril Systems), or 10% bleach. Bleach will leave a residue, so if using bleach, it is important that it be rinsed off afterwards. After cleaning, turn up the temperature in the growth facility as high as possible for 2 to 4 d. Because you have removed the host material, sanitized, and "cooked" the area, it is very unlikely that any pests or spores will survive.
3. Clean the floors of the greenhouse or growth room weekly. Sweep up debris and mop the floors.

2.2. Growth Supplies

1. Always use clean or new pots and trays. For potting soil, use bagged, commercial, peat-based soilless mixes. If pots or flats must be reused, they should be cleaned thoroughly and sterilized by soaking overnight in 10% bleach solution and rinsing with water afterward.
2. Some *Arabidopsis* growers choose to autoclave their soil before planting in order to kill any pests or pathogens that may be present. This is not advisable, however, because it may adversely affect the soil properties. Commercial mixes normally contain a wetting agent and a nutrient charge that may be altered after autoclaving. Autoclaving soil also involves much extra labor.

2.3. Dead Plant Material

1. Do not leave dead or drying plants in the growth area. When plants are no longer needed, bag them securely and remove them from the growth facility. Also, remove cells that do not have any germinated seeds from plant flats. These cells will stay wetter than ones with plants growing in them and can breed fungus gnats and diseases.
2. Plants that are dry and awaiting seed harvest should be moved out of the growth facility. This reduces the chance that pests from potentially heavily infested senescing plants will migrate to vulnerable seedlings. The ideal situation is to have a separate seed harvest room, where senescing or dry plants can be stored at room temperature, away from the growth facility, allowing valuable growth space to be used for actively growing plants.

2.4. Controlling Pest Outbreaks

Avoid introducing and spreading pests.

1. If there are already pests in the growth facility, establish an entry protocol whereby anyone who has entered an infested area refrains from entering uninfested areas later on that same day. Make sure that all the users of the growth facility adhere to this protocol, even if it seems inconvenient.
2. Wear a lab coat when working in the growth facility. Pests are easily spread on clothing, both by people moving within the facility and by people coming in from the outdoors. Dedicate a set of "growth area only" lab coats for all users of the facility. Use sticky mats at the entrance of the growth facility to help prevent pests from being tracked in.
3. Quarantine any new plants to avoid introducing pests from another growth area. Isolate the new material for a few days and carefully inspect it for emerging pests. This waiting period is important because pests that are not visible initially may hatch out.
4. The easiest way to control a pest outbreak is often to dispose of the affected plants. Although this may set back a research project for a few weeks, it is worth considering in order to curtail a more serious outbreak. If the infested plants are essential and must be saved, move them to a separate growth chamber or other isolated area that is away from unaffected plants. When

moving infested plants, prevent pests from spreading by covering the flat with a dome or by bagging the flat. Clean the cart and dome afterwards.

