

Dossier: Impatiens Downy Mildew

Author: Catalina Salgado-Salazar Reviewers: JoAnne Crouch, Margery Daughtrey, Cristi Palmer, Nina Shishkoff Additional Contributors: Amy Abate, Yu-Han Lan

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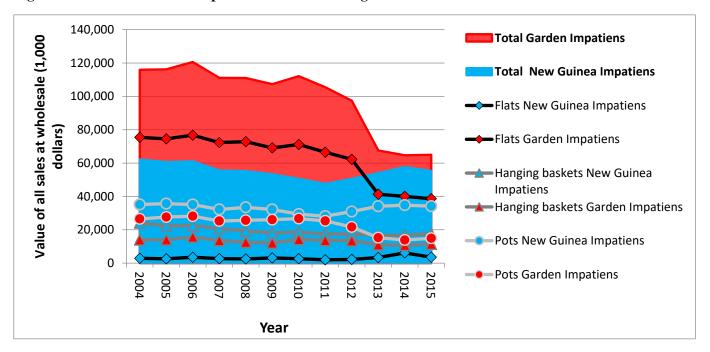
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Introduction and Importance

Impatiens are among the most valuable and widely cultivated floriculture crops in the world [1]. Their popularity is attributed to a wide array of available colors, preference for growing in shade, long flowering season and ease of use as a container plant [2,3]. However, over the last decade, the production of impatiens has been at risk due to the downy mildew disease (IDM) caused by the fungus-like oomycete *Plasmopara obducens*. This disease has significantly affected the bedding plant industry, as the onset of the disease caused a reduction in the production of impatiens plants for flats, hanging baskets and pots. As a result of IDM, the industry suffered a dramatic drop in wholesale values during the last decade, going from \$178 million in 2014 to \$120 million in 2015, or a reduction of 32% in revenue (USDA-NASS 2004 - USDA-NASS 2015 reports; summarized in Figure 1). While the total sales



of New Guinea Impatiens and other impatiens (primarily garden impatiens, *I. walleriana*) have declined greatly since 2004, New Guinea Impatiens dropped initially and then remained relatively constant with a slight increase in 2013 to 2015 for an overall decline in the market of 11%. Other impatiens dropped initially in 2006, levelled off, and then experienced another steep decline in 2013 for a percentage drop of 44%.





Historically, the oomycete pathogen P. obducens had been reported to be present in the United States territory and in other parts of the world since the late 1800's, occurring on native or wild relatives of the cultivated Impatiens walleriana. Plasmopara obducens was first found on I. noli-tangere in Germany in 1877 [4]. Specimens in the US National Fungus Collections show that the pathogen was also found on the North American native *I. pallida* in the US as early as 1884 [5]. Until up to 2003, only sporadic reports of downy mildew disease affecting Impatiens species in the U.S. could be found, mostly in the form of specimen records deposited in herbarium collections. This continued even after the establishment of the steady commercial use of *I. walleriana* in the 1960's and despite of the presence of IDM causal pathogen since the late 1800's. The same sporadic reports of the disease also applied to other parts of the world [5]. The year 2003 marked the initial observation of outbreaks of the disease occurring in greenhouse production of *I. walleriana*. The first report of IDM in the UK in 2003 occurred on a range of cultivars associated with commercial growers around the area, and also in public and private gardens. These outbreaks prompted regulatory actions by the UK Plant Health and Seeds Inspectorate, as the pathogen had never been reported in that country before, even on wild or native relatives of cultivated *I. walleriana* [6]. In the US, IDM is not considered a quarantine pest, as the disease has been present in the US for well over a century. Instead, it is considered a quality pest, meaning that it can affect the quality of finished plants at the grower, landscaper or consumer level [7]. Shortly after the UK disease outbreaks in 2003, IDM was reported at three I. walleriana greenhouse production sites in 2004: California [8], New York [9] and Tennessee [5]. Even though these outbreaks occurred during greenhouse production, no associated landscape outbreaks were reported, and it seemed the disease might be controlled within the greenhouse and not enter other phases of production [9]. However, in 2011 P. obducens was reported as the cause of regional IDM outbreaks for the first time in landscape beds and container plantings in North America [3]. By the end of the 2012 growing season, the disease had been confirmed in 34 states in the U.S. [10-14]. Currently, IDM has been reported from 42 states including Hawaii [10]. In addition, there have been an increasing number of reports of the disease in other countries where I. walleriana or other Impatiens species are produced [15-20] or grow in the wild, including Italy, Serbia, and Taiwan. In 2007, Pseudoperonospora cubensis was reported as causing downy mildew of I. irvingii in Cameroon [21], however no pathogenicity test



