Bacterial Wilt of Toronto Creeping Bentgrass: A Potential Threat to Turfgrass Culture in North America

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Editor's note: D. L. Roberts is currently a Postdoctoral Research Associate at Michigan State University where he is continuing research in turfgrass pathology. He received his B.S. and M.S. from the Ohio State University. His M.S. research involved the study of Rhizoctonia on sugar beets. In December, 1982, he received his Ph.D. from Michigan State University where his dissertation research determined that a bacterium was the cause of the unresolved C-15 problem-decline on Toronto creeping bentgrass. He has published several articles on the discovery, diagnosis, distribution, isolation, pathogenicity, characterization and control of bacterial wilt, the first known turfgrass disease caused by a bacterium. Dr. Roberts will be the speaker at our June meeting.

Dr. Joe Vargas needs no introduction to WGCSA members. He is a long time friend of the golf course industry in Wisconsin.

Introduction

Ever since its selection in the 1930s by the USDA and USGA, Toronto creeping bentgrass (Agrostis palustris Huds. cv. Toronto = C-15) was considered the elite among the bentgrasses for propagation on golf course putting greens. During the 1970s, a disease of unknown origin, commonly known as the C-15 problem or C-15 deadline, devastated many Toronto creeping bentgrass putting greens in the Midwest. The disease gained national recognition when it devastated the Butler National Golf Course two weeks prior to the 1980 PGA Western Open. This unpredictable and uncontrollable epidemic of the disease initiated an intensive study of the problem.

Association of Bacteria with Diseased 'Toronto'

No fungal pathogens, the most common causes of turfgrass diseases, could be consistently isolated from diseased 'Toronto' plants at Michigan State University. Use of transmission electron microscopy (TEM) led to the discovery of bacteria associated with the xylem vessels of diseased plants (Fig. 1). Xylem vessels transport water and fertilizer nutrients from the roots to the crown and leaves. Blockage of these xylem vessels with millions of bacteria readily explained the rapid wilting of infected plants. The discovery of bacteria in diseased Toronto creeping bentgrass was important for the following reasons: 1) it suggested a solution to the unpredictable disease outbreak at Butler National; 2) it provided a pathogenic agent for the unresolved C-15 problem-decline which had affected many midwestern golf courses, and 3) it suggested the first known turfgrass disease incited by a bacterium.

Table 1. Bacterial wilt grasses in Europe. Originally discovered in Switzerland in 1975, bacterial wilt has now been found in numerous genera of grasses in many countries.

Location	Host Grasses
1975	
Switzerland	Dactylis (Orchardgrass)
	Lolium (Ryegrass)
1982	
	Dactylis (Orchardgrass)
Switzerland	Lolium (Ryegrass)
Belgium	Festuca (Fescue)
France	Arrhenatherum (Oatgrass)
Germany	Phleum (Timothy)
Great Britain	Alopecurus (Foxtail)
Norway	Agrosis (Bentgrass
Netherlands	Poa (Bentgrass)
	Trisetum
(New Zealand)	Phalaris (Canarygrass)
	Agropyron (Wheatgrass)

Distribution and Diagnosis

Plants affected with the disease at Butler National exhibited blue-green, shriveled leaf tips indicating a very rapid wilt. At many midwestern locations affected with the C-15 problem, plants were observed to wilt in the manner of a few hours. Usually the plant remains in the wilted stage (blue-green) for a short time before the entire plant turns brown and begins to decompose. This brown, decomposed stage of the disease is most often observed by scientists and superintendents, and has undoubtedly been confused with many other diseases over the past