5. Keep similarly aged plants together. Plants of the same age will be easier to monitor for pests. Older, senescing plants seem to be more vulnerable to pests and it is best to keep them separated from young seedlings. The ideal approach is to coordinate growth of plants in a particular area so that they mature at about the same time, allowing the growth facility to be cleaned out before planting again.
6. A licensed pesticide applicator or greenhouse manager will be able to handle pesticides safely and properly. In the United States, the regulations of the Worker Protection Standard (www.epa.gov/pesticides/safety/workers/workers.htm) must be followed to protect workers and pesticide handlers from exposure. Local rules may vary, and different regulations for pesticide use will apply in other countries. Some of the pesticides mentioned in this chapter are required by law to be applied by someone who has had pesticide applicator training. In some cases it is necessary to restrict access to the growth facility while the pesticides are being applied, as well as for a period immediately afterward. Pesticide labels describe the specific "reentry interval," as well as the required personal protective equipment (nitrile gloves, respirator, etc.).
7. For spraying fewer than 10 flats, a small hand sprayer will be sufficient. For larger infestations, use a pump sprayer. Set the nozzle to a fine setting. It is best to create a "mist" of small droplets that will evenly cover the surfaces of the plant rather than a "rain" of larger ones.
8. Some pesticides, such as those used to treat fungus gnat larvae, must be applied as a "sprench." The goal is to spray and drench the soil at the same time. For fewer than 10 flats, a common laboratory squirt bottle works well for this. For larger numbers of flats, use a pump sprayer with the nozzle set at the coarsest setting.
9. Always check for phytotoxicity on a few plants before undertaking a large-scale pesticide application. Read and follow the directions on the pesticide label, use proper personal protective equipment, adhere to reentry interval guidelines, and keep good records of all pesticide applications.
10. Consider the growth environment before spraying. Greenhouses are usually designed so that they can be sealed off before spraying, but that is not necessarily the case for growth rooms, which may vent into work spaces. Make sure that the air from the growth rooms does not recirculate with air in neighboring labs and offices. Growth chamber manufacturers recommend that pesticides should not be applied in reach-in chambers.
11. Biological pest controls such as predatory mites, parasitoid wasps, nematodes, and the bacterium *Bacillus thuringiensis* can be used on *Arabidopsis*. However, such control methods are not absolute and usually require some tolerance to low-level pest populations. Extremely keen scouting abilities are also important to ensure that biological controls are implemented early in an infestation. Biological pest control is not always appropriate for *Arabidopsis* research because even a low-level pest problem can alter plant phenotypes beyond experimental parameters. Biological control agents work best when applied before a pest outbreak, rather than once it has started. Therefore, weekly releases of biological control organisms are recommended.

2.5. Early Signs of Pest Outbreaks

Learn the early signs of pest outbreaks and monitor for these. Often, damaged or sick plants can be recognized before the pests themselves are observed ***(3)***. Regularly inspect all plants in the growth area. Keep a record of the locations where you suspect a pest outbreak and check those areas closely.

2.6. Maintaining Healthy Plants

1. Provide optimal plant growth conditions (temperature, light, humidity) to generate healthy plants (***4***; *see also* Chapter 1 in this book). *Arabidopsis* grows well at 20 to 23ºC, 50 to 65% relative humidity, and a light intensity of 80 to 150 μE.
2. Do not overwater or underwater plants. Thoroughly wet the soil and then allow it to dry out slightly before watering again. In most situations watering two times per week is sufficient, though the watering schedule will change with the age of the plants. An added advantage to

such a watering protocol is that the incidence of fungus gnats and other underground pests is reduced if the soil is not continuously wet.
3. Ensure that there is good air circulation around plants.

3. Arthropod Pests

Fungus gnats and thrips are the most common pests of *Arabidopsis* in growth chambers, growth rooms, and greenhouses. Aphids and spider mites are less problematic, but can also infest *Arabidopsis*, especially in greenhouses where they may already be established on other plants.

3.1. Fungus Gnats

Fungus gnats (*Bradysia* spp.) are one of the most serious *Arabidopsis* pests. They can appear, seemingly from nowhere, and quickly take hold. Often, fungus gnats are not detected until the adult flies emerge. However, it is the larval stage that does all the damage and it is therefore important to monitor plants for signs of larval feeding. Fungus gnat larvae feed on tender roots, fungi, decomposing organic matter, and decaying plant tissue in the soil. They will also chew on leaves that are touching the surface of the soil. Larvae pupate underground and emerge at the surface as adult gnats. Adult fungus gnats do not harm the plants, except by laying more eggs, which soon hatch into more hungry larvae. The full fungus gnat life cycle takes 3 to 4 wk, which may account for the fact that the heaviest infestations are usually seen on older plants.

Shore flies (*Scatella* spp.*)* are often confused with fungus gnats. These flies do slightly resemble adult gnats, but are stockier, have white spots on their wings, and tend to fly when disturbed. Fungus gnat adults will generally run along a surface when prodded. Shore flies are associated with algae and breed in standing water such as that found in drainpipes and sink traps. They are not a significant pest to *Arabidopsis* and are not covered here.

3.1.1. Fungus Gnat Prevention

1. Fungus gnat eggs can be found in commercial, bagged soilless mixes, especially those that are high in partially decomposed organic matter. Before planting, drench the soil with a suspension containing *Bacillus thuringiensis israelensis*, which is sold as Gnatrol™ (Valent Biosciences). Any fungus gnat larvae that hatch in the soil will be killed after they ingest the *B. thuringiensis* toxin.
2. Allow growth media to dry slightly between watering and do not leave pools or puddles of standing water on the floor or bench. Fungus gnats thrive in damp, humid conditions that also promote fungus growth.