were performed to further test its potential impact on this or other *Impatiens* species. *Pseudoperonospora cubensis* has not been reported as causing downy mildew on impatiens elsewhere.

Plant Description

The genus *Impatiens* is a member of the family Balsaminaceae and is one of the most species-rich genera of angiosperms, with over 1,000 species endemic to tropical, subtropical and temperate regions of Africa, Madagascar and Asia [2-22]. Only two *Impatiens* species are native to North America (*I. capensis* and *I. pallida*), however other temperate (*I. balfourii*, *I. grandulifera* and *I. parviflora*) and tropical species (*I. balsamina*, *I. hawkeri* and *I. walleriana*) have been introduced to the region and constitute annual and in some cases perennial species [23,24]. Out of the cultivated *Impatiens* species, *I. walleriana* is the most commonly grown species in the bedding-plant industry worldwide [24]. While *I. walleriana* had been on the market in the U.S. since the late 1800's, it only became widely popular in 1960's after the introduction of improved hybrids developed in Costa Rica by Claude Hope [24,25]. Since then, *I. walleriana* has become one of the top floricultural crops not only in the United States, but also in the United Kingdom, the Netherlands and Germany, among others [7].

Impatiens walleriana is an annual plant that can reach close to 3 feet tall and spread around 2 feet. These plants bloom constantly once the bloom cycle is initiated [2], and they are easily grown in evenly moist, well drained soils in locations in part or full shade. Wild species or those species that have escaped gardens into nature also prefer moist and shaded habitats of the kinds provided by mountain rainforests, wet grasslands or the banks and shores of bodies of water [23].



Figure 1. Impatiens walleriana

Photo taken from: CC BY-SA 3.0, https://en.wikipedia.org/w/index.php?curid=1027675

Pathogen Description

Plasmopara obducens was first described in 1877 by J. Schröter as *Peronospora obducens* infecting *I. noli-tangere* in Germany. This pathogen is a fungus-like organism, known to belong to the oomycetes or water molds. The pathogen is closely related to other well know downy mildew pathogens such as *P. halstedii* (sunflower and rudbeckia downy mildew), *P. viticola* (grape downy mildew) and more distantly related to other oomycete pathogens such as *Pythium* and *Phytophthora* [3]. Microscopic features of this pathogen include pyriform haustoria and hyaline, monopodial sporangiophores with a slightly swollen base and with three apical branchelets forming right angles to the supporting branches or axis, with no apical thickening. Sporangia are ovoid to globose, hyaline, occasionally with a single pore on the distal ends [20]. Thick walled and slightly dematiaceous oospores can be

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observed sometimes in diseased tissues, both fresh and dry [19,20]. Oospores can be observed not only in diseased tissue of stems but also in leaf tissue, leaf stalks and petals of flower buds [19,20]. *P. obducens* recently been shown to be homothallic or self-fertile, no requiring two mating types to form viable oospores [26].

Pathogen Biology and Disease Cycle

Plasmopara obducens produces two types of spores, zoospores and oospores. Sporangia are the sac-like structures that contain the zoospores. Zoospores are motile spores that are the result of asexual reproduction and are produced on the abaxial surface of infected leaves. Oospores, the second type of spores, are the result of sexual reproduction. Both kinds of spores are able to initiate new infections. IDM is spread mainly through the movement of infected plants within the greenhouse industry or by currents of air or water that move inocula. Transplanting healthy plants into soil containing oospores will also result in disease if environmental conditions are favorable [27,28].

