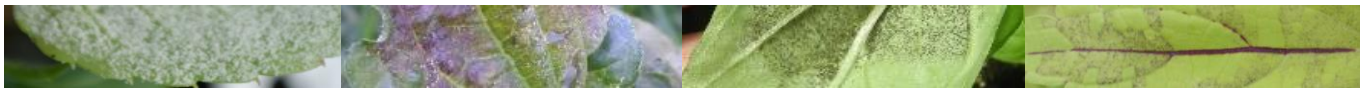

Dossier: Rose Downy Mildew

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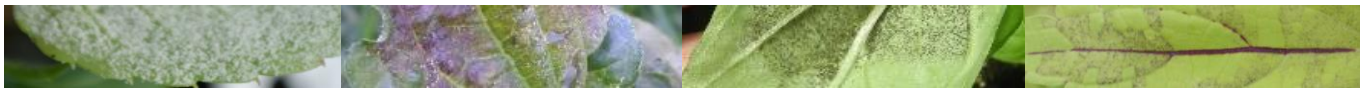
Introduction and Importance

Roses (*Rosa* sp. L.) are widely popular and economically important in the ornamental shrub and cut flower industry, not only in the U.S. but also in other countries such as the Netherlands, Colombia, Ecuador and Kenya [1]. According to the 2015 USDA Floriculture Crops Summary, the potted and cut roses industry in the U.S have a retail value of > $ 42 mill. The downy mildew disease of rose caused by *Peronospora sparsa* was first described in England in 1862 [2], and by 1880 was first reported in Midwestern U.S [3]. Currently, the disease is distributed across the country and rest of North America, Asia, Africa, Europe, Oceania and South America [4,5]. The global spread of rose downy mildew might be due to the commercial trade of plant material, as every single plant part has the potential of carrying the pathogen without symptoms being visible to the naked eye [5]. During the last 20 years, rose downy mildew has become a serious problem for U.S. growers, causing significant loses every year [6]. Although rose downy mildew is more severe in greenhouse conditions, it is also a serious problem in field grown roses when environmental factors favor the development of the disease [3]. All rose cultivars are considered to be susceptible to the downy mildew disease, although they can differ greatly in their sensitivity. The cost of the major diseases of rose, which includes downy mildew, can be up to half of the total cost of the crop [7].

Plant Description

Genus *Rosa*

Rosa is a genus of woody perennial flowering plants in the family Rosaceae and comprises approximately 190 species. Most species are native to Asia, with smaller numbers native to Europe, North America, and northwestern Africa [3]. Rose species are distributed widely in temperate and subtropical habitats in the Northern Hemisphere [8] where they can grow in sites ranging from riparian and swampy to dry, hot deserts. Roses were independently



domesticated in both Europe and China several thousands of years ago for a range of uses including for medicinal, ornamental and fragrance industries [7]. Roses are a group of plants that can be erect shrubs or climbing/trailing vines with stems with sharp thorns that are used to deter predators. The multipetal flowers vary in size and shape and are usually large and showy, in colors ranging from white through yellows and reds [8]. The leaflets of rose plants are ovate and range from medium to dark green in color and grow alternate in clusters varying in number. The flowers of roses can range from simple, five-petaled blooms to large, heavily compacted ones. Rose plants vary widely in size, with miniature roses only reaching a maximum height of 8 inches, and some species of climbing roses reaching up to 50 feet [9]. Species, cultivars and hybrids are all widely grown for their beauty and fragrance, however the system of cultivar classification has become complicated and inexact due to the extensive hybridization and selection which has given rise to thousands of cultivars [3]. Roses can be propagated by rooting cuttings, by budding, or by grafting [3]. Roses thrive in sites with full sunlight or with at least 6 hours of sunlight, and usually are tolerant of most soil types albeit well drained; however, they do better in a relatively fertile soil high in organic matter with a soil pH of 6.0-7.0 [10].