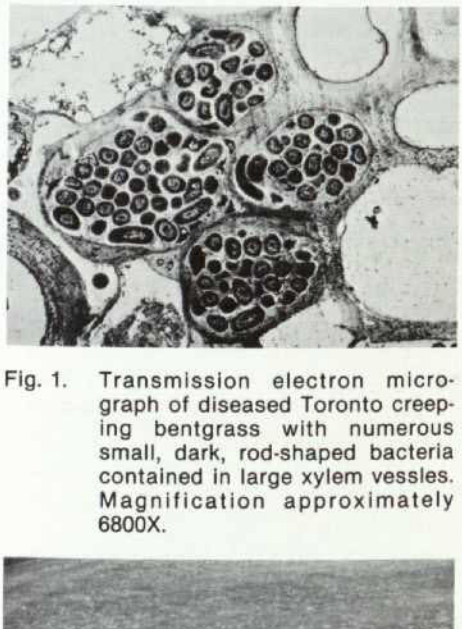
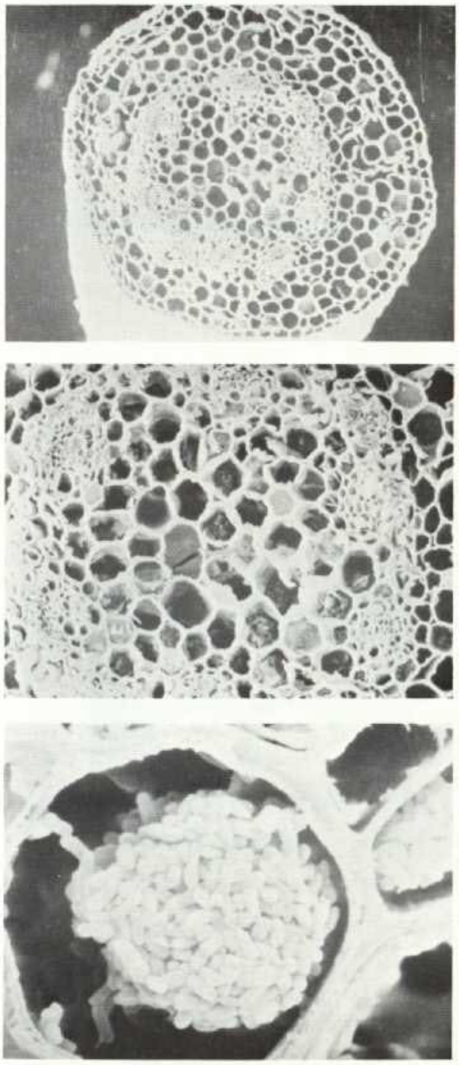


Fig. 1. Transmission electron micrograph of diseased Toronto creeping bentgrass with numerous small, dark, rod-shaped bacteria contained in large xylem vessles. Magnification approximately 6800X.

Fig. 2. Individual selection of Toronto creeping bentgrass plants by the disease resulted in characteristic uneven putting surface.

decade. Random selection of plants by the bacterium produced an uneven putting surface in advanced stages of the disease (Fig. 2). Other grasses were not apparently affected by the disease.

Careful observation of symptoms and examination of diseased plants with electron microscopy has shown the disease to be present at more than 25 locations in Indiana, Illinois, Michigan, Ohio and Wisconsin. Scanning electron microscopy (SEM) proved to be much more rapid in diagnosis of this disease (Figs. 3-5) than transmission electron microscopy (TEM). This widespread geographical distribution presents strong evidence that this bacterium is the cause of the C-15 problem-decline which has affected 'Toronto' for many years.



Figs. 3-5. Diagnosis of the C-15 problem with scanning electron microscopy. Scanning electron micrographs of root from diseased Toronto creeping bentgrass at three magnifications. Bacteria (rod-shaped objects) are readily visible in Fig. 5. Fig. 3, Magnification approximately 150X. Fig. 4, Magnification approximately 2000X. Fig. 5, Magnification approximately 6,600X.

Disease Control with Oxytetracycline

Disease control trials were initiated at several locations where bacteria were associated with 'Toronto' putting greens during the Fall of 1980 and Summer of 1981. The locations included: The St. Charles Country Club, St. Charles, Illinois — Superintendent Pete Leuzinger; Village Links, Glen Ellyn, Illinois — Superintendent Tim Kelley; Bay Pointe Country Club, Detroit, Michigan — Superintendent Don LaFond; and Alpine Country Club, Grand Rapids, Michigan — Superintendent Jeff Carsock.