3.1.2 Fungus Gnat Detection

1. Plants grow slowly and lack vigor when fungus gnat larvae chew their roots. Seedling and early-rosette-aged plants are the most vulnerable.
2. Small round holes will appear on leaves touching the soil surface where the larvae have sampled leaf tissue (**Fig. 1A**). Seedlings are extremely vulnerable to fungus gnat larvae. With severe infestations, whole seedlings can be devoured overnight. If suspected fungus gnat holes are seen, carefully lift up the leaf and look for whitish translucent larvae with black heads (**Fig. 1B**). The larvae will avoid light, and usually respond by burrowing into the soil.
3. Yellow sticky cards (Whitmire Micro-Gen) hung throughout the growth area will catch adult gnats (**Fig. 1C**). These cards work only as a monitoring device and should not be considered a control method. Count the number of gnats on each card and keep records as a quantitative measure of the severity of the infestation. Continue monitoring after treating for the gnats to determine the efficacy of the treatment.
4. Cut a small (half-inch) plug of raw potato and place this on the soil surface. After one day, lift the plug and look for larvae feeding on the potato. This is also a good technique to use for monitoring the success of soil treatment, or as part of a regular scouting of the growth facility. Mark the positions where the potato plugs are and check them every few days. They will need to be replaced as they dry out or become moldy.

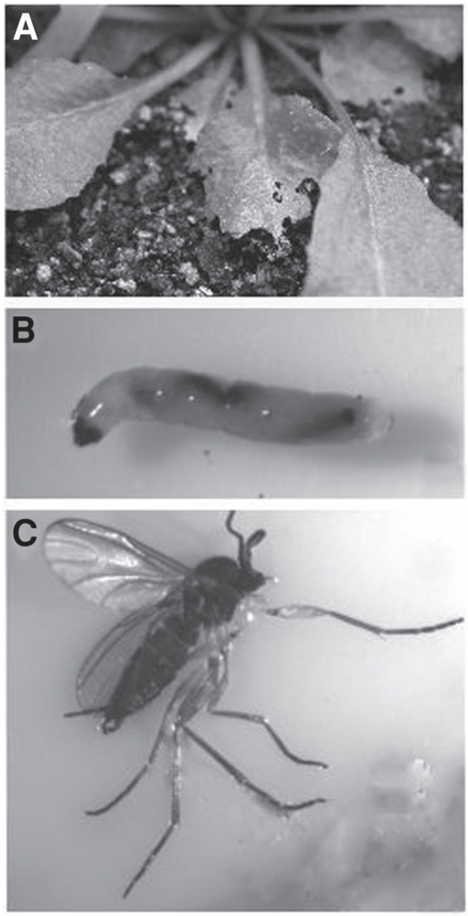

Fig. 1. Fungus gnats (*Braydisia* sp.). **(A)** *Arabidopsis* leaves chewed by fungus gnat larvae. **(B)** Fungus gnat larvae, approx 2 mm. **(C)** Fungus gnat adult trapped on sticky card, approx 2 mm.

3.1.3. Fungus Gnat Treatment

1. Treatment should be targeted at the larval stage. It difficult to control adults because they hide in places where they are not easily reached by pesticides. Fungus gnat larvae live near the soil surface, so the pesticide should be applied to the upper inch of the soil. This can be done using a spray nozzle on a coarse setting and spraying the material onto the soil, or by carefully watering the solution onto the flats. Eggs and pupae are generally not susceptible to pesticides, so it will be necessary to repeat applications, following the directions on the label, every 5 to 7 d until the infestation is eliminated.
2. Gnatrol is not only a good preventive treatment for fresh soil, but also a good pesticide to use on existing fungus gnat infestations. After ingestion of the *B. thuringiensis* bacterial toxin, the gut becomes paralyzed, feeding stops, and the larvae die. The strain of *B. thuringiensis* in Gnatrol specifically targets fly larvae and is not harmful to Arabidopsis or biological control insects that are used in the greenhouse. Gnatrol is effective for only 48 h, so it must be applied repeatedly.
3. Insect growth regulators (IGRs) are effective against fungus gnat larvae. These are pesticides that interfere with the normal development of the insect pest. S-kinoprene, sold as the product Enstar II® (Wellmark International) can be applied as a drench to the soil. Another IGR, azadirachtin, sold as Azatin® XL (Olympic Horticultural Products), is also applied as a drench and is extremely effective against fungus gnat larvae.