In an infected impatiens plant, sporangiophores and sporangia will form during cool and humid conditions $(60^{\circ}\text{F} - 73^{\circ}\text{F})$ and emerge from the internal tissue on the underside of the leaf, sometimes through the stomatas. If water is present on the leaf surface, sporangia can germinate and produce massive quantities of zoospores that can cause secondary infections within the same plant or in other plants after being spread by wind or water. Sporangia can also be dislodged from the leaf tissue and fall to the ground where the zoospores can be released and spread short distances by water splash or be spread longer distances by wind currents to nearby plants [28]. Once on a new plant, zoospores encyst, then form germ tubes that enter stomata and from there invade inner tissues of the plant by growing between the cells. There is a latent period between infection and the appearance of visible plant symptoms of up to two weeks, after which the disease cycle repeats throughout the growing season. Oospores that were formed in previous growing seasons can be induced to form sporangia and release zoospores during cool, wet conditions. These zoospores are rain-splashed into susceptible tissue where they can germinate and start new infections, or can penetrate the plant through roots. The sporangia and zoospores can serve as the source of inoculum for new infections in nearby plants in landscape settings.

The thick-walled oospores that *P. obducens* produces are considered the overwintering structures of the pathogen and can survive in soil for several years and infect healthy impatiens plants in following growing seasons [3,28]. So far, there is no evidence of seed-borne transmission of *P. obducens* in *I. walleriana* [28], but recent research has shown evidence of seed transmission of *P. obducens* in *I. balsamina* [M. Daughtrey, personal communication].

Figure 2. *P. obducens* sporangiophores and sporangia

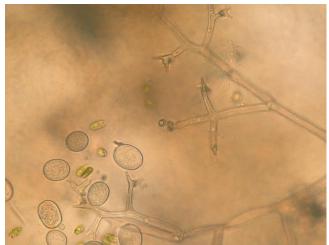


Photo by: Nina Shishkoff

Figure 3. P. obducens oospore

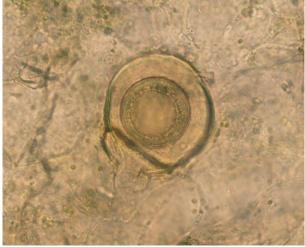


Photo by: Nina Shishkoff



Epidemiology

Most evidence indicates that the development of IDM is favored by cool and wet environmental conditions, which are the very same habitat conditions in which *I. walleriana* plants thrive. Greenhouse experiments showed that the sporulation of *P. obducens* on infected plants is influenced by the interaction of temperature and relative humidity. *P. obducens* showed the highest percentage of spore release when the relative humidity was higher than 75% for at least 24 hours; however, the relative humidity has to be accompanied by milder temperatures, usually below 25° C (77° F) [29]. Higher temperatures usually inhibit the sporulation of *P. obducens*. The release of sporangia also seems to follow a diurnal pattern, with peak releases observed around 12:00, after which sporangia release is reduced [29]. Cold temperatures, for example those present during fall and winter can trigger the formation of oospores in plant tissues; these oospores will overwinter until environmental conditions are favorable again [29]. At the same time, oospores require preconditioning of at least 1 - 2 months of near 0° C (32° F) temperatures to germinate [29]. Once certain weather conditions appear (high moisture and cool temperatures), preconditioned oospores will germinate to produce extra large sporangia to start a new disease cycle [29]. Alternatively, systemically infected but asymptomatic plants can act as early sources of inoculum, as is the case in other downy mildew disease systems [28].

Disease Symptoms

Early symptoms are very difficult to distinguish from water and nutrition deficiency or spider mite injuries [31]. Young plants, new growth (immature leaves) and seedling cotyledons are most susceptible and typically show symptoms first [32]. These early symptoms include yellowing or stippling of infected leaves, which may curl downward at the edges. Plants infected at an early stage of development may be stunted in both, height and leaf size.