Figure 1. Hybrid tea rose

Photos taken from: https://maxpull-gdvuch3veo.netdna-ssl.com/wp-content/uploads/2010/05/red-rose.jpg

Pathogen Description

The rose downy mildew disease is caused by *Peronospora sparsa* Berk (syn. *Peronospora rubi* Rabenh. ex J. Schröt). This species was first described in 1862 [2]. The name of this pathogen reflects the characteristic sparse production of white spores on the infected rose tissues [6]. *Peronospora sparsa* is a fungus-like organism, known to belong to the oomycetes or water molds in the family Peronosporaceae. Microscopic features of this pathogen include intracellular haustoria, hyaline dichotomously branched sporangiophores can be observed emerging from stomata on the undersides of leaves, with straight trunks and branched 3 to 4 times with bifurcated tips. Typically, one branch of each pair curved inward and one reflexed. Sporangiophores ended with sporangia that can be slightly ovoid, colorless to yellowish-brown [11]. Hyaline or pale-yellow colored oospores with thick cell wall can form mostly in the spongy parenchyma of leaves but can also be found in sepals, flowers, buds, stems and petioles [12]. The regular formation of oospores might be an indication this pathogen is homothallic.

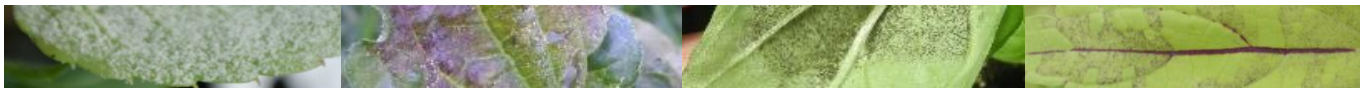
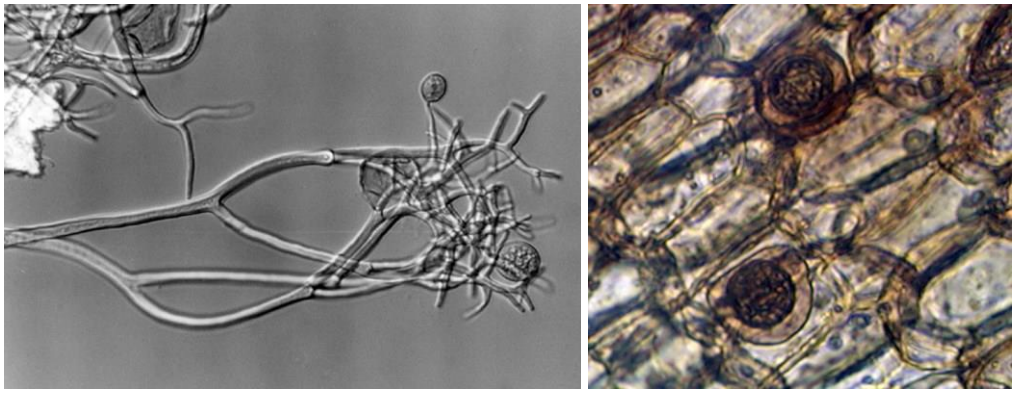


Figure 2. *Peronospora sparsa*: Sporangiophores, sporangia and oospores



Photos taken from: http://www.cabi.org/portfolio/compendia/normal/61169.img and http://www.cabi.org/portfolio/compendia/normal/61174.img

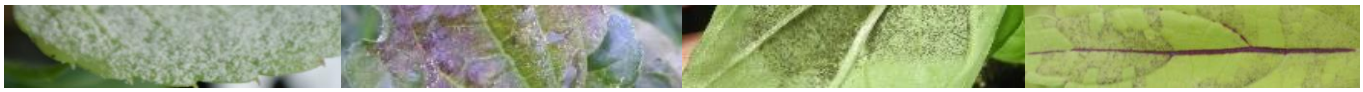
Pathogen Biology and Disease Cycle

According to field observations of roses grown in open fields and greenhouses, *P. sparsa* produces oospores and zoospores. Sporangia are the sac-like structures that where zoospores are formed. Zoospores are motile spores that are the result of asexual reproduction and under the right environmental conditions are produced in the abaxial (lower) surface of infected leaves, stems and flower buds. Oospores are the result of sexual reproduction and are usually produced in the internal leaf tissues. Rose downy mildew spreads mainly through the propagation of infected plants, through the use of infected mother plants and/or by inoculum present on infected plant debris. Zoospores are moved by wind or water currents.

Once a *P. sparsa* zoospore reaches a rose host and environmental conditions are conducive, zoospores germinate and produce appressorium that penetrates the leaf epidermis directly into the periclinal cell wall, in between the anticlinal cell walls or through the stomata. Once inside the leaf tissues, the developing hyphae spread intercellularly forming a cushion of mycelia just below the stomata. After the incubation period, sporangiophores can emerge from the stoma openings or from stem or flower tissues. After emergence, sporulation reaches its final stage and spores are produced. If water is present in the surface of the leaves, sporangia can germinate and produce massive quantities of zoospores that can cause secondary infections within the same plant, or being spread by moist wind or water splash. Sporangia can also be dislodged from the leaf tissue and fall in the ground where the spores can be released and spread short distances by water splash, or longer distances by wind currents to nearby plants where the disease cycle starts again. Oospores can form inside the mesophyll of infected rose leaves and stems and might function as source of inoculum for new infections.