4. In addition to *B. t. israelensis* mentioned above, the predatory mite *Hypoapsis miles* and the predatory nematode *Steinernema feltiae* can be used on *Arabidopsis* for biological control of fungus gnats with good results. Both of these organisms need to be incorporated into the soil at planting time. The nematodes are sold under many brand names and, to ensure obtaining viable nematodes, it is best to purchase them through a reputable biological control supplier. A list of biological control suppliers in North America can be found at the Web site: www.cdpr.ca.gov/docs/ipminov/ben_supp/contents.htm

3.2. Thrips

Thrips, in most cases the species *Frankliniella occidentalis,* are another serious pest of Arabidopsis. In their 3-wk life cycle, these insects pass from eggs through two larval and two pupal stages to become adults. Pupae are generally in the soil, but all other growth stages are found on the plant. Thrips are good at hiding in axils, rosette centers, and even flower buds. As a result, they are difficult to control and early detection is particularly important.

3.2.1. Thrip Prevention

1. Regular, methodical monitoring is important for detecting thrips in the early stage of infestation.
2. Old soil and dead dry plant debris can harbor thrips and thrip pupae. When disposing of plant materials, completely remove them from the growth facility. A trash bag full of plants left by the door can serve as a reservoir for thrips waiting to colonize new plants.
3. Thrip-proof screens made from 0.16-mm mesh can be installed on greenhouse vents. Keep in mind, however, that these screens will severely restrict the air flow in the greenhouse. Additional ventilation may be required if such mesh is installed.
4. Keep greenhouse floors clean and clear of weeds that can harbor thrips.
5. Make preventive releases of the predatory mites *Neoseiulus cucumeris* or *Hypoapsis miles* (discussed below).

3.2.2. Thrip Detection and Monitoring

1. Typically thrip damage will be seen before the insects themselves are detected. Thrips scrape the leaf tissue and then consume the plant juices, which results in silvery-white patches that are usually seen at leaf edges, or near the center of the rosette (**Fig. 2A**). Sometimes the white patches will be covered with black dots of thrip frass.
2. Distorted expanding leaves are often caused by thrip damage.
3. Adult thrips are tiny, slender, and difficult to see. They are 1 to 2 mm long and resemble a small splinter (and are just as aggravating) (**Fig. 2B**). Larvae of thrips are slightly smaller, but can also be seen (**Fig. 2C**). If thrip damage is discovered, blow on the plant gently and then look at the damaged area with a hand lens. In response to the CO_2 in your breath, the adult and larval thrips are likely to come crawling out from the center of the rosette where they are hiding.
4. Tap plants, especially flowering or senescing plants, over a piece of white paper, and thrips will be seen crawling on the paper. In a similar manner, thrips are often found when harvesting seeds.

3.2.3. Thrip Treatment

1. Thrips can be suppressed with insecticidal soaps such as Safer®Brand (Woodstream Corp.), or M-Pede® (DowAgroSciences), but complete control is difficult. If plants are stressed, due to severe infestation or some other factor, soaps can sometimes damage leaves.
2. Thrips are known to readily develop resistance to several pesticides. For this reason it is important to rotate pesticides with different modes of action. Conserve™ (Dow AgroSciences), Mavrik Aquaflow® (Wellmark International), and Avid® (Novartis) all work against thrips on *Arabidopsis*. Plan to make two or three applications 7 d apart.
3. Treat all plants in the infested growing area. Adult thrips fly, and can also be transported on clothing.
4. Biological control is particularly challenging with established populations of thrips. However, the predatory mite *Neoseiulus cucumeris* can be used, and it does have the advantage of being