Figure 4. Downy mildew symptoms: leaf yellowing



Photo by: M. Daughtrey

Figure 5. White downy-like fungal growth on the underside of leaves



Photo by: M. Hausbeck

Under cool and humid conditions a white fuzzy growth composed of spores and sporangia may be visible on the abaxial surface of infected leaves [3,10,11]. Infected plants will stop flowering and exhibit blossom and leaf drop, initially from the lower parts of the plant and then progressing up the stem. As the disease advances, stems eventually collapse, and in very wet conditions infected plant tissues become water-soaked, soft and mushy [33]. *Impatiens balsamina* (balsam impatiens) does not show such dramatic symptoms as *I. walleriana* [34]. Balsams show yellow or brown leaf spots, with white sporulation seen on the underside of leaves, but plants continue to stand tall and to flower. Other species of impatiens typically only show scattered leaf spots [30].



Figure 6. Effects of impatiens downy mildew in the landscape



Photos by: M. Daughtrey

Host Range

Plasmopara obducens shows host specificity, affecting only plants in the genus *Impatiens*. There are no reports of this pathogen affecting plants in other genera. All cultivars of *I. walleriana* (common garden impatiens), including double impatiens, mini impatiens and interspecific hybrids of *I. walleriana* originating from either seed or cuttings, are susceptible to IDM. *Plasmopara obducens* is often found causing disease in *I. balsamina* (balsam impatiens). The North American native impatiens, *I. pallida* and *I. capensis* (jewelweeds) and *I. glandulifera* (Himalayan balsam) are also known hosts for *P. obducens* [31. Downy mildew of *I. sultanii* caused by *P. obducens* was reported as a new disease in Japan in 2012 [18]. In 2012, a study in Riverhead, NY showed that sporulation of *P. obducens* was observed in *I. auricoma*, *I. arguta*, *I. flanaganae* and *I. hochstetteri* [34]. A similar study in 2015 found that *I. briartii*, *I. cinnabarina*, *I. grandis*, *I. irvingii*, *I. laurentii*, *I. repens* and *I. sodenii* var. *uguensis* presented scattered leaf lesions, with some species showing stem discoloration. However, none of these impatiens hosts showed the same devastating disease symptoms as the susceptible *I. walleriana* [34]. New Guinea impatiens (*I. hawkeri*) and interspecific hybrids including Fanfare, Divine, Celebration. Celebrette and SunPatiens® are highly resistant to *P. obducens* and have not been reported as being affected by *P. obducens* [28,31].

Geographic Distribution

It is generally expected that IDM is present wherever impatiens plants are grown. This disease is well known to all impatiens growers in the world; however, not many official reports have been made, so consequently not many reports can be found in databases. According to the fungus-host database of the U.S. National Fungus Collection (USDA – ARS) [5], IDM has been reported present in Australia [35], Austria, Bulgaria, Canada, Czech Republic, China, Costa Rica, Germany, Guatemala, Hungary [20], India, Italy [17], Japan [18], Korea, Norway [19], Pakistan, Poland, Romania, Russia, Serbia [15], Taiwan [16], United Kingdom [6], and Uzbekistan.

In the United States, as of June 2017, IDM has been reported in 42 states including Hawaii (Figure 7, Table 1)



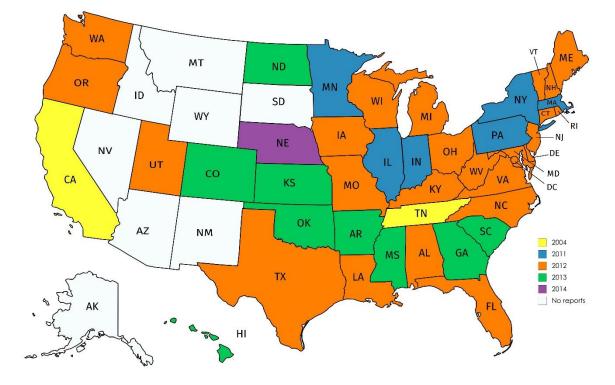


Figure 7. Current distribution of impatiens downy mildew disease in the United States

Table 1.	Current distribution of impatiens downy mildew disease in the United States including year of
first repo	rt and reference