Epidemiology

Under humid, cool conditions, sporangiophores and sporangia could appear in great numbers on the lower surface of leaves, and in more severe infections, on flower buds and stems. Under less favorable conditions, the production of sporangiophores and sporangia on any part of the rose plant is sparse and difficult to detect [3]. Roses are almost always unaffected when the environmental humidity is less than 85% [3]. Growth chambers experiments have shown that when the temperature is optimal, as little as 4 h of leaf wetness were sufficient to start the infection process on rose by *P. sparsa* [13,14]. For temperatures near 10°C at least 8 h of leaf wetness are required for significant levels of infection [14]. Sporangia of *P. sparsa* usually do not sporulate at temperatures lower than 6°C, however sporangia can survive exposure to such temperatures, highlighting the importance of the correct handling of infected plant material to avoid the continuous carry over of the disease [14]. Optimum temperatures for infection range from 15°C to 20°C, and at this range significant proportions of leaves can be symptomatic after 4 days [14]. The spore germination is reduced drastically or the pathogen is being killed at temperatures above 25°C [3,14]. Some studies have indicated that *P. sparsa* isolates from different hosts and/or geographic locations may vary in their response to environmental temperature [15] possible showing geographic adaptation that influences



the temperature ranges for the germination [14]. The recurrence of the disease in successive years and the tendency for severe symptoms to appear very quickly after favorable environmental conditions appear to suggest that *P. sparsa* might overwinter as dormant mycelia in rose cuttings and plant roots [16]. Sporangia can survive and be viable on dried, fallen leaves for as long as one month [3]. Oospores can be formed soon after the colonization by *P. sparsa* starts and can be found in infected leaves and stems. The role of the oospores in disease overwintering is not well understood but some studies indicate that oospores are likely the primary overwintering inoculum [3,16].

Disease Symptoms

Different species and varieties of roses respond differently to the downy mildew disease, making difficult an accurate diagnosis [6]. Downy mildew symptoms can occur not only on leaves but also in stems, peduncles, calyxes and petals. Early symptoms are very difficult to distinguish from nutrition deficiency, spray injury or black spot disease caused by fungi [17], and usually include purplish-red or brown colored leaf spots that can appear squared as the pathogen may be restricted to areas between major veins [17]. As disease develops, leaves may turn yellow and drop, sometimes causing severe leaf drop [6]. *P. sparsa* does not always sporulate on the underside of the leaves like other downy mildew pathogens, however under very humid conditions, downy mildew mycelia will form under the purplish-red or brown colored spots [17]. Collapsing buds or cankers can form on newly emerging rose canes [6].

Figure 3. Rose downy mildew disease symptoms

Photos taken from: https://www.apsnet.org/publications/imageresources/PublishingImages/1998/Rose27.jpg and http://www.gpnmag.com/wp-content/uploads/RoseDMZappingBuds.jpg

Host Range

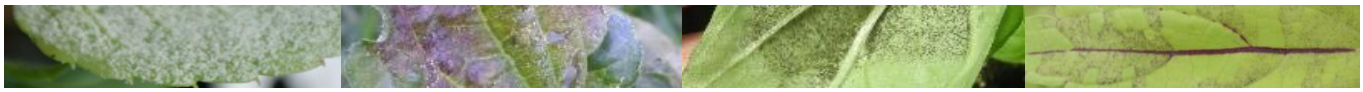
Peronospora sparsa is the causal agent of downy mildew disease in roses (*Rosa* sp.), blackberries, raspberries and dewberries among others (*Rubus* sp.), and *Prunus laurocerasus* (cherry laurel) [18]. *Peronospora* on *Rosa* sp. and *Rubus* sp. were once believe to be separate species (*P. sparsa* and *P. rubi*), however cross-inoculations preformed in vitro with isolates from rose and blackberry indicated that isolates of either host can infect the other [15].

Geographic Distribution

Rose downy mildew is considered widespread, occurring in all regions where roses are found. A complete list of the geographic distribution can be found at CABI Invasive Species Compendium (http://www.cabi.org/isc/datasheet/39730).