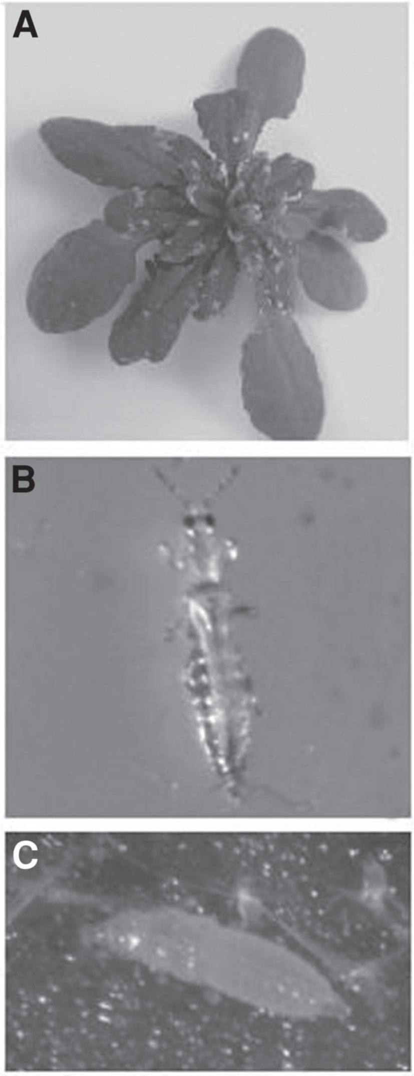

Fig. 2. Thrips (*F. occidentalis*). **(A)** *Arabidopsis* leaves damaged by thrip feeding. **(B)** Adult thrip, approx 1.5 mm. **(C)** Thrip larva, approx 0.5 mm.

able to reach into the thrips' hiding spaces (axils, flowers, etc.) where pesticides may not reach. This mite is especially good at attacking first instar larvae. Use of this mite does require a lot of attention to scouting and monitoring of both the predator population and the thrip population. Several releases of *N. cucumeris* will be required, and, if chemical pesticides are used, they must be compatible with this mite. Ask the biological control supplier about pesticide compatibility when purchasing mites. The soil-dwelling mite *Hypoapsis miles* will attack thrip pupae as well as fungus gnat larvae.

3.3. Aphids

Aphids are common greenhouse pests that also infest *Arabidopsis*. Aphid outbreaks in growth chambers or growth rooms occur less frequently, but there can be transfers of aphids from nearby greenhouses. The green peach aphid, *Myzus persicae,* is the species that is most frequently found on *Arabidopsis*, though other species (e.g., melon aphids and root aphids)

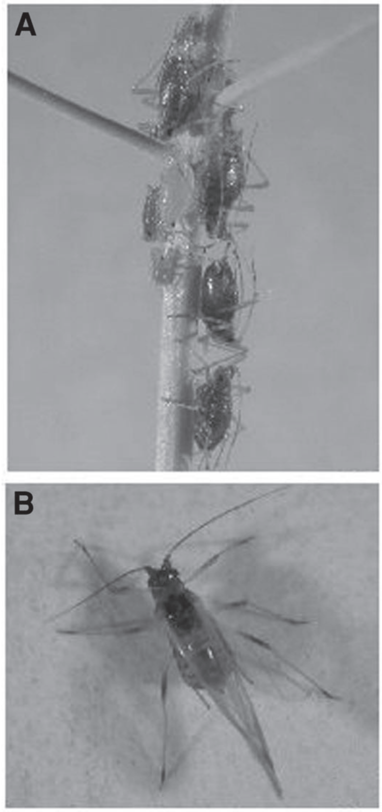

Fig. 3. Aphids (*M. persicae*). **(A)** Nymphs and adults feeding on an *Arabidopsis* stem, 0.5 to 2 mm. **(B)** Winged adult aphid, 1.5 to 3 mm.

have been reported. *M. persicae* are soft-bodied, yellow, green, red, or tan pear-shaped insects, 1 to 3 mm long, that are usually seen in clusters on the undersides of leaves or along the stems. Aphids reproduce by parthenogenesis, with females giving birth to genetically identical first-instar offspring. Because of this, even a single aphid can quickly become a large infestation. Winged asexual female aphids are produced when food becomes limiting—for instance, when plants start to go to seed or are drying down. These winged parthenogens can spread quickly and invariably produce wingless female offspring. Aphids can also reproduce sexually, but this is initiated under cool temperature (15°C) and short daylight conditions that are not commonly found in *Arabidopsis* growth facilities.