State	Year	Source	
Alabama	2012	https://doi.org/10.1094/PDIS-12-13-1248-PDN	
Alaska	*		
Arizona	*		
Arkansas	2013	https://www.uaex.edu/farm-ranch/pest-management/plant-health-clinic/	
California	2004	https://doi.org/10.1094/PDIS.2004.88.8.909B	
Colorado	2013	http://www.ballseed.com/pdf/ImpatiensDownyMildewGrowerGuidelines.pdf	
Connecticut	2012	http://www.ladybug.uconn.edu/FactSheets/impatiens-downy-mildew.php	
Delaware	2012	http://extension.udel.edu/blog/tag/downy-mildew/	
Florida	2012	https://doi.org/10.1094/PDIS-08-12-0705-PDN	
Georgia	2013	http://plymouthnursery.net/blog/12297/impatiens-disease-downy-mildew	
Hawaii	2013	https://doi.org/10.1094/PDIS-10-13-1017-PDN	
Idaho	*		
Illinois	2011	http://web.extension.illinois.edu/state/newsdetail.cfm?NewsID=28862	
Indiana	2011	http://ballpublishing.com/GrowerTalks/ViewArticle.aspx?articleid=18921	
Iowa	2012	https://hortnews.extension.iastate.edu/2012/8-29/mildew.html	
Kansas	2013	https://agriculture.ks.gov/	
Kentucky	2012	https://doi.org/10.1094/PDIS-10-12-0973-PDN	
Louisiana	2012	http://www.lsuagcenter.com	
Maine	2012	http://www.maine.gov/dacf/php/horticulture/idm.shtml	
Maryland	2012	https://extension.umd.edu/hgic/invasives/impatiens-downy-mildew	
Massachusetts	2011	https://ag.umass.edu/home-lawn-garden/fact-sheets/impatiens-downy-mildew-in- home-gardens	
Michigan	2012	http://msue.anr.msu.edu/news/downy_mildew_on_impatiens_becomes_widespread_i n_michigan_landscapes	



State Year Source		Source	
Minnesota	2011	https://www.extension.umn.edu/garden/yard-garden/flowers/managing-impatiens downy-mildew-in-landscape/	
Mississippi	2013	Balbalian, C., Mississippi State University, personal communication	
Missouri	2012	https://ipm.missouri.edu/MPG/2013/1/Downy-Mildew-on-Impatiens/	
Montana	*		
Nebraska	2014	K. Broderick, University of Nebraska Diagnostic Clinic, personal communication	
Nevada	*		
New Hampshire	2012	https://extension.unh.edu/resources/files/Resource003821_Rep5440.pdf	
New Jersey	2012	http://www.state.nj.us/agriculture/news/hottopics/approved/topics120706.html	
New Mexico	*		
New York 2011 http://www.ballpublishing.com/GrowerTalks/ViewArticle.aspx?articleID=19112			
North Carolina 2012 https://projects.ncsu.edu/cals/plantpath/extension/clinic/news-alerts.html		https://projects.ncsu.edu/cals/plantpath/extension/clinic/news-alerts.html	
North Dakota	2013 https://doi.org/10.1094/PDIS-12-14-1312-PDN		
Ohio	2012	https://doi.org/10.1094/PDIS-06-12-0545-PDN	
Oklahoma	2013	http://entoplp.okstate.edu/pddl/2013/PA12-18.pdf	
Oregon	2012	https://pnwhandbooks.org/node/2920/print	
Pennsylvania	2011	http://extension.psu.edu/plants/green-industry/news/2012/alert-downy-mildew- impatiens-this-year	
Phode Island 2012 http://www.ecolandscaping.org/05/pests-pest-management/impatiens-down		http://www.ecolandscaping.org/05/pests-pest-management/impatiens-downy-mildew- alert-uri-plant-protection-clinic/	
South Carolina	2013	http://plymouthnursery.net/blog/12297/impatiens-disease-downy-mildew	
South Dakota	*		
Tennessee	2004	http://www.thepaginator.com/Uploadfile/44/1085/pdf/20130513073808414.pdf	
Texas	2012	http://plantclinic.tamu.edu/2012/05/25/impatiens-down-mildew/	
Utah	http://utabnests.usu.edu/files_ou/un_newsletter/2013/LitabPests_Newsletter-		
Vermont	2012	http://www.uvm.edu/pss/EIPM/IPMreport2012.pdf	
Virginia	2012	https://www.pubs.ext.vt.edu/content/dam/pubs_ext_vt_edu/PPWS/PPWS-19/PPW 19-pdf.pdf	
Washington	2012	https://pnwhandbooks.org/node/2920/print	
West Virginia	2012	M. M. Rahman, West Virginia University, personal communication	
Wisconsin	2012	https://datcp.wi.gov/Documents/PIBAnnualReport2013.pdf	
Wyoming	*		

