Pythium and Recycled Irrigation Water

Reclaiming water may be cost efficient, but are you doing the right things to prevent a Pythium outbreak?

By Ping Kong, Patricia A. Richardson and Chuan Hong

Pythium species are a group of important pathogens of floral crops. Pythium diseases are typically soilborne and are difficult to control once a plant is infected or soil/soilless mixes are contaminated. These diseases usually attack root systems and progress gradually without apparent foliar symptoms until a plant is near collapse. As a result, they often receive less attention, are not likely controlled in a timely manner and may result in more crop losses than those diseases displaying more obvious symptoms. The best strategy for managing Pythium diseases is prevention by eliminating or minimizing inoculum from all possible sources.

Among numerous sources of inoculum, irrigation water can be an important one for Pythium species, as already demonstrated for Phytophthora species. Species of Pythium and Phytophthora are closely related to each other morphologically and genetically; they are in the same family taxonomically. Pythium species are 10-100 times more populous than Phytophthora species in irrigation water. But unlike Phytophthora pathogens, Pythium species in irrigation water are poorly understood. This is partly due to the extreme difficulty in species identification of Pythium. Basic information is lacking. For example, what and how many species of Pythium are present in irrigation systems? Are these species plant pathogenic?

Chlorination is an integral part of crop production in many enterprises. A very practical and important question is whether current chlorination protocols recommended for Phytophthora management can control Pythium pathogens in irrigation water.

IN THE GREENHOUSE

Capturing surface water for irrigation is critical to sustain the production and growth of individual enterprises and the floral industry as a whole, as global water scarcity and pollution rapidly spreads. How to minimize disease pressure associated with recycling irrigation water is an emerging and serious question all growers face today. An immediate benefit of this study is provision of a procedure to ensure efficacy of chlorination for controlling Pythium pathogens in irrigation water. This study clearly demonstrates the diversity of Pythium species present in irrigation water and the need for water decontamination in greenhouse and nursery production, especially those recycling irrigation water.

You can use these results and start developing lines of defense

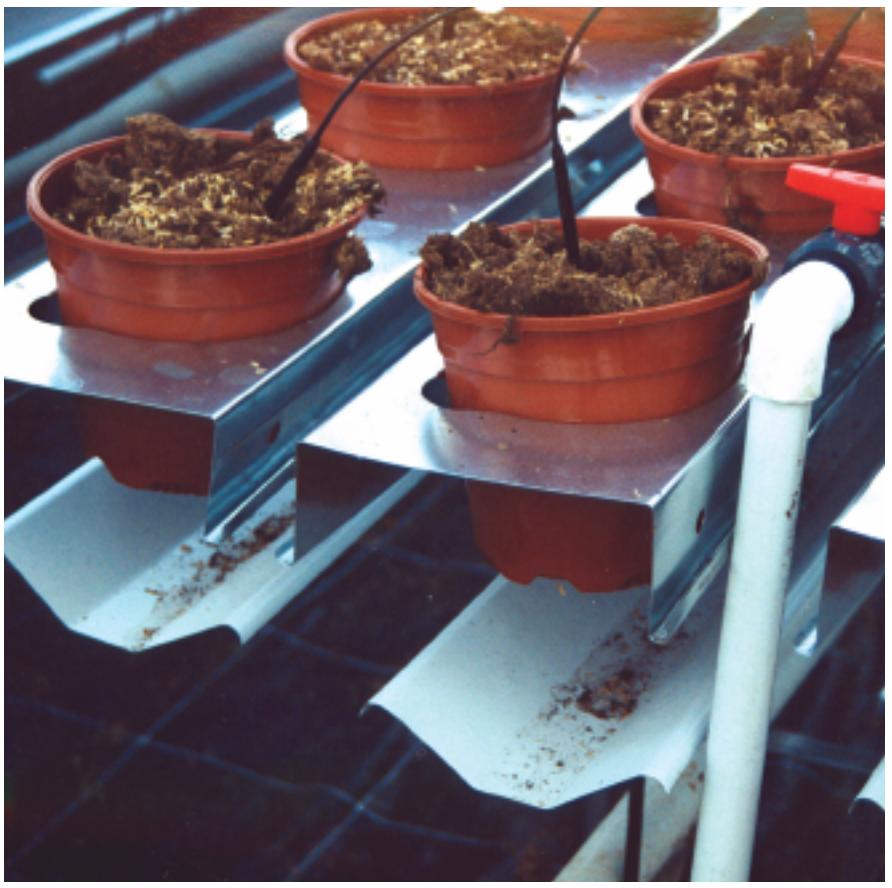
along with chlorination against pathogen build-up in recycling irrigation systems through modifications or new designs. You can also use these results to prioritize production expansion and research agenda. This study developed a molecular fingerprinting technique for rapid and accurate species identification. This will greatly facilitate future research on biology and management of Pythium species and diagnosis of plant diseases caused by these species, and will help you improve disease management in the long run.