3.3.1. Aphid Prevention

Screens on greenhouse vents, diligent inspection of nearby host material, and good housekeeping in the growth facility will help deter aphids.

3.3.2. Aphid Detection and Monitoring

1. Aphids can be found on flower stalks, on young leaves in the rosette center, and on the undersides of leaves (**Fig. 3A**). Winged aphids (**Fig. 3B**) are highly mobile and typically can be found on the rosette leaves. They are often trapped on the yellow sticky cards used to monitor fungus gnats.
2. A sticky, moist "honey dew" will coat the plant surfaces when aphid infestations are severe. Sometimes this honey dew will turn black with sooty mold.

3.3.3. Aphid Treatment Suggestions

1. Insecticidal soaps such as SaferBrand or M-Pede are effective for treating aphids on *Arabidopsis*, as long as the plants are otherwise reasonably healthy. Early detection of aphid infestations is important because soap treatment can burn the leaves of stressed plants. As with any chemical treatment of *Arabidopsis*, it is always a good idea to do a test spray on a few plants first.
2. Other suggested aphid pesticides are Mavrik Aquaflow® and Enstar II® (both from Wellmark International).
3. *Aphidius colemanii* is a small parasitoid wasp that sometimes enters greenhouses without assistance, or can be purchased from a biological control supplier. Female wasps lay eggs in adult aphids, which hatch and feed on the dying aphid. Adult wasps emerge and go on to parasitize more aphids. Evidence of wasp predation can be seen as tan-colored "shells" within clusters of aphids, sometimes referred to as "aphid mummies."

3.4. Spider Mites

The two-spotted spider mite, *Tetranychus urticae,* is a common greenhouse pest that can be found on *Arabidopsis*. Like aphids, spider mites are sap-sucking and can cause leaves and flowers to wilt and die. The entire mite life cycle takes about 3 wk: Eggs hatch into hungry nymphs, the nymphs go through several cycles of feeding followed by dormancy, and adults hatch after the third dormancy period.

3.4.1. Spider Mite Prevention

1. Spider mites thrive in hot, dry conditions. Maintaining a high relative humidity (greater than 50%) will deter spider mites.
2. Sanitation is important for preventing spider mite infestations, in particular if there has been an outbreak recently or nearby. Spider mites are good at hiding in cracks in the floor and in plant debris. Diapause, a type of hibernation, can occur when temperatures drop, days get shorter, or when the food supply gets low, such as when plants are senescing. Fertilized adult females will stop feeding and laying eggs, and will hide in cracks until conditions improve.

3.4.2. Spider Mite Detection

1. All life stages of spider mites are usually found on the undersides of leaves, which makes early detection difficult. Once established, however, they can sometimes be seen crawling across the leaf surface or near the center of the rosette (**Fig. 4A**).
2. Mite feeding causes a stippling effect, in which the leaves become covered with white or yellow chlorotic spots. In early infestations, the stippling tends to be more concentrated near the leaf margins (**Fig. 4b**).
3. When the infestation becomes severe, spider mites will spin threadlike webbing that covers the inflorescences. Mature flowering plants and senescing plants are especially vulnerable.

3.4.3. Spider Mite Treatment

1. Only adult spider mites and feeding nymphs are susceptible to pesticides. The eggs and dormant nymphs will not be harmed by chemical treatment. Therefore, it is important to plan on multiple pesticide applications 5 to 7 d apart. Insecticidal soaps such as Safer Brand or the pesticides Avid and Mavrik Aquaflow are effective chemical controls.
2. Several predatory mites that consume spider mites are available from biological control suppliers. Some will provide more immediate control, whereas other types will establish themselves and provide control over the longer term. One caution is that some species of predatory mites will feed on pollen when they run out of prey species. This could potentially cause crosspollination, which most *Arabidopsis* researchers want to avoid. Ask the supplier for a mite that does not feed on pollen.