* Not known

Disease Management

Management of IDM should be focused on prevention rather than trying to control active infections, as no fungicide has been proven effective curing downy mildew [3,9,27,28,31]. Preventive measures include cultural and chemical practices:

Cultural practices (Greenhouse and landscape)

- Start with propagation material free of disease.
- Growers should inspect plant material upon arrival for any sign of disease problems.
- Do not carry over impatiens in the greenhouse for more than one season.
- Minimize greenhouse humidity and leaf wetness for extended periods of time.
- Irrigate early in the day and avoid overhead irrigation in the late afternoon or evening. This would help limit secondary infections.
- Ensure good air circulation and water drainage. Keep plants well spaced.
- In the landscape, avoid if possible overcrowded plant beds or a monoculture of *I. walleriana*.
- Scout frequently for detection of early symptoms
- Keep seed-propagated plants separated from cutting-propagated plants.

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- Remove suspect plants and send samples to a diagnostic lab for prompt identification
- Remove infected plants to prevent disease from spreading. Infected plants can act as sources of inoculum. If sporulation on the abaxial side of leaves is observed, bag plants, collect fallen leaves and plant debris and seal the bags before removing from greenhouse. Infected material should be disposed of offsite.
- Do not leave diseased plants in the landscape to avoid triggering oospore production.
- Disinfest the affected greenhouse area (floor or bench) before placing another impatiens crop.
- No volunteer impatiens plants should be allowed to grow near production facilities
- Do not compost infected plant material on premises.
- If IDM has been previously observed in the landscape, there is a high chance the disease will show up in the next growing season and may enter local production greenhouses the next spring. For nearby commercial greenhouses and retail locations, growers should assess consumer demand and crop risks and consider preventive programs for growing *I. walleriana*. For landscapes, options may include planting other annual plants such as New Guinea impatiens or knowingly planting susceptible impatiens assuming they will need to be replaced at the start of decline.

Fungicide Management

Current recommendations for IDM 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. Fungicides can provide very good control of IDM only when applied preventively to healthy plugs, cuttings, young and mature plants prior to transplanting into the landscape [28].

The application of fungicides should start before symptoms are seen. Several IDM management programs are available which recommend preventive treatments with rotations among different fungicides. It is highly unlikely that applications of fungicides after symptom development will provide effective management, and this has potential to increase the risk of fungicide resistance.

As with any other pathogen with similar characteristics, *P. obducens* can develop resistance to previouslyeffective fungicides. Since the disease pressure has increased gradually since the start of the landscape outbreaks, the increased use of fungicides for disease control has provided the opportunity for the development of fungicide resistance. The use of a single fungicide repeatedly over a prolonged period of time is <u>not</u> recommended, as it can select for and favor populations of the pathogen that are resistant to that particular fungicide and all others in the same class [32]. The fungicides for which fungicide resistance has been observed in *P. obducens* in at least one location include mefenoxam, fluopicolide, boscalid + pyraclostrobin, and phosphonate [36]. Rotation among different fungicide modes of action will allow continuing use of these products and minimize risk for developing resistance to other fungicides.

From 2011 to 2016 the IR-4 project initiated research aimed to determine the efficacy of several fungicides on downy mildews, IDM was one of the ones studied [37]. For this project, 41 products with distinct active ingredient were tested. Table 2 contains a summary of the products with excellent or good control of IDM. For more detailed information consult the IR-4 Downy Mildew Efficacy Summary and Literature Review (https://www.ir4project.org/ehc/environmental-horticulture-research-summaries/) [37].