Cultural Practices

- Inspect plant material upon arrival for any sign of disease problems.
- When growing in greenhouses, minimize humidity and leaf wetness for extended periods of time, if feasible.
- Ensure good air circulation and water drainage with well-spaced plants.



- Scouting frequently for detection of early symptoms and remove infected plants to prevent disease spreading.
- Practice good sanitation, removing fallen leaves and debris.
- Do not compost infected material onsite.

Cultivar Disease Resistance

Multiples sources of downy disease resistance have been identified in wild species of roses. The presence of resistance in wild roses could serve as a valuable resource for resistance breeding [19], but the markers have not been well characterized, which prevent their use in breeding programs [7]. Marker assisted selection, RNAi technologies, genomic and transcriptomic studies of resistance related genes in both plants and pathogen, could help to identify useful genes that can be used to improve downy mildew disease resistance in roses [7].

Fungicide Management

Current recommendations for rose downy mildew management rely on protective fungicide treatments with applications as soon as environmental conditions are favorable for disease development and before noticeable symptoms and inoculum are present. For outbreaks that occur annually under favorable conditions, it is possible to time for preventive applications. Contact fungicide treatments, such as Protect (manoczeb) and Daconil (chlorothalonil), combined with systemic active ingredients, such as Heritage (azoxystrobin) and Subdue Maxx (mefenoxam) provide multiple options. For additional options, please review the IR-4 Downy Mildew Efficacy Summary and Literature Review (https://www.ir4project.org/ehc/environmental-horticulture-research-summaries/) [20]. Disease management is more effective when full coverage of the underside of the leaves is achieved during contact fungicide treatments or when systemic products such as those in FRAC groups 4 (mefenoxam), 33 (phosphorus acid generators) and 49 (oxathiapiprolin) are employed in rotation.

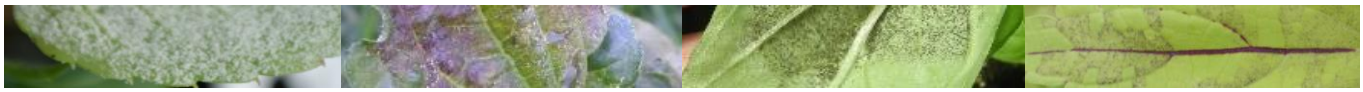
To slow the development of resistance, it is highly recommended that label directions are followed closely and that rotations occur among different modes of action as established by the Fungicide Resistance Action Committee (FRAC).

Genetics and Genomics

A technique utilizing the polymerase chain reaction (PCR) has been developed to detect *Peronospora sparsa* in rose tissues [21].With this method, DNA of *P. sparsa* was detected in the cortex of stem and root tissues of symptomatic plants. The pathogen's DNA was also detected in tissues of asymptomatic mother plants used as a source of propagation materials.
Efforts to sequence the rose genome are underway [22].

Knowledge Gaps

- **Breeding for resistance:** Even though downy mildew is an economically important disease of roses, the majority of the breeding for resistance has been focused on powdery mildew disease caused by *Podosphaera pannosa*. Marker assisted selection, RNAi technologies, and genomic and transcriptomic studies of resistance related genes in both plants and pathogen could help to identify useful genes that can be used to improve downy mildew disease resistance in roses (Debener and Byrne 2014).
- **Cultural control**: There is some evidence that reducing plant density may reduce disease, but this needs further valuation (O'Neill et al 2002). Role of crop debris in overwintering/spring inoculum has not been determined; some recommend that fallen leaves/debris be collected (Bertus 1973) and buried or burned. There is some evidence that shifting crop row orientation to take advantage of prevailing wind movement reduces disease (O'Neill et al 2002).
- **Genomics:** The pathogen's genome has not been sequenced yet.
- **Genetics:** Mating types for *P. sparsa* are unknown.
- **Molecular Biology:** Plant-pathogen interactions are not well understood.
- **Disease forecasting**: A grower-accessible tool is needed to predict when management options need to be implemented.



- **Biology:** Are there specific triggers for oospore formation? Does *P. sparsa* form zoospores (increasing inoculum potential) under certain conditions? Are infection parameters (temperature, humidity) the same for shrub roses as for hybrid teas? Can fungicide treatments be halted once the growing temperature reaches a certain point? Are oospores formed in shrub roses as readily as in hybrid teas?

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