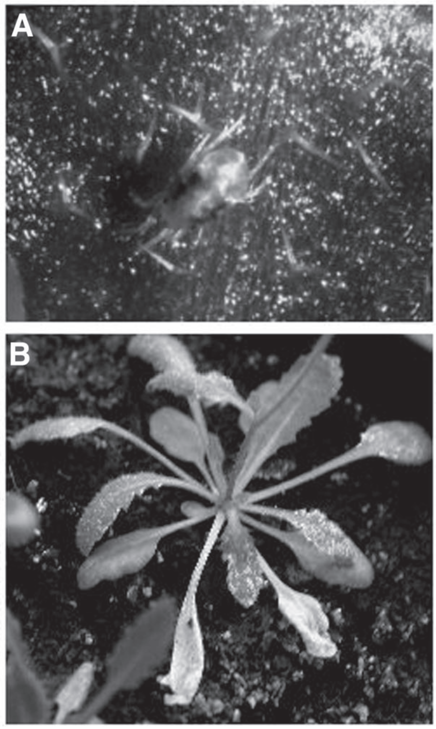

Fig. 4. Spider mites (*T. urticae*). **(A)** Spider mite adult, approx 0.5 mm. **(B)** *Arabidopsis* plant with leaf stippling caused by spider mite feeding.

4. Diseases

The incidence of severe diagnosed microbial diseases in laboratory-grown *Arabidopsis* is relatively uncommon, at least in part due to the short life cycle of *Arabidopsis*, the controlled conditions that most researchers use to grow their plants, and the fact that *Arabidopsis* plants are always grown from seed. In greenhouses, fungal and bacterial pathogens typically occur in situations where there is high humidity, poor air circulation, or continuously wet soil. Viruses are usually spread either by insects or when plants are propagated from cuttings. However, *Erysiphe* sp. (powdery mildew) and *Botrytis cinerea* (gray mold), two fungal pathogens that are familiar in horticultural settings, do occasionally cause spontaneous *Arabidopsis* infections, and are described below.

On the other hand, it is likely that many cases of pathogens infecting *Arabidopsis* remain unidentified, because it is often easier to dispose of a set of sick-looking plants and start over, rather than trying to identify the potential microbial agent causing the problem. We know from studies of plant–pathogen interactions that *Arabidopsis* is susceptible to many pathogens, including a variety of viruses, bacteria, and both necrotrophic and biotrophic fungi. Almost everyone working with *Arabidopsis* has encountered unexplained symptoms such as wilting, yellowing of the entire plant, chlorosis of leaves, anthocyanin accumulation, or general lack of vigor. Often, this is attributed to a "bad batch of soil," but it is possible that these sickly plants are infected with an unidentified pathogen.

*4.1. Powdery Mildew (*Erysiphe *sp.)*

Powdery mildew is an obligate biotrophic pathogen that infects many agricultural crops. Species in the genus *Erysiphe* commonly infect *Arabidopsis* in greenhouses and growth cham-

Fig. 5. Powdery mildew (*Erisyphe* sp.) on *Arabidopsis* leaves.

bers. Airborne fungal spores settle and germinate on living plant tissue. After germination, the mycelium grows over the surface of the leaf, projecting food-absorbing haustoria into the epidermal cells. The pathogen develops in this manner, invisible to the naked eye, until spore-producing conidiophores are formed. This reproductive phase is visible on the leaf as a patch of white "powder" that consists of the conidiophores and the multitudes of spores ready to become airborne, continuing the cycle.

4.1.1. Powdery Mildew Prevention

1. Reduce the relative humidity immediately surrounding the plants by allowing good air circulation around plants. Keep plants spaced apart and use fans to keep air circulating throughout the growth area. Although powdery mildew spores can germinate at a lower humidity than most fungi, the progression of the disease on the plant is increased in high humidity environments. Do not leave water standing in the trays beneath the pots, as this increases the humidity immediately surrounding the plants.
2. Control the temperature and humidity in the growth facility. Warm days with low relative humidity combined with cool nights with high relative humidity favor development of powdery mildew. These conditions are common in winter in greenhouses that do not have good temperature control.
3. Clean the growth area between plantings. After removing all plants, wash all surfaces (walls, floors, benches, etc.), and then turn up the temperature as high as possible for a few days. Cleaning will remove all remnants of plant material on which spores might survive. Heating the area will dry out and kill any remaining spores.