Three chemicals were applied in 1000 and 1500 parts per million (ppm) solutions at 1 gal per 20 ft^2. The chemicals included oxytetracycline (Mycoshield, Pfizer Corp.), streptomycin sulfate (Agrimycin 17, Pfizer Corp.) and cupric sulfate (Kocide 101, Kocide Corp.). Excellent control was achieved with oxytetracycline at all locations (Fig. 6). Streptomycin sulfate and cupric hydroxide did not inhibit disease development. While oxytetracycline provided excellent control of the disease, the high rates (approximately 2.5 lbs/1000 ft^2) used in the experiments are expensive and require large amounts of water and time for application. Further research is in progress for lessening the application rates of oxytetracycline and water, and for possible application in sand top dressing. This research is extremely important since oxytetracycline may be the only chemical means of inhibiting this disease.

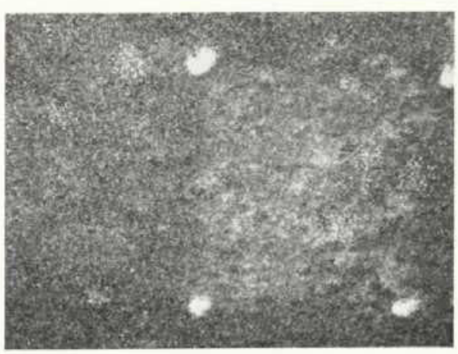


Fig. 6. Oxytetracycline (Mycoshield-Pfizer Corporation) at 2.5 lb formulation in 50 gallons water per 1000 ft^2 inhibited disease development (non-treated control plot within white markers). Streptomycin sulfate, cupric hydroxide and common fungicides were ineffective for control of this disease.

Isolation and Pathogenicity

A series of scientific procedures must be followed to determine with absolute certainty that a bacterium is the cause of the C-15 problem-decline. After careful grinding of diseased 'Toronto' plants and culturing on specialized growth media, a bacterium exhibiting yellow pigmentation was isolated in high numbers (Fig. 7) from plants at locations in Illinois, Indiana, Michigan, Ohio and Wisconsin. Hygrothermograph data from golf course locations showed that warm temperatures (about 80°F) and high humidity (above 95% for 24-48 hours) favor infection of 'Toronto' by the bacterium. Laboratory inoculation of 'Toronto' creeping bentgrass in conducive environments resulted in wilt symptoms similar to those observed in the field (Fig. 8). Examination with scanning electron microscopy proved that high numbers of the bacterium were limited to the xylem vessels of experimentally inoculated plants (Fig. 9). The consistent isolation and pathogenicity confirmed that the bacterium was the cause of the widespread C-15 problem-decline. Based upon wilt symptoms and the bacterial incitant, the disease was named "bacterial wilt of Toronto creeping bentgrass", the first known bacterial disease of turfgrass.

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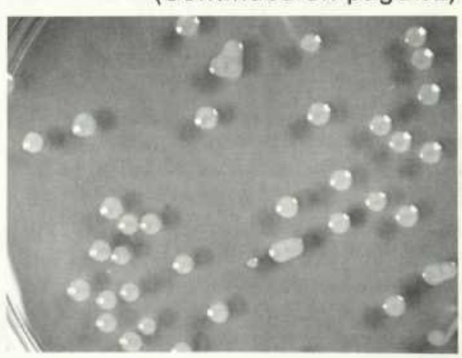

Fig. 7. Bacteria were isolated in high numbers in the laboratory form diseased Toronto creeping bentgrass. Bacterial colonies (yellow spots) measure approximately 1/8 inch in diameter and contain millions of microscopic bacteria.

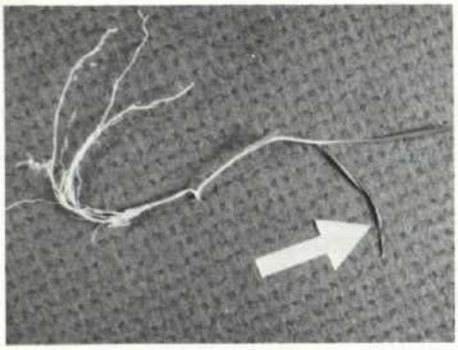

Fig. 8. Laboratory inoculation of Toronto creeping bentgrass with the bacterium resulted in wilt symptoms (arrow) similar to those observed ion the golf course, proving that the bacterium is the cause of the C-15 problem-decline. The disease was named "bacterial wilt of Toronto creeping bentgrass".