Table 2. Products that showed good to excellent control in efficacy trials for the control of IDM.	Table 2.	Products that showed	good to excellent control in efficat	cy trials for the control of IDM.
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Product	Active Ingredient(s)	Rate(s) T		Efficacy	Manufacturer
Adorn (V-10161)	Fluopicolide*	Foliar	1 fl oz per 100 gal 2 fl oz per 100 gal 4 fl oz per 100 gal	Excellent control	Malant
4FL (Presidio)		Drench	1 fl oz per 100 gal 2 fl oz per 100 gal 4 fl oz per 100 gal	Excellent control	Valent
Micora (NOA 446510)	Mandipropamid	Foliar	4 fl oz per 100 gal 8 fl oz per 100 gal	Excellent control	Syngenta
Orvego (BAS 651)	Ametoctradin + Dimethoate	Foliar	11 fl oz per 100 gal 14 fl oz per 100 gal 28 fl oz per 100 gal	Excellent control	BASF
Pageant 38WG	Boscalid + pyraclostrobin *	Foliar	6 oz per 100 gal 12 oz per 100 gal 12.5 oz per 100 gal 18.5 oz per 100 gal	Excellent control	BASF
Segovis (SYN546539)	Oxathiapiprolin	Drench	3.2 fl oz per 10 gal	Excellent control	Syngenta
Stature DM	Dimethomorph	Foliar	9.6 oz per 100 gal 12.3 oz per 100 gal 12.8 oz per 100 gal	Excellent control	BASF
Stature SC	Dimethomorph	Foliar	6.12 fl oz per 100 gal 12 fl oz per 100 gal	Excellent control	BASF
Subdue MAXX 2E	Mefenoxam*	Foliar	0.5 fl oz per 100 gal 1 fl oz per 100 gal 2 fl oz per 100 gal	Excellent control	Syngenta
		Drench	0.5 fl oz per 100 gal 1 fl oz per 100 gal	Excellent control	
Alude	Potassium phosphite	Foliar Drench	64 fl oz per 100 gal 2.5 qt per 100 gal	Good control	Cleary/ NuFarm
			0.75 pt per 100 gal 2.5 qt per 100 gal	Good control	
Inosco (A14658C) ** not available commercially	Potassium phosphite	Spray	4 pt per 100 gal 8 pt per 100 gal	Good control	Syngenta
Vital	Potassium phosphite	Foliar	4 pt per 100 gal	Good control	Luxemburg
		Drench	1.25 pt per 100 gal 4 pt per 100 gal	Excellent control	Chemical

* Fungicide resistance has been observed for this fungicide in a minimum of a single location but treatments may still provide control of susceptible populations in other geographic locations. Ensure fungicide rotation to minimize resistance development [36].

Host Resistance Efforts

In the long term, the best strategy to overcome the effects of IDM is breeding new varieties of impatiens with resistance to *P. obducens*. There is a need to develop *I. walleriana* (or similar plants) with long-lasting disease resistance against *P. obducens* that will give long term protection and ensure sustainable production to satisfy future customer demands. Breeding programs should not be limited to development of variously colored impatiens, but should also focus on disease/pest resistance. Currently, Mark Bridgen at Cornell University is looking to test *I. walleriana* hybrid germplasm, hoping to find plants with increased disease resistance (M.



Daughtrey, personal communication). It is also safe to assume that seed companies are already conducting breeding efforts to find disease resistant impatiens.

Genetics and Genomics

Since there were no reports of IDM on *I. walleriana* until 2003 and *P. obducens* had been described as present in North America since the late 1880s on native, wild relatives of the cultivated impatiens, the first stages of research tried to determine if it was possible that a new strain or pathotype of the pathogen or even a novel mildew species was responsible for disease outbreaks. Initial efforts at understanding the genetics and origins of IDM outbreaks were based on single nucleotide polymorphism (SNP) analysis (J. Crouch, personal communication. For this study, five SNPs in the rDNA ITS region of *P. obducens* were identified and used to genotype samples of the pathogen. Samples included pre-epidemic populations (i.e. samples of the disease found prior to 2003), the first IDM sample from *I. walleriana* in 2004, and post-epidemic IDM samples (i.e. IDM samples collected after 2004). Using these five variable SNPs, nine haplogroups could be detected. However, there were not many genetic differences among the haplotypes, and few differences were seen between *P. obducens* and other downy mildew pathogens such as *Bremiella megasperma* and *P. constantinescui*. The pre-epidemic samples clustered in two haplogroups, which also contained modern samples. Since these SNPs showed little power to discriminate differences between the diversity of *P. obducens* isolates, more powerful molecular markers were needed.