4.1.2. Powdery Mildew Detection

1. White patches of mildew will first become visible at the tips of the upper surfaces of older rosette leaves (**Fig. 5**). When the powdery mildew infection is advanced, cauline leaves, stems, and siliques will also show these patches.
2. Powdery mildew is most easily detected on plants that have reached the large-rosette stage. Plants that are much younger and otherwise appear to be healthy can still have a powdery mildew infection that has not yet reached the reproductive phase and is thus invisible to the naked eye.

4.1.3. Powdery Mildew Treatment

1. Early treatment is important because the airborne spores are easily spread. Avoid spreading powdery mildew by not entering other plant growth areas after exposure to infected plants.

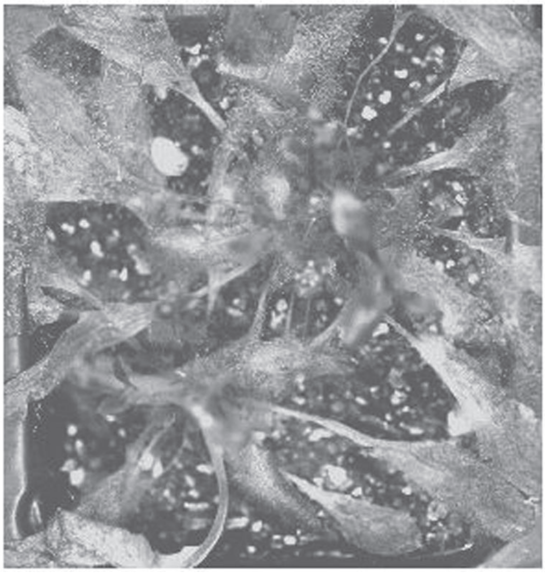

Fig. 6. *Botrytis* infection on an *Arabidopsis ein2* mutant plant. Under normal growth conditions, *Botrytis* infections are uncommon and do not result in extensive lesions. However, some commonly used *Arabidopsis* mutants, such as *ein2*, are hypersusceptible to *Botrytis* infection. *Botrytis* can also cause extensive damage to plants that are grown under high-humidity conditions.

2. When powdery mildew is first discovered, cover the affected plant with a damp paper towel, which reduces the spread of the spores. Spraying plants or moving infected plants drives spores onto neighboring plants.
3. Sulfur-based fungicides sold for use in house and garden are effective against powdery mildew. Use according to the directions on the label. In some cases, however, sulfur sprays may cause older leaves to turn yellow or senesce prematurely. Do a test spray on a few plants.
4. A fungicide that is effective against powdery mildew is 3336F (Cleary Chemical Corp.).

*4.2. Gray Mold (***Botrytis** *sp.)*

Botrytis is not very common on *Arabidopsis*, but in high-humidity conditions, plants that are already weak, stressed, or compromised in some way can become infected. *Arabidopsis* mutants in the ethylene signaling pathway (for example, the commonly used *ein2* mutant) are particularly susceptible to *Botrytis* infections.

4.2.1. Botrytis *Prevention*

1. Keep the growth area clean and remove plant debris that can harbor *Botrytis* spores.
2. Healthy, vigorously growing plants are unlikely to become infected with *Botrytis*. Avoid wounding or damaging plants when handling flats. Wound sites are frequent areas of infection.
3. Keep the humidity low around plants by providing good ventilation and not overcrowding plants. In a greenhouse, when night temperatures drop, a thin film of moisture can condense on plant surfaces, creating an ideal condition for *Botrytis* spores to germinate. Airflow around the plants helps to keep leaves dry. Water plants early in the day, and do not leave water standing in the tray.

4.2.2. Botrytis *Detection and Treatment*

1. *Botryis* appears as a gray mold on older leaves (**Fig. 6**).
2. The fungicide 3336F is reported to be effective against *Botrytis*.

North American Biological Control Suppliers can be found at the Web site: www.cdpr.ca.gov/docs/ipminov/ben_supp/contents.htm.

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