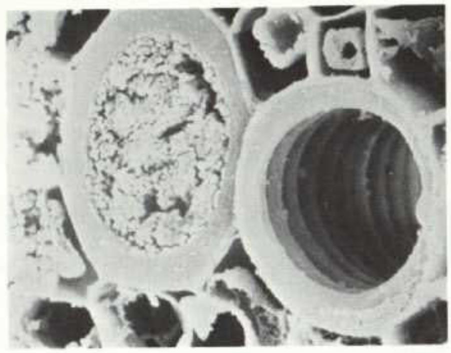

Fig. 9. Scanning electron microscopy proved xylem limitation (ribbed tunnels) of bacteria (small rod-shaped objects) in laboratory inoculated plants. Magnification approximately 2600X.

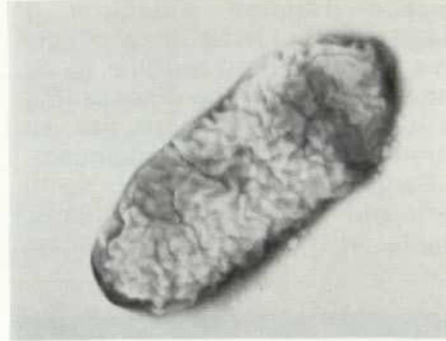

Fig. 10. Transmission electron micrograph of individual bacterium showing morphological characteristics of rod-shaped, rippled external surface and small size (1/25,000 inch in length). Magnification approximately 50,000X.

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Preliminary host range studies indicate that the 'Toronto' bacterium does not infect other turfgrasses, ie. 'Penncross' or 'Penneagle' creeping bentgrasses and Poa annua, which were turfgrasses commonly found on or overseeded into putting greens where bacterial wilt has destroyed the 'Toronto' creeping bentgrass.

Identification and Characterization

Bacteria are characterized according to morphological (size and shape), biochemical-physiological (nutritional) and serological (immunology) criteria. Morphological studies with electron microscopy has shown that the Toronto bacterium is rod-shaped, possesses a rippled external surface and measures approximately 0.5 by 1.0 microns (1 micron = 1/25000 inches) (Fig. 10). Based upon morphological and nutritional evidence, the bacterium must be classified as the bacterial species Xanthomonas campestris. Strains of this bacterial species attack many other plants particularly fruits, vegetables and field crops, but have never been reported to cause disease on turfgrasses in North America.

The New Threat — Bacterial Wilt

Serology is a highly specific immunology technique which can identify closely related bacteria. Until this time, serology has not been used to any extent in turfgrass science. Tests at Michigan State University showed that 'Toronto' bacteria from different states are very similar to each other but unlike other bacteria that commonly cause plant diseases in North America (Fig. 11).

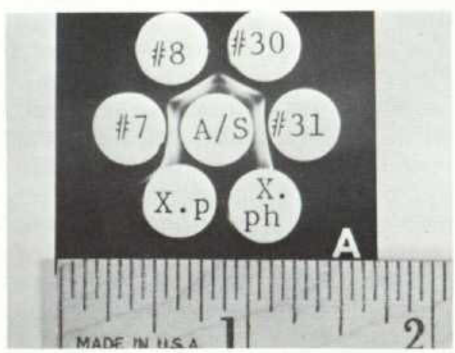

Fig. 11. Serology, a highly specific immunology technique, demonstrated that Toronto bacteria #7, #8, #30, #31 from different states were identical as evidenced by white precipitate line formation. The Toronto bacteria are apparently unrelated to other common North American bacteria represented by X.p. and X.ph. (without line formation).

