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



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.

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.

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 ft2. 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 ft2) 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



Fig. 6. Oxytetracycline (Mycoshield-Pfizer Corporation) at 2.5 lb formulation in 50 gallons water per 1000 ft² inhibited disease development (non-treated control plot within white markers). Streptomycin sulfate, cupric hydroxide and common fungicides were ineffective for control of this disease. 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.

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.

(Continued on page 32)



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 rodshaped 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.

(Continued from page 31)

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 biochemicaland shape), biochemical-physiological (nutritional) and serological (immunology) criteria. Morphological studies with electron microscopy has shown that the Toronto bacterium is rodshaped, 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



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).

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 North America (Fig. 11).

examples of introduced diseases include Dutch Elm Disease and the Chestnut Blight which have virtually destroyed the American elm and



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.

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 turforass 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

The authors would like to express gratitude to Dr. Karen K. Baker and the Center for Electron Optics for advice and cooperation. Sincere appreciation is expressed to the Michigan Turfgrass Foundation, the primary supporter of this research.

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.



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WISCONSIN GOLF COURSE SUPERINTENDENTS ASSOCIATION **1984 MEETING SCHEDULE**

DATE	LOCATION	HOS
May 21	Riverside Golf Club Janesville	Bruc
June 18	Quit-Qui-Oc Golf Club Elkhart Lake	Paul
July 9	South Hills Club Fond du Lac	Pat
August 13	Stevens Point Country Club Stevens Point	Jeff
September 17	Milwaukee Country Club Milwaukee	Danr
October 18	Lawsonia Links Green Lake	Tom
November 5	Tumblebrook Country Club Pewaukee	Bob

Bruce Schweiger	SPEAKER Bruce Williams
Paul Miller	Dr. David Roberts
Pat Norton	Paul Boizelle
Jeff Bottensek	Paul Bando
Danny Quast	Dr. Bill Daniel
Tom Spaulding	Jerry O'Donnell
Bob Belfield	Business Meeting



WGCSA MAKES RESEARCH GRANT TO WISCONSIN TURFGRASS ASSOCIATION



The membership of the Wisconsin Golf Course Superintendents Association unanimously passed a motion at the April meeting to donate \$1500 to the Wisconsin Turfgrass Association. The money will be used for turfgrass research at the University of Wisconsin-Madison.

WTA President Tom Harrison expressed gratitude for the gift and assured WGCSA members that they played a very important part in the success of the Wisconsin Turfgrass Association.

The \$1500 grant will be funded from ad revenue of the GRASS-ROOTS.



	Elkhart Lake
)	South Hills Club
	Fond du Lac
st 13	Stevens Point Country CI
	Stevens Point
mber 17	Milwaukee Country Club
	Milwaukee
per 18	Lawsonia Links
	Green Lake
mber 5	Tumblebrook Country Clu
	Pewaukee