Simple sequence repeat (SSR) markers were developed from a whole genome sequence assembly to study the population structure and evolution of *P. obducens* [38]. With the analyses of 18 SSR markers it was shown that the pre-epidemic and modern samples of this disease were divided into seven unique subgroups (J. Crouch, personal communication). In these analyses, it was observed that the pre-epidemic isolates clustered only in one subgroup, and that modern IDM samples were found as part of all seven subgroups.

A fluorescence in situ hybridization (FISH) tool has been developed for the detection and visualization of *P. obducens* from plants and soil (J. Crouch, personal communication). FISH is a sensitive and robust method that uses sequence-specific, fluorescence-labeled oligonucleotide probes to detect target organisms from the environment. A FISH assay for *P. obducens* that targets a portion of the rDNA-ITS region resulted in good fluorescence signal compared to those of the negative controls. The standardized protocol can be used to directly visualize *P. obducens* in situ from plant and soil samples [39].

Knowledge Gaps

- <u>Disease forecasting tools.</u> Disease management procedures for IDM could be highly improved by having a disease forecasting or disease modeling procedure. This could help improve the disease management recommendations for a particular state or area, targeting their specific needs.
- **Epidemiology**. As with any other disease, control measures depend on the proper identification of contaminated plants and plant related material, like soil. There is little knowledge about the origin of the pathogen contamination that is creating the disease outbreaks: is it the plant propagation material? Is it the soil? Is *P. obducens* surviving in soil season after season? Good cultural practices intended to help in the management of this disease recommend that impatiens cuttings and seed-produced material be free of downy mildew contamination. A molecular diagnostic tool could be a valuable way to address these questions.
- <u>Plant health improvement.</u> We need a better understanding of the role of phosphorus in the phosphorus acid generators that appear to improve plant health even when *P. obducens* is established within the plant.
- <u>Pathogen lifecycle.</u> A better understanding of factors triggering (and releasing) latency in impatiens downy mildew infections would be beneficial to those trying to manage the disease. How do environmental conditions, particularly temperature, affect oospore formation? Are there biological, solarization, or chemical treatments that could reduce the survival of oospores in garden beds? How is the disease lifecycle different in native hosts vs. the bedding plant *I. walleriana*?



• <u>Host resistance</u>. What are the genetic determinants of tolerance/resistance in various impatiens species? Can these genes be bred into *I. walleriana*? Can they be inserted into modern cultivars via CRISPR?

Glossary

Abaxial (leaf): the lower surface of a leaf.

Dematiaceous: denoted the dark color, usually brown or black, of the pathogen's growing structures. **Germplasm**: in the case of plants, genetic resources such as seeds or tissues that are collected and maintained for the purpose of breeding, preservation or other research uses.

Haplogroup: a group of similar haplotypes that share a common ancestor with a shared nucleotide polymorphism mutation.

Haplotype: group of genes in an organism that are inherited together from a single parent.

Haustoria: a slender projection from a parasitic plant, such as dodder, or from the hyphae of a parasitic fungus, enabling the parasite to penetrate the tissues of its host and absorb nutrients from it.

Hyaline: having a glassy, translucent appearance.

Pathotype: A variant of a microorganism that is distinguishable from other members of its species by its virulence.

Simple Sequence Repeat (SSR): also called microsatellites or short tandem repeats, SSRs are short (1–6 bp) DNA repeat motifs that show a high level of length polymorphism due to insertion or deletion mutations of one or more repeat types.

Single Nucleotide Polymorphism: variation at a single nucleotide position in a DNA sequence among individuals.

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