RESEARCH RESULTS

Genetic codes of biological agents are stored in chemicals called deoxyribonucleic acid (DNA) or ribonucleic acid (RNA). For Pythium pathogens, these genetic codes are stored in double stranded DNA. In this project, we developed a DNA fingerprinting technique for rapid and accurate species identification. The underlying mechanism of this technique is to separate the two complementary DNA strands, then let individual strands form secondary structures (conformations). Under prescribed fingerprinting conditions, formation of secondary structures depends on the DNA sequence thus can be a reliable character for species identification. The fingerprinting technique is called single-stranded conformational polymorphism (SSCP) analysis. With a single DNA fingerprint, we were able to differentiate 36 Pythium species assessed so far.

With this DNA fingerprinting technique, we have identified more than 20 species of Pythium from irrigation water. The most abundant species include *P. dissotocum*, *P. torulosum*, *P. sulcatum* and *P. porphyrae*. All identified species are plant pathogenic but vary in degree of aggressiveness. These results indicate the diversity of Pythium species present in irrigation water and the need for water decontamination in greenhouse and nursery production, especially those recycling irrigation water.

A total of 15 isolates were tested with zoospores for chlorine sensitivity and were assigned a number/letter combination. These isolates included six from nonchlorinated irrigation water, five from chlorinated water and four from diseased plants. Chlorine assay of each isolate was repeated twice. Efficacy of chlorine treatment was measured by the average number of colonies per petri dish with the most colonies per dish for control (0 ppm). Numbers of colonies varied among the repeated tests of the same isolates. They also differed with isolates from the same sources (nonchlorinated water, chlorinated water or plants). Differences among the isolates within the three sources remain unclear. Figures 1 and 2, below, illustrate the data; detail assay data of the 13 other isolates were omitted for simplicity. ➧



Recycling water may put you at risk of a Pythium outbreak, but there are steps you can take to prevent it.

Figure 1. Chlorine sensitivity of Pythium aphanidermatum *(#5J5) isolated from a*

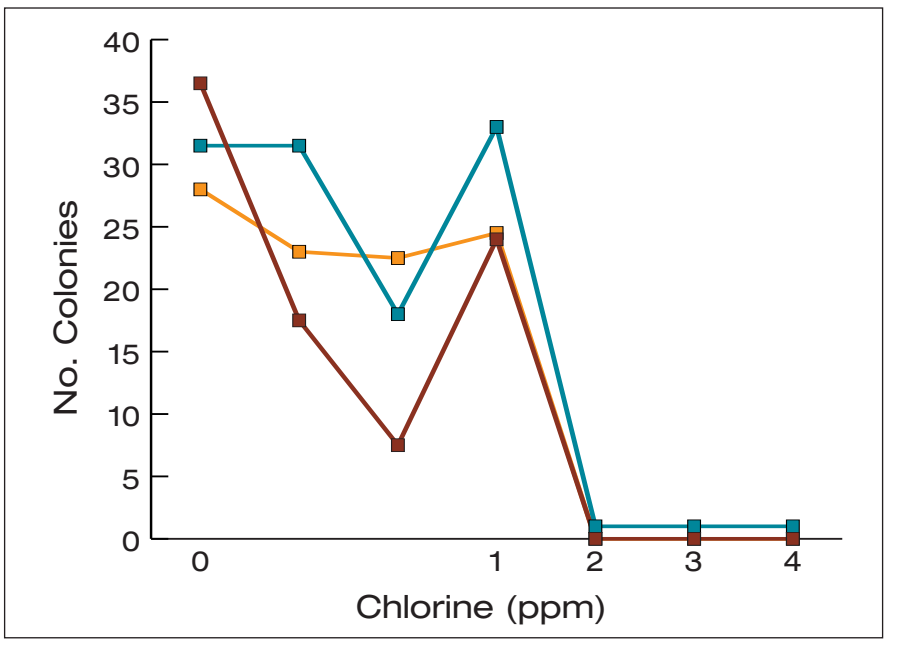
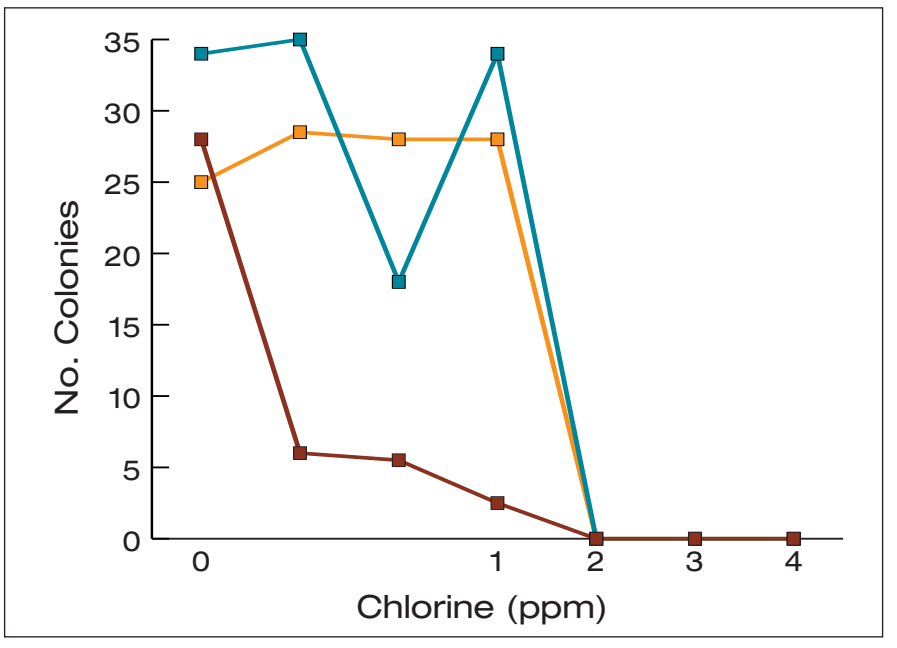