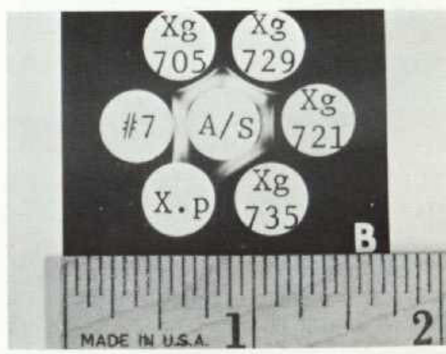

Fig. 12. Isolates Xg 705, Xg 729, Xg 721 and Xg 735 cause bacteria wilt of grasses in Europe and are related to the Toronto bacterium as evidenced by white line precipitation between them and Toronto bacterial isolate #7. This suggests that the origin of the Toronto bacterium was Europe where bacterial wilts of many grasses were known.

Further characterization of the 'Toronto' bacterium with serology has revealed startling information. The 'Toronto' bacterium is closely related to Xanthomonas bacteria that causes wilts of grasses throughout Europe (Fig. 12). The close serological relationship of the 'Toronto' bacterium and Xanthomonas bacteria from Europe suggest that the 'Toronto' bacterium was introduced into the United States from Europe. Other examples of introduced diseases include Dutch Elm Disease and the Chestnut Blight which have virtually destroyed the American elm and chestnut species throughout North America.

Bacterial wilt of grasses in Europe was originally discovered in Switzerland in 1975 (Table 1). Since that time the disease has been found in France, Germany, Great Britain, Norway, Netherlands, Belgium, and New Zealand. Various strains of the bacterium attack a wide variety of host grasses including Lolium (ryegrasses), Poa (bluegrasses), Festuca (fescues), and Agrostis (bentgrasses) (Table 1). Many of the grases attacked by the bacterium in Europe represent genera from which most of our American golf course varieties are derived.

The American turfgrass industry, turf managers, and scientists must be aware of the potentially devastating threat that these bacterial diseases represent. If other bacteria are introduced into North American or if the 'Toronto' bacterium changes in its host specificity, other widely cultivated varieties of turfgrasses such as the 'Penncross' and 'Penneagle' creeping bentgrasses, Kentucky bluegrasses, fescues, bermudagrass, ryegrasses, etc. may be attacked. Such important grasses are cultivated throughout large regions of North America. Bacterial wilts of grasses in Europe are regarded as so potentially dangerous that resistance to the bacterium is now incorporated in breeding programs. In the United States, renovation and replacement of 'Toronto' greens with other turfgrasses has only temporarily decreased the threat of bacterial wilt. Because very little effort is being exerted at the study of bacterial wilt control with oxytetracycline, bacterial spread, the infection process and plant host range, another major disease epidemic in addition to the C-15 problem-decline may occur without warning. Potential widespread epidemics may pose serious threats to the culture of specific turfgrasses in North America in years to come.

Acknowledgements

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WGCSA MAY MEETING
Janesville
Riverside Golf Club
May 21

BELFIELD, TUMBLEBROOK HOST SPRING BUSINESS MEETING

A pleasant aspect of the annual meeting of the WGCSA was our site — Tumblebrook Country Club. The March meeting has been held at a hotel/convention center frequently in past years, and returning to a golf club was appreciated by all of those in attendance. The comfortable surroundings at Tumblebrook created the atmosphere for a very enjoyable and productive meeting. Bob Belfield was our host.

Officer and committee reports informed members of the Board's activities during the winter months. Refinement of office procedures, improved relations with allied professional associations, a new direction for the GRASSROOTS and the 1984 meeting schedule and speakers were discussed. THE WGCSA 1984 Directory was introduced and distributed by Dale Marach, Bill Roberts and Randy Smith. Rod Johnson reported on his survey of members and indicated there is a lot of interest in scheduling a WGCSA dinner-dance.

New business acted on by the membership dealt with by-law changes defining membership categories and some needed changes in the nominating committee procedures.

Bob Welch was kind enough to bring the entertainment — a film of the 1983 Masters Tournament in Augusta. It was a great way to end the meeting and to set the stage for the 1984 golf season in Wisconsin.