Figure 2. Chlorine sensitivity of Pythium sulcatum *(#15J9) isolated from irrigation water. Each line represents results of three repeated tests.*



No colonies were recovered at 2 ppm or above for the majority of isolates. Four isolates produced a few colonies at 2 ppm or above in one or two of the three repeated tests. Isolate 17C2 produced two colonies at 2 ppm in one test. Isolate 23J3 produced 0.3 colonies at the same concentration in two tests. Isolates 4E1 and 5J5 produced a few colonies at 8 ppm in one test, but none at 2 ppm or above in the other two tests (see Figure 1, page 33). Isolates producing colonies at 2 ppm or above produced colonies only at 1 ppm or lower in additional tests. These results suggest that the previously recommended 2 ppm free chlorine at discharge points (risers or sprinklers) for control of Phytophthora species also will effectively control Pythium zoospores in irrigation water.

It must be noted that substantial numbers of colonies were recovered at 1 ppm for a majority of the isolates, and there

Floriculture Industry Research and Scholarship Trust (FIRST), a combination of Bedding Plants Foundation and Ohio Floriculture Foundation, supports floriculture scholarships and research, such as this article, across the United States and Canada. More research results, scholarship winners and applications for scholarships or research funding are available at www.firstinfloriculture.org. How can you help in FIRST's drive to improve the production and marketability of plants? Here are some ideas:

- Review scholarship and research grant applications
- Purchase Gator Giveaway Raffle Tickets
- Participate in the Dramm 5k Relay or DS Cole Hockey in Columbus events at the annual OFA Short Course
- Become a board member
- Participate in our fun trade show booths!!
- Set up a named scholarship or research fund
- Purchase Reverse Raffle tickets for a chance to win $10,000 and other cash prizes
- Catch some laughs at the Paul Ecke Jr. Floral Design Contest during the annual OFA Short Course

For more information, you can also contact FIRST, P.O. Box 280, East Lansing, MI 48826-0280; phone (517) 333-4617; fax (517) 333-4494 or E-mail: first@firstinfloriculture.com.

essentially were no differences in colony recovery between 0.25 and 1.0 ppm of chlorine for most isolates. This chlorine response is considerably different from that of Phytophthora species reported previously. The mechanism behind this is unknown. But these results suggest that any failure to maintain recommended 2 ppm chlorine may expose entire crops in the production facility to Pythium diseases.

Pythium diseases are a major limiting factor affecting floral crop production. This problem will continue to aggravate with growing global water scarcity and pollution. Identification along with pathogenenicity tests of Pythium species present in irrigation water suggested that decontamination of recycled water before use is required to produce quality plants. Growers should use this recommendation as a guide when building a new production facility (e.g., greenhouse) or modifying an existing facility to minimize Pythium disease risk.

IDENTIFICATION

Identification of these species is the first and most important step towards development of effective pathogen management strategies. This research also made specific recommendations on using current chlorination protocol for waterborne Pythium control. Use of these recommendations will reduce the risk of incomplete water decontamination, which subsequently reduces crop losses and improves plant quality. In addition, this research overcame a major difficulty in identification of Pythium isolates into species, and produced a molecular technique that with a single DNA fingerprint steadily separated dozens of species. Accurate species identification is foundational for any biological study. Lack of effective identification tools has long prevented Pythium disease control research from progressing as needed. This research provides researchers with an effective tool for disease management studies, improving research quality and productivity and helping the industry on Pythium management in the long run. GPN

Chuan Hong is an assistant professor of Plant Pathology, Ping Kong is research scientist and Patricia A. Richardson is research specialist at Virginia Tech's Hampton Roads Center, Virginia Beach, Va.

This article was reprinted with permission from Floriculture Industry Research and Scholarship Trust.

LearnMore
For more information related to this article, go to www.gpnmag.com/lm.cfm/gp050404